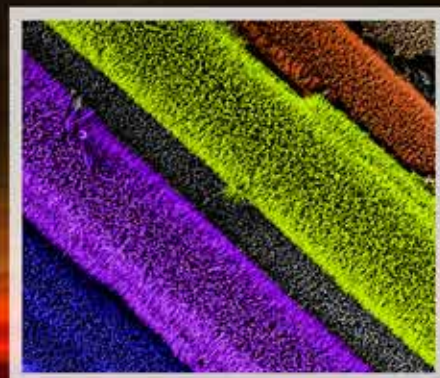


2016 Annual Report

Laboratory Directed Research & Development
at Pacific Northwest National Laboratory

DISCOVERY

in Action




Pacific Northwest
NATIONAL LABORATORY

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Laboratory Directed Research and Development Annual Report

Fiscal Year 2016

March 2017

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Pacific Northwest National Laboratory
Richland, Washington 99352

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Laboratory Director's Message

For Pacific Northwest National Laboratory advances the frontiers of science and delivers technological innovation to address some of the world's most challenging problems in energy, earth sciences and national security.

Key to advancing the Department of Energy's missions today, and well into the future, is strong stewardship of our scientific and engineering capabilities. As part of our annual strategic planning process, we determine where to invest our discretionary R&D funding with an eye toward specific outcomes—transformational science and technology, accelerated innovation, new partnerships and enhanced core capabilities. Each year, we select a portfolio of Laboratory Directed Research and Development projects that explore new concepts in alignment with these priorities.

This report summarizes our FY 2016 LDRD program, highlighting more than 200 projects that span fundamental discoveries to applied technologies. We are proud to showcase the creativity of our scientists and engineers and the resulting accomplishments that reflect the breadth and depth of PNNL's research. The report also describes how the program is managed and aligned with DOE's strategic objectives.

Thank you for taking the time to learn about the transformational science and technology underway at Pacific Northwest National Laboratory.



Steven F. Ashby
Dr. Steven Ashby
Director, PNNL

Advanced Sensors and Instrumentation



Advanced Sensor System to Preserve U.S. Food Security: Determining Early Onset of Bovine Respiratory Disease

Ethan D. Farquhar

This project aims to build a sensor network capable of measuring the physiological traits of cattle to predict early onset of bovine respiratory disease (BRD). This system will revolutionize the way that ranchers manage their herds and assist in early detection of BRD, both to increase the security of the nation's food supply and to prevent billions of dollars in annual economic losses to the U.S. cattle industry. Our project diverges from previous attempts at bovine instrumentation by integrating an array of the newest, most innovative sensors and miniaturized power systems that have been recently developed at PNNL, and it will make use of a spatial data infrastructure analytical framework to apply big-data approaches to determining disease signatures.

Early detection of disease is essential to mitigate the devastating effects of an epizootic or zoonotic event on humans or the U.S. food supply. Whether a result of agro-terrorism or natural viral evolution, such events have disastrous impacts to national security in terms of loss of life, immediate depletion of national food stores, and massive devastation to economic resources. Of all diseases affecting the U.S. cattle industry, BRD imposes the largest losses, which total about \$5 billion annually. Further motivation for such monitoring systems comes from the long-standing, widespread use of antibiotics on animals that result in significant pools of resistance genes among bacteria, including human pathogens. Such rampant use of antibiotics markedly increases the likelihood of a pandemic event.

To date, a first version of the candidate wearable system has been developed. Many different sensors, as well as locations to place those sensors, have been investigated.

In discussions with subject matter experts, we became aware of several potential targets that would be suitable for monitoring in a passive, non-invasive way. track the general activity levels of the animal, as well as the relative angle between the head and the neck (using two inertial measurement units).

Pulse oximetry was thought to be a leading candidate for a respiratory disease, as blood oxygen saturation would be reduced for animals struggling to breathe. However, discussions with veterinarians have led us to believe that both melanin and the thick hides of cows combine to make pulse oximetry using standard sensor systems a difficult task. While we do not plan to abandon this modality, it has been reduced in its priority because of the difficulty in obtaining this data. However, further discussions with collaborating veterinarians have revealed a desired ability to simply acquire this data and, thus, may indicate a future study area.

Audio is a promising modality for the capture of many activities that are of interest to researchers of BRD. Respirations, ruminations, coughing, sneezing, and other such noises are all events of significance that are easily captured by applying microphones in strategic locations on the animal.

Of course, body temperature is a vital element, and this can be captured through use of integrated temperature systems.

In FY 2016, 15 systems that combine the above sensor systems and an integrated battery, as well as full internet connectivity (allowing for researchers to monitor systems from remote locations), have been developed and are ready for deployment in future animal-based studies. Further efforts to increase the robustness of the system are ongoing.

While the system has been planned and specifically incorporates the above sensors, what has really been designed is a method for deploying a wide variety of sensors in a connected platform, suitable for targeting many other application spaces.



Basic sensor platform (not including sensors).



Sensor platform encased in 3D-printed enclosure with attached sensors.

Combined Microscale ^{13}C and ^{18}O Measurements at Cutting-Edge Sensitivities and Spatial Resolution

Jim Moran

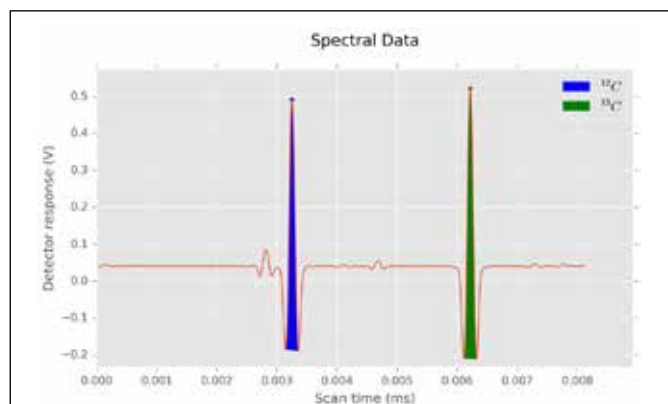
We are employing a capillary absorption spectroscopy platform to enable cutting-edge stable isotope measurement sensitivity. The resulting sensitivity improvements will enable a range of new sample introduction techniques, thereby advancing research in areas including such diverse fields as microbial nutrient cycling, forensics investigations, and ecological studies, among others.

This project seeks to harness emerging advances in laser technologies to enable truly cutting-edge measurement sensitivities for performing stable isotope analysis of carbon (^{13}C). The capillary absorption spectroscopy (CAS) system we are developing will provide orders of magnitude measurement sensitivity improvement over traditional analytical platforms, predominantly isotope ratio mass spectrometry (IRMS). Recent work with laser ablation sampling coupled to IRMS demonstrated spatial resolution of approximately $50\mu\text{m}$ when measuring ^{13}C over a solid surface. Ultimately, however, spatial resolution is controlled by the amount of sample required for analysis, whereby higher measurement sensitivity would enable improved spatial resolution. While IRMS is traditionally the workhorse of light stable isotope analysis, it requires tens of nmoles of carbon for analysis. The emerging CAS system, in comparison, has significantly higher sensitivity and requires less than a picomole per analysis, translating to over four orders of magnitude less or approximately one ten-thousandth the amount of sample required by IRMS. The vast sensitivity improvements inherent in the emerging CAS system can translate to unprecedented spatial resolution of samples or analysis of extremely small bulk samples. By combining laser ablation with a CAS-based measurement approach, this project seeks to enable spatially specific isotope measurement (in both natural abundance and labeled studies) at scales not previously feasible and to thereby enable a suite of potential stable isotope applications. Such a capability can provide new opportunities for exploring nutrient cycling in microbial communities, investigating very small samples for forensic analysis, or studying small growth rings in animal tissues (e.g., fish otoliths or hairs).

In order to be successful, the instrument we are constructing first requires a CAS unit able to interrogate a sample with infrared light to measure respective ^{12}C and ^{13}C content and then interpret the resulting absorbance measurements to

provide a final isotope content. We are developing the needed CAS hardware and software in parallel efforts. While many aspects of this system can be commercially supplied, the CAS measurement platform and the software to extract isotopic information from resulting absorption spectra is being pioneered at PNNL.

A central concern in effective implementation of the CAS hardware is identification of suitable absorption targets and then the removal of optical interference that may obscure accurate peak measurements. We identified a portion of the CO_2 absorption spectrum containing nearly adjacent, similar intensity optical transitions correlating to both ^{12}C and ^{13}C that can be interrogated using a tunable quantum cascade laser. This region is largely devoid of interference from other molecules (e.g., H_2O , N_2 , typical volatile organic compounds, etc.), with the few additional peaks being linked to neighboring CO_2 transitions. Still, optical feedback in the system was initially a problem, and we used a series of techniques, including different focusing approaches and the use of a coiled, tapered capillary fiber to minimize this feedback and thereby increase baseline stability.



This project is targeting stable isotope analysis of CO_2 using a capillary absorption spectroscopy technique, whereby selective absorbance bands of CO_2 containing ^{12}C and ^{13}C at discrete wavenumbers enables isotopic quantification. A software system developed for this project facilitates both rapid data collection and analysis to report a sample's measured isotopic abundance, with the above example showing the software-identified peaks relating to ^{12}C and ^{13}C transitions within the CO_2 analyte. The data shown above was collected using scanning of a tunable quantum cascade laser over a range of wavenumbers (chosen to include the targeted spectroscopic transitions) and then compiling the resulting 6,000 scans (collected at 122 Hz) into a representative spectrum of the sample for isotope determination.

An additional method employed to reduce optical feedback, as well as other sources of interference, is the software development undertaken by this team. The software contains a number of data processing steps that first help deconvolute a series of raw data points to reduce aberrant background/noise from the signal and secondly performs data integration to collect, process, and collate thousands of laser scans—generated on the order of seconds—and report a measured isotope ratio for the sample. The graphical user interface is designed for rapid data analysis by the user.

IRMS has traditionally served as the primary instrumentation for stable isotope ratio measurements. Sensitivity limits and mass interference issues confine the application of IRMS to scientific questions that would otherwise benefit from stable isotope analysis. The CAS system being developed here offers vastly improved measurement sensitivity, while also circumventing mass interference issues associated with IRMS and should enable stable isotope application to a wide variety of new scientific investigations.

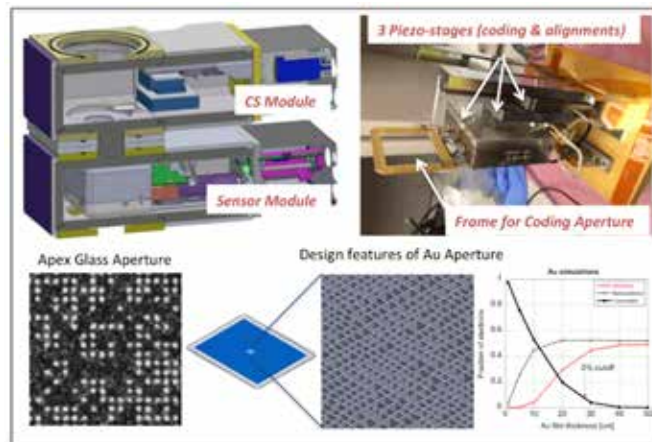
Design and Development of Coded Aperture for Video Compressive Sensing Acquisition System for Environment Transmission Electron Microscope

Libor Kovarik

The objective of this project is to develop a temporal compressive sensing (CS) acquisition system in order to improve temporal resolution, or correspondingly reduce the electron dose, by an order of magnitude in comparison with conventional sensing.

Environmental Transmission Electron Microscopy (ETEM) represents a powerful capability for resolving the structure of materials with atomic-level resolution during exposure to gaseous environments. While modern, aberration-corrected environmental TEM instruments provide atomic-level resolution, the speed and sensitivity of the acquisition system represents the main limiting factor for studying the transformation dynamics. In an effort to improve temporal resolution, it was recently proposed that the concept of CS has the potential to improve both the temporal resolution and sensitivity of the recording system. Compressive sensing is a fundamentally new concept for enhancing temporal resolution. It is based on the idea that images/data can be well represented in a much sparser (compressed) form using a suitable basis set and that this suitable basis set can be fully recovered from significantly fewer data samples than in conventional, full-pixel acquisition. For video CS, the sparsification along the temporal dimension is accomplished by modulation, with a coded aperture (mask) placed in front of the detector, and subsequent computer reconstruction.

To pursue the goal of improving temporal resolution for TEM imaging, the current project focused on developing a temporal CS acquisition system. In collaboration with Direct Electron LP, we developed and built a CS video acquisition camera prototype. The CS camera was built to be modular, with one module housing a coded aperture controlled by piezo-stages, and second module housing the electron pixel sensor. We spent a significant portion of this project on the design and development of the coded aperture, which is the most critical component of the CS acquisition system. For TEM electron optics, the coded aperture must be designed as a fully open, self-supported aperture, with 50% transitivity at random pixel locations. The challenge is in designing and manufacturing the code aperture at micrometer size and with well-defined features.



Prototype of CS acquisition camera for ETE, which consists of upper module for coded aperture and piezo stage control, and lower module for the sensor. There is a detailed view depicting the manufactured apex glass aperture, as well as the design of Au aperture where webbing in the aperture is an active blocking component of the aperture.

We have followed several manufacturing processes for fabrication of the coded aperture, including glass etching, Au electroplating, and mechanical drilling of brass. In collaboration with 3D Glass Solutions, Inc., we manufactured the aperture based on etching of Apex glass. A custom-made aperture, 200 μm in thickness and incorporating opening features of 33.6 μm , was manufactured. The testing of the aperture revealed that, with the coded apertures, the acquisition is fully capable of modulating the video signal; it was successfully confirmed that the acquisition with the coding apertures does not lead to any undesired image artifacts associated with translating stage in front of the detector. However, due to the limitations associated with the design and manufacturing process, the code aperture had low transmissivity, which led to a video reconstruction that did not meet the design performance.

Prototype of CS acquisition camera for ETE, which consists of upper module for coded aperture and piezo stage control, and lower module for the sensor. There is a detailed view depicting the manufactured apex glass aperture, as well as the design of Au aperture where webbing in the aperture is an active blocking component of the aperture.

To address the limitations of an Apex-glass-coded aperture, we have redesigned the effective transmissivity by including the webbing in the aperture as an active blocking component and also changed the fabrication process, which enables us to use reduced thickness. In collaboration with the University of Louisville Micro/Nano Technology Center, we started a fabrication of coded aperture based on electroplating of 30 μm Au. The aperture features were defined in photoresist and Si mold. For electroplating of the aperture in 30 μm photoresist, we have encountered several obstacles from cracking and aperture-feature rounding. For electroplating in Si mold, the obtained aperture features were shown to maintain much

better definition, but the final step involving planarization of the aperture led to mechanical failure. The aperture is currently being fabricated using a modified protocol, which would avoid the planarization step altogether and, thus, avoid the mechanical failure during preparation.

In summary, we have built a coded aperture acquisition system for CS acquisition in the optical column of the environmental electron microscope. We also developed design principles and a manufacturing process for the coded aperture, given the constraints of TEM applications.

Development of an Ultra-Small Volume Detection and Sample Delivery System for Exploring Microscale Heterogeneity with NMR

Karl T. Mueller

We have developed methods and instrumentation that will integrate high-resolution nuclear magnetic resonance (NMR) spectroscopy and magnetic resonance imaging (MRI) microscopy with lab-on-a-chip technology for studying sub-nanoliter samples.

Microscale heterogeneity plays a key role in determining the outcome of attempts to decrease pollution, optimize industrial production, or understand cellular-level processes. Because of the unique capability of NMR to characterize biological and chemical systems in detail without significantly perturbing them, we seek to extend the reach of NMR spectroscopy to routine analysis of microscopic samples. To this end, we have developed methods and instrumentation that enable high-resolution NMR spectroscopy and MRI microscopy as a tool for studying microscale heterogeneity. The current generation of commercial, small-volume NMR detectors requires sample volumes of 5-10 μL , and we are now showing that high-resolution NMR can be used for practical studies of volumes that are several orders of magnitude smaller, allowing for novel studies that address individual microsystems such as cells and aerosols.

We have designed, optimized, and characterized a microstrip-based NMR probe for application on (sub-)nanoliter samples.

Two different approaches have been undertaken. Our first approach preserves spectroscopic resolution with small-volume detectors and involves two steps. The first is development of a detector with near-optimal sensitivity for sub-nanoliter samples with high filling factor, and we use pulse sequences to recover resolution. The advantages of this design include 1) robust and simple scalability (especially compared to microcoil detectors), 2) better magnetic field homogeneity than microslot detectors, and 3) a single-sided design, simplifying sample loading and allowing for integrated applications with lab-on-a-chip devices.

Our second approach is to obtain high-resolution NMR with high sensitivity by optimization of the microstrip detector combined with a novel sample loading strategy. The dimensional parameters of the detector were optimized using numerical simulations with regard to radio frequency sensitivity and homogeneity, with additional considerations of design. The influence of copper surface roughness and grain structures on the spectral resolution was investigated. The microstrip board was easily batch-fabricated using conventional printed circuit board techniques at low cost. With an appropriate sample preparation technique, high sensitivity, high radio frequency homogeneity, and high resolution (0.8–1.5 Hz, depending on sample positioning) were achieved for 1D and 2D NMR spectroscopy on 1 nL of a water and sucrose mixture.

Development of New High-Temperature Piezoelectric Materials for In-Core Diagnostics and Prognostics

Tiffany C. Kaspar

Safe operation of next-generation structural materials requires real-time, local material prognostics and diagnostics probes. We will develop new, high-temperature, and radiation-resistant piezoelectric sensor materials to acquire local diagnostic data in extreme environments such as nuclear reactor cores.

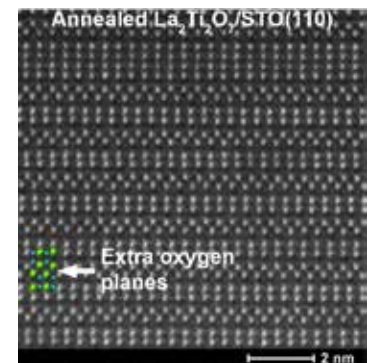
The acquisition of local diagnostic data in extreme environments for *in-core* material prognostics and diagnostics requires the development of new piezoelectric sensor materials that operate at high temperatures with adequate radiation damage resistance. In particular, *in-core* ultrasonic techniques, such as classical ultrasound and nonlinear ultrasound, have the advantage of being able to detect dislocations, defect clusters, and precipitates that form during material use, which makes them particularly strong candidates for improved prognostics. $\text{Pb}(\text{Zr,Ti})\text{O}_3$ (PZT) is the piezoelectric material of choice in low-temperature and moderate-radiation-dose environments. However, the effective operating temperature of PZT is limited to about 200°C. No piezoelectric material has yet emerged that exhibits both a high Curie temperature and a strong piezoelectric response. We propose to develop such a material by tailoring the properties of an existing high-temperature piezoelectric, layered-structure perovskite, $\text{La}_2\text{Ti}_2\text{O}_7$, through doping and epitaxial strain.

Thin films of $\text{La}_2\text{Ti}_2\text{O}_7$ were deposited via pulsed laser deposition (PLD) on lattice-matched substrates. Control of crystalline orientation through epitaxial stabilization was demonstrated for $\text{La}_2\text{Ti}_2\text{O}_7$ films deposited on various single crystal substrates. The deposition on rutile $\text{TiO}_2(110)$ is one not previously reported in the literature. For all films, crystalline orientation and phase were confirmed by both x-ray diffraction (XRD) and scanning transmission electron microscopy (STEM); in all cases, the deposition and post-growth annealing conditions were optimized such that the majority phase was the desired orientation.

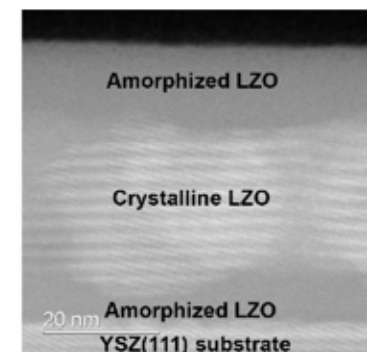
During PLD, films of $\text{La}_2(\text{Ti}_{0.9}\text{Zr}_{0.1})\text{O}_7$ were also synthesized, and XRD and STEM testing indicated that Zr substituted for Ti in the lattice, as intended, and did not segregate to form Zr-rich secondary phases.

A collaboration was initiated with Dr. Seungbum Hong at Argonne National Laboratory to perform piezoelectric characterization of the $\text{La}_2\text{Ti}_2\text{O}_7$ and $\text{La}_2(\text{Ti}_{0.9}\text{Zr}_{0.1})\text{O}_7$ films. Dr. Hong is an expert in piezoresponse force microscopy (PFM), a piezoelectric measurement technique using an atomic force microscope. Preliminary characterization by PFM at PNNL indicates that $\text{La}_2\text{Ti}_2\text{O}_7/\text{SrTiO}_3(001)$ is piezoelectric. Dr. Hong will characterize the in-plane and out-of-plane components of the piezoelectric response of $\text{La}_2\text{Ti}_2\text{O}_7$ thin films as a function of crystalline orientation. PFM will also be used to assess the effect of Zr doping on the strength of piezoelectric coupling.

The radiation resistance of pyrochlore $\text{La}_2\text{Zr}_2\text{O}_7$ was investigated by sending a thin film sample to the Radiation Effects Facility at Texas A&M University, where it was irradiated to simulate the radiation environment *in-core*. Pyrochlores are known to exhibit high radiation tolerance, and are of interest as a potential waste form for radionuclides. In contrast to previous published reports, the thin film of $\text{La}_2\text{Zr}_2\text{O}_7$ remained partially crystalline to a dose of 10 displacements per atom (dpa), as measured by Rutherford backscattering spectrometry in the random and channeling geometries collected as a function of dose. Amorphization as a function of dpa does not follow the direct amorphization model proposed for these materials. Post-irradiation analysis revealed that the irradiation-induced amorphization occurred from the film surface down and from the interface up, leaving a crystalline core in the center



STEM image of $\text{La}_2\text{Ti}_2\text{O}_7$ deposited on $\text{STO}(110)$ by PLD and post-annealed at 1100°C for 4 hours in air. The extra oxygen planes in the PLS structures are indicated; the piezoelectric coupling occurs along this direction.



STEM image of $\text{La}_2\text{Ti}_2\text{O}_7$ (LZO) film on $\text{YSZ}(111)$ substrate after 1 MeV Zr^+ irradiation to a dose of 10 dpa.

portion of the film. In addition to providing insight into the amorphization mechanism of pyrochlores, this thin film sample serves as a baseline for a subsequent irradiation study of undoped and Zr-doped $\text{La}_2\text{Ti}_2\text{O}_7$.

In summary, the deposition of piezoelectric $\text{La}_2\text{Ti}_2\text{O}_7$ thin films was optimized with desired crystalline orientations on various single crystal substrates. Collaborative, PFM measure-

ments will investigate the piezoelectric response of undoped and Zr-doped $\text{La}_2\text{Ti}_2\text{O}_7$ films. The radiation resistance of pyrochlore $\text{La}_2\text{Zr}_2\text{O}_7$ was investigated, providing insight into the radiation response of this material and also serving as a baseline for subsequent irradiation studies on undoped and Zr-doped $\text{La}_2\text{Ti}_2\text{O}_7$ films.

EUV Laser Ionization Mass Spectroscopy

Andrew M. Duffin

The objective of this project is to research the application of state-of-the-art, extreme ultraviolet (EUV) lasers for rapid elemental analysis, with particular focus on actinide materials.

Efficient elemental and isotopic analysis with nanoscale spatial resolution has the potential to open new forensics capabilities not realized with current techniques. This project is pursuing EUV laser ionization for this purpose, as well as to dramatically improve the state-of-the-art in laser ablation coupled mass spectroscopy.

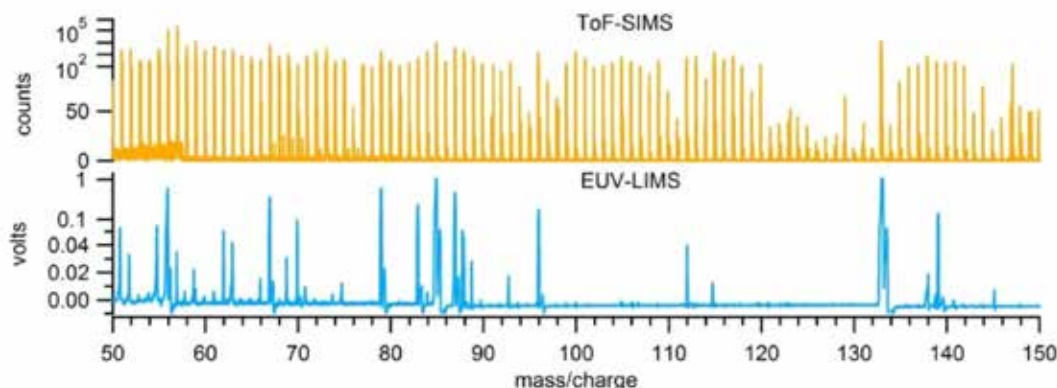
The recent development of reliable, tabletop EUV lasers (mainly from the Engineering Research Center for Extreme Ultraviolet Science and Technology at Colorado State University [CSU]) has enabled performing laser ablation mass spectroscopy in a fundamentally different mode. While all laser ablation relies on multiple photons imparting sufficient energy to a solid surface to remove material, EUV laser light (46.9 nm or 26.4 eV) has at least four times more energy per photon compared to traditional laser ablation light (190-1080 nm or 6.5-1.1 eV). That is, EUV light is energetic enough to break even the strongest chemical bonds with a single photon. In addition, EUV laser light can be focused down to below 100 nm spots, a roughly one to two orders of magnitude improvement over most commercial laser ablation systems.

This project received initial seed funding during the final month of FY 2014, which was used to meet with our collaborators at CSU to align the expectations of the two research groups, develop a research plan, coordinate schedules for

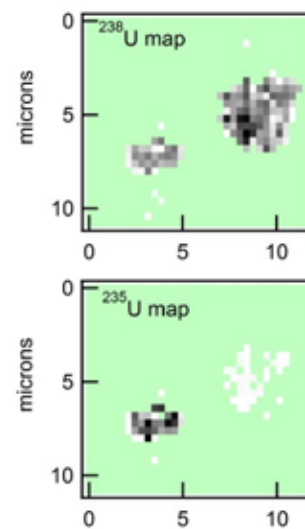
future experiments, and familiarize ourselves with EUV laser operation.

During FY 2015, we carried out many of the experiments planned during the tail end of FY 2014. Specifically, we investigated the ability of EUV laser ionization mass spectroscopy (EUV-LIMS) to perform elemental analysis on a commonly used laser ablation standard, NIST 610. The NIST 610 glass is doped with 61 elements each at approximately 500 µg/g. Consequently, it presents a complex mass spectrum with many possible molecular interferences. The main finding from NIST 610 analysis was that EUV-LIMS produces a relatively interference-free mass spectrum wherein many elemental peaks are observed. Qualitatively, the detection limits for EUV-LIMS are tens to hundreds of parts-per-million, depending on the element. In addition, the ratio of elemental ions to molecular interferences can be increased with laser intensity.

During FY 2016, we conducted experiments to determine sample utilization efficiency, measure spatial resolution, and map uranium particles for isotopic abundance. We were able to measure approximately 0.01% of the atoms removed from the sample, a respectable number for mass spectroscopy and one that is likely limited by the TOF system and not the EUV ionization source. We determined the spatial resolution to be about 70 nm, which is on par with the best nano-sims capabilities. Finally, we demonstrated the ability to isotopically map uranium particulate samples with high spatial resolution. We conclude that EUV laser ionization is a promising new technique for high spatial resolution elemental and isotopic analysis.



Mass spectrum of NIST 610 collected with ToF-SIMS (top plot) and EUV-LIMS (bottom plot).



Isotope maps of low enriched and natural uranium particles.

Integration of an Atmospheric Flow Tube Ionization Source with a Novel Ion Mobility Analyzer

Robert G. Ewing

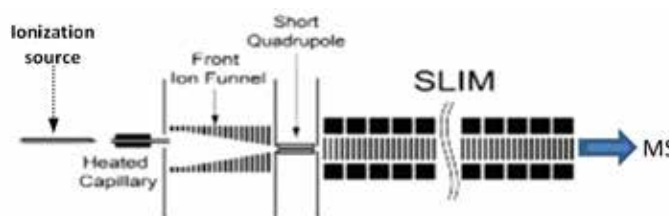
Field detection of illicit substances important to national security issues requires smaller instruments that are robust, have minimal power requirements, and are, preferably, hand-held. This project will investigate the development of a new generation of field-deployable detection technologies with improved sensitivity and selectivity over current capabilities.

An Atmospheric Flow Tube (AFT) has demonstrated sub-parts-per-trillion detection levels as an ionization source for a mass spectrometer (MS). Structures for lossless ion manipulations (SLIM)-ion mobility separations (IMS) devices provide much improved resolution over standard IMS, yet can operate at pressures above those required by mass spectrometry, allowing for reduced pumping requirements. This project will integrate an AFT ionization source with a SLIM-IMS device, both recent inventions at PNNL. The AFT-MS provides the first ever detection of illicit substances at parts-per-quadrillion levels in real-time. Many observers of this technology have asked if it could be made smaller, specifically if it could be integrated into an IMS. The SLIM-IMS device shows the potential to both accumulate/concentrate ion signal and provide improved resolution over standard IMS units. This effort leverages and builds upon existing PNNL technology to achieve this goal. A field portable device capable of detecting illicit substances is relevant to both counter terrorism and non-proliferation needs, resulting in significant impacts to national security applications.

In FY 2016, a SLIM-IMS module was assembled and interfaced to an Agilent quadrupole time-of-flight mass spectrometer as the ion detector for initial testing. The SLIM-IMS device consists of two mirror-image sections where the combination of fields applied to the two sections allow ions of specific mobility to be transmitted through the device. The overall device is 30 cm long and maintained at a pressure of 4 Torr, which is suitable to the level of confining radio frequency field.

Ions were generated with a dielectric barrier discharge source (the same source used in the AFT), transmitted to SLIM through a large inlet capillary (1 mm inner diameter) and an ion funnel for maximum ion utilization and improved sensitivity. Voltages and other parameters were adjusted

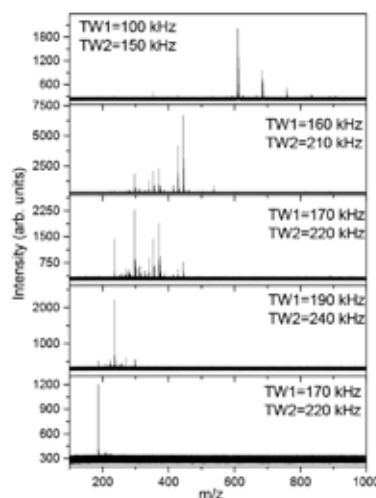
to optimize ion flow through the system, as monitored by the mass spectrometer. The ion densities were also compared to those generated with a standard electrospray ionization source. Towards the end of FY 2016, successful ion transmission was achieved with ion intensities from the dielectric barrier discharge source similar to those observed from the electrospray ionization source.



Schematic diagram of the full instrumental arrangement. Ions are generated by either electrospray ionization or distributed plasma ion source (DPIS).

Using the optimized ion transmission, parameters of the SLIM-IMS device were adjusted to demonstrate mobility-based selective ion transmission. Selective transmission of high to low mobility ions was demonstrated with masses ranging from m/z 110 to 2,500, as measured by the mass spectrometer.

Efforts planned for FY 2017 include the following: 1) integration of the AFT with the SLIM-IMS device, 2) collection and accumulation of low density ion currents, 3) replacement of the mass spectrometer with a Faraday plate detector, and 4) demonstration of the ability to detect and identify various threat substances from a room air sample. These steps provide a logical progression toward the successful integration and demonstration of the detection capabilities of the AFT-SLIM-IMS technology.



Mass spectra obtained from ionization of 10 μ M tributylamine by DPIS at different TW1/TW2 frequencies. A different set of ions is observed as the frequency window moves.

Low-Background Liquid Scintillation Counter

John L. Orrell

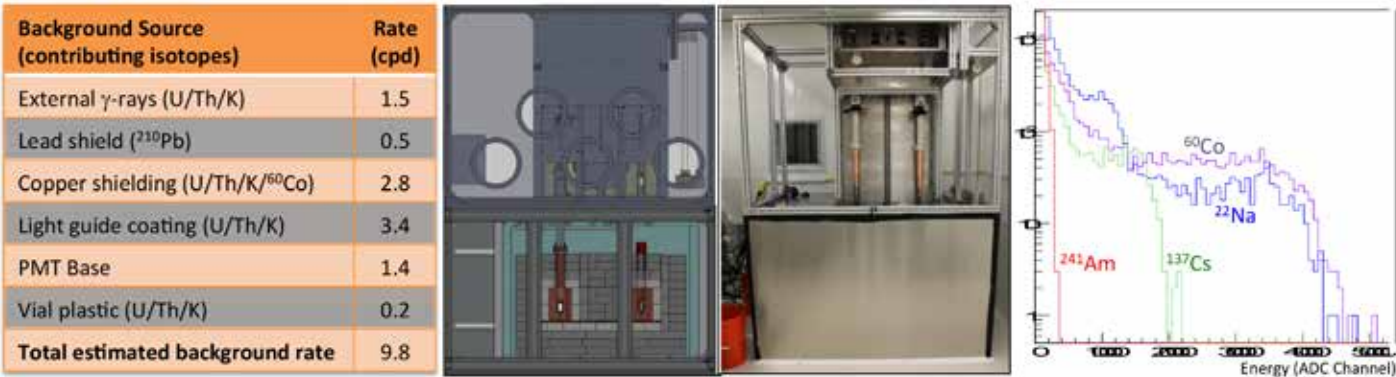
This project has developed a unique, low-background liquid scintillation counter by taking advantage of the PNNL shallow underground laboratory's cosmic ray shielding. The system is sensitive to β and α emitting nuclides at concentration levels approximately 10 times lower than commercially available instruments.

Pacific Northwest National Laboratory's shallow underground laboratory is a resource for measurement of low-concentration levels of radioactive isotopes in samples collected from the environment. The development of a low-background liquid scintillation counter is now complete and further augments the measurement capabilities within this underground laboratory. Liquid scintillation counting is especially useful for measuring charged particle (e.g., β , α) emitting isotopes with no (or very weak) γ -ray yields. The combination of high-efficiency detection of charged particle emission in a liquid scintillation cocktail coupled with the low-background environment of an appropriately designed shield located in a clean, underground laboratory provides the opportunity for increased-sensitivity measurements of a range of isotopes, including tritium (^3H), strontium ($^{89,90}\text{Sr}$), or α -emitters in the naturally occurring uranium/thorium (U/Th) decay chains.

The third year of this research and development project has involved the following two distinct activities: 1) system construction/installation, and 2) initial measurement of a suite of radionuclides demonstrating the performance of the system.

The low-background design follows a standard principle of nested active and passive shielding. From outermost to innermost, the shield is composed of a radon exclusion box, plastic scintillator panels for cosmic ray rejection, neutron absorbing 30% borated polyethylene, 8-inches of progressively lower ^{210}Pb concentration lead bricks to shield against external γ -rays, and a final inner, 2-inch-thick copper liner that also serves as the scintillation light collection guide. The novel light collection guide is designed as a hollow, highly reflective channel/guide milled within the inner copper liner. The reflective channels guide scintillation light from decays measured in the scintillation cocktail vial to photomultiplier tubes (PMTs) that are "around a corner" within the shielding so as to guard against trace radioactivity in the PMTs contributing to the background of the instrument. The included figure shows radiation transport simulation results in the form of a table of anticipated background contribution rates, based on an improved, high-fidelity simulation model developed in FY 2016. The figure also presents an engineering cut-away view of the final design, as well as the assembled system in the shallow underground laboratory. On the far right of the figure, initial calibration results are shown for four externally located radioisotopes, demonstrating the operation of the detection system.

The system developed will now transition to an initial operational phase where samples of opportunity for ongoing radiochemical programs are measured to determine the utility of the system to specific isotopes in low-concentration scenarios. Demonstration measurements of the radioisotopes of ^3H , $^{90}\text{Sr}/^{90}\text{Y}$, and ^{241}Am will be used to explore the detailed perfor-



A high-fidelity Monte Carlo simulation of the radiation transport of a variety of different background sources was employed to evaluate the liquid scintillation counter system design. The table shows the contributions for each of the backgrounds studied. The final design and photograph of the assembled system are shown in the center. The far-right energy spectra demonstrate operation of the system during an energy calibration test employing four external radiation sources.

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mance of the system, with respect to low-energy detection, spectral deconvolution, and alpha-particle discrimination. The background of the system will be characterized and evaluated in comparison to the table of estimated backgrounds

shown in the figure. This project has successfully added to the repertoire of low-level radiation measurement systems located in the PNNL shallow underground laboratory for use in environmental sampling.

Platform for Large-Scale Determination of Protein-Ligand Binding

Ryan T. Kelly

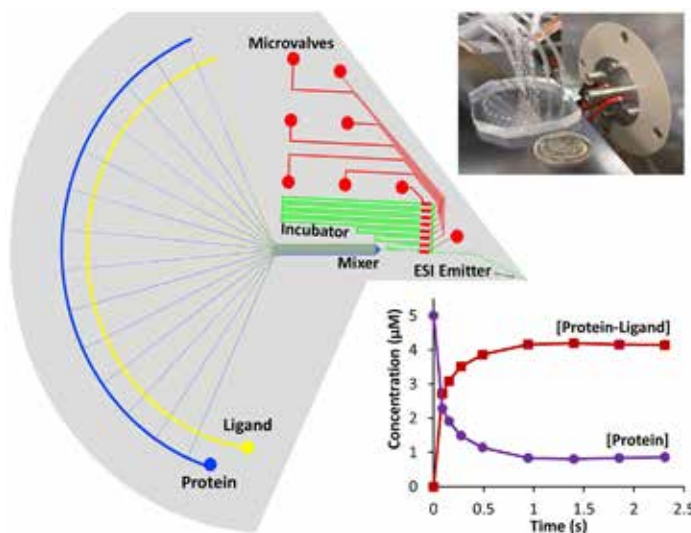
The objective of this proposal is to develop a microfluidic platform and combine it with ion mobility spectrometry-mass spectrometry (IMS-MS) for the rapid determination of binding affinities and kinetic parameters associated with noncovalent protein-ligand interactions. The platform is among the first to provide both label-free and solution-based association (k_{on}) and dissociation (k_{off}) rates and, thus, avoids the label, surface, and mass-transport-related artifacts of existing measurements.

Noncovalent protein-ligand interactions are fundamental to many biological processes, including signal transduction, enzymatic catalysis, and immune response. Determination of protein-ligand binding affinities and kinetics is critically important for drug discovery and understanding protein function. A number of analytical methods have been developed for protein-ligand studies, including equilibrium dialysis, liquid chromatography, capillary electrophoresis, surface plasmon resonance (SPR), spectroscopy, and calorimetry. Many of these methods, however, require labelling or immobilization of one of the binding partners to a solid substrate, which can substantially interfere with the binding and can introduce additional complications such as costly assay development.

Mass spectrometry is increasingly being used for protein-ligand studies, as it provides precise mass information and detailed quantitative information of unbound proteins and protein-ligand complexes without the need to incorporate a label or immobilize one of the binding partners. While the use of MS for measuring equilibrium binding affinities continues to mature, application of MS for determining the dynamics of such interactions has scarcely been undertaken. We have developed a microfluidic platform that incorporates protein and ligand introduction channels, a

multi-lamellar flow mixer, an automated volume-adjustable incubation chamber, and an integrated electrospray source for time-resolved, MS-based monitoring of protein-ligand binding kinetics. The device is fabricated using multilayer soft lithography and consists of three layers: a flow layer, a control layer, and a cover layer. The multi-lamellar flow mixer contains 28 co-flowing channels of alternating composition that merge into a single narrow channel. The protein introduction channel is on the flow layer and directly connects to the long co-flowing channels, while the ligand introduction channel is part of the cover layer and connects to shorter co-flowing channels via holes punched on the control layer.

To monitor protein-ligand binding kinetics, the protein and ligand solutions were initially split into two sets of co-flowing channels. Mixing was triggered at the merging triangle and completed in the narrow channel. After rapid mixing, the solution was directed to the incubation channels. The incubation time was determined by the volume of the given flow path, which was selected through the actuation states of the integrated pneumatic microvalves. The mixture was subsequently electrosprayed and detected by an IMS/time-of-flight MS instrument, which was optimized to preserve fragile noncovalent interactions.



By measuring the ratio of unbound protein to complex based on acquired mass spectra, the relevant concentrations can be obtained at each time point. This assumes that ionization efficiency is not changed upon binding of the ligand, which is reasonable for a large protein and a small ligand, as is the present case, and it is possible to verify this assumption or correct for discrepancies as needed with the incorporation of a non-

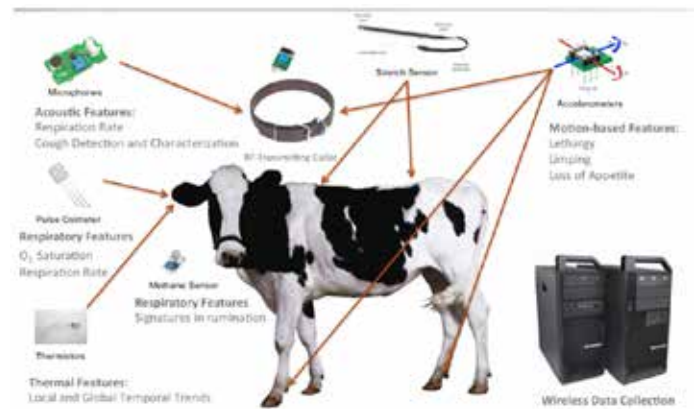
binding internal standard. With the current system, the K_D of carbonic anhydrase-furosemide binding interactions is calculated as 6.35 μM (standard error of the mean of 1.5 μM), which is similar to that obtained by SPR. On and off rates were also obtained using nonlinear curve fitting.

To summarize, under this project we have designed a novel platform for IMS/MS-based monitoring of protein-ligand binding dynamics.

Signatures for Early Disease Detection: Application of a Non-Invasive Multi-Modal Sensor System for Bovine and Swine

Luke J. Gosink

This research will develop new statistical models and integrate these models with next-generation biosensor platforms to monitor and detect early indications of disease through a passive, wearable monitoring system called Yellow Jacket. This system will be independently evaluated by research universities in independent studies to detect early indications of Bovine Respiratory Disease (BRD) in cattle and porcine influenza in swine.



Yellow Jacket Wearable Sensor System

Exposure science is the collection and analysis of quantitative and qualitative data needed to understand the nature of contact between receptors (e.g., people, animals, and ecosystems) and physical, chemical, or biologic stressors. It also plays a key role in the development and application of epidemiology, toxicology, and risk assessment and provides critical information for protecting human and ecosystem health. Infectious diseases are an especially concerning class of exposures. Whether a result of agro-terrorism or natural viral evolution, such events have disastrous impacts to national security in terms of loss of life, immediate depletion of national food stores, and massive devastation to economic resources.

In every disease outbreak, early detection is recognized as the single most important factor in disease mitigation and management. The objective of this work is to demonstrate new advancements in early disease detection capabilities through a wearable, real-time monitoring system called Yellow Jacket. This system will be independently evaluated and used to detect early indications of BRD in cattle and porcine influenza in swine. The challenge with detecting diseases in early stages is that there is rarely any single indicator or symptom of the infectious agent. As a result, diseases can rapidly spread to other individuals long before more obvious indicators are visible. To date, contemporary research expounds on the benefits of using a single sensor modality for monitoring diseases in livestock (e.g., accelerometers or acoustic devices) but have yet to address how to aggregate/fuse information from multiple sensor types to provide more accurate and earlier disease detection capabilities.

The Yellow Jacket system addresses this challenge by aggregating multiple types of data obtained from varied, noninvasive, passive sensor instrumentation. In the Yellow Jacket system, each sensor targets a specific physiological indicator (e.g., thermal signatures, oxygen saturation in blood, acoustics in upper and lower lungs, respiration rates, activity levels, strenuous recovery rates, pressure around lymph nodes and chest cavity, etc.). Through the aggregation of this information, diseases can be detected based on combined sources of early evidence long before contemporary methods would indicate the disease.

During FY 2016, challenge studies were conducted by Mississippi State School of Veterinarian Medicine that focused on early onset of BRD in young cattle. These studies, which were not funded by DOE or PNNL, used the Yellow Jacket system to collect data and monitor the condition of the animals.

Based on our initial research, we have identified several new, early indicators of respiratory disease that have not previously been known based on physical sensor measurements and observational data.

In FY 2017, we will complete the development and validation of these physiological signatures and integrate them with biological signatures that are currently being developed based on microbiomes existing in the animals' stool, respiratory tracts, and metabolite analysis performed on the animal's saliva and blood. A final task will present the compounded evidence of biological and physiological signatures that will form a comprehensive roadmap for detecting the earliest indications of respiratory disease in cattle.

Smart Node – A Highly Adaptable Passive Acoustic Receiver System

Z. Daniel Deng

This project has developed a highly adaptable, passive acoustic receiver (termed “Smart Node”) that has applications in many environments such as detecting, decoding, and localizing fish implanted with acoustic transmitters in rivers; detecting and localizing vocalizing marine animals in the ocean; and monitoring noise from human activities such as blasting and pile driving.

Hydrokinetic and hydropower turbines often sweep into their flow or expose fish and other aquatic life to the risk of injury or death. They also impact the aquatic environment beyond their immediate physical presence by generating noise that may detrimentally affect the behavior or damage sensory hearing of fish and other animals. Information about the mechanics of the risk to individual fish, populations of fish, and other aquatic animals that may result from the presence of these machines is necessary to pursue modification to their structure and operation to mitigate risk.

Currently available passive acoustic receivers are designed to accomplish a specific task such as detection of the transmission from an acoustic transmitter or to record the sound occurring in an aquatic environment over a specific band of frequencies. They typically do not permit users to modify their operation in any significant manner, other than selection of parameters that manage the function of the instrument within its restricted range of design operation. In consequence, acquisition of data needed to assess the risk to fish and other animals requires the purchase of several expensive pieces of equipment, time investment in learning how to use the different devices, and accommodating differences in deployment requirements, plus other miscellaneous costs. However, the most significant problem with currently

available passive acoustic nodes is that they cannot be easily assembled into arrays to perform demanding tasks such as 3D tracking of acoustically tagged or vocalizing animals or to assess the 3D structure of a noise field. All tracking and mapping tasks that utilize passive acoustic techniques require that the time of arrival of all signals received at each node in an array be known with very high accuracy (i.e., microseconds). This is currently done by using a common clock, such as a global positioning system receiver, and linking all receivers to the clock so that they all share the same time.

The hardware design uses innovative analog processing techniques to reduce system complexity and power consumption. The hydrophone signal is first amplified using a variable-gain amplifier, then passed through a quadrature demodulation circuit. This circuit mixes the signal with the 416.7 kHz modulation frequency at both 0-degree and 90-degree phase shifts. The two outputs are each passed through a low-pass filter and sampled by a 16-bit, dual-channel, analog-to-digital converter at 250 kHz. Performing the demodulation in hardware rather than software allows the use of a much lower sample rate and relaxes the downstream signal processing requirements. On the digital side, a Microsemi SmartFusion2 Microcontroller/Field-Programmable Gate Array (FPGA) hybrid receives the digitized signals and correlates the waveforms with a Barker code to detect transmissions. This operation is performed continuously on FPGA fabric. Detected waveforms are saved in a sequence of rolling buffers and passed to the microcontroller for further decoding. The overall design is very flexible and can be reconfigured to record waveforms with or without demodulation.

Initial tests demonstrated that the circuit would require higher gain on the first-stage amplifier to detect actual tag codes, possibly in conjunction with additional noise filtering. However, the system successfully detected and decoded signals from an acoustic beacon in lab testing.

Universal Liquid TEM Microfluidic Cells Based on SALVI for Predictive Materials

Xiao-Ying Yu

We have developed a unique vacuum compatible microfluidic device, System for Analysis at the Liquid Vacuum Interface (SALVI), which enables direct imaging of liquid surfaces by multiple analytical platforms.

Transmission electron microscopy (TEM) is a widely used technique for material characterization with high spatial resolution at the nm level; thus, it is one of the Department of Energy's (DOE's) most important tools for predictive material sciences. The surging interest in the ability to study liquids using TEM has resulted in several companies offering specialized TEM holders and chips for liquid TEM imaging. These holders are expensive, costing more than \$300 thousand each, compared to about \$40 thousand for a standard TEM holder. In addition, liquid TEM holders are not compatible with standard TEM chips. The specialized chips needed for liquid TEM studies often have limited dimensions, thus limiting the type of samples can be analyzed. Although improvement has been made to reduce sample bulging under vacuum by designing different window sizes and shapes, there is a strong need for a universal liquid TEM device, compatible with standard TEM holders and with diverse applications and low cost, eliminating the need to purchase specialized sample holders and expensive TEM chips.

In response to this need, we have developed a unique vacuum compatible microfluidic device, SALVI, which enables direct imaging of liquid surfaces by multiple analytical platforms. Extending SALVI to TEM enables analysis of the same sample with nm spatial resolution, without altering the sample through methods such as freezing or drying. This innovation is a novel, universal tool that is suitable for multimodal imaging for advanced chemical imaging platforms and significantly improves our capability to investigate predictive material sciences. SALVI will make liquid TEM analysis accessible to anyone with a TEM instrument for *in situ* dynamic material characterization, without needing to acquire expensive sample holders and holder-specific chips.

The potential accessibility of SALVI-TEM cells to any TEM will ultimately popularize dynamic imaging and analysis of materials of interest to the general community and significantly increase our knowledge of the evolving interfaces involving liquids, a grand science challenge outlined in the DOE science mission.

We conducted two tasks to achieve our research objective of providing a universal SALVI-TEM platform suitable for liquid

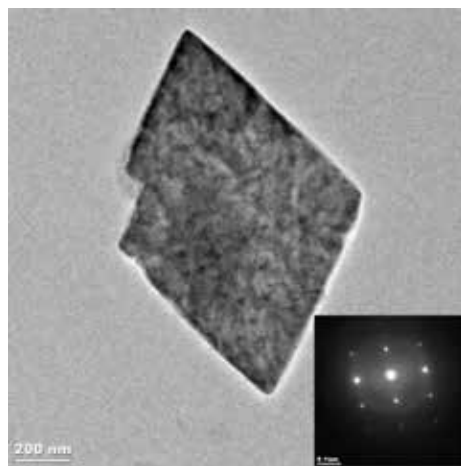
imaging, using a standard TEM holder with much reduced cost and increased flexibility.

In FY 2015, we completed the first task—to demonstrate the SALVI-TEM chip feasibility using a standard TEM holder. After developing a SALVI-TEM stationary device suitable for a standard TEM holder (e.g., Gatan holder), experiments were conducted with an aqueous solution containing different particles. Several TEM silicon nitride (SiN) windows, each with different dimensions in window size, window shape, and spacer thickness, were tested to determine the conditions for enclosing liquid and TEM liquid imaging. An optimized TEM condition was identified.

In FY 2016, we accomplished our second task—to optimize the SALVI approach for TEM applications using standard TEM chips. We also conducted dynamic drying experiments of aluminum hydroxide using a standard TEM heating holder

The innovation achieved in this project will appeal to not only the microanalysis and microscopy community, but also anyone with research interests in material analysis involving liquid phases. As liquid TEM technology advances, it is anticipated that more people will begin to use this unique, yet universal tool to study materials *in situ* in dynamic conditions. Moreover, it will allow more diverse studies in mesoscale imaging of material interfaces toward predictive material sciences for DOE.

The SALVI-TEM cell will fulfill the scientific challenge of unique characterization capabilities for multimodal imaging, significantly enhance our abilities to study materials from different space scales involving multiple phases, and ultimately address the grand scientific mission of predictive material synthesis and energy production, based on improved understanding at the atomic and molecular level.



The TEM image of a single AlOOH particle in water. The insert shows the x-ray diffraction pattern of this particle.

Biological Sciences



Advancing Ecosystem Understanding of Carbon Turnover and Storage through Molecular Characterization

Kirsten S. Hofmockel

Using a long-term continental scale experiment, we will test if increases in nitrogen inputs will augment the accumulation of plant and microbial residues onto mineral soil surfaces. This research will provide the first continental scale analysis of nitrogen effects on molecular biogeochemistry, which is necessary for managing soil fertility.

The coupling between concurrently changing carbon (C) and nitrogen (N) cycles remains a key uncertainty in understanding feedbacks between the terrestrial C cycle and climate change. Existing models do not consider the full suite of linked C-N processes, particularly belowground, that could drive future C-climate feedbacks. The recent focus on C and N interactions stem from the fact that N determines how much CO₂ natural ecosystems can absorb (via photosynthesis), versus how much soil organic matter (SOM) will be mineralized (to support heterotrophic metabolism). Decades of empirical studies addressing N effects on soil C storage have not revealed predictable relationships between N additions and ecosystem C cycling. This is, in part, because soil harbors a wealth of diverse organic molecules, most of which have not been measured in hypothesis-driven field research. For the first time, we will systematically assess the chemical composition of SOM and functional characteristics of the soil microbi-

ome to enhance our understanding of the molecular underpinnings of ecosystem C and N cycling. Our work aims to 1) quantify how changes in N inputs affect SOM formation and fate in six replicated ecosystem experiments from across the United States, and 2) identify microbial traits and/or chemical biomarkers that change in response to N inputs.

To this end, we have acquired soils from 6 ecosystem experiments across the United States that have been subjected to 8 years of N addition treatments. These soils have been analyzed for chemical composition and prepared for a long-term laboratory incubation experiment designed to identify how the soil fertility and stability is altered by N fertilization. We have also been developing novel functions and statistical code for handling soil chemistry data (Fourier transform ion cyclotron resonance) and mapping to metabolic maps for easy visualization. This aspect of the research bridges biology and data science by creating user-friendly data tools. It will enable analyzing diverse data to address our central research hypothesis.

Preliminary results suggest distinct carbon signatures from our field experiments and shifts in soil chemistry in response to 8 years of N fertilization. Using our new analytical tools, we will determine the abundance, composition, and stability of C compounds before and after our incubation experiment. This work has the potential to identify key molecules, or biomarkers, which are central to the long-term storage of soil C.

Aperture

Christer Jansson

This project aims to understand how parameters such as CO₂ levels, light, soil moisture, and temperature affect the aperture of the stomata by regulatory mechanisms in the guard cells that make up the stomata.

Plants are sessile organisms, and their survival depends on efficient perception and response to the constantly changing environment. A major interface between plants and their surroundings is represented by stomatal pores formed by pairs of highly specialized guard cells. To maximize CO₂ uptake for photosynthesis, and at the same time minimize water loss, guard cells sense various signals and adjust the stomatal pore size accordingly. The way in which stomatal aperture is adjusted by the rapid movement of guard cells has fascinated plant biologists for decades.

How abscisic acid (ABA) induces stomatal closure and how light induces opening have been studied extensively. ABA is a plant hormone synthesized during drought, and it prevents water loss by inducing rapid stomatal closure. ABA triggers a signaling network in guard cells that results in loss of turgor and reduction of the stomatal aperture. Blue light triggers stomatal opening in most plants by a signaling network that involves protein kinases. The mechanisms for regulating stomatal closure and opening involve distinct proteins and kinases that regulate ion traffic across the plasma membrane.

The purpose of the Aperture project is to leverage the Environmental Molecular Sciences Laboratory (EMSL) 'omics and imaging capabilities to study how the plant stomata is regulated by environmental factors in *Brachypodium distachyon*. This will lead to better understanding of how parameters such as CO₂ levels, light, soil moisture, and temperature affect the aperture of the stomata by regulatory mechanisms in the guard cells that make up the stomata.

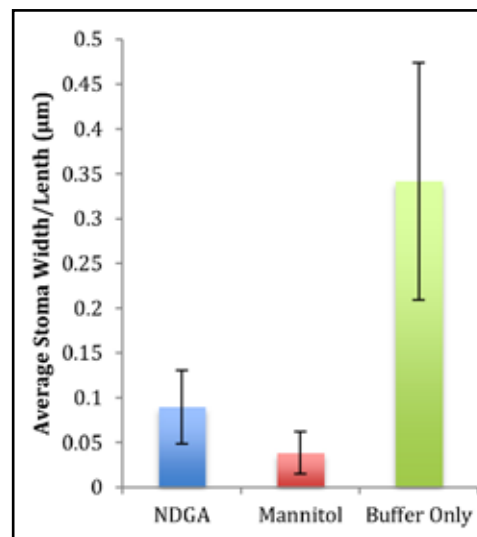
The objectives during the first 5 months of the project were two-fold: 1) to develop methods for imaging *Brachypodium* stomata in by confocal and helium ion microscopy (HeIM) in order to enable characterization and capture of *Brachypodium* stomata in the living state, and 2) to develop the technology for single-cell transcriptomics of *Brachypodium* guard cells.

HeIM allows for sub-nanometer resolution of uncoated biological tissues. In combination with high-pressure freezing, HeIM enabled high-contrast, high-resolution imaging of stomata on the surface of drought resistant *Brachypodium* leaf tissue in the living state, including intact leaves and epidermal. Future studies will delve into plants exposed to drought and/or ambient and elevated CO₂ levels for different periods of time.

Single-cell analysis (SCA) will elucidate cellular functions within individual cells in response to changes in their environment that are not accessible from bulk measurements of heterogeneous populations. We were able to demonstrate our ability to isolate individual guard cells from whole plant tissue, a critical step prior to extraction of ribonucleic acid (RNA) and generation of a cDNA library from individual cells for RNA-seq.

We investigated phenotypic responses to both phytohormones and inhibitors of stomata function to understand how the plant responds to environmental stress. *B. distachyon* was grown with or without the phytohormone gibberelin to survey all strains for differences in stomata concentration. We found gibberelin caused not only significant increases in stomata concentration in all lines, but also resulted in irregular stomata morphology. ABA induces stomata closure, so we exposed leaf tissue to nordihydroguaiaretic acid (NDGA), a known ABA inhibitor, to compete and block ABA activity and force stomata to remain open. We used mannitol to induce stomata closure by osmotic stress.

Leaf tissue was cut from the central portion of the leaves, dissected according to published procedures, and the samples were imaged in the Zeiss LSM 710 Confocal. Transmitted light and chlorophyll A autofluorescence (red channel) images were collected in two channels and overlaid.



Average stomata width by length of exposed leaf tissue.

PN15084/2759

High-pressure freezing (HPF) of *Brachypodium* leaf tissue was carried out on a Leica HPM 100. Following HPF, leaf samples were fixed by the 2-hour Fast Freeze Substitution method (Electron Microscopy Sciences), washed, all water removed, then dried. The samples were purged, brought to supercritical pressure and temperature (42°C, 1200 psi) for incubation and equilibration, then pressure was slowly reduced (100 psi/min), and, after the bleeding process, dried samples were transferred to HeIM holders with carbon tape and stored until imaged on an Orion helium ion microscope (Carl Zeiss Microscopy), and images were imported into ImageJ software, version 1.50a (NIH), and features were measured using the scale bar and line tool.

For isolation of guard cells by flow cytometry, leaf blades from *Brachypodium* were gently scraped with a razor blade to produce a fine pulp, which was subsequently placed in plant digestion Enzyme, vortexed slowly for 1 hour, and sonicated. The sonicated digest was washed and resuspended, then filtered through a 100 µm nylon net filter prior to performing flow cytometry for further cell isolation.

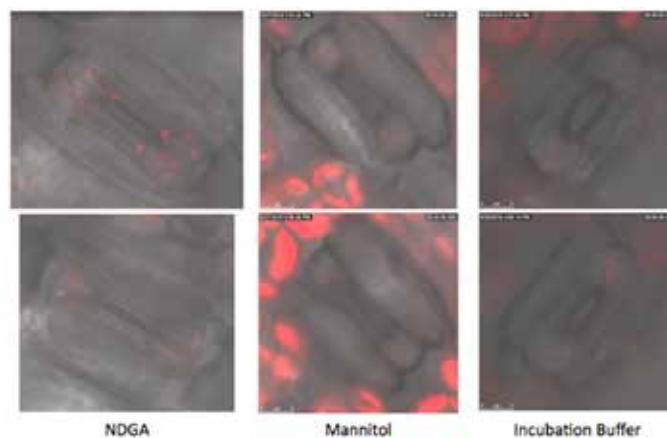
Leaf tissues isolated from the central portion of the leaves were dissected according to published procedures, and laser capture dissection microscopy (LCDM) was performed using a Zeiss PALM MicroBeam.

High-resolution microscopy is dependent on the treatment of sample prior to imaging. One of our initial goals was to preserve the leaf tissue in a living state that would reveal native cellular structures. Prior to treatment of our leaf tissues, we verified preservation of the stomata and plant cells by confocal fluorescence microscopy.

HeIM reveals the surface architecture of the *Brachypodium* leaf tissue in fine detail. The mesophyll side of the guard cells is more clearly defined in the abaxial layer as opposed to the outer epidermal layer, where they appear camouflaged by the surrounding tissues. HPF clearly preserves the natural architecture of the leaf tissue surface and allows long-term storage of different treatments and sample modalities. Ideally imaging the native state of the leaf tissue would require additional adjustments to the fixation methods, as well as eliminating the need for conductive coatings of the leaf tissue. However, our results clearly demonstrate that we can attain sub-micrometer resolution of the leaf tissue surface.

Further experiments will utilize tissue-sectioning methods following HPF treatment to reveal the native organization of organelles within the stomata and surrounding guard cells. Moreover, analysis of varying light, temperature, and calcium will capture the stoma in the varying open conformations in high resolution that will reveal further details of the *Brachypodium* and other plant stomata.

ABA is used by plants to instantaneously signal stomata closure. We surmised that inhibiting this signal could maintain the stomata open long enough to gain high-resolution images of the stomata in an open state. We found that leaf tissue viability is severely hindered after 1 hour of dissection. However, we were able to observe responses to NDGA and Mannitol by high-resolution microscopy. We were able to collect 25 different z-stacks of stomata from each condition. We measured the width and length of each stomata to obtain a measure of stomata size relative to each condition. Unfortunately, the incubation buffer contained high potassium that caused an influx of potassium ions into the guard cells, resulting in open stoma and significantly masked function of the phytohormones. Future studies will involve additional phytohormones, as well as plants exposed to drought and/or ambient and elevated inorganic carbon (i.e., CO₂, bicarbonate) levels for different periods of time to understand how *Brachypodium distachyon* responds to environmental stress.



Confocal fluorescence microscopy imaging using transmittance of leaf tissue that has been peeled. The stoma can be seen surrounded by the guard cell in the abaxial layer of the leaf tissue (center, yellow circle). The bright line section (from left to right on the lower half of the image) is the phloem of the leaf surrounded by chlorophyll containing mesophyll cells (red channel is the autofluorescence from chlorophyll a).

At the Fringe of a Shifting C Paradigm with Climate Change: Unlocking the Organo-Mineral Controls on the Bioavailability of C at the Terrestrial-Aquatic Interface

Ryan S. Renslow

Through our creative experimental approach of simulated pedogenesis and unique combination of technologies that span across fields and disciplines, this work will advance our understanding of the mineralogical and biological mechanisms that regulate carbon cycling along the terrestrial aquatic continuum in soils and sediments.

Recent evidence highlights the importance of organo-mineral interactions in regulating the source or sink capacity of soil. High surface area soils, such as allophane-rich or clay-rich soils, retain organic matter (OM) via sorption to mineral surfaces which can also contribute physical isolation in inter-layer spaces.

In addition to the mineralogical influence, stabilization of OM can be influenced by development of reduced conditions (high water content and solubility and transport limitations on oxygen availability) that inhibit decomposition. Though recent literature for soils suggests that mineral interactions are important in governing the fate of OM, the capillary fringe can be permanently or seasonally inundated with water, making it hydrologically similar to marine sediments where mineral adsorption is considered the governing process for persistence of OM.

Since near-surface capillary fringe sediments would be subject to pedogenic processes, it is unclear what the contribution of surface area and adsorption is to stabilization of OM in these sediments. Most of the work to date, which relates surface area to stabilization of OM, has focused on investigations of natural soils where OM has been incorporated onto mineral surfaces before sampling, and details on the mechanisms for this dynamic system can only be inferred, limiting our ability to properly predict the source sink capacity of soil. This research is taking methodical steps towards elucidating these mechanisms.

To test the mechanisms of mineral surface area protection of OM, we are facilitating secondary precipitation of aluminosilicates in the presence of OM held at two different temperatures in natural Nisqually River sediments (Mt. Rainier). Recently, a three-month reaction was completed, which is intended to simulate early pedogenesis.

This research is being conducted with the hypothesis that increased soil temperatures will thermodynamically favor weathering and resultant precipitation of secondary minerals generating micro-environments, coatings, and micro-aggregates trapping OM. Following pedogenesis, we used an isotopically labelled substrate to prime the native microbial communities and induce biodegradation of native OM. We tracked biomass, microbial respiration, and OM transformations during a short-term incubation.

Using mass spectrometry technologies in tandem, we will identify changes to mineral-linked OM from before and after the incubation. We also will measure OM chemical adsorption/desorption, biodegradation, and the molecular composition of mineral-associated OM, both prior to and following the temperature manipulation. To simulate the saturation of capillary fringe sediment and associated transport and reaction of OM, column experiments will be conducted using the reacted sediments.

The results from the static experiments will then be used to model the transport phenomena from the column experiments using COMSOL Multiphysics, which will yield a user-friendly carbon transport and environmental attenuation (C-TEA) predictive computer application.



Sediment and groundwater sampling at Nisqually River (Mt. Rainier).

Automated Biofilm Imaging with White Light Interferometry

Raymond S. Addleman

A new microscopic imaging method was developed to non-destructively monitor the activity of microbial communities and biofilms with white light interferometry (WLI). The technology enabled in situ observations on the growth of a single bacterium into a microcolony with unprecedented precision.

Biofilms form when microorganisms settle on and colonize a surface. This biological process is non-linear and results in heterogeneous, structured communities that have features across micro- and macro-scales. Traditional imaging methods, such as bright field microscopy, confocal microscopy, electron microscopy, or atomic force microscopy, must necessarily limit the field of view in order to achieve fine resolution. Further, these techniques are destructive to the biofilm and prevent continuous monitoring and study of the biofilm as it develops.

WLI is a specialized optical microscopy technique that can generate 3D topographical maps of a sample surface. WLI has not been used extensively for biological imaging because of the need for a highly reflective surface. However, WLI is a non-destructive and non-contact technique—its advantage lies in its optical resolution, which can be as low as 3 nanometers in the vertical direction. Imaging bacterial communities with this technique offers a unique opportunity to monitor structural, topological, and volumetric changes with unprecedented precision. The technique can resolve volume changes as low as approximately 500 zeptoliters (10^{-21} liters), which is four orders of magnitude smaller than the volume of a typical bacterium (1 femtoliter).

A flow cell was designed and built to specifically accommodate the optical demands of interferometry, while also providing a platform for controlled bacterial growth. Flow cells were rapidly prototyped and manufactured with a 3D printer. Flow cells were then used to monitor the development of bacterial biofilms over time. The structure of the biofilms was explored as a function of conditions, including when the system was exposed to a common disinfectant.

The biofilm imaging method was adapted to accommodate the common 96-well plate. This advancement will enable imaging of biofilms under a variety of growth conditions or under exposure to chemicals of varying concentrations.

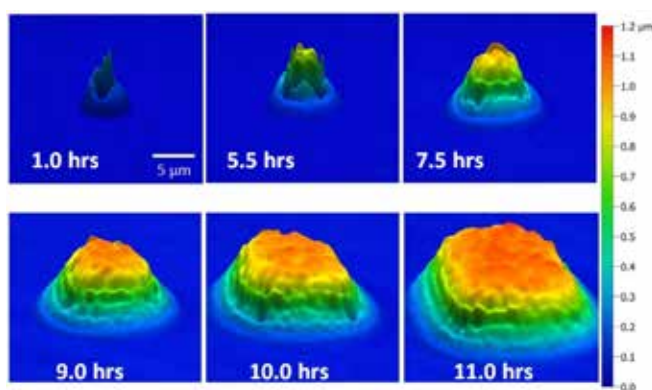
In the future, the method may be used for: rapid diagnosis of bacterial infections, environmental evaluations of potentially toxic materials, and biomedical testing of therapies, coatings, and chemical agents that aim to disrupt bacterial biofilms.

Finally, WLI imaging of microbial growth was used to accurately enumerate bacteria in a plate culture. WLI was used to identify individual bacteria on an agar plate. At high resolution, a single bacterium was tracked and monitored as it grew into a microcolony. Subtle changes in topology revealed the emergence of a protective extracellular layer over the colony. In future work, this technique may be used to identify key transition points during which cells experience phenotypic transformations as a result of spatial crowding (i.e., quorum sensing).

WLI is unique in that the vertical resolution is independent of magnification. Even at low magnification, the technique is sensitive to topological changes on the order of 3 nm. Large area images of bacterial culture plates enabled parallel measurement of growth rates of about 500 colonies. This data may be used to monitor growth dynamics in bacterial communities.

Several of the technologies developed under this project may have commercial applications in the areas of biomedical research and drug development. A patent was filed and the team began to engage external partners. The research was also presented to the scientific community in presentations at national and international conferences.

Future efforts in this area will be focused on clinical applications and on the fusion of WLI with other imaging techniques that are germane to the biological sciences.



An interferometric microscope was adapted to enable non-destructive monitoring of a bacterial colony in the early stages of development.

Biological Threat Signatures for *Bacillus anthracis*

Cindy Bruckner-Lea

Being able to distinguish laboratory-adapted pathogens from those that are naturally occurring remains a challenge for the research and biodefense communities. This work will enable a more rapid identification of the source of disease outbreaks and a more effective response.

Previous work at PNNL has been focused on identifying and characterizing protein, carbohydrate, and other cellular signatures of pathogens grown in laboratory media. In addition, we have shown that the commonly used genomic tools for identification and characterization of pathogens are insufficient for providing information regarding whether an isolated pathogen has been cultured in a laboratory, which would be a likely prerequisite to its use as a bioweapon. This project was initiated to provide an in-depth investigation of both wild and laboratory strains of *Bacillus anthracis* using proteomics to understand the protein signatures that differentiate benign from possibly intentional infections.

In FY 2015, two of our team members traveled to the Emerging Pathogens Institute at the University of Florida to meet with Dr. Jason Blackburn and compile a list of global wild and laboratory *B. anthracis* strains from his collection, to be grown for proteomic analysis for this research project (a total of 80 samples). Dr. Blackburn also visited PNNL in July 2015 to give a seminar and have additional project discussions. His laboratory prepared the *B. anthracis* samples for proteomic analysis at PNNL and verified that the samples were sterile through extensive testing. However, a Centers for Disease Control and Prevention transport moratorium on *B. anthracis* delayed the shipment of samples to PNNL until the current fiscal year.

Also in FY 2015, a team member traveled to Ventura, CA, for the Gordon Research Conference on Chemical and Biological Terrorism Defense and presented data about *Yersinia pestis* threat signatures, which were generated during a previous PNNL effort that led directly to this project. During the meeting, a partnership with Northwestern University was established that expanded our project capabilities and included a proteomic characterization of *Y. pestis* and *Y. pestis* infection

and allowed additional proteomic studies with *Y. pestis* while we were awaiting *B. anthracis* samples.

We were provided samples of lung fluid from *Y. pestis*-infected and uninfected mice, as well as cultured *Y. pestis* samples for proteomics analysis (all samples were inactivated and non-infectious prior to sending to PNNL). This work investigated proteomic signatures of *Y. pestis* isolated from mouse lungs after infection and *Y. pestis* processed without any laboratory culture steps. As expected, we observed significantly increased protein abundance in virulence determinants in the *Y. pestis* isolated from mouse lungs after infection compared to the cultures grown under laboratory conditions, and we also observed unexpected changes in central metabolism and stress response. The host response to *Y. pestis* infection was also investigated and can serve as a baseline for future proposed studies using additional strains of *Y. pestis* from different evolutionary lineages. These data offer additional insight into protein expression in laboratory-adapted pathogen strains during infection, which also informs our main efforts investigating *B. anthracis*.

At the end of FY 2015, we invested a significant amount of time further analyzing existing unpublished data that supports project goals and drafting manuscripts. Specifically, we drafted two manuscripts describing the work to study long-term adaptation of *Y. pestis* to laboratory conditions; we modified a manuscript and added data that investigated the use of proteomic signatures of *Y. pestis* grown in different laboratory media for forensics; and we revisited work that examined the differences in proteomic signatures of *B. anthracis* spores produced in laboratory media versus soil. The two manuscripts discussing adaptation of *Y. pestis* to laboratory conditions were ultimately merged into a single manuscript, which was published in December 2015.

In FY 2016, we received the delayed 80 autoclave-inactivated *B. anthracis* biomass samples and began trial proteomic preparations to determine optimum experimental conditions. Samples were processed in randomized batches over the course of about 3 months, beginning in January 2016, and analyzed using liquid chromatography (LC)-mass spectrometry (MS)/MS bottom-up proteomic techniques. All 80 samples were run on the LC-MS/MS instrument in duplicate by July 2016. During the periods of downtime, Dr. Owen Lei-

ser trained extensively in tissue culture techniques at Biosafety Level 2 (BSL2) using commercially available human primary and immortalized cell lines and an attenuated strain of *B. anthracis*. This training provided the foundation for future work at both BSL2 and BSL3 controls and safeguards, which will be critical to PNNL success in pathogen signature science.

Dr. Leiser also gave an invited plenary talk at the July 2016 Cascadia Proteomics Symposium in Seattle, WA. He presented preliminary findings from this project and discussed the utility of proteomics techniques for forensic applications. Over the course of two days, he also had the opportunity to meet and interact with other regional proteomics experts, several of whom expressed interest in future collaborative efforts.

This LDRD project has also led to collaboration in FY 2016 with another LDRD project as a part of the Signature Discovery Initiative. Drs. Landon Sego, Eric Merkley, and others had developed an approach for using proteomics data to train a

machine-learning algorithm, the Lasso regression classifier, to distinguish wild from laboratory strains of *Y. pestis*. Dr. Sego applied the same approach to proteomics data generated from the *B. anthracis* cultures. The classifier was able to differentiate wild and laboratory strains of *B. anthracis* with over 90% accuracy.

In addition to training and data production, Dr. Leiser has begun the task of *B. anthracis* data analysis and manuscript preparation. These tasks will continue into FY 2017, during which time we expect to submit at least two manuscripts for publication: one manuscript will discuss proteomic signatures distinguishing wild from laboratory strains of *B. anthracis*, and a second will present proteomic data regarding a recently identified Nigerian strain of *B. anthracis* possessing qualities relevant to vaccination efforts in the region. Our collaboration with Drs. Sego and Merkley may also yield further publications.

Cultivation-Independent Untangling of Microbial Gene Regulation Networks

Steve Lindemann

We designed an approach to identify the genomic targets of transcription factors in uncultivated microbes using molecular probes that mimic metabolites of interest.

Despite over a century of attempts, less than 1% of the world's microbes are currently thought to have been isolated and cultivated in the laboratory. Our inability to separate the remaining more than 99% stems largely from the fact that, in nature, microbes are almost never found on their own but rather exist as members of diverse microbial communities. As community members, they typically interact strongly with each other, exchanging resources as they cooperate and compete with one another. In contrast to macroorganisms, interactions in microbial communities occur at the molecular scale and are difficult to decipher. The mechanisms by which species and the community's function changes overtime remains an ongoing investigation.

Microbes regulate their gene expression in response to environmental cues and the presence or absence of specific metabolites—chemical compounds involved in growth and survival. This regulation occurs by the binding of specific metabolites to proteins known as transcription factors. Once the transcription factor binds its respective metabolite, it changes shape and can turn “on” or “off” gene expression. When changes in gene expression are detected by metatranscriptomics, it is difficult to tease the exact mechanistic cause or transcription factors involved in the change. This limits our understanding of gene regulation in microbial communities and hampers our ability to predict how these communities will respond to perturbation.

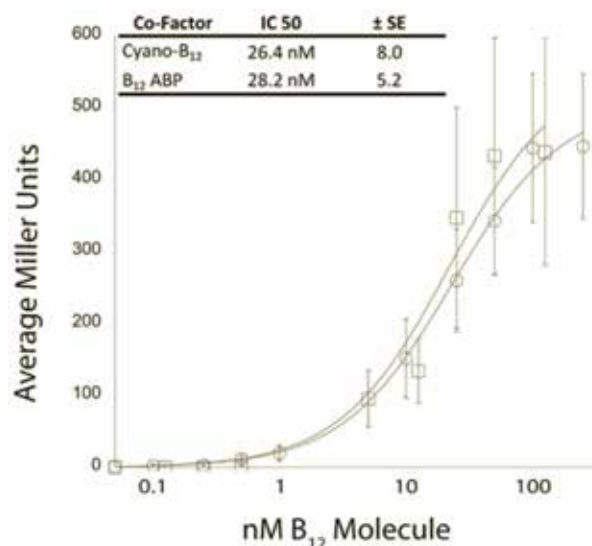
In this project, we are developing a system using metabolite-based probes to identify both transcription factors and the genes they regulate. These molecular probes are designed to mimic metabolites but are modified to allow irreversible binding to transcription factors. This allows us to isolate the ternary probe-protein-DNA complexes. The probe-bound proteins are identified using proteomics, while the genes regulated by the regulators are determined using DNA sequencing. Using a metabolite-based molecular probe approach facilitates the study of highly diverse microbial species, from the human microbiome to extreme environmental communities.

To demonstrate the feasibility of our approach, we have examined two well-known cases of metabolite-mediated gene regulation in *Escherichia coli*—repression of tryptophan

(Trp) biosynthesis and activation of ethanolamine utilization. The transcription factor TrpR is a transcriptional repressor of genes encoding Trp biosynthesis when the intracellular concentration of Trp is high. Using a bioassay, we demonstrated that several Trp probes are transported into living cells, bind TrpR, and repress gene expression. This bioassay helped us to identify the best probe structure and how modifications of the Trp probe are tolerated by TrpR. Final Trp probes will be tested for their ability to enrich TrpR-probe-DNA complexes.

We also have used a similar bioassay to demonstrate that our vitamin B₁₂ molecular probe (B₁₂-ABP) binds the transcription factor EutR to induce expression of genes in the ethanolamine utilization pathway. Results demonstrate that the B₁₂-ABP is inducing gene expression in *E. coli* cells similar to the unmodified Cyano-B₁₂ vitamin. This is the first time ever that a molecular probe has been shown to form the ternary protein-metabolite-DNA complex and be transcriptionally active. With this validation of our B₁₂ probe, we can now enrich the DNA that is bound to the transcription factor and probe.

In FY 2017, we will employ the Trp and B₁₂ probes to enrich probe-DNA complexes to show that they bind correctly to their DNA targets via DNA sequencing and proteomics. Once the expected *E. coli* targets of the probes have been successfully identified, we will apply the probes to different model microbes and experimental communities. We also will synthesize additional probes that mimic other metabolites of interest (e.g., sugars and antibiotics) to determine which proteins recognize these metabolites and what pathways they regulate. Overall, this approach will help predict how microbes will respond to perturbations.



Results demonstrate that the B₁₂-ABP is inducing gene expression in *E. coli* cells similar to the unmodified Cyano-B₁₂ vitamin.

Deciphering Microbial Communication through Metabolites

Thomas O. Metz

This project will establish a Metabolomics Center at PNNL to support the Microbiomes in Transition (MinT) Initiative. The Metabolomics Center will develop the advanced measurement capabilities necessary to define the metabolic processes of microbiomes and determine how these processes change due to perturbation.

Scientific questions to be addressed by the PNNL MinT Initiative include 1) what are the impacts of perturbations on microbial community metabolic processes, 2) what are the impacts of changes in microbial community metabolic processes on ecosystem function and health, 3) can a systems-level understanding of microbial community interactions and metabolic processes be developed, and 4) can improved models be developed to better predict the impact of perturbations on microbial community metabolic processes.

With these questions in mind, understanding microbial metabolism in a community context is essential. Metabolomics measurements will play a key role in defining the metabolic processes of microbial communities and in determining how these processes change due to perturbations. Examples of microbiome perturbations include transitions of the human microbiome due to industrial processes associated with energy extraction and production (e.g., exposures to radiation, oil spills, pesticides, and fertilizers for biofuel feedstocks) and transitions of environmental microbiomes due to climate change.

A related aspect of microbial metabolism is communication via metabolites and other small molecules. These molecules are used by microbes to communicate with each other, their environments, and their hosts. However, the majority of microbial metabolites have not yet been identified, nor have their roles in interactions between community members been characterized. The objectives of the Metabolomics Center are to 1) determine the relative influence of direct (e.g., via secondary metabolites) versus indirect (e.g., via changes in microbial community metabolite pools) microbial communication and interaction through metabolites and other small molecules; 2) increase the identification coverage of the

metabolomes of microbial communities, their hosts, and their environments; and 3) identify the mechanisms by which metabolites involved in communication and interaction are generated, transported, and sequestered within microbial communities.

In FY 2016, this project had the following goals: 1) develop the capability for targeted, quantitative analysis of metabolites involved in central carbon metabolism at the community level; 2) establish the capability for comprehensive, untargeted analyses of lipids and polar and secondary metabolites; and 3) create a knowledgebase of identified metabolites, unidentified metabolite features, their chemical information, and associated microbiome metadata.

Progress toward the first goal included the establishment of analytical methodology for quantitative analysis of central carbon (e.g., glycolysis, citric acid cycle) metabolites in both soil and gut microbiome samples using both gas chromatography-mass spectrometry (MS) and nuclear magnetic resonance spectroscopy (NMR) in a combined, complementary workflow. This methodology was then applied in studies of both wetland and permafrost soils, as well as a humanized mouse model of autism. The goal of the latter study was to determine the role of the gut microbiome and its metabolism in the development of autism.

Work on the second goal focused on incorporating liquid chromatography (LC) and tandem mass spectrometry (MS/MS) into an existing Fourier transform ion cyclotron resonance MS-based metabolomics workflow for characterization of polar and secondary metabolites. The addition of LC and MS/MS will provide much higher coverage of metabolomes in microbiome samples and much higher confidence in metabolite identifications, respectively.

Progress on the third goal included establishing standard ontologies for microbiome experimental meta data, as well as MS and NMR data formats, and establishing the initial computational infrastructure of a Microbiome Metabolomics Knowledgebase.

In addition to the above progress, several new collaborations were established with microbiome investigators, both within PNNL and at external institutes. These collaborations have resulted in the securing of new programmatic funding.

Decomposers in Transition

Kirsten S. Hofmockel

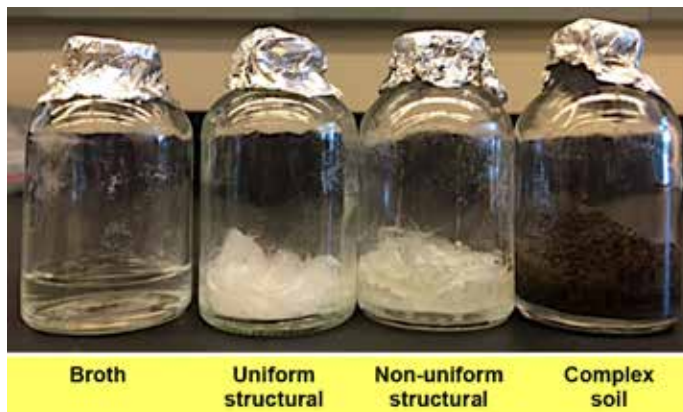
Microorganisms played a central role in forming the atmosphere of the earth, yet we are unable to accurately model how microbes affect current and future climates. This work will provide fundamental biological information required to understand and model how microorganisms transform plant residue from the soil to the atmosphere.

Microbially explicit carbon cycling models are based largely on homogenous liquid cultures that exclude the spatial components of the soil matrix. We have developed an experimental platform that varies the spatial complexity of the microbial habitat to test how connectivity of microbes to each other and to substrates affects microbial metabolism of soil carbon. Using activity based protein profiling (ABPP), in combination with metatranscriptomic analysis, will enable us to identify the key organisms and enzymes regulating carbon decomposition and accurately model carbon transformations in the soil habitat.

Testing Soil Microbial Community Incubations with Varying Degrees of Spatial Heterogeneity: We have developed growth conditions for soil microbes cultured in broth, uniform structural, non-uniform structural, and complex soil in order to evaluate the how spatial heterogeneity and nutrient diffusion affect microbial community composition, extracellular enzyme production, and carbon substrate metabolism. Many experimental factors, including culture apparatus, media volume, growth rate, approximate biomass recovery, and structural matrix components, were optimized.

Model Development: JAM (Just Another Microbial) model has been modified to reflect the batch experiments, with parameter sets drawn from the literature. Through collaborative interactions with data scientists, statisticians, microbial ecologists, chemists, and modelers, we have designed empirical experiments to determine growth and individual carbon source consumption rates to constrain parameters of the existing JAM model. These experiments were designed so that mixed carbon sources could be used for incubations in order to stimulate growth and enhance community diversity to reflect a population that mirrors the native soil, while generating data required to improve predictive modeling. We have

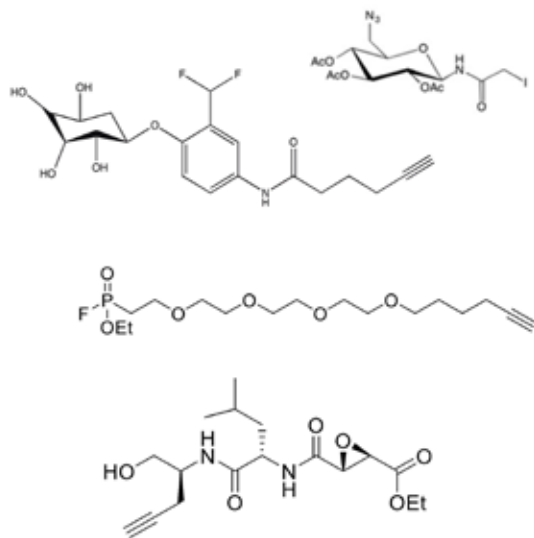
also developed bioinformatics tools and tested methods to rapidly and accurately annotate microbial DNA sequence data. This software has increased the speed and accuracy with which we can characterize microbial community responses.



Example of varying degrees of spatial heterogeneity—broth, uniform structural, non-uniform structural, and complex soil—for soil microbe incubations.

Activity Based Protein Profiling Approach for Assigning Hydrolytic Enzyme Activities to Soil Microbial Members: ABPP is a technique that utilizes small molecule chemical probes to label and enrich enzymes based upon a particular metabolic function. For this research, we are using probes designed to label functionally active glycoside hydrolases and proteases. Probe targets can then be isolated via enrichment techniques and analyzed using high-resolution liquid chromatography-mass spectrometry so that the probe targets can be interrogated and assigned protein identification and microbial origin. We have probe-labeled both microbial cells and extracellular enzymes produced in our tester cultures and visualized the probe-labeled enzymes with sodium dodecyl sulfate polyacrylamide gel electrophoresis by appending fluorophores derivatized to include moieties that react with probes through a copper-catalyzed cycloaddition. Scaling up soil microbiome cultures have historically been an issue for soil scientists, especially when the primary carbon substrate for feeding the microbes is the recalcitrant—yet highly relevant—cellulose, which is the focus of our experiments. Low biomass presents a significant challenge for accurate and successful peptide identification. We are working with PNNL scientists to develop new strategies for addressing proteomics analysis of low

Activity Based Probes for Hydrolytic enzymes



Activity Based Probes for glycoside hydrolases and proteases for profiling enzymes involved in decomposition reactions.

protein samples. These strategies include optimized copper-catalyzed cycloaddition of reporter molecules, streamlined enrichment techniques, nanowell sample preparation, and eventually, the utilization of the new analytical technique tool called Structures for Lossless Ion Manipulations.

Determining Mechanisms of Microbial Metal Mobilization in Coastal Wetland Environments

Rene M. Boiteau

The purpose of this project is to understand feedbacks between climatic change and metal availability that impact the rates at which organisms produce and break down organic carbon in sediments and coastal wetlands.

Many biologically essential trace metals, such as iron, cobalt, nickel, copper, and zinc, are scarcely soluble in aquatic environments. Previous studies have demonstrated that organisms regulate the solubility and bioavailability of these metals through the production of redox shuttles and organic metal chelating agents with a range of metal binding strengths and specificities. Determining what compounds are produced where and when is central to understanding how microbial communities adapt to changes in metal supply that result from external environmental factors such as hydrology or pH.

Climate change is expected to significantly alter patterns of terrestrial temperature and hydrology, which affect the availability of micronutrient metals. The goal of this project is to characterize metal active compounds directly from wetland and soil environments using hyphenated liquid chromatography mass spectrometry (MS) and determine which organisms are utilizing them by identifying the microbes that possess the biosynthetic pathways for these compounds in order to address the following questions: 1) What metabolites bind and solubilize metals in soil/waters across environmental gradients, and how do they impact metal bioavailability? 2) What organisms produce and take up these metal species in the environment? 3) What chemical or environmental triggers activate specific metal acquisition strategies? 4) What metabolic pathways are co-regulated with metal metabolism, and what is the potential impact of these changes on rates of organism growth and carbon cycling?

During year one of this project (which began August 2016), the capability of analyzing metal speciation at PNNL using chromatography hyphenated with inductively coupled plasma mass spectrometry has been established. During FY 2017, this analytical tool will be used to detect metal binding species (Fe, Cu, Ni, Zn) in environmental samples and optimize their chromatography. These compounds will then be characterized by coupling the same chromatography to high-resolution electrospray ionization mass spectrometry (Orbi-

trap or FTICR) to obtain parent ion and fragmentation information, as well as by capturing the fractions and analyzing the isolated compounds by direct infusion with FTICR-MS to obtain molecular formula based on accurate mass. The masses of metal binding compounds will be searched against in-house libraries of characterized metal chelates that have been compiled from scientific literature. For matching masses, authentic standards of the chelate will be obtained (i.e., purchased or isolated from cultures) and analyzed on the same instrumentation to positively validate the identity of the compounds found in natural samples.

In addition, microbial metagenome mining is being used to investigate which organisms produce these compounds.

During FY 2016, metagenomes from Kansas Native Prairie soils were screened for the presence of genes involved in metal metabolite production. Numerous gene homologs to siderophore pathways were found, including bacillibactin, pyoverdine, enterobactin, staphylobactin, and aerobactin. An entire assembled cluster containing multiple pyoverdine biosynthesis genes was found. This gene cluster closely matches those of *Pseudomonas*, an abundant taxa within these soils, suggesting that pyoverdines may potentially be important for iron acquisition in these environments.

In addition to metal chelators, many redox active molecule (phenazine) biosynthesis pathways were observed. These genes are phylogenetically similar to several *Acidobacteria* and *Proteobacteria* genes, raising the intriguing question of whether iron-reduction-based pathways are used by certain microbes under some conditions, while siderophore-based pathways are used under other conditions.

Since oxygen can quench reactive phenazines, our hypothesis is that siderophores are used for iron acquisition under oxic conditions that likely dominate in dry soils, while reductive mechanisms are used under low oxygen conditions that are likely to exist in waterlogged soils. Further MS-based analysis for the detection of siderophores and phenazines in Kansas prairie soils will be carried out during FY 2017 in order to determine whether these metabolic pathways are active under different conditions.

Finally, the transcriptional activity of these pathways across a wet/dry gradient will be investigated to provide additional insight into which microbes are responsible for active production of these compounds. Similar analyses will be conducted on other metagenomes from other soil systems, including permafrost and agricultural land.

Dynamic Multiscale Modeling of Complex Biosystems: A Framework for Multiscale Metabolic Modeling

Garrett B. Goh

This project envisions creating a multiscale modeling framework for simulating cell-level metabolic processes from first principles, while maintaining sensitivity to molecular-level perturbations. The target application for this work is to enable rational computational metabolic engineering, such as in designing low-recalcitrance biomass.

Until recently, we have lacked the mathematical framework to model metabolic processes, which often encompass modeling thousands of reactions. As rate parameters necessary for solving the differential equations used in existing kinetic simulations are difficult to obtain, the most widely adopted methods for metabolic modeling currently rely on fitting experimentally determined growth parameters to a set of pathways consistent with the measurements, an approach known as constraint-based flux modeling. One shortcoming of this approach is its lack of predictive power, as it is simply an interpolation of experimental data.

The objective of this research is to advance metabolic modeling methods based on first principles, using the thermodynamic metabolic modeling framework developed by Cannon and co-workers. Our research differs from existing metabolic modeling methods, as it is based on first principle statistical thermodynamics theory and eschews kinetic parameterization entirely. Instead, standard free energy of reaction are used as input parameters that can be easily computed from computational chemistry methods and integrated in a multiscale fashion.

Prior work reported by Cannon and co-workers has only demonstrated the ability to use thermodynamic metabolic models in toy systems, and no evaluation as to its predictive capability had been completed. Therefore, we evaluated its performance for modeling glycolysis in yeast and compared it to experimental metabolite concentration and fluxes. A reasonable agreement was achieved with a correlation of $R^2 > 0.8$ and a RMSE < 1.0 in log concentration units. The most important finding from this work is that regulatory interactions needed to be modeled in order to achieve any reasonably predictive results. This observation is consistent with

established kinetic models of glycolysis in existing literature. These results were presented at an oral presentation at the International Conference on Intelligent Systems for Molecular Biology, the flagship conference for the International Society of Computational Biology, and two other smaller qBio conferences. We are currently preparing a manuscript for publication in FY 2017.

The importance of accurately representing metabolic regulation in our model led to expanding the scope of the existing study to include an uncertainty quantification analysis. Using simple toy models, our key findings indicate that the predictions of the thermodynamic metabolic model are most sensitive to external boundary conditions (i.e., growth conditions) and the network topology, which includes regulation. Interestingly, the accuracy of the input parameters (standard free energy values) had comparatively less effect on the quantifiable predictions of the model (metabolic concentration and fluxes). The significance of this study is that the primary determinant of the accuracy of thermodynamic metabolic models, network topology, is an empirical property that cannot be predicted from first principles. This project is currently ongoing, and a manuscript is expected to be completed in FY 2017.

A key objective of this research is metabolic modeling with as little reliance on experimental data as possible, and because the sensitivity of thermodynamic metabolic models are highly subjected to empirical data on metabolic network topology, we altered the scope of the study to investigate the efficacy of using machine learning models and “big data” to extrapolate missing information. Due to the lack of sufficient high-quality data and biological complexity of metabolism, we decided to proceed with the next phase of the project using model systems obtained from the chemistry domain, where less complicated systems exist with greater “ground truth” certainty. Nevertheless, we expect that the methods developed for machine learning in computational chemistry, once validated, will be applicable to the computational biology domain.

The final project in FY 2016 was a survey of the current state of using machine learning for scientific research. We identified deep neural networks (deep learning) to be the most promising algorithm, as unlike conventional machine learning algorithms, they have the ability to perform automatic feature engineering with little or no domain knowledge. This

means that deep neural network models can potentially infer representations (i.e., concepts) given a sufficiently rich dataset. A comprehensive review of deep learning for computational chemistry applications is currently underway.

In FY 2017, we will be focusing our efforts on evaluating and developing deep neural network algorithms for computational chemistry applications. Specifically, we have selected

toxicity prediction as the model system. From this, we will address questions related to 1) the extent of domain knowledge required to develop a predictive model, 2) the interpretability of deep neural networks and its potential for re-discovering existing chemical concepts, and 3) uncertainty quantification of deep neural networks.

Establishing New Standards for Automated Digital Biofouling Analysis

Curtis J. Larimer

We developed a novel method to monitor, quantify, and prevent the unwanted accumulation of biological materials on surfaces. The technology can quickly and accurately measure biofilms and biofouling on most surfaces.

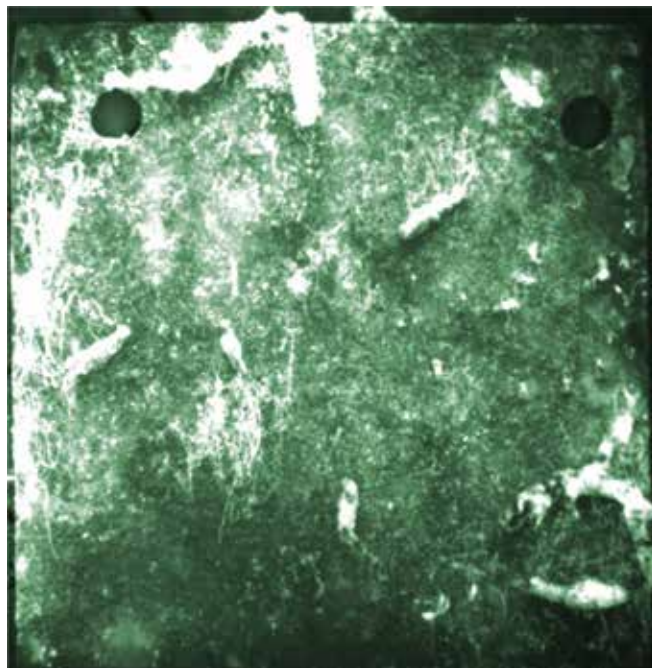
Biofilms form when microorganisms settle on and colonize a surface. Biofilm formation creates a wide range of problems in society, including contamination and nosocomial infections in medical facilities, reduced efficiency of heat exchangers, transport of invasive species on boat hulls, and increased fuel consumption due to drag on ships. Existing methods for evaluating biofouling rely on qualitative assessments rather than objective and quantitative image analysis. Modern tools to monitor, quantify, and prevent the unwanted accumulation of biological materials on surfaces are missing.

In this project, a unique methodology for biofouling analysis was developed. The procedure is divided into three steps: first, the sample of interest is dyed with a specially formulated mixture of biomolecular stains in order to enhance contrast of organic fouling. Then the stained sample is photographed with either a digital camera or with a fluorescence imaging system. The final step is to process the data with a software package that analyzes the image and quantitatively evaluates the extent of biofouling on the sample surface. The software produces a single value for biofouling growth intensity that is representative of the total amount of biological material attached to each sample. The results of this analysis method can be used to assess and compare the effectiveness of coatings and cleaning methods that may be used to prevent or control biofouling.

The primary focus of this project was to expand the types of images that could be analyzed with this method. In particular, we aimed to expand the selection of stains that could be used in the first step to include fluorescent stains. This also required development of a fluorescent imaging procedure and modification of the analysis software to include fluorescence images. The analysis of fluorescent images is important when samples have a high-contrast background (e.g., black paint).

The most important finding from this year was the successful implementation of fluorescent staining into the procedure. This result will broaden the potential application of the technology and encourage adoption in the scientific and industrial biofouling communities through commercialization. A patent was filed for this technology and results were published on the cover of *Analytical and Bioanalytical Chemistry*.

In future work, we aim to submit the method for validation as a standard by the American Society for Testing and Materials. Such a standard could be used to validate the performance of new biomedical devices and anti-fouling coatings, as well as other technologies. The rapid nature of the method developed in this project may help to speed development in these areas.



Development of a new biofouling analysis method can easily assess and quantify the amount of biological material that has accumulated on a surface. Fluorescence imaging can be used to distinguish biofouling on a dark background.

Exploring and Engineering Phototrophic-Heterotrophic Partnerships

Hans C. Bernstein

This project is identifying controllable, cooperative ecological phenomena employed by phototrophically driven microbial communities for conceptualizing and engineering multispecies biocatalytic platforms. The target application for this work is energy capture and transfer from renewable resources, light, and CO₂.

Microbial consortia engineering has become an established scientific discipline populated by interdisciplinary biologists, engineers, and ecologists. The methodology is based on assembling microbial communities through enabling, encouraging, or enforcing interactions between distinct cell populations and their environment. Applications have the widely accepted potential to contribute technology toward key social benefits such as biofuel production, carbon sequestration, and environmental remediation. The soundness of the consortia concept for biotechnology applications is supported by observations in nature: naturally occurring ecosystems optimized by eons of evolution are almost ubiquitously organized as interacting mixed communities. Of these, photoautotrophic microbial consortia are of keen interest to chemical and biological engineers as promising catalytic systems capable of utilizing the renewable resources light and CO₂.

The objective of this research is to dissect metabolically coupled interactions within naturally occurring biofilms from unique environments (i.e., high temperature and hypersaline) to discover ecological cooperation strategies to build engineered microbial consortia. The focus will be on identifying natural biological phenomena between photoautotrophic and heterotrophic microorganisms that can be controlled and harnessed. Our research differs from previously reported microbial community studies because it will advance technical understanding in the context of fundamental and applied science.

Phototrophic-heterotrophic communities were investigated in three separate experiments. The first study used data provided (partially) by external university collaborators and established that both photoautotrophic biofilm communities

were capable of producing biofuel “precursors.” We also established the spatially resolved kinetics of photosynthesis and respiration in these systems, which was published in the *Journal of Bioresource Technology*.

The second biofilm community study employed a novel flow-cell bioreactor designed and built for this project to culture and maintain benthic, hypersaline microbial mats. Advanced oxygen microsensor methods were combined with the novel PNNL technology laser ablation isotope ratio mass spectrometry (LA-IRMS) to resolve spatially photosynthetic oxygen evolution, heterotrophic respiration, and carbon fixation. This work has also been published in multiple articles.

A high-temperature “hot-spring” community was cultured in an advanced turbidostat bioreactor and tested for its ability to overcome light and oxygen stress as a community. Preliminary results suggest that the artificial phototroph-heterotroph community performs marginally better (in terms of measured specific growth rate) than the axenic, thermophilic cyanobacteria controls. The results from this portion of the project were presented at an international conference at the end of FY 2015.

The physiology of marine cyanobacteria as potential drivers of synthetic photoautotrophic-heterotrophic consortia was investigated in greater detail by studying axenic growth and photosynthetic performance, for which two studies were completed. The first evaluated the growth and photosynthetic performance of *Synechococcus* sp. strain PCC 7002 under mono- and dichromatic light regimes, which found distinctions in photoautotrophic functional capacity based on the spectral quality of incident light. These findings were published in *Frontiers in Microbiology*. A second similar study employed an advanced feedback-controlled turbidostat photobioreactor to investigate/compare the growth and photosynthetic performance of *Synechococcus* sp. strain PCC 7002 to its transcriptional response during variable incident light intensities and oxygen tensions. Additional and related studies have also been published in FY 2016 as a result of this project, including articles in *mBio*, the *Journal of Cellular Physiology*, the *ISME Journal*, *Wound Repair and Regeneration*, *Biotechnologies for Biofuel Production and Optimization*, and *Science Report*.

Global Forensic Chemical Exposure Assessment for the Environmental Exposome

Justin G. Teeguarden

Our goal is to perfect and apply technology that allows measurement of thousands of chemicals important to public and environmental health in blood and other fluids used for biomonitoring in less than 5 minutes. The technology will revolutionize the way the effects of chemicals are studied, how safer energy technologies are developed, and how we predict and respond to the effects of global climate change.

Human and ecological health is widely recognized as the product of the genome and its environmental analogue, the exposome: the comprehensive history of exposures to natural and manmade agents within individuals and populations. One ambition of the Department of Energy's Human Genome Project was to use genomic information to predict the health of humans and the environment. However, without corresponding knowledge regarding human chemical exposure and its interaction with the genome, information from the Human Genome Project was not sufficient for making these predictions.

Advanced analytical instruments developed by PNNL offer the chance to provide this missing information by performing measurements that were previously not possible (i.e., quantifying hundreds to thousands of very structurally diverse chemicals, simultaneously, within single samples of blood or other fluids used for biomonitoring of exposure). This project was initiated to perfect the application of PNNL's advanced instruments in making these measurements. The analytical approach used is called ion mobility spectrometry-mass spectrometry (IMS-MS).

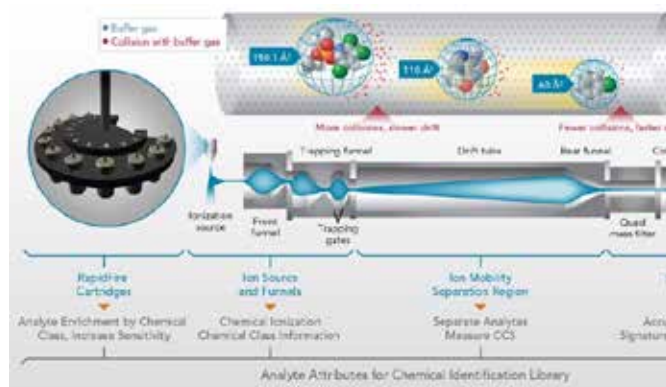
Developing a reference library of chemicals and their unique chemical signatures measured by IMS-MS, a central project goal, continued in the third year using industry standard methods PNNL helped establish. These chemical signatures include the IMS drift times and accurate masses, which become the basis for the high throughput, accurate quantification of the chemicals. To date, we have developed a reference library and chemical signatures for approximately 500 chemicals (lipids and chemicals). A novel computational algorithm that automates development of the library was significantly improved in this year's effort, now providing more accurate assembly of data and calculation of entries for the library. The manuscript describing this method is undergoing internal review before submission. We expanded our collaborations with instrument developers to improve software systems for extracting and pro-

cessing information from the IMS-MS for chemical identification. The team completed a pioneering proof-of-concept and application of the tools in the analysis of blood and urine samples, identifying previously unidentifiable environmental degradation products of polycyclic aromatic hydrocarbons, potential new biomarkers of diabetes, and demonstrated unprecedented sample throughput. Now submitted for publication, this work has seeded collaborations with several academic and non-academic institutions, supporting new work in fields as diverse as microbiology, plant science, and natural product chemistry.

Major progress was also made in the development of advanced computational approaches for the prediction of the unique chemical signature for chemicals without the need for the use of chemical standards. Overall, the research produced innovations that create a high throughput computational method for the accurate identification of unknown analytes, transforming the field of chemical identification from one limited by standards and libraries, to one with almost no limitations, to identifications of compounds present above detection limits. The overall IMS-MS computational library workflow for the exposome was submitted for publication.

During FY 2016, we established new partnerships with the U.S. EPA., Texas A&M University, Iowa State University, other PNNL DOE programs, and several U.S. corporations. These partnerships apply the combined IMS-MS and computational chemistry platforms to forensic chemical analysis of microbiome, human, environmental, and other samples to address emerging questions related to chemical exposures, as well as plant and microbe biology as they relate to energy production and global climate change.

Two of four expected papers were submitted for publication, with two additional papers targeted for November 2016 and February 2017. Three grants were submitted to support continued development of the tools and methods.



Rapid global chemical forensics is possible with the unique combination of IMS-MS and computational chemistry developed at PNNL.

GoBrachy: Developing a Metabolite-Trait Association Network Model for Carbon Allocation in *Brachypodium*

Christer Jansson

We will develop a Metabolite Trait Association Network—a predictive model that links metabolite profiles in the grass model plant *Brachypodium* to quality traits, such as aboveground biomass (AGBM) and belowground biomass (BGBM), using high-coverage metabolomics.

Thirty different *Brachypodium sylvaticum* and *Brachypodium distachyon* lines were selected for this study. Experiment 1 was conducted with 30 *B. sylvaticum* lines and experiment 2 with 30 *B. distachyon* lines. Each seed was planted in a 3-inch pot and was grown in a controlled environment at 22°C with 16:8-hour light dark cycles and 400 parts-per-million CO₂, 60% relative humidity, and 350 µmol-1 light. Each plant received 50 ml of water every other day from start to end for the control samples. Experiment 1 was harvested at 8 weeks of development and experiment 2 was harvested at 4 weeks of development. Plant height and fresh weight of aboveground and belowground tissue was manually recorded. Samples were flash frozen in liquid nitrogen and stored for further processing.

Drought was imposed by withholding water from the experimental samples for 6 days in experiment 1 and 7 days in experiment 2. Drought samples were re-watered once after the set drought period and allowed to recover for 24 hours before harvesting.

Experiment 1 samples are currently being processed for dry weight and experiment 2 data are shown below.

Some wilting and leaf rolling was observed in some accessions following the 7-day drought treatment. These changes were subtle and were difficult to capture at the macroscopic level. Some lines appeared smaller in stature compared to the respective well-watered controls. However, the effect of drought treatment was clearly visible with the dry biomass. The total dry biomass of the 30 accessions ranged between

0.2 g to and 0.7 g. Drought treatments resulted in significant changes in total dry biomass in lines 1, 4, 5, 7, 17, 22, 27, 28, and 30.

Biomass partitioning was significantly altered in lines 1, 4, 5, 16, 17, 18, 19, 22, 25, 27, and 28 for AGBM and lines 1, 2, 5, 7, 9, 15, 18, 20, 21, 24, 25, 28, and 30 for BGBM. Water use efficiency for each line was calculated by dividing the total dry biomass by the total volume of water used. Ten lines from experiment 2—lines 4, 5, 6, 10, 13, 18, 19, 25, 26, and 27—were selected for further characterization using metabolite profiling.

Lyophilized aboveground and belowground tissue was ground to a fine powder using a tissue lyzer, and 40 mg from each sample was extracted with 80:20 methanol:water for 1 hour. Samples were centrifuged and the supernatant was split and transferred into two new HPLC vials. Three hundred µl was used for gas chromatography-mass spectrometry (GC-MS) analysis and 400 µl were saved for future liquid chromatography-mass spectrometry analysis.

The entire metabolome of *B. distachyon* obtained by GC-MS was subjected to a PERMANOVA analysis using Bray-Curtis distance to test for differences in metabolomes between genotypes and treatments (control and drought). The number of permutations was set at 10,000.

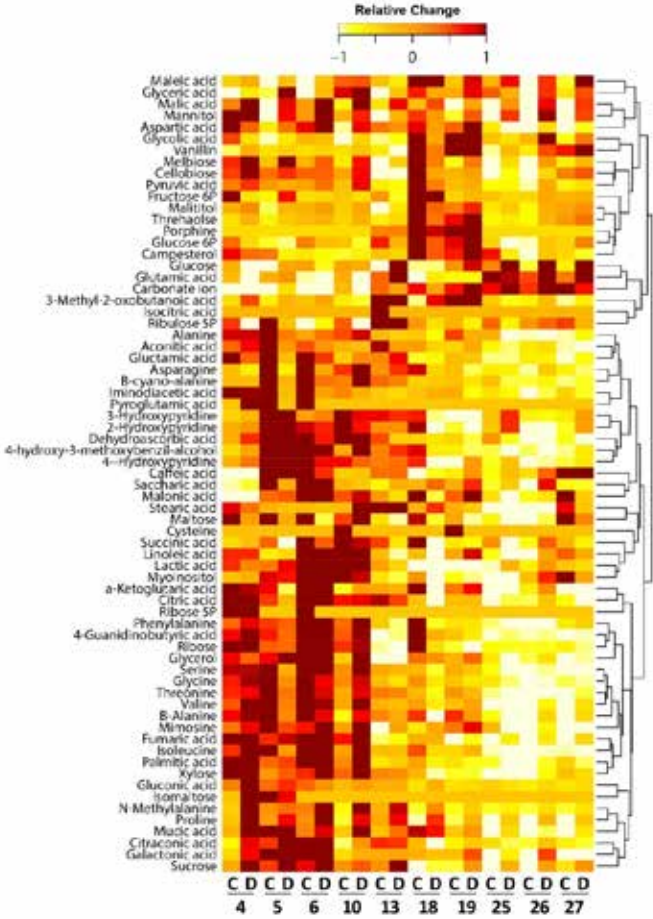
Both genotype and treatment had a significant effect ($P < 0.05$) on the overall metabolomes of plants. Moreover, the interaction between genotype and treatment also showed a statistical significance indicating that the effect of drought was statistically different with each genotype.

All of the GC-MS metabolomes were subjected to principal component analysis (PCA), as well, to determine the natural variability among the samples.

Principal component (PC) 1 chiefly separated the genotypes 4, 5, 6, and 10 from the rest. Interestingly, PC2 separated the drought effect on plant metabolomes and all of them, with the exception of genotype 18, followed the same trend. According to this PCA, the metabolome of genotype 18 did not change between control and drought treated plants.

A heat map of all identified variables with more than 0.8 of matching scoring was plotted to observe the relative change in concentration of each metabolite between genotypes and treatments using the aboveground tissue samples. The relative concentrations for each variable were scaled to the same level before plotting the heat map. Dark red and light yellow correspond to the highest and lowest concentration for the specific metabolite, respectively. Different numbers at the bottom of the figure represent different genotypes. C and D represent the two treatments, control and drought, respectively.

In FY 2017, we will continue to obtain metabolite profiles for the *B. sylvaticum* and *B. distachyon* lines and develop the Metabolite-Trait Association network models. We will also screen the same accessions at elevated CO₂ levels and high temperatures, mimicking future climate scenarios to build predictive models for plant-atmosphere-soil interactions.



Gut-on-a-Chip for Multi-Omic Studies of the Gut Microbiome

Joshua N. Adkins

There is a great need for reproducible, comprehensive characterization of biomolecules from complex microbiome research. The development of a gut-on-a-chip with coupling to biomolecular “omics” measurements will allow greatly improved microbiome studies and hypothesis development.

Alteration of the human gut microbiota composition and function has been associated with many human pathologies. The impacts of perturbation resulting from pathogens, antibiotics, or toxic chemicals on the microbiome, despite great interest, remain largely unknown.

A critical challenge to effectively study the effect of perturbations on microbiota is the lack of well-controlled methods to sustain bacteria in direct contact with a living epithelium. The “gut-on-a-chip” is a microfluidic technology that provides a microenvironment mimicking many aspects of the gut’s environment *in vitro* and in a small, controllable platform. Made of a clear, flexible polymer, this organ-on-a-chip consists of three microfluidic channels that enable low flux of media, chemical, and/or microbes with peristalsis-like motions, inducing the production of polarized differentiated intestinal.

Host cells grown on this type of chip exhibit phenotypic traits of an intestinal epithelium such as mucus production and innate immune responses. Further, in contrast to other *in vitro* methodologies, the gut-on-a-chip can support stable microbial communities for long periods of time.

We are introducing and greatly extending this capability at PNNL to enable characterization of the microbiome in the context of perturbations. Different perturbations will be tested, including the effects of antibiotic treatment and a pathogenic infection. We will develop a refined set of gut-on-a-chip models to allow reproducible analysis of gut microbiomes in transition in a time-dependent manner. The microbiota will be characterized at the molecular level by both microscopic imaging and multi-omics technologies. The analysis of microbiomes in transition by this combination of cutting-edge technologies will allow PNNL to demonstrate an area of excellence in the molecular characterization of gut microbiota.

This year, we had a month’s allocation of funding to provide initial consumables and small equipment purchases while assembling the project team. We produced and tested prototype chips for appropriate mechanical functions and have revised the chip and manipulation components.

During the next year, we will be producing chips with host mammalian cells. These initial cell culture methods will be evaluated for appropriate cell morphology and mucous layer production. Upon successful cell development within the chip, we will perform initial biomolecular measurement to understand quantities available. In parallel, we will begin to introduce complex microbial communities to the chips to understand the conditions that promote growth, development, and maintenance of the microbial community.

Impact of Environmental Stressors on Complex Biological Systems

Karin D. Rodland

This project integrates data from multiple imaging and mass spectrometry (MS)-based technologies to predict complex biological system response to external stressors. In addition to furthering PNNL's capabilities in systems biology, we can also explore applications relevant to human health.

The concepts of stability and resilience are key to promoting ecosystem sustainability in the face of significant climate change. The same fundamental principles are relevant to the maintenance of homeostasis (health) in complex organisms subjected to environmental perturbations. Our objective is to specifically target the flow of information, from the genome to the functional level defined by proteins and metabolites, with emphases on the dynamic response to environmental perturbations and the communication mechanisms that allow complex systems—whether communities of microbes or organized tissues in multi-cellular organisms—to adapt to and mitigate perturbation. We will achieve these goals by applying systems biology approaches to quantitative data generated with unique PNNL instrumentation in MS, nuclear magnetic resonance, and imaging to understand the effects of environmental stressors on multi-cellular systems. We will select novel model systems that represent unique challenges in understanding the effects of environmental stressors on the health and resilience of complex biological systems.

Examining the effect of carbon source on gut microbiome dynamics using model consortia. This task addressed the following two, crucial questions: 1) how the diversity of the natural gut microbiome in mice changes during *in vitro* incubation using common carbon sources and the effects of culture conditions and initial diversity on the final consortium complexity, and 2) the effect of diet induced obesity (DIO) on the gut microbiome and the stability of those changes outside the gut. Beta-diversity was determined over sequential passage samples originating from lean and obese mice to determine how well the consortia represent the original diversity, as well as whether the final consortial composition is governed by initial diversity or culture conditions. Comparisons were made between consortia derived from lean vs. DIO mice. Because the cultures used prebiotic carbon sources, which are more similar to the carbon sources fed to lean mice, the gut microbiomes from DIO mice showed larger changes and moved toward a species makeup resembling that of lean mice.

Identification of systemic components associated with preconditioning induced protection against ischemic tissue injury.

Collaborating with Oregon Health and Science University (OHSU), we established the OHSU-PNNL Co-Laboratory for Integrated 'Omics to tackle problems related to functional proteomics in complex model systems. One such model system is the use of toll-like receptor (TLR) agonists to induce tolerance to ischemic injury in mouse models of brain, kidney, heart, liver, and lung ischemia. The Stenzel-Poore group at OHSU has shown that the combined induction of circulating factors and a local tissue response are required to mediate the protective effects of TLR agonists; however, the identity of these factors is currently unknown. We are currently providing a comprehensive global proteomics analysis of samples provided by the group to identify the TLR-mediated responses in the systemic circulation and in the brain that may orchestrate ischemic protection with the goal of new therapies for ischemic injury.

Metagenome generation enabling multi-omics analysis of community establishment and re-establishment after disruption. A manuscript titled, "Functional Response within a Cellulosic Degrading Anaerobic Bioreactor Measured During Community Establishment and Re-establishment After Perturbation" was submitted to Applied and Environmental Microbiology in 2015, and accepted pending revisions. The work utilized a multi-omics approach to evaluate the hypothesis that, within an engineered bioreactor degrading cellulose, deterministic factors significantly influence community re-establishment after perturbation. The multi-omics measurements acquired as part of this study showed that deterministic factors had a greater influence than stochastic factors on community establishment as predicted by ecological succession theory rather than ecological neutral theory. In the earlier work, protein identification was based on genomic databases from near neighbor organisms; the objective of this project was to conduct metagenomic sequencing of the actual rumen microbiome, to provide more accurate identification of functional proteins. The work was accomplished, the manuscript re-submitted, and it was accepted for publication.

A human primary triple co-culture lung platform for characterizing influenza A virus infection. Translating the intimate discourse between viruses and their host cells during infection is a challenging but critical task for development of antiviral interventions and diagnostics. Advances in MS-based proteomic technologies are enhancing studies of viral pathogenesis by identifying virus-induced changes in the protein repertoire of infected cells or extracellular fluids. This project developed a novel 2D triple co-culture system reproducing normal respiratory epithelium and immune cell components, which was infected with influenza A viruses to study the effects of immune cell contributions on the response to viral infection.

Integrated *In Situ* Chemical and Topographical Optical Imaging of Live Microbiomes in Transition

Curtis J. Larimer

With this project, we aim to develop a novel “bioscope” that combines white light interferometry and hyperspectral optical microscopy to simultaneously capture physical (three-dimensional topology) and chemical (spectrally resolved absorbance micrographs) images of microbes and their communities.

Many microbial communities form elaborate multicellular structures, but the rules governing their architecture are not well understood. Quantitative microscopy of microbiomes in transition requires next-generation instruments that are capable of generating label-free cell-level maps of host-microbe interactions in a non-destructive fashion. We will develop a unique capability for *in situ* imaging, allowing multidimensional topographic and chemical analysis of live microbiomes in transition over a broad range of length and time scales.

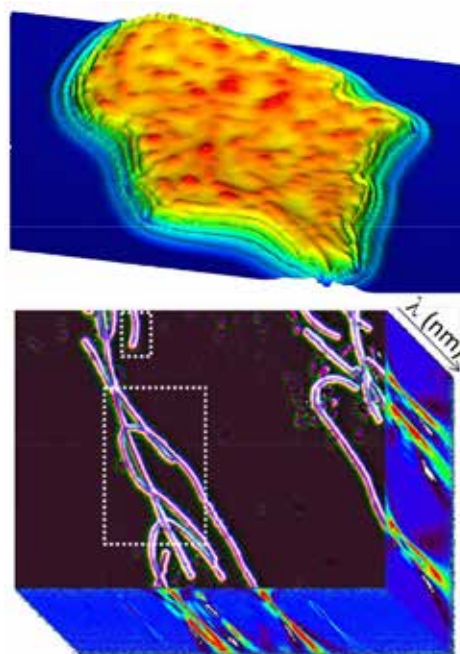
White light interferometry (WLI) has the highest resolution of any optical imaging technique (3 nm axial resolution) and can measure volume changes with femtoliter precision, but its use in bioimaging has been very limited. Hyperspectral optical microscopy (HOM) will complement WLI, because it can assess chemical composition and—like WLI—is a non-destructive technique with a fast acquisition time that is well-suited to imaging unlabeled samples.

Our goal is to provide a key technology that will enable non-destructive monitoring of microbial systems in order to identify the key transition period(s) in microbiome dynamics and to provide cell-level maps that enable noninvasive imaging of host-microbe interactions. We will focus on demonstrating system capabilities by first resolving intracellular and community-wide redox states. To accomplish this we will develop a multimodal noninvasive nanoscale physical and chemical imaging instrument with speed, throughput, and dynamic ranges that are unattainable, using current generation bioimaging techniques. We will develop a unique capability for *in situ* imaging, allowing multidimensional topographic and chemical analysis of live microbiomes in transition over a broad range of length and time scales.

This project started near the end of the 2016 funding cycle. We initiated the team’s instrument design efforts and purchased hardware and software that will enable instrument construction and data collection in following years.

Future research in this project will be divided into two major focus areas. First, we will focus on development of a new optical imaging system based on the fusion of WLI, HOM, and fluorescence. Then we will test, validate, and demonstrate the new imaging capability with high-impact systems to investigate biological hypotheses. We aim to test our new “bioscope” with a simple model microbial system grown on defined media. We aim to observe the morphology and absorption characteristics of microcolonies as they grow into a microbiome community. Fusion and subsequent analysis of WLI and HOM data will be used to study growth rates and redox states in microbial communities.

At the conclusion of our instrument development and testing efforts, we aim to demonstrate the new instrument’s imaging capability by applying it to the *in vitro* gut-on-chip system being developed by another project.



Development of a new bioimaging microscope will provide a capability for non-destructive imaging of the topology (above) and chemical composition (below) of microorganisms.

PN16109/2886

Microbial Community Dynamics and Plant Phenomics with Single-Cell Gene Expression and Imaging Mass Spectrometry

Galya Orr

The inherent heterogeneity of microbial communities limits the questions that can be resolved by averaged cell population measurements, which lump the behavior of an entire population of cells or organisms into a single average value. It is, therefore, possible that the processes deduced from the whole population average are, in actuality, not employed by any of the individuals within the population. The goal of this proposal is to establish new approaches using existing instruments to enable the investigation of individual cells within complex cell populations. The new capabilities will be applied to decipher interactions and mechanisms underlying the function of complex microbial communities in C cycling, biomass degradation, plant resistance, and in the release of volatile organic compounds.

Two specific aims have been pursued under this project. The first was designed to establish a novel approach using mass cytometry (CyTOF) to quantify the expression of multiple genes in individual cells from complex microbial communities in high throughput. The second aim was designed to establish a novel approach using nanoscale secondary ion mass spectrometry (NanoSIMS) to image bacterial cells by the expression of specific genes. These approaches will be used to understand mass and energy fluxes within complex microbial communities and identify the critical players at the cellular and molecular levels with sub-micrometer spatial resolution. Together, our approaches will gain new understanding of the metabolic and regulatory principles underlying the function of the community as a whole.

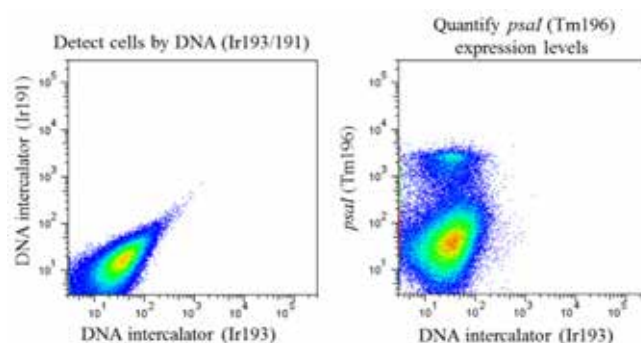
We have established novel approaches to quantify the expression of multiple genes or proteins in individual bacterial cells using mass spectrometry approaches. Mass cytometry has been used to analyze single cells labeled by antibodies tagged with rare earth elements, mostly to detect the expression of specific cell-surface receptors in mammalian cells. However, no study so far has applied mass cytometry to quantify gene

expression in single cells using an *in situ* hybridization approach in high throughput.

Based on our solid experience in the design and application of fluorescence *in situ* hybridization (FISH) probes, we have designed multiple probes and established the protocol for tagging them with different rare earth elements. Our approach relies on multiple sub-probes (about 20 base each), spaced along the mRNA molecule by 3-5 bases. Each sub-probe is tagged with 20 atoms, carried by the commercially available metal-chelating polymers. We have established the protocol for reacting the maleimide groups on these polymers with SH groups that we added to each sub-probe.

This approach ensures that each probe is tagged with sufficient number of atoms for mRNA detection and quantification, using both the CyTOF and the NanoSIMS. Using the CyTOF, we have optimized the above tagging approach and demonstrated the detection and quantification of the probes in bacterial cells with no detectable background.

Further, we have established the application of the technique to detect expression of multiple mRNA species in a single bacterial cell. We also have optimized NanoSIMS analysis protocols and demonstrated single cell imaging of bacteria expressing the universal bacterial 16S rRNA (EUB), as well as the mRNA for the photosystem I reaction center subunit PsaI using thulium 169 labeled probes. We are currently pursuing imaging of bacterial cells where three different subunits of the photosystem I reaction systems are tagged with three different isotopes.



The gene expression level of photosystem I reaction center subunit PsaI was quantified in thousands of cells in high throughput using CyTOF. **Left:** DNA intercalator tagged with iridium was used to identify the bacterial cell population. Each dot in the bivariate plot represents a cell. **Right:** The expression of PsaI was quantified by tagging *in situ* hybridization probes with Thulium 196 (Tm196), showing an average of 500 mRNA copies per cell.

Microbiome-Exposome Interactions

Aaron T. Wright

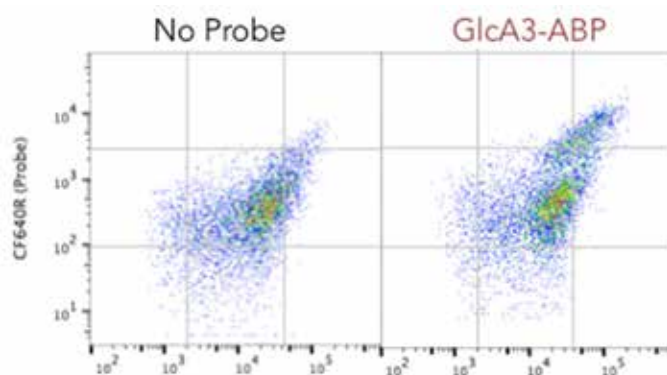
This project is addressing key gaps in our understanding of how the composition and function of mammalian microbial communities (microbiomes) are impacted by exposures to environmental agents and how these changes impact host susceptibility to the agents.

The broad diversity of microbes residing in the body greatly expands the synthetic and metabolic capacities of the human genome. The microbiome produces essential amino acids and vitamins, metabolizes otherwise indigestible dietary compounds, and shapes patterns of host immunity and tolerance. Emerging studies also show that the microbes within the intestinal tract can mediate the chemical transformation of xenobiotics such as pharmaceuticals and environmental contaminants associated with energy production. The impact that these environmental exposures have on the microbiome, and in turn, how the perturbed microbiome influences host susceptibility to environmental agents, is poorly understood. Work in this area is complicated by the large number of microbial species and enzymes capable of performing certain metabolic functions. A better understanding of these functions will be critical to the development of microbiome-based therapeutic strategies. In order to gain this understanding, innovations in approaches to identify the specific proteins associated with a given biochemical activity are needed.

This work aims to go beyond current metagenomic strategies to gain a mechanistic understanding of the specific organisms and proteins involved in a given microbiome activity. To achieve this goal, we are employing activity-based probes (ABPs) and proteomics profiling, coupled with new computational approaches for prediction of microbiome functions. This will allow us to address two interrelated aims: 1) determining the impact of environmental exposures (e.g., chemicals, diet) on the composition and diversity of gut microbiomes using mouse intestinal microbiomes as a model, and 2) developing and applying activity-based protein profiling (ABPP) methods to enable identification of proteins that have metabolic activity toward foreign chemicals (xenobiotics) and the microbes that express these activities.

Activity-based cell sorting of the gut microbiome. Within the past year of work, we have developed a pipeline for isolating, probing, and identifying microbes based on their enzymatic activity. To demonstrate the power of this approach, we have developed a probe to identify microbes that produce enzymes capable of reversing glucuronidation, a major step

in xenobiotic metabolism. Glucuronidation, or the addition of a glucuronic acid to a molecule, occurs in the liver to process a large number of drugs ranging from chemotherapeutics to antivirals for excretion. By removing the glucuronic acid, gut microbes can re-activate the compound, leading to increased side effects and toxicity. We have been able to sort out such microbes and are in the process of using DNA sequencing to identify them. This will be the first instance of using a chemical probe to isolate and identify microbes based on a specific metabolic activity.



Example flow cytometry analysis of mouse large intestine microbiome after labeling with either no probe (control) or a glucuronidase ABP.

Activity-based protein profiling of microbiome metabolic activities. To enable identification of specific metabolic activities, proteins, and microbial community members that express these, we are developing ABPs that target enzyme active-site chemistries for common xenobiotic metabolism activities. Our initial focus has been on the development, optimization, and testing of ABPs that target monooxygenases and glucuronidases that represent major pathways of phase I and phase II metabolism of environmental chemicals. Design and synthesis of first generation ABPs has been completed, and we successfully demonstrated the ability of the ABPs to label (bind) proteins selectively within the large and small intestines of untreated mice, providing a first-time demonstration of *in vivo* labeling of specific microbiome enzymatic activities. Through the use of click-chemistry, we demonstrated that fluorophores can be conjugated to the probes after binding to target *in vivo* and used to sort microbes based on their targeted metabolic activity by flow cytometry. Initial 16-sequence analyses from ABP-sorted samples have also confirmed ABP binding to specific microbes within the intestine. Thus, our early results provide proof-of-principle evidence for a new technological strategy that will enable rapid identification and isolation of specific microbial components (proteins, organisms) based on targeted metabolic activities.

Microbiome Models Across Scales – From Metabolism to Succession: A Framework for Modeling, Simulation, and Theory Development for Microbial Ecology

William R. Cannon

We are developing new computational and theoretical methods that are more predictive than the current state of the science to allow the understanding of adaptation, acclimatization, and collapse of microbial communities in a changing environment.

This project is modeling the dynamics of microbes in complex environments, soils, and the gut to understand how a changing environment affects the microbes residing in it and how these microbes feed back to and shape the environment.

As reported last year, we developed a hybrid approach to functional guild modeling that does not require a fixed yield to relate the energy supplying process to the growth process. This was done by splitting the summary reaction that summarizes metabolism into a catabolic reaction used for energy production and auxillary functions and an anabolic pathway used for cell growth. However, to implement this approach means that we have to be able to quantify the energy needed strictly for cell growth. In other words, we need to know how much work, in kilojoules per mole, is needed to make a cell.

The traditional approach to estimating the work required to make a cell is to inventory all of the parts of a cell—the metabolites, the proteins, the nucleic acids, and the fatty acids—and to tally, roughly, the energy required to put them into their final forms. Estimates using this approach vary widely and are, at best, educated guesses.

During FY 2016, we realized that there is a much more direct and rigorous way to determine how much work is required to produce a cell. The method takes advantage of specific chemostat experimental designs in which the nutrient concentrations are strictly controlled; the product concentrations, including cell biomass, are precisely measured; and the growth rate is increased at a fractional rate, such that linear growth is maintained.

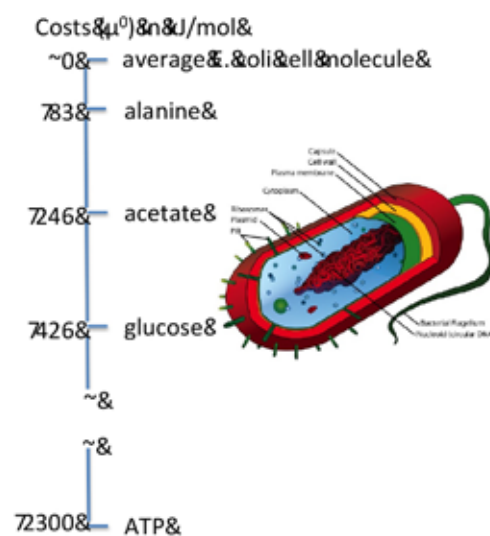
We then developed a method that tests for a linear free-energy relationship between the nutrients and products (including cell counts). Once the linear free-energy relationship is found, the standard chemical potential of the cell can be accurately estimated. The standard chemical potential can then be related to the work required to produce a cell when the cell birth rate matches the cell death rate. The value inherently includes contributions from all individual cell

components, from metabolites and proteins to nucleic acids and fatty acids. The estimated value of the standard chemical potential of an average metabolite in a cell is much higher than that for individual metabolites, which are measured in dilute aqueous solution. This high value reflects the significant amount of work required to organize even a bacterial cell.

This value is directly related to the concept of cell maintenance energy, which is the minimal work to maintain a cell. The value determined for bacteria growing in minimal media is much more favorable than previously thought and predicts that some bacteria can be maintained under mild non-equilibrium conditions. This value also implies that life may not be as improbable as we had previously anticipated. The creation of a primordial cell from an abiotic environment may be inevitable if the environmental conditions are right.

In FY 2017, we will be working to create the functional guild models of a microbial community with the gut environmental model that includes mucin. Mucin is the network of complex carbohydrates in the gut (excreted by goblet cells in the epithelium) on which microbes reside. Our long-term goal is to model radiation effects on the gut microbiome and how radiation impacts the host.

The scale of chemical potentials found for common metabolites in the cell, as well as the chemical potential of an average metabolite in a bacterial cell. The chemical potentials for metabolites are determined in dilute, aqueous



solution, while the average metabolite in a cell is in a highly organized environment. It takes a significant amount of work to organize this environment, which is reflected in the high value of the chemical potential of the average metabolite shown at the top of the scale. However, this rigorously estimated value from experimental data is significantly lower than previous estimates.

Microbiome Responses to Hydrologic Regime Shifts and Subsequent Alteration to Ecosystem Function

Vanessa L. Bailey

The overarching goal of this project is to determine how changing redox conditions (i.e., hydrologic change) impact the coupling of soil biogeochemical cycles and whether there are broad rules that transfer between ecosystems. Understanding these processes in soils under changing hydrologic regimes is critical to improving process-rich models of climate change effects on the land-climate system.

Soil microbiomes strongly influence the functioning of terrestrial ecosystems, from the pore-scale to the global-scale, by transforming materials in the environment. The availability of water strongly influences the rate and nature of microbiome-driven material transformations. Soil water content is a dynamic variable influenced by shifting hydrologic regimes due to natural variation (e.g., seasonal and weather-driven precipitation) and human-influences (e.g., land-use and climate change). Determining how soil water content dynamics—and changes in those dynamics—impact soil microbiome composition and function is fundamental to DOE's Biological and Environmental Research missions related to global carbon and water cycles. This project seeks to discern the sensitivity of soil microbiome molecular processes to altered hydrologic regimes.

Filling this knowledge gap is critical, as it currently limits our ability to robustly predict changes in ecosystem function as a consequence of altered hydrologic regimes. The proposed project aims to fill this knowledge gap through hypothesis-driven experiments coupled to multi-omic characterization and stable isotope probing of key metabolic pathways. Changes in the mean and temporal variability of soil moisture will alter the balance among aerobic and anaerobic metabolic pathways, but underlying dynamics of soil microbiome functional potential are currently unknown.

The research focuses on a tidally influenced wetland near the mouth of the Columbia River in Washington State. Climate change is altering the hydrologic regime in this and similar watersheds, such that historically snowmelt-driven moisture inputs are being replaced by rain events, and the timing of snowmelt and precipitation is changing. Aquatic-terrestrial transition zones like wetlands will experience the immediate

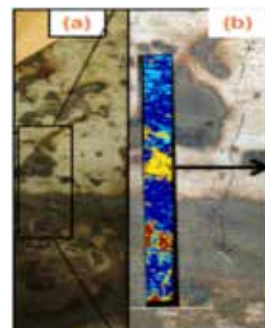
consequence of changes in the magnitude and dynamics of hydrologic flow. Coastal wetlands are particularly susceptible to storm events, salt-water intrusion, and increasing water levels. Pulsing hydrology can activate and deactivate microbial populations that quickly alter the nature and rate of biogeochemical transformations; extended saturation conditions can drive methane production, whereas drying will expose soil organic matter to rapid microbial decomposition. As fluctuating redox conditions alter terminal electron acceptor availability and recovery strengths in soil, a disproportionate release of carbon dioxide can also stem from alternative anaerobic degradation processes like sulphate and iron reduction, in addition to methanogenesis.

We hypothesized that extreme moisture regimes will select for microbial taxa with life-history strategies adapted to desiccation or moisture stress. We conducted a microcosm study subjecting soils to drying (oxic) and saturation (anoxic) for an extended period (75 days) at 21°C to 1) interrogate changes in the microbial community composition in the light of ecological theory, and 2) to elucidate the relationship between community response and geochemical signatures.

We also conducted analyses (bacterial composition and chemical profile) on samples of varying magnitude to capture the distributed microsites with distinctive functions within the soil core. A significant number of rare bacteria were uniquely revealed in smaller-size samples that may represent higher diversity in the soil, compared to those revealed by bulk samples. This included a greater relative abundance of specialized members such as sulfate reducers.

The probable association of these kinds of organisms with specialized microenvironments within the soil matrix is supported by silver-foil-based spatial assays for sulfate reduction that were conducted on the same soils. It is likely that unique microbes and processes that are heterogeneously distributed through soil profiles exert a greater influence on the biogeochemical cycles in a soil than are suggested by traditional bulk-volume analyses.

(a) Silver foil incubated in a wetland core. Dark areas record sites of sulfate reduction. The colored rectangle in (b) shows an area that was mapped using C60 FTICR-SIMS with a spatial resolution of 40 μm . Like colors (maroon, blue, and yellow) indicate pixels whose mass spectrum are similar using a hierarchical cluster analysis.



Multi-Scale Processes Controlling Spatial Variation in Greenhouse Gas Emissions in a Subarctic Watershed

James C. Stegen

This project combines field measurements with simulation modeling to deepen understanding of and reduce uncertainty in predicted greenhouse gas emissions across permafrost to non-permafrost transition zones. This is vital, as increased greenhouse gas emissions due to permafrost thaw could produce positive feedbacks that accelerate the rate of climate change, yet there is a high-degree of uncertainty in model predictions, because fundamental processes are poorly represented in climate models.

Microbial communities play a central role in the functioning of natural ecosystems by heavily influencing biogeochemical cycles. A major scientific challenge is to use knowledge of those influences to improve the ability of Earth System Models to robustly predict future climate change by incorporating feedbacks between environmental change and biogeochemical rates. Understanding how shifts in the environment are tied to shifts in biogeochemical rates via changes in microbial communities is particularly relevant in high latitude terrestrial systems underlain by permafrost due to vast carbon stocks currently stored within thawing permafrost.

The primary research objective of this study is to gain new knowledge of the factors that govern observed patterns in the rates of greenhouse gas (CO₂) emissions associated with permafrost to non-permafrost transition zones. The project further aims to use that knowledge to improve representation of soil microbiology in the Community Land Model (CLM). The strategic goal of the project is to better position PNNL for securing new DOE-BER programmatic funds.

To pursue these objectives, field studies are being used to generate data needed to first test CLM predictions and, in turn, provide hypotheses for how the model may be improved through inclusion of additional biotic (e.g., microbial community composition) and abiotic (e.g., organic carbon composition) features. The field site, near Fairbanks, AK, is characterized by spatial transitions in the presence/absence

of permafrost and in active layer depth (the depth of soil overlaying permafrost). Across these spatial transitions, greenhouse gas emission rates are being directly measured and biotic/abiotic features of the associated soil environment are being characterized.

From an initial sampling trip to the Alaska field site in 2013, CO₂ emissions were characterized across spatial gradients in active layer depth (ALD). Results show a strong relationship between ALD and CO₂ emission rates, but only across the lowest elevation sites with the thinnest ALDs. This result points to a highly non-linear influence of ALD on CO₂ emissions at the landscape scale. Multi-scale field sampling designs were implemented in the 2014, 2015, and 2016 field seasons to better characterize this non-linear relationship, understand how other environmental features contribute to the non-linear behavior, and to evaluate the degree to which primary drivers of CO₂ emissions are maintained across spatial scales.

To go beyond the influence of ALD, soil cores were taken across the spatial gradients in active layer depth. Soil core material from 2013 has been characterized in terms of numerous biotic and abiotic characteristics, including microbial community composition and organic carbon composition. These data resulted in a peer-reviewed publication that aims to overturn existing approaches for characterizing microbial diversity and linking the resulting measurements to ecosystem properties. Soil cores from 2014 sampling have been processed using similar characterization methods; a portion of characterization was in collaboration with EMSL and an additional portion in collaboration with a Science Undergraduate Laboratory Internship student. Coupling belowground data to CO₂ flux from 2014 samples revealed that the relationship between active layer depth and CO₂ flux is indirect. The best predictor of flux rates—and presumably the variable that most directly indicates the control point over CO₂ flux—was a proxy for the degree to which soil C has been microbially processed. CO₂ flux was also coupled with aboveground variables, and tree-stand basal area showed the strongest relationship with CO₂ flux, though this relationship was scale-dependent.

In addition, CLM has been set up, and simulations have been run for a variety of conditions informed by field observations. For example, we observed that a key plant functional trait (specific leaf area) varies significantly with ALD. Simulations are being run that manipulate this functional trait to evaluate the ecosystem-scale implications. A manuscript describing these field observations is currently in review. The next modeling step is to run CLM across multiple spatial scales across the field system. This approach takes CLM—a global-scale model often run on 10km² grid cells—down to the scale of point field measurements, thereby reaching across an extreme disparity in scale. To support this integration, we quantified spatiotemporal variation in CO₂ emissions—using

the 2015 and 2016 field seasons to sample at a larger spatial scale relative to 2014—and net primary production and ongoing efforts (associated with soil core processing) are characterizing vertical profiles of numerous key variables such as soil moisture, pH, texture, C content/composition, and microbial biomass, composition, and potential enzyme activities. These data will enable a multi-scale comparison between CLM predictions and empirical observations.

Results have been presented at national and international meetings, formed the basis of two invited seminars, and have generated two papers and two additional manuscripts.

Permafrost Microbiome Responses to Hydrologic Perturbation and Subsequent Alteration to Ecosystem Function

James C. Stegen

This project uses laboratory incubations and molecular characterization of field-collected permafrost to deepen understanding of how permafrost microbiomes respond to hydrologic perturbations and connects these responses with shifts in greenhouse gas emissions. This is vital, as increased greenhouse gas emissions due to permafrost thaw could produce positive feedbacks that accelerate the rate of climate change, yet there is a high-degree of uncertainty in model predictions, because fundamental processes are poorly understood.

Microbial communities play a central role in the functioning of natural ecosystems by heavily influencing biogeochemical cycles. A major scientific challenge is to use knowledge of those influences to improve the predictive ability of microbially explicit models by incorporating feedbacks between environmental change, microbiome responses, and biogeochemical rates. Understanding how shifts in the environment are tied to shifts in biogeochemical rates via changes in microbial communities is particularly relevant in permafrost systems due to vast carbon stocks currently stored within thawing permafrost.

The primary research objective of this study is to reveal processes governing the response of permafrost microbiomes to hydrologic perturbation. The project further aims to provide that knowledge and associated data to modeling efforts within the Microbiomes in Transition (MinT) initiative. These modeling efforts aim to build a predictive modeling framework that explicitly represents key members of soil microbiomes and the redox-associated processes they drive and feedback with.

Field-collected permafrost is being characterized using advanced molecular tools (multi-omics) and incubated in the laboratory under different redox and hydrologic conditions.

In addition, permafrost is being thawed *in situ* across a naturally occurring hydrologic gradient. Laboratory experiments are specifically designed to link permafrost microbiomes and associated biogeochemical processes to shifts in soil moisture conditions.

Recommendations from external advisors to conduct an initial scoping experiment were followed and this experiment provided key insights into microbiome response-to-thaw and other information (e.g., variability across replicates and the time-scale of microbiome responses) that is being used to design follow-on experiments.

The initial experiment did not attempt to manipulate moisture, but instead thawed permafrost under different redox conditions (either aerobic or anaerobic conditions). The reason was to mimic redox conditions that might arise under different moisture conditions; drier soil generally leads to aerobic conditions, while saturated soil generally leads to anaerobic conditions.

Permafrost cores were incubated across a time series that allowed biogenic greenhouse gases to be repeatedly sampled. Replicate cores were also destructively harvested at multiple time points. The results showed that redox conditions had a strong influence over microbiome response-to-thaw and biogenic gas emissions. In particular, under aerobic conditions, the microbiome became dominated by one species: *Rhodoferrax ferrireducens*. This organism can grow across a broad range of conditions and likely contributes significantly to system-scale biogeochemical function. One important aspect is that it can couple fermentation products to the reduction of Fe(III). This suggests that it has the potential to inhibit production of CH₄—a process that also relies on fermentation products. In turn, conditions that favor *R. ferrireducens* likely shift biogenic gas production toward CO₂ and away from CH₄. This provides new insight into factors governing the balance between CO₂ and CH₄ following permafrost thaw, which is a critical uncertainty due to CH₄ being a far more potent greenhouse gas than CO₂.

In addition to the microbiome itself, we observed significant shifts in metabolite profiles following permafrost thaw. The data revealed a significant increase in protein-like metabolites associated with phosphorylation molecular transformations under conditions where *R. ferrireducens* became dominant. This suggests that *R. ferrireducens* is likely a dominant driver of biogeochemical cycling and microbe-microbe interactions in thawing permafrost, at least under conditions that facilitate its growth. We have hypothesized that conditions in which there is significant spatial or temporal variation in redox potential facilitate *R. ferrireducens* due to its ability to grow both aerobically and anaerobically.

In addition, recent field efforts provided soil for vertical profiling of both seasonally thawed soil and its associated permafrost. One motivation for this vertical profiling is to determine what field conditions facilitate *R. ferrireducens* and to link variation in its activity to the activity of other microbial groups, such as fermenters and methanogens.

Results have been presented at national and international meetings, are generating two manuscripts, and provided the foundation for a FY 2017 DOE Early Career proposal.

PhenoAccess: Physiological Phenotyping of *Brachypodium* Accessions

Amir H. Ahkami

We are developing experimental capabilities to obtain a better understanding of available phenotypic diversity of model grasses for bioenergy crops in a fluctuating environment like drought, heat wave, and elevated CO₂ levels.

The main goal of this work is phenotypic analysis of the genetic variability in model grasses by systematic quantification of phenotypic traits. This will provide a fundamental basis for development of a Virtual Plant-Atmosphere-Soil System (vPASS) model, which will enable a better predictive understanding of how the molecular, cellular, and organismal traits of plants are shaped as a function of plant genotype and its ecosystem environment.

The focus will be on the non-invasive monitoring of the most important structural and physiological traits at the organ and whole organism level, as well as physiological screening at the tissue and cellular level. PhenoAccess aims to allow physiological characterization of natural accessions, inbred lines, and selected mutants of the model grass *Brachypodium* under different climate scenarios. It will accomplish this by extracting an extensive list of phenotypic traits in a high-throughput (HTP) and non-invasive way from EcoSimulators, which are dynamic plant growth chambers equipped with different automated imaging systems, including visual RGB (Red-Green-Blue), thermography, and fluorescence cameras.

In collaborating with other projects, the plant phenotypes associated with a cellular structure, plant gene, or a biochemical marker will be identified in a fluctuating environment (e.g., drought, heat wave, elevated CO₂ levels), and this will eventually allow us to take steps towards bridging the genotype-environment-phenotype gap.

Recently, several HTP phenotyping platforms that include controlled-environment growth facilities equipped with different imaging systems have been developed to measure plant growth, development, and responses to the environment. Nevertheless, we are still far from understanding the complex interactions between plants and their dynamic environment. In light of high-performance computing, the combination of high-resolution internal with HTP external phenotyping will provide new opportunities to obtain detailed and complementary information on plant physiology when interacting with its environment.

General knowledge gaps to be considered in this project include 1) how do we dissect the complex genetic architecture of biomass development in plants, and 2) how are changes in complex and quantitative agronomic traits (as external phenotypes) linked to cellular and molecular alterations (as internal phenotypes) in developmental responses to environmental stimuli.

In particular, the central research hypotheses include 1) manual phenotypic measurements correlate with image derived phenotypic data, 2) the rate of above- and belowground biomass of each individual *Brachypodium* genotype is variable under drought stress, and 3) this phenotypic plasticity is predictable and can be linked to cellular structures (sugar transporters and/or aquaporins) and molecular profiles (stress-responsive transcription factors and/or cross-talk between phytohormones). PhenoAccess will apply HTP non-invasive, image-based plant phenotyping approaches to test these hypotheses.

We expect to see a large phenotypic variation in *Brachypodium* natural accessions and selected mutants in response to environmental changes, which potentially, in collaboration with other iPASS projects, can be used 1) as essential input for developing a computational model—vPASS—describing how the plant genotype is linked to the ecosystem phenotype in a larger context, 2) to unravel key genes and metabolites (biomarkers) controlling complex traits like drought tolerance or responses to elevated atmospheric CO₂ levels, and 3) to describe plant, stress-related, cellular physiology responses, which forms a key interface between genotype and phenotype.

In FY 2016, we mainly focused on providing the basic facilities and infrastructures needed for the EMSL Plant Ecosystems Lab. To this end, plant growth chambers were purchased and set up, followed by growing sample *Brachypodium* plants under controlled conditions.

In FY 2017, we will continue building a prototype for plant phenotyping by focusing on image-based approaches using visual and thermography cameras, optimizing data analysis, and correlating with manually obtained data. We will also develop EcoSimulators as novel capabilities in plant phenotyping at PNNL/EMSL for HTP and non-invasive phenotyping. We will collaborate with other projects to provide a framework for linking organismal, cellular, and molecular physiological traits in plants.

Probing Complex Microbiomes Using Mass Spectrometry and Sequencing Capabilities to Understand How Microbiomes are Influenced by their Environment

Stephen J. Callister

This project is developing next-generation sequencing and mass spectrometry capabilities to address key knowledge gaps in our functional understanding of how complex microbiomes influence and are influenced by their surrounding environment.

Microbiomes, defined as the totality of microorganisms and their collective genetic material present in a specific environment, are responsible for key processes such as carbon and nutrient cycling. Understanding the mechanistic role of microbiomes in these processes, so that predictive models concerning these nutrient cycles can be improved, requires a molecular look at their function. This molecular investigation can only be achieved through the development of sophisticated sequencing and mass spectrometry capabilities.

Unfortunately, more than 90% of microbial community members are uncultivated, with functions and identities unknown. The high diversity and lack of representative assembled genomic databases is an impediment to molecular analysis using high-throughput sequencing platforms and other “omics” technologies.

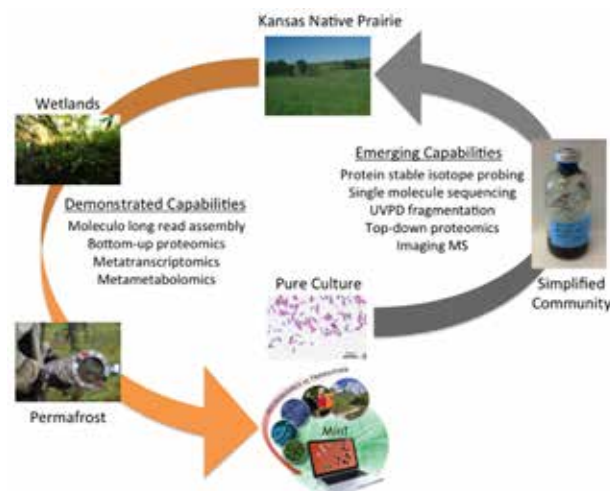
To address this challenge, we are developing novel metagenomics and metaproteomics workflows, combining both demonstrated and emerging bioinformatics and empirical capabilities. These workflows are being systematically evaluated—first on simplified microbiomes, and then on complex model microbiomes, such as the soil microbiome found in the Konza Prairie Biological Station (KPBS) Long-Term Ecological Research (LTER) site. After initial evaluation, these enhanced capability workflows will be applied to microbiomes found within boreal permafrost soils and Pacific Northwest wetland soils.

Progress was made during FY 2016 towards development and enhancement of several capabilities designed to 1) increase the amount of DNA sequence information about microbiomes, which provides a measure of potential microbiome function; 2) measure actual/realized function; and 3) characterize the diversity of populations within the microbiome. Two of these capabilities are described below.

Increasing the amount of DNA sequence information about the microbiome. The assembly of small pieces of DNA into larger pieces, and the functional annotation of these assembled pieces, is a major hurdle in the measurement of the functional potential of the complex microbiome. During FY 2016, this project developed a DNA sequence assembly and annotation software tool that provides bioinformatics support to a novel molecular technique (termed Moleculo) for generating larger DNA pieces from the complex microbiome. This tool was demonstrated on extracted DNA sequenced for the Konza soil microbiome. This tool yielded more than 10,000 assembled pieces of DNA (contigs) over 10 kilobase pairs, a significantly larger number than other tools used to perform a similar task. From these assembled contigs, 354,863 genes were identified, with 32% of these classified as having known enzymatic function. These results were recently published in the peer-reviewed scientific journal *mSystems*.

Measurement of actual/realized function. While the number of genes identified above is a significant advancement in the measurement of potential function, a greater challenge is measurement of these genes/enzymes as proteins (realized function) using mass spectrometry. The number of mass spectrometry measured microbiome proteins is often a small percentage of what is predicted. Two possibilities for this are 1) not all proteins are produced by the microbiome at any one time, and 2) the redundancy of amino sequences across the protein domain can make it difficult for mass spectrometry to distinguish one protein from another.

To evaluate these possibilities, use of ^{18}O (in the form of H_2^{18}O) is being investigated as a way to separate the active functionally relevant populations from those that are dormant (i.e., not expressing genes as proteins). The use of H_2^{18}O (required for all active microorganisms) for targeting the active metagenome has been demonstrated, and the capability is being adopted by PNNL as part of this project. Novel, however, is the measurement of *in vivo* incorporation of ^{18}O within amino acid sequences making up proteins. Initial pilot studies, using pure culture soil microorganisms, have been successfully undertaken, revealing that ^{18}O incorporation into proteins is measureable using mass spectrometry, and preliminary results suggest that a select type of amino acid incorporates this stable isotope (allowing for predictive modeling). This result is being verified using a simplified community of four populations that are experiencing perturbation (i.e., culture being exposed to light after growing in the dark) in order to separate the perturbation associated functional proteome from the non-perturbed proteome. If successful, a high-level publication has been planned for FY 2017, and the stable isotope method will proceed to the application phase using the Konza soil microbiome.



Metagenomics and metaproteomics workflows are being developed on demonstrated and emerging capabilities. These workflows are being applied to complex soil microbiomes found within Kansas native prairie soil, Pacific Northwest wetlands soil, and boreal permafrost soil. Overall, this project is supporting the larger objective of understanding how microbiomes transition in response to perturbation.

RhizoControl: Does the Rhizospheric Microbiome Influence the Plant Metabotype? A Plant Gnotobiotics Approach

Christer Jansson

*This project is building a knowledge base to understand how the rhizospheric microbiomes in the grass *Brachypodium distachyon* control the synthesis of volatile organic compounds and other metabolites, as well as how these interactions affect drought tolerance in the plant.*

Understanding plant-microbe interactions and their impact on plant metabolotypes is highly relevant to DOE's missions in sustainable biofuel production, understanding carbon cycling, and mitigating negative effects of climate change. In addition, the knowledge is of importance to a new EMSL focus on plant phenotyping, where the plant microbiota and their interactions with the plant host are of central interest.

Many studies in the last decades have shown that plant-microbe interactions are key to understanding plant growth and health, as well as sustainable crop production. There is growing evidence that a significant number of plant metabolites are, in fact, produced by associated microbes or through interaction with their host. However, a thorough investigation of plant-microbe interactions in plant metabolite profiling is lacking, which indicates a knowledge gap that critically limits the ability to improve plants for renewable energy and chemicals production.

The objectives of this project are three-fold: 1) to establish the workflow and infrastructure for plant gnotobiotics work; 2) to apply gnotobiotic approach to gain further understand-

ing of rhizospheric microbiome influences and controls of plant metabolites (i.e., its metabotype) using the model grass *Brachypodium distachyon*; 3) establish a model rhizospheric community to provide the rhizospheric microbiome for downstream and future experiments in model grasses (e.g., *Brachypodium distachyon*, *Panicum virgatum*, *Miscanthus*).

To date, this project has successfully established working protocols and workflows for future gnotobiotic plant work at PNNL. Metabolite profiling clearly showed that the plant metabolotypes differed between Microbiome-plus and Microbiome-minus plants ($P < 0.0005$).

Additionally, a principal component analysis was performed to the same metabolomic dataset. Case plot of the principal component analysis demonstrated that both groups of plants (Microbiome-plus and Microbiome-minus) are separated in the multidimensional space, showing different metabolome composition.

This project also sought to completely characterize the rhizosphere microbial communities within a specific set of apple orchards and successfully accomplished this goal. Data integration of the multiomic data is ongoing. The final data integration should lead to submitting high-quality and high-impact manuscripts for publication in FY 2017.



An example of the *Brachypodium distachyon* this project is studying.

Rhizosphere Underground: Unraveling the Role of Microbes in Stabilizing Carbon Pools in Soils

Alice C. Dohnalkova

This project's goal is developing a multi-capability platform for correlative imaging and analyses, in order to investigate microbial and mineral association and interactions in the rhizosphere. This multi-modal, multi-scale imaging and analytical platform will be applicable to a variety of other environmental sciences systems.

Microorganisms are fundamental to biogeochemical processes, as microbial metabolism significantly contributes to the regulation of global carbon and nitrogen cycling. Rhizosphere, the zone immediately surrounding the plant roots, plays an important role in production and stabilization of soil organic matter, with considerably more abundant microbial population than in the adjacent bulk soil environment. However, the impact of the community of soil microbes within the rhizosphere has not been examined in detail due to the difficulty of tracking microscale processes in natural microbial communities in complex soil habitats.

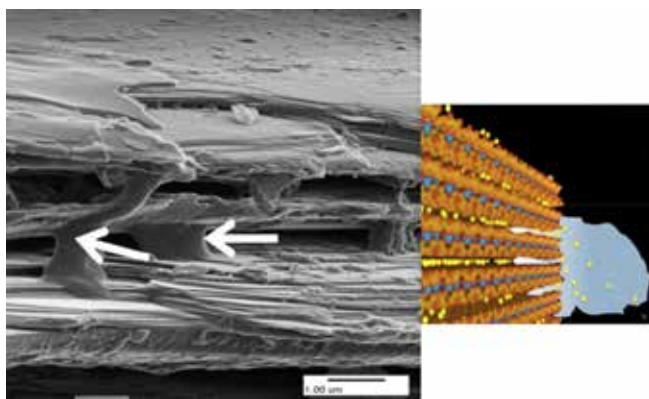
This platform will be used on a model ecosystem to identify interactions of individual microcosm components: roots, microbial communities, and soil minerals. Special emphasis is given to formation of microbial extracellular polymeric substances that may significantly contribute to persistent C pools. In particular, we will examine the current hypothesis that preferential association with mineral surfaces and soil pores is an important mechanism of carbon stabilization. Our tools will include high-resolution electron microscopy (EM) imaging and EM-coupled minerals analyses, X-ray diffraction (XRD), and Fourier transform ion cyclotron resonance (FTICR) for identifying the newly formed organic C compounds.

Seedlings of *Pinus resinosa*, a widespread North American pine forest ecosystem, was cultivated under experimental conditions in well-defined soil mineral mix (quartz, biotite, and anorthite) with bacterial inoculum to develop biofilms-minerals associations. The mesocosms were destructively sampled at time points and investigated through the suite of imaging and analyses for extracted labile and persistent soil organic matter (SOM). Datasets were correlatively evaluated, and a mechanistic model will be created from the integrated datasets.

We designed a multi-modal, multi-scale imaging and analytical system to provide an integrated workflow with seamless consecutive steps in multi-scale investigations, from millimeter-scale to atomic level in imaging, including EMSL's state-of-the-art, high-resolution EM suite for microbial-mineral interactions visualization and analyses, as well as micro-XRD for mineral analysis. We are coupling imaging with consecutive extractions of root microbial system for FTIR-MS, to identify carbon species involved in the process.

FY 2015 accomplishments included applying this platform to address the following specific hypotheses in the *Pinus* sp. ecosystem: 1) the FTICR showed spatial differences in the rhizosphere soil systems; 2) we observed a pattern in preferential bacterial attachment and coverage to minerals; 3) Micro-XRD revealed differences in minerals with time, suggesting microbially induced minerals weathering; 4) we extracted DNA to identify the key microbial players in the rhizosphere processes of mineral weathering and soil formation.

In FY 2016, we correlated the initial lab microcosm data with a system in the field experiment setup, to find relatedness of the features such as microbial mineral weathering, SOM-specific association with minerals, microbes phyla identification by 16S and ITS2 sequencing, and identification of the SOM components by FTICR.



Scanning electron microscope image of a clay mineral with microbially produced amorphous organic matter. The illustration depicts how extracellular polymeric substance penetrates between the mineral sheets, enabling cation extraction during the mineral weathering.

Signatures of Environmental Perturbation – Microbial Community and Organic Matter Resilience

Vanessa L. Bailey

The overarching goal of this project is to demonstrate capabilities developed by the Signatures Discovery Initiative to identify one or more signatures that indicate impaired resilience of the soil carbon biogeochemical system following a system-wide perturbation such as simulated climate change.

Transformation of soil carbon by microbes leads to the release of greenhouse gases into the atmosphere, which directly impact global climate by exerting a strong positive feedback effect on temperature (i.e., global warming). Currently, analytical tools do not assess the vulnerability of soil carbon reservoirs to changing climate, but it is likely that microbial transformations of soil carbon will increase with temperature, resulting in even greater gas emissions. As part of the terrestrial ecosystem carbon cycle, the soil microbial community deposits new soil carbon, as well as respiring existing soil carbon. By integrating chemical and molecular signatures of the soil system, these microbial community interactions with vulnerable soil carbon may be predicted with molecular resolution.

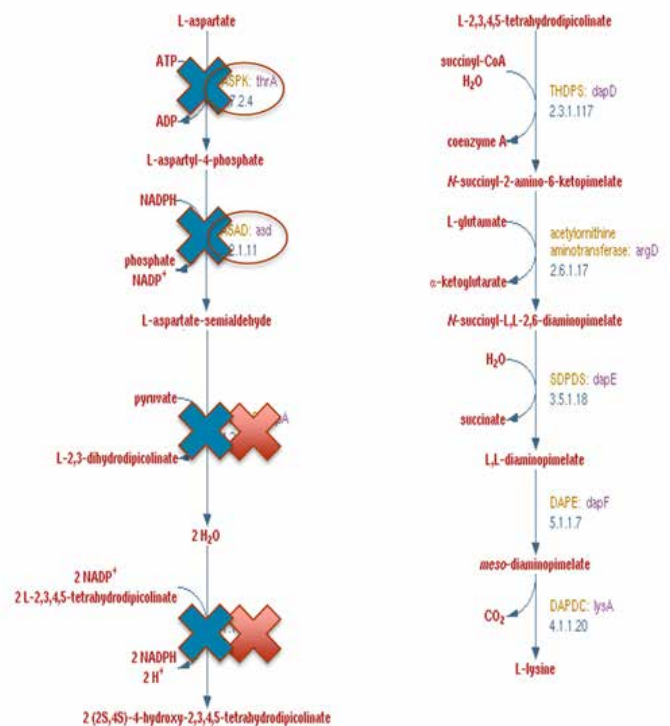
The focus on ecosystem stress and climate change is currently relevant, as researchers and policymakers strive to understand the consequences of climate change. Successful development of chemical/molecular profiles that link soil microbiology with soil carbon to ascertain ecosystem vulnerability and resilience would have great impact on assessments of soil ecosystems in response to global climate change. These efforts align directly with research interests of DOE, USDA, and the NSF.

This project aims to improve our understanding of soil biogeochemistry and ecology, as well as the relationships between carbon chemistry, microbial community structure and function, and ecosystem perturbations. We leveraged an existing long-term experiment at the Arid Lands Ecology Reserve in Richland, WA, in which soil cores were reciprocally transplanted between the cooler, moister upper slope position and the hotter, dryer lower slope position in 1994. This transplant was designed to mimic the stress of climate change with natural temperature and moisture gradients

provided by elevation. A previous study of these soils suggested that one of these core soils lost robust microbial community function as a result of this transplant.

In 2016, we focused on linking chemical and biological data sets to identify the biogeochemical processes that separate our soil treatments—specifically the native soils from the lower and upper sites, the disturbance control soils at each site, and the lower-to-upper and upper-to-lower site transplants.

The chemical profiles of the soil carbon were generated by Fourier Transform Ion Cyclotron Resonance (FTICR) mass spectrometry and yielded 96,000 unique features across all samples. Chemical transformations were inferred from the chemical data using Kendrick mass defect analyses, and these transformations were matched to chemical reactions and the enzymes that perform them using the KEGG database. The microbial functional potential was profiled using Illumina



A lysine biosynthetic pathway (KEGG Module 00016: Lysine biosynthesis, succinyl-DAP pathway, aspartate to lysine) showing enzymes that were not observed in the microbial functional profile (metagenome; red X), as well as those not observed in the chemical profile (FTICR; blue X).

HiSeq DNA sequencing and yielded 726 million sequence reads. These sequence reads were mapped to greater than 19,000 protein families, from which we inferred function based on the Enzyme Commission (EC) assignments for the protein families.

For both data sets, the EC assignments were used to identify specific biochemical pathways, and statistical analyses revealed fewer than 30 biochemical pathways differentiated the treatments in both the chemical and biological datasets. An intriguing observation was the significant decrease in lysine biosynthesis pathways in soils from the lower site,

compared to the upper site. Lysine metabolism has been linked to osmotic tolerance in microbial communities. Our integrated analyses of chemical and biological shotgun datasets identified seven of the nine enzymes required for a particular lysine biosynthetic pathway in the metagenome, and five of the nine steps of the synthetic pathway were detected in the chemical profile. From this integrated analysis, we infer that lysine synthesis was impaired in the lower site, and this may have resulted in a decreased ability of soils from this site to adapt to new conditions.

Spatially Resolved, Consortial Approaches Toward a Mechanistic Understanding of Soil Nitrogen Fate

Steve Lindemann

Through this research, we aim to generate a species- and spatially resolved model of microbial soil nitrogen (N) cycling that will enable identifying the human-controlled variables that affect microbial carbon- (C-) N cycling in lands under cellulosic biofuels feedstock production, as well as other agricultural ecosystems. This work will contribute towards developing methods to reduce N₂O emissions from globally increasing agricultural activities.

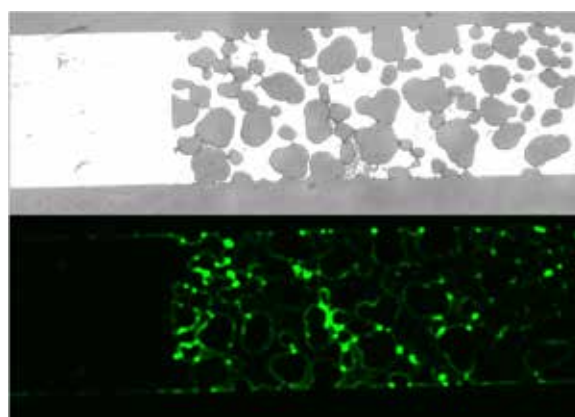
Soil mesocosm experiments have been traditionally used to examine environmental controls on microbial process rates, but these experiments are generally hampered by soil species richness, thereby making it hard to uncover mechanisms operating within microbial communities. In contrast, single-species models are more tractable but unable to describe higher-order, community-based effects operating between interacting species. To help fill this needed capability gap, we aim to generate consortial models of soil nitrogen cycling for species-resolved, genome-enabled experimentation and design a pore-scale microfluidic model to examine the impact of microbial behavior upon nitrogen immobilization.

Root bioreactors have been developed and initial colonization and biofilm formation experiments were successful. Inoculation, growth, and sampling methods were prototyped with *Pseudomonas*. Root exudates were acquired from collaborators and characterized using a Fourier transform ion cyclotron resonance (FTICR)-mass spectrometer (MS). Switchgrass rhizosphere soil was acquired from Texas biofuel field sites, including an alluvial silt loam and a black-land prairie soil.

Concurrent with the development of the consortial systems, we have developed a variably saturable microfluidic device with surface chemistries relevant to nitrogen immobilization at the soil pore scale. While silicon or glass micromodels are often used to study subsurface flow and transport processes, because of their ability to provide a well-defined micrometer-scale pore structure, the cation exchange capacity provided by the native silanol groups on their surfaces poorly represent that of native soil. This becomes limiting for understanding nitrogen cycling in the rhizosphere within pore-scale micromodels. To overcome this inherent limitation, we developed two generations of pore-scale micromodels that provide a

more accurate surface chemistry for studies involving soil-based microbes. Surface treatment involves introducing a slurry of clay particles (such as montmorillonite or kaolinite) into a microfluidic system. These devices are then dried to form a surface clay coating on the micromodel surfaces. For preliminary evaluation, a green-fluorescent-protein-expressing strain *Pseudomonas fluorescens* were cultured within the test devices containing a homogeneous pore structure. An increase in fluorescence over time demonstrated microbial adhesion and growth on the clay coated surfaces in these models. We have since transferred coating methodology to a variably saturable inhomogeneous micromodel, which will enable direct testing of the hypothesis that heterogeneities in wetting governs denitrification by controlling carbon flux.

Continued work on this project will include using root and soil bioreactors with switchgrass communities and exudate C inputs to test how C inputs from plants affect microbial nitrogen metabolism and the production of N₂O versus other nitrogenous species. We are using a series of fluorescent and isotope labeling experiments to trace N cycling through the constructed, spatially organized microbial systems and how this structure directs parallel C transformations with implications for greenhouse gas production. By manipulating nutrient (e.g., C, N, etc.) and culture conditions (e.g., pH, temp, desiccation, ionic strength, etc) we will investigate the impact of microbial community interactions on microbial metabolism and the repercussions for system-level biogeochemistry (export or dissolved or gaseous nutrients). This work has important implications for understanding how C-N metabolism affects greenhouse gas production and leaching of nitrate and dissolved organic C at terrestrial-aquatic interfaces.



Micromodels with clay surfaces (top). Fluorescent *Pseudomonas* grown for 32 hours prefer clay surfaces of soil micromodels (bottom).

Spatially Resolved Quantitative Gene Expression Analyses Applied to Transitioning Mouse Gut and Soil Microbiomes

Galya Orr

We will establish quantitative gene expression analysis approaches in individual and small clusters of bacterial cells, while preserving their spatial context within their native environments.

The function of individual organisms within a microbial community is governed by the local environment and interactions with neighboring organisms. In complex spatial organizations of communities that consist of hundreds or thousands of different species, individual cells of a given clonal population of organisms can experience vastly different local environments and are likely to express very different genes in response. The spatial relationships between individual organisms within the community are, therefore, key to understanding the role of the individual organism and, ultimately, to deciphering how the community functions as a whole.

Meta-transcriptomic analyses have gained important insights into processes that take place by the community as a whole and suggested functional roles for different species. However, this averaged-cell-population approach lumps the behavior of an entire population into a single average value, while ignoring the impact of the diverse local environments on the responses of individual organisms and the complex spatial relationships between them. Adding a spatial context to gene expression analyses will be vital to understanding fundamental rules by which complex communities function and evolve under changing environments.

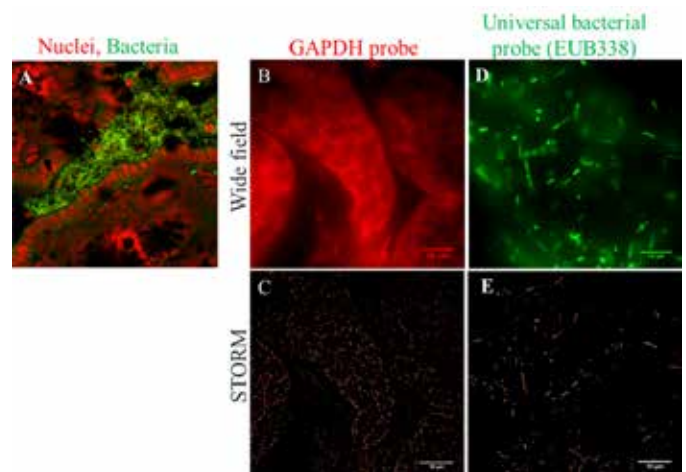
Recent advances in single cell analysis have enabled quantitative gene expression analyses in individual cells, while preserving their spatial context within the tissue. To date, these approaches have been demonstrated only in eukaryotic cells. The translation of these techniques to bacterial cells presents multiple challenges due to their small size and content, as well as their complex environments within microbiomes.

Building on our experience in single cell RNA-Seq in mammalian cells, we will apply methods and reagents developed specifically for small amounts of starting RNA to small clusters of bacterial cells, isolated by laser capture microdissection (LCM) from mouse gut cryo-sections. We will also apply quantitative fluorescence *in situ* hybridization (FISH) to enable multi-gene expression analysis in single cells, while preserving the spatial context of the individual cells. When applied using single molecule fluorescence imaging techniques, FISH enables the

quantification of single transcripts in intact cells with high accuracy. Further, by combinatorial barcoding of the FISH probes, it will be possible to significantly increase the number of genes that can be quantified within a cell simultaneously.

Our project is designed to establish, for the first time, two challenging techniques for quantitative multi-gene expression analysis in individual or small, spatially defined clusters of bacterial cells, while preserving the context of their local environments and spatial relationships within the microbiome. These include 1) quantitative FISH by super resolution fluorescence imaging (STORM/PALM) and combinatorial barcoding of the FISH probes for quantifying the expression of multiple genes in intact cells simultaneously, and 2) the isolation of small, spatially defined microbial clusters using high magnification LCM, followed by meta-transcriptome analysis using RNA-Seq. The techniques are being established in the mouse gut microbiome, integrating with current efforts utilizing activity-based probes and proteome analysis of the microbiome as it is perturbed by antibiotics.

Ultimately, this effort will identify fundamental rules and mechanisms by which perturbed microbiomes operate with details that cannot be achieved by averaged cell population measurements, which ignore the local environments and spatial organization among the community members.



A: Mouse gut cryo-section, showing the host intestinal epithelial cell nuclei (red) and the bacteria in the lumen (green). **B, C:** Cryo-section processed for FISH, targeting GAPDH mRNA molecules (red), imaged by wide-field microscopy (B) and by STORM (C). Each spot in the STORM image represents an mRNA copy. **D, E:** Cryo-section processed for FISH, targeting bacterial rRNA molecules (green) using the universal probe, imaged by wide-field microscopy (D) and by STORM (E). Unlike mRNA, the number of rRNA copies in the bacterial cell is extremely high and is targeted for establishing the technique.

Statistical Integration of Omics Data from Microbiomes

Joseph M. Brown

We will develop an infrastructure and computational tools designed to process, analyze, and visualize microbial community sequence data to address the challenge of computational analysis.

Microbes exist and function in communities. Studies of natural environmental or human-associated sites often reveal hundreds to thousands of microbial species co-existing (i.e., a microbiome). The interactions between microbes within such a microbiome are often complex, as is the impact the microbiome as a whole has on its environment. Deciphering microbial activities in their natural environments is a prerequisite to understanding their functional role in environmental processes. Research efforts to understand the vital roles these communities of microbes play in human health and environmental sustainability have thus turned to high-throughput technologies to measure the various biomolecules extracted from a sample (e.g., nucleic acids, protein, and metabolites) to infer the functional capability and emergent properties of the microbiome.

One of the most readily available data types for microbiome research is nucleic acid sequence data. It is feasible to sequence the DNA extracted from natural microbial communities consisting of many species. However, such DNA samples consist of a composite mixture of the genetic material of all of the community members (i.e., a metagenome), and inferring the functional capability or assigning taxonomy from complex mixtures of genetic material has proven challenging. Further, sequencing technologies are capable of generating approximately a terabyte of data in a week, requiring the need to develop novel analytical tools. Robust analysis of complex data from microbial communities in human hosts and in the environment will be key to understanding their vulnerability to change.

We have built a sequencing capability and data delivery process at PNNL around industry best practices. Data obtained from our instrument go through rigorous quality scrutiny, facilitated by an interactive and extensible dashboard that we have developed. Our automated protocol allows us to utilize institutional computing for archiving, processing, and data delivery to accelerate the pace of data distribution to researchers.



Screenshot of FQC dashboard. Displays several quality metrics, including mean read quality within interquartile ranges.

Sequencing fragments of an rRNA marker gene as a means of capturing community structure and dynamics is standard practice in microbial ecology. We developed a protocol to complement our sequencing capability that enables rapid quantification and classification of the marker gene amplicon data (16S rRNA, ITS, and 18S). An average run of the full protocol takes less than 30 minutes and includes common output file formats compatible with downstream analysis tasks.

We also developed a method of predicting functional attributes from raw metagenomic sequence data using hidden Markov models (HMMs) of protein families (i.e., functionally related proteins). Our method leverages the following public HMM libraries: Pfam, PhyloFacts, TIGRFAM, and FOAM, as well as high-performance computing, to process hundreds of millions of sequence reads and produce high-quality, functional assignments. Using data from well-characterized microbes, we were able to make functional assignments to each HMM library and calculate a positive predictive value of this approach for each dataset. The results in the included table show the consistently high positive predictive value of this approach.

Future work for this project includes greatly expanding our capabilities associated with processing metagenomic and metatranscriptomic data. This work will include reproducible workflows for quality control, assembly, and annotation, in addition to planned visual analytics tools to be applied to these data.

Understanding Cellular Communication and Controlling Directional Flow of Nutrients

Julia Laskin

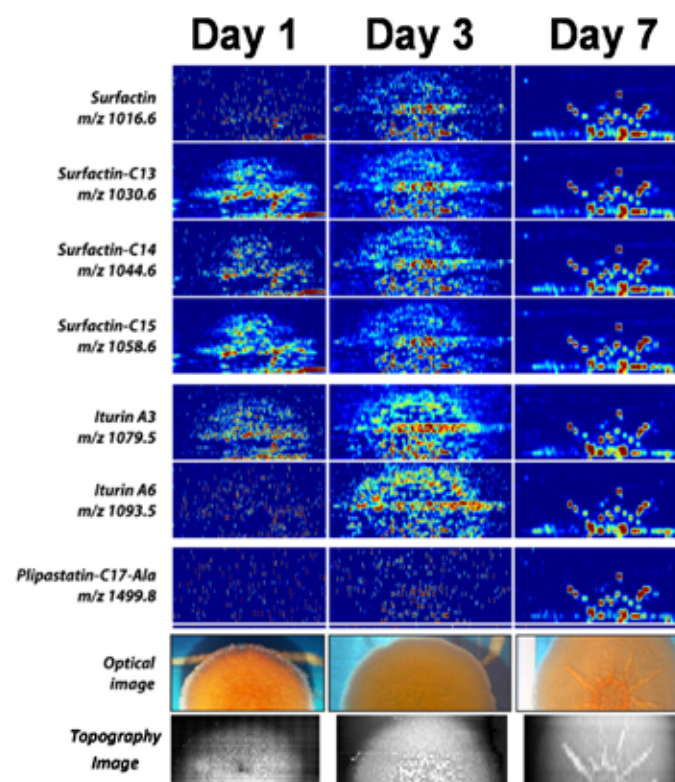
We have developed unique capabilities for molecular imaging of metabolites involved in cellular communication using mass spectrometry.

Understanding and controlling intra- and intercellular communication in diverse biological systems is one of the grand challenges in biology. New capabilities for quantitative imaging of chemical gradients in biological systems with high spatial resolution and high chemical specificity will facilitate understanding cellular communication. This project is focused on developing mass spectrometry imaging (MSI) approaches capable of measuring chemical gradients of hundreds of metabolites in biological samples of arbitrary topography. Chemical gradients are examined using the novel nanospray desorption electrospray ionization (nano-DESI) MSI capable of label-free detection of individual molecules released by cells. The advanced development of nano-DESI MSI conducted under this project enabled robust automated imaging of living microbial communities directly from nutrient agar plates, which had never been done before.

In FY 2016, we continued developing robust tools for imaging biological samples of arbitrary morphology. We created two unique approaches for constant-distance-mode nano-DESI MSI using either shear force or ion conductance measurement as a feedback. Both approaches were implemented by incorporating a nanopipette-based distance probe into the nano-DESI source. In a first approach, when the nano-DESI probe approaches a sample, several vibrational modes of a nanopipette are affected by the shear force with the sample. In a second approach, the ion conductance between two barrels of the nanopipette provides a feedback signal. Using these methods, we examined, for the first time, molecular signatures of living *Bacillus subtilis* colonies at different stages of growth on agar plates. The feedback system allowed us to image an approximately 800 μ m-tall colony, while maintaining a constant distance between the nano-DESI probe and colony. The constant-distance mode nano-DESI MSI enables imaging of many metabolites, including non-ribosomal peptides (e.g., surfactin, plipastatin, and iturin) on the surface of living bacterial colonies, ranging in diameter from 10 mm to 13 mm, with height variations up to 0.8 mm above the agar plate.

Furthermore, sample topography is recorded simultaneously with nano-DESI MSI. The resulting ion images allowed us to identify molecular signatures of both fully and partially developed biofilms. The new multimodal imaging approach developed in this study is ideally suited for correlating sample phenotype to its chemotype. A patent application and a manuscript describing this experimental approach have been submitted, and the results were presented at the annual American Society of Mass Spectrometry Meeting.

Although the second approach based on the ion conductance measurement is still in an early development stage, it has already shown great promise for coupling electrochemical microscopy and MSI. The combination of these techniques will be used in future studies for a detailed understanding of cellular communication in several model systems, including well-characterized microbial and yeast colonies, along with plant tissues and individual cells.



Molecular signatures of growing *Bacillus subtilis* colonies on agar plates (top) acquired simultaneously with topography images (bottom).

Using Modified Proteins for Forensic Deconvolution of Xenobiotic Dose Quantitation and Timing

Jordan N. Smith

A novel biomonitoring approach utilizing modified-protein fingerprints to reconstruct dose and timing of dose is being developed, and a multipronged technique to identify protein modifications caused by environmental chemical exposures was established. This biomonitoring approach could allow for dose reconstruction with greater precision and accuracy than current methods.

Humans are exposed to a barrage of chemicals from occupational settings, consumer products, accidental releases, and the environment. Many chemicals are known to cause or have been linked to adverse health effects. To better understand the potential risks associated with these exposures, efforts are underway to biomonitor targeted human populations for chemical exposures, as highlighted by the National Research Council of the National Academies report, *Exposure Science in the 21st Century: A Vision and a Strategy*.

Current state-of-the-art methods to predict how much of a chemical or when exposure occurred (i.e., dose reconstruction) require *a priori* knowledge of one of two factors: dose or timing of dose. Blood or urine samples are analyzed for biomarkers of exposure (the chemical of interest or a metabolite of that chemical). Computational models are used to reconstruct a dose that may have caused a measured biomarker concentration. Biomarker concentrations are highly dependent on time and dose; without some information about one of these factors, the most sophisticated dose reconstruction strategies are reduced to simply concluding that exposure occurred. For compounds that are rapidly cleared from the body, even robust *a priori* information may not be sufficient to reconstruct doses without a high level of uncertainty, since biomarkers can only provide adequate information over a few biological half-lives.

We are developing a new approach for reconstructing dose and timing of dose using chemical adducts to proteins. Many highly reactive chemicals (which are typically chemicals of toxicological significance and are difficult to biomonitor) will react with proteins in blood to form chemical-protein adducts. Since there are many proteins in blood, exposure to a chemical will result in the formation of chemical adducts on many proteins. Different proteins have different natural

turnover rates, which means each chemical exposure will create a modified-protein fingerprint that is unique by dose and time after the dose occurred. The goal of this project is to identify, measure, and utilize modified-protein fingerprints for dose reconstruction.

An approach utilizing modified-protein fingerprints could revolutionize human health biomonitoring. Modified-protein fingerprints would allow for dose reconstruction of greater precision and accuracy than currently available methods. Additionally, information on timing and dose could be obtained from one sample without any required *a priori* knowledge. For rapidly cleared chemicals, this approach may be the only practical option to confirm that exposure occurred, as other biomarkers may be too low to measure.

In our first year of the project, we developed a multipronged approach to identify protein adducts. First, we used global proteomic measurements to search for protein adducts in purified proteins, plasma, and whole blood treated with reactive chemicals. Custom bioinformatics software was developed to aid in verification of protein adducts from mass spectrometry data. Second, we utilized activity-based protein profiling to search for protein adducts. This technique utilizes chemical probes designed to enrich specific protein targets, which we used as an inverse indicator of targets for protein adduct formation. Activity-based protein profiling was used in tandem with global mass spectrometry to identify and confirm protein adducts.

Our research strategy identified protein adducts caused by different chemicals. We identified protein adducts caused by organophosphate pesticides, herbicides, fumigants, and other environmental chemicals. Chemical adducts were identified on serine and cysteine amino acid sites on proteins.

In the upcoming year, we plan to develop quantification methods for protein adducts and computational models for dose reconstruction. Targeted mass spectrometry methods will be developed to quantify adducts, and turnover rates of proteins and adducts will be measured. These measurements will be integrated into a computational model to predict dose and timing of dose from modified-protein fingerprints. We anticipate that this new approach can be broadly applied for biomonitoring toxic chemicals that are traditionally difficult to monitor in occupational settings, deployed armed forces, and the general public, including chemical exposures from consumer products, accidental chemical releases, and terrorist attacks.

Visualizing Viral and Nanoparticle Interactions with Cells Across Scales and Modalities

James E. Evans

This project is focused on combining biochemical, physical, and multimodal imaging to visualize iron oxide nanoparticle (NP) interactions with cells and guide the optimization of surface coatings for efficient and targeted magnetic particle imaging (MPI) of tumors.

Understanding the interactions between biological cells and nano-sized objects has implications among many fields but has inherent limitations due to mismatch in size. Nanoparticles are few to tens of nanometers in diameter, whereas cells are typically few to tens of microns in each dimension, providing a thousand-fold difference in scale. This natural discrepancy hinders sampling efficiency and reproducibility, especially for rare cell-nanoparticle interactions. New methods are, therefore, required to improve throughput, and this project aims to augment methods and instruments developed and utilized by past projects to apply it to understanding nanoscale object interactions with biology.

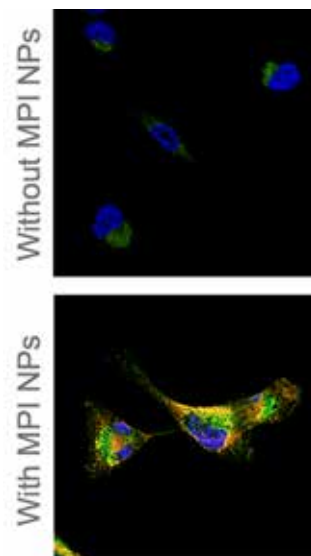
We have completed the synthesis, conjugation, and characterization of 27-nm diameter iron oxide cores with several different surface functionalities to understand biocompatibility with cells. We have also performed systematic multimodal imaging of the uptake and interactions of these nanoparticles with cells to visualize and quantify their localization, abundance, and effects on cell morphology. Specifically, we are interested in understanding the answers to the following questions:

- ▶ Are nanoparticles quickly breaking down or losing biocompatible coating once internalized?
- ▶ Where are nanoparticles localized once internalized?
- ▶ Are nanoparticles adhering to the surface of cells?
- ▶ Do different surface coatings and conjugated ligands affect internalization and fate?

In FY 2016, we initiated a comprehensive study comparing various iron oxide nanoparticle surface coatings on their internalization by human fibrosarcoma cells. To date, we have identified several surface chemistries that are favorable for magnetic particle imaging, with evidence of cellular internalization and inert interactions. We have also identified several undesirable formulations that show potential nanoparticle degradation/dissolution following cellular uptake.

Taken together, this study is illuminating how surface chemistry modifications determine iron oxide nanoparticle biocompatibility, and we are utilizing this physicochemical information to guide the next generation of functionalized nanoparticles for efficient magnetic particle imaging. We have also expanded the study to other cell lines to understand how different cell types respond to the various nanoparticle formulations and evaluate directed targeting to cancer cells.

Confocal microscopy results clearly show internalization by fibrosarcoma cells as seen by the lack of red signal for the control samples (without MPI NPs), but bright red (and yellow when overlapping green signal is present) signal distributed throughout the cell in the presence of NPs.



vPASS 1.0: Quantifying Signatures of Phenomic Expression of a *Brachypodium* Ecosystem as a Function of Genomic and Environmental Variables

Daniel C. Fortin

*We are faced globally with the challenge of increasing essential plant ecosystem services; critical to this challenge is understanding how the plant phenotype is shaped as a function of its genotype and environmental interactions. This project will perform statistical analyses of experimental data generated by plant experiments and develop empirical models relating *Brachypodium* ecosystem phenome expression as a function of controlled environmental conditions and variations in the plant genome.*

The ability to discover statistically significant correlations between variations in the plant genome and *Brachypodium* ecosystem phenome expression requires a robust exploratory data analysis (EDA) framework that can inform the development of appropriate statistical models. EDA is of paramount importance for the purposes of assessing data quality, determining relationships among explanatory variables, assessing relationships between explanatory and outcome variables, and making a preliminary selection of appropriate models. The multi-omics measurements that will serve as predictive variables for plant phenomic expression require novel data integration and EDA approaches.

This research project will lead to the development of a statistical framework that can perform exploratory data analysis (EDA), data integration, discover biologically meaningful relationships, and explore the appropriateness of candidate statistical models on data to inform downstream plant models. This research will also result in empirical models of *Brachypodium* phenome expression that will identify meaningful correlations between phenome expression and variation in the plant genome, which will in turn inform plant model parameters used in vPASS 2.0.

In FY 2016, we have identified robust computational tools that will be used as a framework to for quality control (QC) and exploratory data analysis methods for relevant plant experiments. We have completed a comprehensive review of statistical techniques used on plant genetic experiments and identified potentially useful analytic approaches. We have been meeting with members conducting preliminary experiments with *Brachypodium*. These experiments will serve to refine analytic approaches and identify sources of variation, which will guide the experimental design in future experiments.

In FY 2017, we plan to finalize the initial data processing framework (i.e., normalization, QC, and EDA). This framework will leverage data normalization methods, algorithms for multidimensional outlier detection, and filtering methods for various data (e.g., peptides, metabolites) based on objective measures. The first step in data preprocessing involves filtering of spurious metabolites, peptides, etc. Additionally, methods for objectively identifying sample outliers are leveraged. Finally, data normalization algorithms and methods for objectively choosing a normalization algorithm based on how much bias a method introduces in the data are available and key for ensuring valid results and conclusions in the data.

We will explore and analyze the experimental data from other relevant projects early in FY 2017 and use the initial results to develop statistical models to investigate the covariance between plant genotype and environmental conditions, and quantitative higher-order plant phenotype. We will combine insights from the exploratory data analysis and a review of current statistical methods used in FY 2016 to construct a multi-source modeling framework to investigate the covariance between plant genotype, environmental conditions, and scored higher-order plant phenotype.

Yarrowia Lipolytica Platform for Biobased Chemical Production

Scott E. Baker

We are developing *Yarrowia lipolytica* as a platform for production of commodity chemicals. In this project, we expressed a polycistronic RNA that contained all of the genes needed for production of itaconic acid, an important biologically derived commodity chemical.

Currently, there is an urgent need for novel biomanufacturing platforms. *Yarrowia lipolytica* is a promising organism with potential for biomanufacturing. There is a continually growing literature of research focused on the use of the yeast, *Yarrowia lipolytica*, as a protein production host and lipid producer. PNNL has a distinguishing capability in fungal biotechnology focused on non-traditional but genetically tractable organisms.

We have developed the molecular genetics needed to manipulate the organism *Yarrowia lipolytica*, putting us in a position to 1) insert pathways for biosynthesis of a variety of industrially relevant compounds, and 2) demonstrate proof of concept that this organism has potential in biomanufacturing. In this project, we will express the itaconic acid biosynthetic gene cluster (consisting of a cis-aconitate decarboxylase, mitochondrial transporter, and major facilitator superfamily [MFS] transporter) in *Yarrowia lipolytica*. We will accomplish this via an expression construct encoding a polycistronic ribonucleic acid (RNA), with each open reading frame (ORF) separated by viral 2A peptide sequences. These sequences cause co-translational cleavage of each gene product from a single RNA. We will use a colorimetric assay to assess the amount of itaconic acid produced.

Initially, we designed the parts of our polycistronic RNA expression construct. Each of the genes need for itaconic acid biosynthesis were codon-optimized for expression in *Yarrowia lipolytica* and subsequently synthesized. We took a modular approach for our construct so we could “mix and match” a variety of open reading frames to test the effect on itaconic acid biosynthesis. Each open reading frame was separated by a sequence encoding the viral 2A peptide, which leads to co-translational cleavage and allows multiple proteins to be pro-

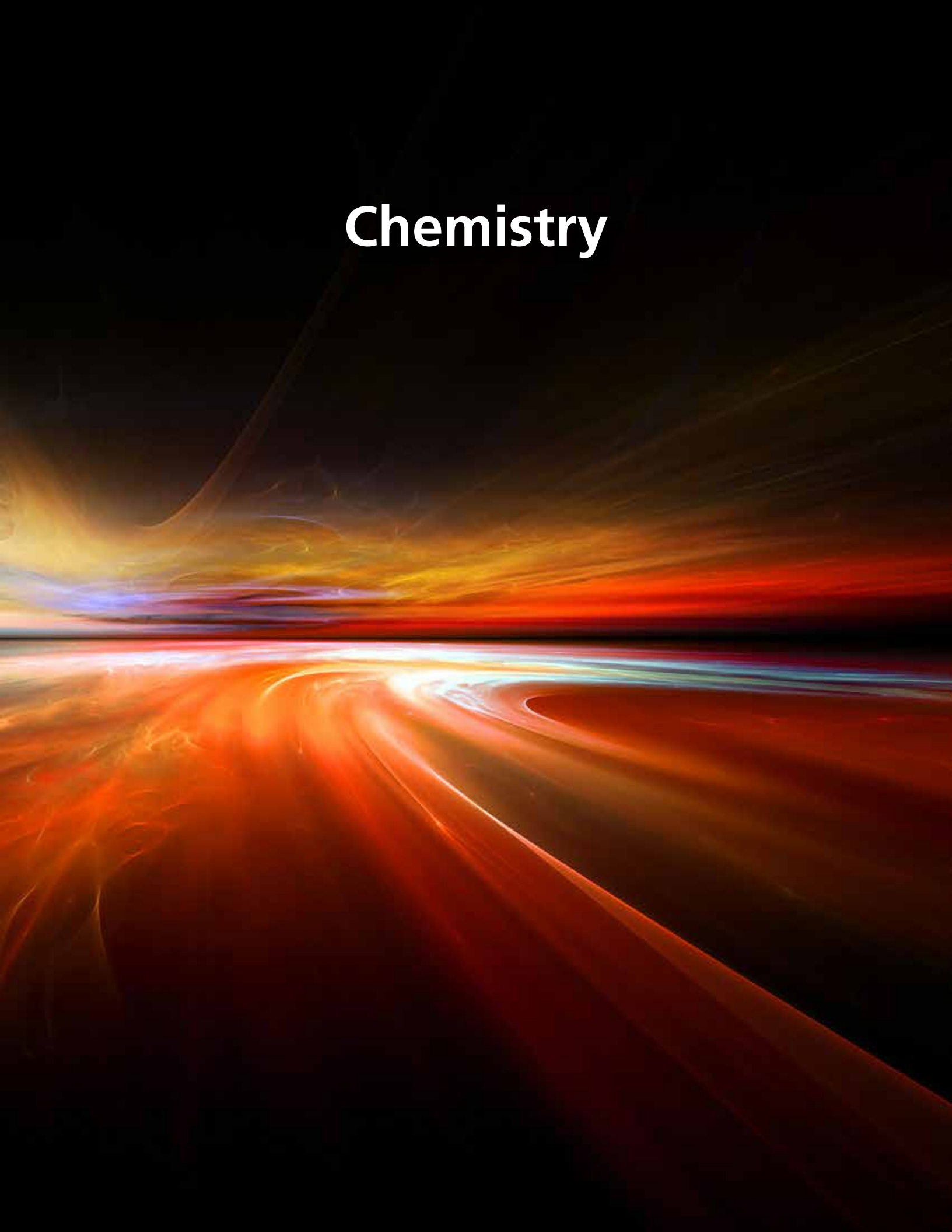
duced from a single RNA. To assess whether the whole polycistronic RNA is transcribed, the green fluorescent protein (GFP) ORF was inserted. Strains with GFP constructs were assessed microscopically. Transformants were generated that expressed some or all of the itaconic acid biosynthetic cluster. Itaconic acid was assayed from the spent media using a colorimetric assay, as well as by high-performance liquid chromatography and mass spectrometry methods. Transformants that incorporated the cis-aconitate decarboxylase, mitochondrial transporter, MFS transporter, and GFP were selected. Then, GFP expression was assayed using fluorescence microscopy and isolated several positive strains. Itaconic acid production was detected in the positive strains at the level of approximately 50mg/L, which indicated that our method for generating multiple proteins from a polycistronic RNA worked.

Previously published studies have shown that the cis-aconitate decarboxylase is essential for itaconic acid production. We tested several different constructs that included the cis-aconitate decarboxylase encoding ORF with and without the transporters. Our findings showed that, even without the transporters, cis-aconitate decarboxylase can produce detectable itaconic acid. Our results also indicated that the expression of the mitochondrial transporter enhanced itaconic acid production.

The final experiments we performed utilized the GFP containing full gene cluster (i.e., cis-aconitate decarboxylase, mitochondrial transporter, and MFS transporter) construct. We used ultraviolet light to mutagenize a strain that was able to make about 50 mg/L itaconic acid and then used a fluorescence-activated cell sorter to isolate cells with higher GFP expression. Several strains were isolated and then assayed for itaconic acid production. We detected increased itaconic acid production in two independent lineages of mutagenized *Yarrowia lipolytica* cells.

Our results indicate that polycistronic RNAs can be expressed and translated into multiple proteins in *Yarrowia lipolytica*. Furthermore, we show that we can isolate strains with higher expression of our biosynthetic cluster by selecting for mutant strains with increased fluorescence.

Chemistry



Bio-Inspired Selective Conversion of Methane to Methanol

Abhijeet J. Karkamkar

To maximize the United States' remote natural gas resources, such as off-shore methane, we seek to develop the new catalysts needed to transform methane into liquid fuels like as methanol.

Natural gas can be found in abundance throughout the United States and is often used for heating, cooking, and electrical power generation. Natural gas is composed primarily of methane but is not widely used for transportation. Currently, there are no commercially viable, direct approaches to convert methane into liquid fuel, and synthetic approaches are expensive and inefficient at small scales.

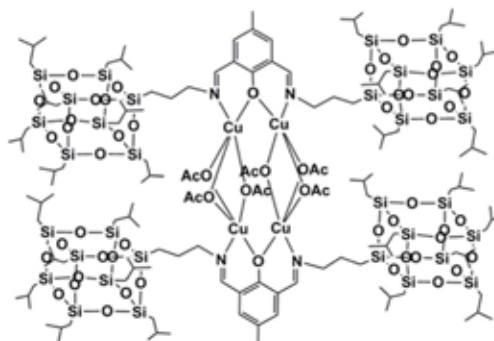
In this project, we are developing a synthetic pathway to prepare copper cluster of controlled nuclearity. We are also developing catalytic pathways to study the structure of these clusters as they undergo transitions during the catalytic cycle. Our objective is to synthesize and characterize an efficient site for the chemo-selective activation of the carbon (C)-hydrogen (H) bond in methane and its selective oxidation to methanol (C-OH). The selective oxidation of C-H bond by an oxidant under mild conditions to produce a liquid fuel is a desirable technical accomplishment.

Thermodynamic considerations show that steam reforming should be favorable only near 1,000 K, but that direct oxidation of methane should be favorable at room temperature. The equilibrium constant indicates that conversion of 33% at 298 K is achievable. Direct, low-temperature conversion to methanol will be competitive, with steam reforming if we achieve a single pass conversion of 5.5% with 80% selectivity toward methanol.

Enzymes, such as methane monooxygenase (MMO), are comprised of multinuclear copper clusters that are the proposed active site. There is considerable interest and debate in the literature, however, regarding the exact nature of the active site.

We have designed and synthesized complex copper clusters with controlled nuclearity. By changing the supported ligand design and metal precursors, we have been able to introduce structural variation in the catalysts. The structure of the ligand and catalysts was characterized by various techniques such as nuclear magnetic resonance and mass spectrometry. These techniques revealed the exact structure of most of these complexes. We also studied the impact of the structure of this copper cluster on the catalytic activity for alkane ox-

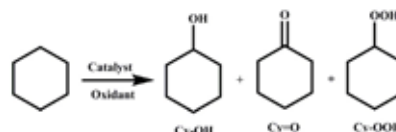
idation. We have synthesized and characterized several of the mono-nuclear and multinuclear copper clusters stabilized in a mesoporous organo-functionalized silica matrix and a polyhedral oligomeric silsesquioxane (POSS) framework.



Structure of tetranuclear oxy-bridged copper cluster linked to POSS core.

Subtle changes at the metal center have a significant impact on the activity of the catalyst. This finding is of particular importance, because we have a very simple method of controlling and tweaking the structure and probing the effect of structure on catalytic activity.

We have evaluated the oxidation of cyclohexane to cyclohexanone and cyclohexanol by a variety of oxidants.



Equation: Cyclohexane oxidation to cyclohexanol and cyclohexanone.

Oxidation of cyclohexane using different copper catalysts.

Catalyst	Oxidants	Cyclohexanol (% Sel.)	Cyclohexanone (% Sel.)	Total conv (%)
2,6-DFPS Cu ₄ Ac ₆ (2,6-DFPS) ₂	H ₂ O ₂	-	-	-
	H ₂ O ₂	0	0	0
		19	81	8.6
Cu ₄ Ac ₆ O(2,6-DFTMS) ₂ @ MCM-41	H ₂ O ₂	0	0	0
	TBHP(W)	18	82	8.6
	TBHP(D)	28	72	8.4

Climate-Related Chemistry of Internally Mixed Atmospheric Particles

Alexander Laskin

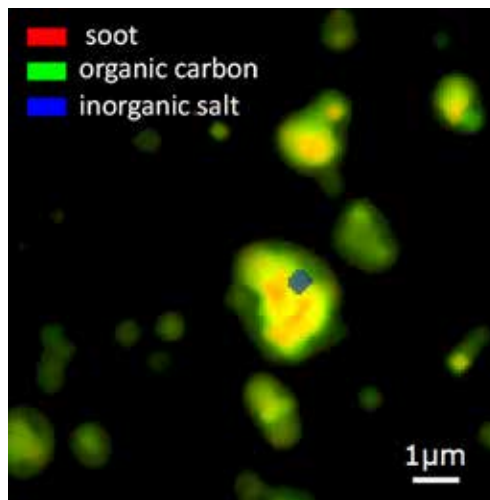
This project applies novel chemical imaging platforms for in-depth characterization of atmospheric particles and develops unique capabilities and methodologies for collaborative research in complex particle chemistry and its multi-phase reactivity relevant to climate change.

Understanding and mitigating the environmental impacts of energy production and use has been identified as one of DOE's important priorities. The production and use of energy often leads to the generation of aerosol particles, either through direct emission or through secondary condensation of oxidized gas-phase emissions. Of primary importance is understanding the role of particles in a broad range of environmental issues, including atmospheric composition and chemistry, assessment of the earth's radiation balance, ozone recovery, and health effects. To address DOE concerns about the safe production and use of energy, it is necessary to develop and apply novel experimental approaches for improved characterization of particle chemical composition and physical properties, understanding their atmospheric evolution and fate.

The intent of our multi-modal collaborative research is to study the chemical and physical properties of atmospheric particles, in order to understand a fundamental relationship between the composition of particles and their atmospheric impact. The ultimate goal is to provide relevant data for use in climate modeling. Our specific research efforts are focused on chemical imaging of individual particles, predictive understanding of their internal structures (mixing states), effects of liquid-liquid and solid-liquid phase separations, and optical properties. We use a combination of X-ray spectro-microscopy and electron microscopy techniques to investigate particle composition, their propensity for phase separations, and mixing states at atmospherically relevant conditions.

Occurrences of phase separations depend on particle size, composition, and reactivity of their internal components that can be probed by chemical imaging. These experimental data are of critical importance to elucidate the molecular-level processes governing mixing states of individual particles. The occurrences of phase separations and mixing states have important implications for particle optical properties, their atmospheric aging, and particle-cloud interactions—the most challenging problems relevant to understanding and mitigation of aerosol effects on climate change. To this end, we conducted a number of laboratory studies focused on the chemistry of internally mixed organic/inorganic particles and effects of their mixing states on hygroscopic properties,

heterogeneous reactivity, and ice nucleation properties. Depending on their composition and ambient conditions, particles exist in liquid, amorphous semi-solid, or solid (glassy) phase states that have important implications in various atmospheric processes.



An X-ray microscopy map of internal chemical components in individual atmospheric particles collected in a tropical forest.

We discovered unique atmosphere-land-surface interaction resulting in generation of solidified airborne soil organic particles (ASOP) composed of soil organic matter. Our study suggests that ASOP are efficiently formed after strong rain events in ecosystems where wet soils are exposed to the ambient air such as agricultural systems and grasslands. Chemical imaging and micro-spectroscopy analysis of ASOP physico-chemical properties suggests that they have important impacts on cloud formation and efficiently absorb solar radiation.

We used chemical imaging to study atmospheric transformations of airborne biological spores at elevated relative humidity. We observed rupturing of fungal spores and subsequent release of hygroscopic inorganic salts that may potentially influence cloud formation. Rupturing of fungal spores at high humidity may explain the bursting events of new particle formations in the ecosystem of a tropical forest.

We examined the mixing state and morphology of atmospheric particles influenced by combined biogenic and anthropogenic emission sources. Observations of soot and inorganic core morphologies and thick organic coatings (illustrated in the figure) provide experimental means to detect particle aging. These measurements provide important constraints for the calculations of morphological effects on particle optical properties expected in their atmospheric aging processes.

Data from our studies can be used to begin systematic evaluation of the next generation of atmospheric models with sufficient fidelity to obtain model-based predictions of the environmental impact by particles.

Development of a Novel Microscopy Platform for Fundamental Studies of Ice Nucleation on Atmospheric Particles

Alexander Laskin

The ultimate goal of this project is establishment of a new experimental microscopy platform to unravel physicochemical processes of heterogeneous ice nucleation on surfaces of individual atmospheric particles.

The present understanding of the role that atmospheric particles play in formation of ice clouds is limited to the extent that their impact on the environment and climate cannot be accurately predicted by models. Atmospheric particles are complex in nature (multicomponent and multiphase) and exhibit different ice nucleation efficiencies that need to be probed with individual particle specificity. This project focuses on the development and applications of a novel experimental platform for fundamental studies of ice nucleating particles (INPs) with microscopic determination of physicochemical properties of individual INPs that govern ice formation propensity. The novel platform is hosted by an environmental scanning electron microscope (ESEM) and allows *in situ* experimental studies resembling fundamental processes of aerosol-ice cloud interactions, which are one of the most challenging problems for predictive understanding of the earth's climate change.

Our project focuses on heterogeneous ice nucleation, which is governed by the physical and chemical properties of INP that can vary from particle to particle. Previous studies suggested that the following properties may control the efficiency of INPs: insolubility, solid particles such as mineral dust; size, a higher nucleation probability for particle with a larger surface area; chemical bonds and their crystallographic arrangement on particle surface, the ability to form hydrogen bonds; and/or active sites, characteristic locations initiate nucleation such as cleavage and growth steps, crack, cavities, and inclusions.

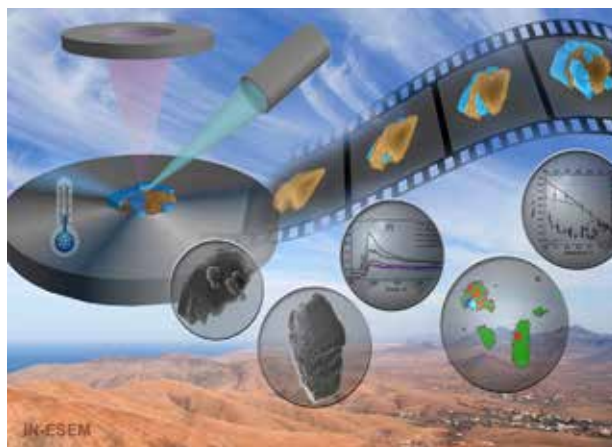
Although these guidelines help to identify which materials are likely to act as INP, they cannot explain the contradicting results observed for real-world atmospheric particles, because a microscopic understanding of the ice nucleation events on the INPs was lacking.

We designed and constructed the cryogenically cooled ice nucleation cell that allows experimental observation of ice nucleation (IN) inside an ESEM instrument over individual INPs at temperatures and relative humidity typical for the upper troposphere/lower stratosphere. We employ the IN-ESEM platform to investigate ice nucleation of complex atmo-

spheric particles relevant to anthropogenic and biogenic emissions. Combined with theoretical chemistry calculations, the experimental data is analyzed to gain a better understanding of INPs and parameterizations for cloud models.

We reported microscopic observations of ice nucleation events on kaolinite particles at the nanoscale and demonstrated the capability of direct tracking and micro-spectroscopic characterization of individual INPs within an ensemble of authentic atmospheric particles. We demonstrated dynamic observations of individual ice formation events over particles in isobaric and isothermal experiments. Isothermal experiments on individual kaolinite particles demonstrated that ice crystals preferably nucleated at the edges of the stacked kaolinite platelets, rather than on their basal planes. These experimental observations indicated that the OH terminated edge surfaces of kaolinite platelets were preferred ice nucleation sites. The chemical differences between the edge and the basal plane surfaces contributed to their different propensities to nucleate ice. The observations of the location of ice nucleation provided direct information for further theoretical chemistry predictions of ice formation on kaolinite.

The IN-ESEM platform provides a new experimental tool to probe effects on the nanometer scale of various materials in response to low temperatures and concomitant variation in relative humidity with previously unachieved precision and resolution. This technique opens a pathway to new physical chemistry endeavors to study ice formation processes relevant to atmospheric science, cryobiology, and material science.



Artwork highlighting nanometer scale observations of ice nucleation on the surface of mineral dust particles in experiments conducted inside an ESEM. This novel experimental platform opens a pathway to new physical chemistry studies of ice formation with applications to atmospheric science, cryobiology, and material science.

Dynamics of Supported Noble Metal Nanoparticles in the Presence of Oxidizing Environment: Application of Compressive Sensing in TEM

Libor Kovarik

The goal of this project is to develop and apply the novel capability of compressive sensing (CS) acquisition in order to enable investigation of the structure and property of catalytic nanoparticles during exposure to elevated temperature and reactive environment, and establish structure-property relationship.

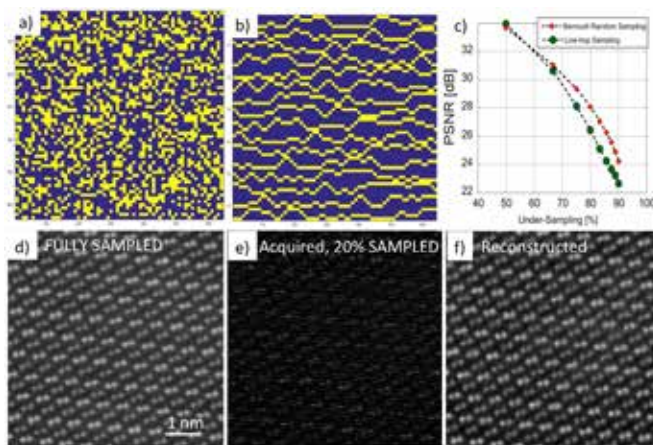
Noble metal nanoparticles are used extensively in catalytic applications due to their highly advantageous catalytic properties. During exposure to high temperatures and a reactive gas environment, the nanoparticles can undergo a range of morphological, structural, and surface transformations, and understanding these transformations is critical for rationalization of functional properties of the catalyst. *In situ* transmission electron microscopy (TEM) provides unique atomic level capability for the analysis of catalysts under *in situ* conditions; however, its application is limited by electron beam damage. Prolonged electron beam exposure under *in situ* conditions leads to a damage/alteration, including reduction, encapsulation by the support material, alloying by the substrate, etc. The objective of this project was to develop and apply the novel techniques of compressive sensing in order to lower the effect of electron beam dose during imaging. Compressive sensing is a method based on the foundation that images/data can be well represented in a much sparser form using a suitable basis set, and that this suitable basis set can be fully recovered from significantly fewer (sparse) measurements than conventional pixel-by-pixel acquisition (hence reducing the acquisition time and dose).

To achieve this goal, we developed sparse sampling methodology for scanning TEM (STEM) imaging by exerting controls over scans coils. Scan coils have significant induction delays, which complicates the implementation of STEM sparse sampling. We designed sparse sampling strategies that are not affected by the scan coils induction delays and demonstrated that this sampling strategy has comparable properties to optimal Bernoulli random sampling. In order to implement this sparse sampling method, we have developed diagnostics procedures for evaluations of scan coil dynamics, and we custom-built field-programmable gate array controls and

software for acquisition (synchronizing the scan coils and annular dark field detector). The viability of the newly developed sparse sampling was rigorously tested, and it was shown that this approach is very effective and can decrease the dose and acquisition time by factor of five times, relative to conventional acquisition, permitting imaging of beam-sensitive materials to be obtained.

Comparison of Bernoulli random pixel sampling (a), and line-hopping sampling (b). Theoretical peak signal-to-noise ratio for reconstructions with Bernoulli random sampling and line-hop sampling (c). Comparison of fully sampled image (d) with sparsely sampled (e) and CS reconstructed image (f) of NiTiO₃.

While the capability of CS was under development, we have continued our work with conventional acquisition setup with the state-of-the-art atomic resolution environmental TEM to study the stability and dynamic behavior of Pd nanoparticles during oxidation/reduction conditions. Most notably, we identified new a mechanism of hollow Pd nanoparticle formation during reduction treatment. The atomic level imaging of Pd under oxidizing/reduction conditions also enabled us to quantify nucleation and growth of the forming phases. In addition, we studied the mechanism of Pt dispersion during high-temperature oxidation treatment. Overall, the work of this project enabled us to gain a new level of understanding of metal supported catalysts.



Comparison of Bernoulli random pixel sampling (a), and line-hopping sampling (b). Theoretical peak signal-to-noise ratio for reconstructions with Bernoulli random sampling and line-hop sampling (c). Comparison of fully sampled image (d) with sparsely sampled (e) and CS reconstructed image (f) of NiTiO₃.

Electrocatalytic Reduction of Phenols and Ethers

Donald M. Camaioni

In this project, we are examining the pathways and challenges of catalyzing low-temperature electrochemical processes for electrical energy storage in the form of hydrocarbons suitable for transportation fuels derived from the lignin components of biomass.

Biomass is the oldest renewable energy supply; however, many biomass and bio-oil feedstocks are not used for modern transportation due to high levels of oxygenation. A wealth of catalysis research exists to thermally convert aqueous bio-oils into hydrodeoxygenated liquid hydrocarbons through reaction with hydrogen. However, for locations where high temperatures and high-pressure hydrogen are not available, the best route to convert biomass to hydrocarbons is unclear. Previous work has shown that model biomass compounds, such as phenol, can be electrochemically converted to products like cyclohexanone, cyclohexanol, and cyclohexane using metal electrocatalysts. Our project was initiated to provide understanding of the mechanism, rate-limiting steps, and energies required for electrocatalytic reductions in comparison to thermal hydrogenation. We will gain insight to the potential of electrocatalytic processes to produce transportation fuels sustainably using renewable feedstocks at a wide scale of plant sizes, even in decentralized locations. It is anticipated that our work will contribute to energy security by developing processes for producing liquid hydrocarbon transportation fuels on demand, in a renewable manner, without greenhouse gas emissions, and also absent the risks associated with using a dwindling fossil fuel source with extremely volatile availability.

During FY 2015 we determined that the mechanism for the electrocatalytic hydrogenation (ECH) of phenol on platinum group metals is the same as that for thermal hydrogenation (TH), where the adsorbed phenol is reduced via sequential addition of adsorbed hydrogen atoms. The difference between ECH and TH observed thus far is the origin of the hydrogen reduction equivalents (hydrogen or water) and the hydrogen coverage on the catalyst surface, which is con-

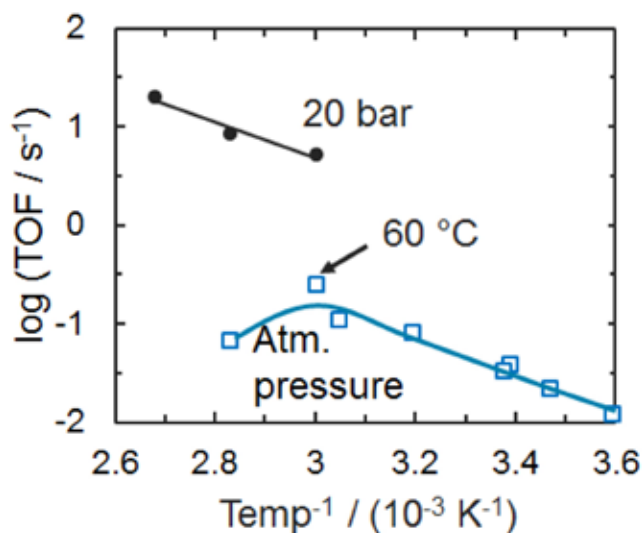
trolled by hydrogen pressure in the thermal process and by the electrochemical potential in the electrocatalytic process. We determined that Rh/C and Pt/C are more active for electrocatalytic reduction of phenol than Pd/C, with Rh/C being the most active at two to three times higher activity than Pt/C.

In FY 2016, we explored the effects of increasing temperature on phenol conversion and determined that, at ambient pressure, there was deactivation of the catalyst at 60 to 80°C for both TH and ECH. We conducted TH measurements at 20 bar hydrogen, where the deactivation was not observed even at 100°C. A pressure of just 6 bar H₂ at 80°C was sufficient to prevent catalyst deactivation during TH. We extended the results of phenol electrocatalytic reduction to diaryl ethers, which were also reduced, but additionally saw C-O bond cleavage. The diaryl ethers were converted at lower rates than the unsubstituted phenol. Measurements of the dependence of TH rates on hydrogen pressure indicate that Pt is zero order in hydrogen from 6 to 20 bar, while Rh is first order in hydrogen under these same conditions.

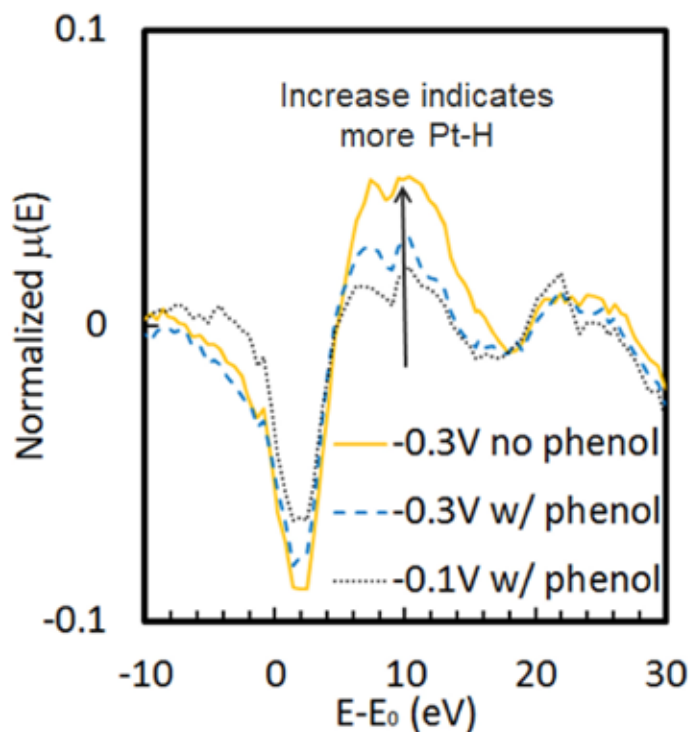
Additionally, we developed an electrochemical cell for *in situ* X-ray absorption spectroscopy measurements and detected a change in the hydrogen adsorption on platinum with the applied potential. At more reducing potentials, more hydrogen was adsorbed onto Pt, while in the presence of phenol, the hydrogen adsorption was decreased. We also detected a Pt-C interaction at a distance that matches what is expected for phenol adsorption on platinum, indicating X-ray absorption spectroscopy can be used to measure coverages of hydrogen and phenol during catalysis.

A major finding was the observation that catalysts show signs of deactivation when the reductions were run at temperatures above 60°C. We hypothesize that this deactivation is due to insufficient hydrogen at the catalyst surface, which allows dehydrogenation of adsorbed organic species (e.g., phenol and/or partially reduced intermediates), which fouls the metal surface. We found that operating at higher H₂ pressure during thermal hydrogenation prevents deactivation of the catalysts. The observation suggests that operating electrocatalytic reductions under several bar H₂ may well be a remedy for the deactivation of the electrocatalysts.

Next year, we plan to investigate the effect of increased pressure on the electrocatalytic reduction, with the hypothesis that this will increase rates and prevent deactivation at higher temperatures. Preliminary results indicate going above 1 bar H_2 increases the rate of reaction, but further increases in pressure are needed to prevent deactivation. We intend to continue investigation of the substituted phenols and diaryl ethers to determine the reason for their lower activity relative to unsubstituted phenol. We also plan to continue with X-ray spectroscopy to characterize the catalysts and their surface coverages under reaction conditions, and we will employ scanning tunneling electron microscopy to investigate the nanoscale morphologies of the catalysts.



Turnover frequencies for the thermal hydrogenation of phenol on Pt/C versus temperature. TH conducted at atmospheric pressure of hydrogen and 20 bar hydrogen. The deactivation at >60 °C observed at atmospheric pressure was not observed at 20 bar. 172 mM phenol in water, pH 5.



Background subtracted X-ray absorption near edge structure (XANES) spectra of Pt/C at different potentials in 50 mM acetate buffer. Potentials are vs. Ag/AgCl. 100 mM phenol was added for tests with phenol.

Electrochemical Processes to Transform Waste CO₂ to Fuels and Chemicals

Asanga B. Padmaperuma

This project has demonstrated electrochemical reduction of waste CO₂ to fuels and chemical products. Electrochemical technologies to convert waste CO₂ to valuable intermediates and products will improve carbon efficiency, improve biorefinery economics, and reduce greenhouse gas (GHG) emissions.

The objective of this project was to develop electrochemical methods to efficiently convert waste CO₂ to fuels, chemicals, and chemical intermediates. Waste CO₂ is generated in both petroleum-based and biobased processes accounting for large losses in valuable carbon that might be put to beneficial use. For example, many biobased processes have only moderate carbon efficiencies, with the lost carbon ultimately ending up in CO₂ waste gas emitted into the atmosphere such as by burning coke or lignin (and possibly natural gas) for process heat. The extent to which carbon efficiency can be improved and GHG emissions can be reduced depends on the efficiency of electrochemical CO₂ reduction, the products formed, and the source of electrons.

An emerging technology for recapturing valuable carbon is electrochemical reduction (ECR) of CO₂ in which intermediates, including ethylene, CO, and formic acid, can be produced. Co-processing ethylene with high-pressure CO₂ in organic solvents is reported to offer routes to value-added products such as succinic and adipic acids. Selectivity depended on the CO₂ to ethylene ratio, with longer chain diacids formed at higher ethylene concentrations. This chemistry occurred at a stainless steel electrode.

CO₂ radical anion is reported to form by ECR in ionic liquids (ILs). In this project, the possibility of reducing CO₂ in ionic liquids to formic acid, oxalic acid, and dicarboxylic acids, by coupling CO₂ radical anion with ethylene, was investigated.

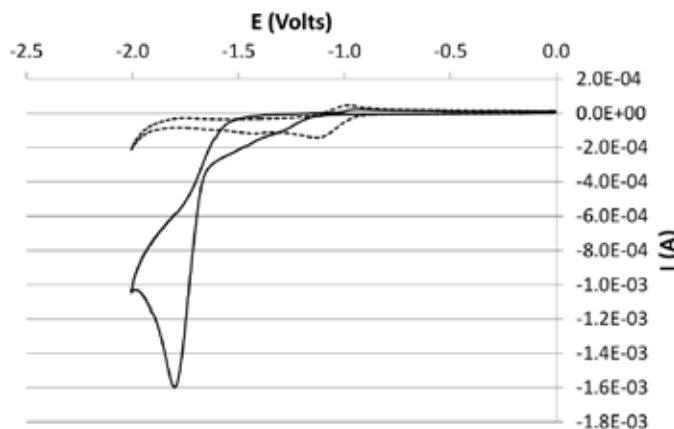
Initial studies were conducted using cyclic voltammetry to determine potentials for CO₂ reduction in hydrophilic and hydrophobic ionic liquids. Five electrode materials were tested in [EMIM][MeSO₃]. A silver electrode gave a distinct, irreversible CO₂ reduction wave at -1.8 V vs an Ag wire reference electrode. CO₂ reduction was also observed in cyclic voltammetry (CV) using Pt, Cu, and Ni electrodes. By analogy

to the literature, the reduction forms CO₂ radical anion.

The effect of added water was investigated by repeating the CV experiments in [EMIM][MeSO₃] containing 10 wt% H₂O. Peak currents were found to be about twice those in the dry IL solvent. It is known that CO₂ solubility is influenced by water content, and it is likely that the increased current results from an increased CO₂ concentration in the IL. For Ag and Cu electrodes, the cathodic limit was unaffected, but for Pt, this limit was decreased.

The use of the phosphonium IL trihexyltetradecylphosphonium chloride, [P(14)666][Cl], was not advantageous. While CO₂ reduction was observed, the voltammetry was highly variable and scans were irreproducible. A high solvent viscosity and low conductivity were serious problems for this solvent.

Controlled potential coulometry was conducted in [EMIM][MeSO₃] with (10% water) and without added water to determine product composition, selectivity, and current efficiency. A potential of -1.7 V vs Ag wire was applied to the cathode in an H-cell in which CO₂ and ethylene were bubbled through the IL solvent. The anode compartment contained 10% H₂SO₄ and a Pt electrode. The compartments were separated by a Nafion membrane. The major products formed were formic and oxalic acids. Succinic acid was not formed, indicating that increased ethylene pressures are required to intercept the CO₂ radical anion. Current efficiencies were found to be higher in the IL with no added water (approximately 30%) than in the water-added solution (approximately 10%). This system could be useful for producing formic acid as an H₂ carrier or for fuel cell use.



CV at a silver electrode in [EMIM][MeSO₃] under CO₂ (solid) and N₂ (dotted).

Fundamental Understanding of Nucleation Processes to Assess Solution Stability and Phase Growth and Genesis

Gregg J. Lumetta

We are developing a physicochemical framework for predicting and manipulating precipitation reactions relevant to nuclear materials processing and nuclear forensics.

Nucleation and crystal growth are fundamentally important to processing nuclear materials and radioactive wastes, but information is lacking regarding the basic science underlying these transformations. Many chemical processes used in the nuclear industry—both for recovery of special nuclear materials and for disposal of radioactive wastes—involve manipulating highly concentrated salt solutions. Precipitation of solids from such solutions is often a concern. In this context, precipitation might be desired (e.g., of plutonium oxalate during plutonium oxide synthesis) or not (e.g., during ion exchange or solvent extraction processes).

In this project, we are developing a framework based on recent developments in crystal growth that have been providing insight into two distinct approaches. In the “classical” pathway, nucleation occurs through ion-by-ion addition to a growing cluster with increasing free energy of formation until a critical size is reached. At this point, the free energy change for further growth becomes negative and occurs via ion-by-ion addition to kink sites at atomic steps on the crystal surface. For both nucleation and growth, morphologies and dependencies of rates on applied supersaturation are well understood and are distinct from those of crystals that form through the second, “non-classical” pathway. In the latter scenario, nucleation and growth occur through the addition of higher order species ranging from oligomeric complexes to liquid droplets to amorphous or crystalline nanometer-scale particles. Detailing the morphologies, internal structures, and dependencies of growth rates on supersaturation allows the formation mechanism to be determined and related to process conditions. Moreover, rate measurements vs. supersaturation and temperature will allow us to determine the activation barriers that control the nucleation and growth processes.

Our research is focused on the precipitation of oxalate salts. In particular, we are investigating nucleation and crystal

growth of Pu(III) oxalate, $\text{Pu}_2(\text{C}_2\text{O}_4)_3 \cdot 10\text{H}_2\text{O}$, because the way this compound forms is of interest to the nuclear forensics community. From the challenges of working with the highly radiotoxic plutonium, initial investigations have used Ce(III) and Eu(III) as surrogates for Pu(III), which allows the development of the experimental techniques to examine the plutonium system safely. Investigations into the precipitation of sodium oxalate from high-salt alkaline solutions will also be pursued because this system is critically important to operation of the Hanford Tank Waste Treatment and Immobilization Plant.

A combination of theoretical and experimental techniques is being used to develop a framework for explaining nucleation and the subsequent crystal growth of the metal oxalate systems from the atomistic to hundreds of micrometers scale. Both classical and *ab initio* models are being used to provide direct but complementary information on the mechanisms underlying phase transformations. Experimentally, these systems are being investigated using advanced *in situ* electron microscopy techniques to gain insight into the nucleation mechanisms involved. This work uses techniques previously demonstrated through *in situ* transmission electron microscopy (TEM) investigations of CaCO_3 , gold nanoparticles, and iron oxide nanoparticles. In addition, cryo-TEM is being used to provide “snapshots” of particle growth with minimal disturbance to the system. Complimentary optical spectroscopic techniques are being used to provide additional insight into metal oxalate crystal growth.

Four different computational models representing the Eu(III) oxalate salt were examined. Two of these considered completely deprotonated oxalate ligands (which is known to be the case for the bulk $\text{Eu}_2(\text{C}_2\text{O}_4)_3 \cdot 10\text{H}_2\text{O}$ compound): $[\text{Eu}(\text{C}_2\text{O}_4)_3(\text{H}_2\text{O})_3]^{3-}$ (**A**) and $[\text{Eu}_2(\text{C}_2\text{O}_4)_5(\text{H}_2\text{O})_6]^{4-}$ (**B**). The other two models considered some degree of protonation of the oxalate ligands to account for the charge neutrality in the crystallographic system: $[\text{Eu}(\text{HC}_2\text{O}_4)_3(\text{H}_2\text{O})_3]$ (**H₃A**) and $[\text{Eu}_2(\text{HC}_2\text{O}_4)_4(\text{C}_2\text{O}_4)(\text{H}_2\text{O})_6]$ (**H₄B**). The computed infrared (IR) spectrum from the dimeric cluster B and H4B was compared to the Fourier transform infrared spectroscopy (FTIR) spectrum experimentally determined for $\text{Eu}_2(\text{C}_2\text{O}_4)_3 \cdot 10\text{H}_2\text{O}$. For **B** and **H₄B**, excluding the exterior, under-coordinated oxalate moieties from IR

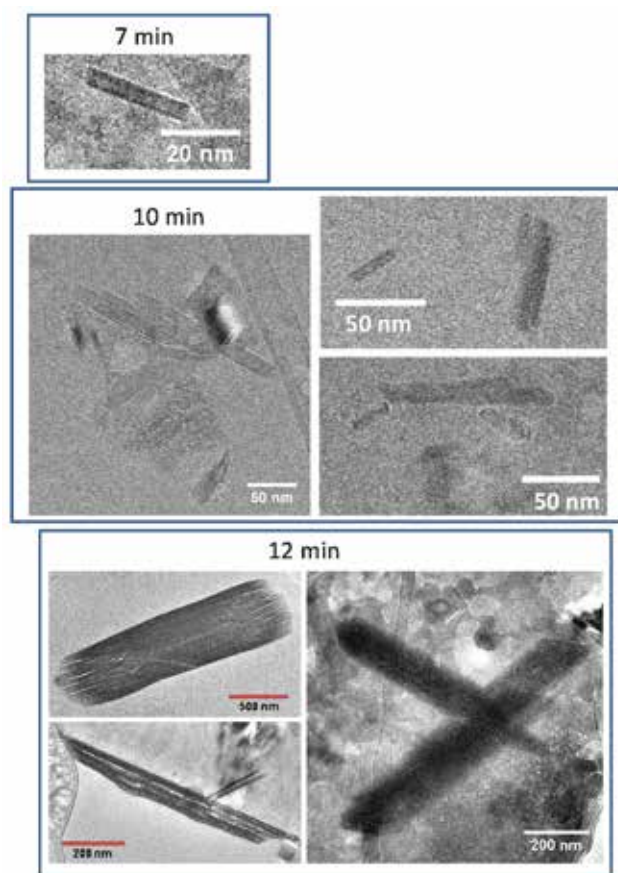
predictions results in a spectrum that matches the experimental FTIR spectrum. Inclusion of the exterior oxalates results in the prediction of additional vibrational bands that are not observed experimentally.

The Raman spectrum measured in this work displayed bands not previously observed. Computational models are working on a hypothesis to explain the appearance of additional bands. The above model for the dimeric cluster that accurately reproduce the IR spectrum do not predict bands where the new Raman peaks are observed. Two scenarios that would account for the observed vibrational bands are currently under investigation. The first scenario is that the material exhibits significant populations of the surface exposed oxalate ligands, where the oxalate ligands at the surface of the particle would attract protons in highly acidic media. The second scenario is that the material includes noticeable populations of a twisted, non-planar oxalate, which has been previously observed in solution to exhibit the same IR spectrum at planar oxalate, but has additional Raman peaks.

Advanced electron microscopy tools were used to examine the formation of $\text{Eu}_2(\text{C}_2\text{O}_4)_3 \cdot 10\text{H}_2\text{O}$ from solutions of Eu(III) nitrate and oxalic acid. Cryo-TEM images were determined for europium oxalate crystals vitrified at several time points after the mixing of the reactants. Vitrification during the specimen preparation process arrested the reaction and preserved the particles in their in aqua conformation. Early in the crystal growth process (e.g., samples vitrified 7 and 10 min. after mixing), lone rods and small assemblies of parallel rods were observed. Rods varied in length from tens of nm to approximately 150 nm. Rod size increased to hundreds of nm in the sample vitrified at 12 seconds after mixing, and the first clusters of rods were observed. More complex clusters, comparable in morphology to the final product but less than one-tenth of the size, were seen in the sample vitrified 20 min. after mixing. Highly poly-disperse lone rods were observed in the same sample.

The capability to monitor the formation of europium oxalate *in situ* was demonstrated using a scanning electron microscope fluid cell. Images were taken every 2 minutes. For the most part, increase in the size of the lath-like crystals was observed over time. But in some cases, the solid crystals appeared to re-dissolve with time. These microscopy tools provide unique insight into the formation and growth of europium oxalate and hold great potential for extension to study of the radioactive plutonium analog.

In FY 2017, work will continue on developing the theoretical framework for obtaining cluster thermodynamics and rates of formation to model Pu(III) oxalate nucleation. The approach will include calculating the electronic and vibrational spectra of the hydrated Pu(III) cation and its oxalate complexes and developing the interaction potentials for large-scale simulation of crystallization. *In situ* microscopy and cryo-TEM methods, coupled with optical and high-resolution ultrasonic techniques, will be used to determine the mechanisms and pathways to nucleation of f-element oxalates and relate these to the morphology of the resulting solids. A key emphasis of our work in the next year will be the transition to using these techniques with Pu.



Cryo-TEM images of the information of europium(III) oxalate crystals.

In-Line, On-Demand Increase of Fuel Octane and Cetane Numbers

Tim Bays

This work will lead to the development of an inexpensive, on-board, in-line, solid-state catalyst that can continuously optimize the fuel octane/cetane rating in response to engine performance demands, using currently available hydrocarbon fuels.

Strategies for increasing light-duty vehicle fuel economy include two coupled approaches: vehicle engine light-weighting and the development of fuels with higher research octane numbers (RON). These strategies meet in modern engines that have increasingly high compression ratios, which require higher RON fuels (e.g., less reactive and more resistant to autoignition). Producing higher RON fuel will solve this problem, but it may be more desirable to change fuel reactivity in response to engine operating conditions.

Most approaches to changing fuel reactivity in real time require a two-tank solution or introduction of a chemical additive. Additives are undesirable because they increase cost and may have unintended consequences. In a two-tank approach, one tank would contain a low reactivity fuel and the other tank would contain a high reactivity fuel. Generally, this approach is not considered to be palatable to consumers, who would need to ensure that both tanks were filled for proper vehicle operation.

A related approach is onboard fuel separation, where a single fuel is separated into high and low octane portions by concentrating either ethanol or aromatics into the higher octane fraction.

The approach explored here is similar to onboard fuel separation, except the chemical composition of a commercially available fuel would be altered between the fuel tank and the engine, such that higher octane or cetane fuel would be provided to the engine on demand, without the need to separate the fuel into different components. We seek to develop a system that provides an inexpensive, on-board, in-line, solid-state catalyst that can continuously optimize the fuel octane/cetane rating in response to engine performance demands and will improve engine performance and fuel efficiency, without the need for special additives or changes to

the fuel refinement process. The result would be a higher octane/cetane fuel that doesn't change the energy requirements for fuel processing.

Over the course of this project, model fuels were passed through a catalyst and examined to assess changes in chemical composition and resistance to autoignition. These fuels were comprised of combinations of *n*-hexadecane, 2,2,4,4,6,8,8-heptamethylnonane, mesitylene, decalin, and 2-octadecene, and were selected because they are representative of those found in both gasoline and diesel fuels. By limiting the number of chemicals in the fuel, it is easier to assess changes that occur as a result of the catalytic process carried out on the fuel.

Catalysts were produced from precursors loaded on a Cordierite substrate. Model fuels were then passed over the catalyst under varying conditions such as flow rate, reactor temperature, and catalyst loading. The resulting fluids were analyzed using gas chromatography (GC) using a mass selective detector (GC-MS), and ignition quality testing (IQT).

A comparison of GC-MS chromatograms of the model fuel and the reacted fuel showed the products resulting from the decomposition of the surrogate fuel components. The quantities and compositions varied based upon the model fuel tested and the reaction conditions. Those products that could be identified were associated with fragments of the

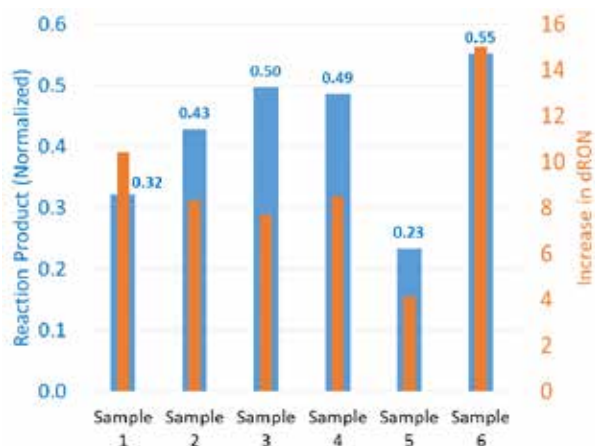


Chart showing the change in RON achieved by sample in comparison to the RON of the original surrogate fuel. Changes in RON are also compared to changes in peak area of a product peak observed in the respective GC chromatogram.

fuel components. Many other products were present, but could not be positively identified.

IQT measurements were carried out according to ASTM D6890. IQT measures ignition delay, providing an indication of the susceptibility of the fuel to autoignition. Ignition delay is mathematically related to derived cetane number and RON. The results of these tests showed increases in RON from

4 to 15 units, depending on the model fuel and reaction conditions. These results very loosely correlated with the normalized quantities of some of the observed reaction products. For other products there was no obvious correlation.

Overall, proof-of-concept was demonstrated for a system that could increase the RON of a currently available commercial fuel in response to changing engine demands.

Localized Surface Plasmon Resonance Spectroscopy, Microscopy, and Sensing

Patrick Z. El-Khoury

This project combines nanoscopic (chiefly surface- and tip-enhanced Raman) and theoretical (static and dynamic quantum chemical simulations) tools to probe the interplay between a single molecule and its local environment.

Efforts to reach the ultimate detection limit (i.e., the sensitivity required to probe 1.66 yoctomoles [$1/N_A$]) of a substance rendered the art and science of engineering plasmonic nanojunctions popular over the past few years. This is a consequence of the extreme localization of electromagnetic fields between plasmonic nanostructures, whereby the detection of the feeble optical response of a single scatterer is feasible. The effect is most evident in surface- and tip-enhanced Raman scattering (SERS and TERS, respectively) from molecules coaxed into plasmonic junctions. Through this project, a combination of experiment and theory is aimed at understanding the operative mechanisms that govern optical spectroscopy at plasmonic nanojunctions. The acquired knowledge is then used to rationally design and fabricate ultrasensitive plasmonic sensors.

Our FY 2013 works revealed that inelastic photon scattering at plasmonic junctions not only retrieves molecular polarizabilities, but also broadcasts intimate details about the environments in which the molecular scatterers reside, as well as the interplay between molecules and plasmons. We employed tools of density functional theory to compute the Raman spectra of 1,3-propanedithiol molecules in various media. A remarkable agreement between experiment and theory not only allowed for reliable spectral assignments, but also introduced an intuitive theoretical framework that accounts for commonly observed phenomena in the single molecule realm (e.g., spectral fluctuations).

Throughout FY 2014, we built on and expanded the concepts set forth in the previous year. Four key reports were published in the *Journal of Physical Chemistry Letters*, *Journal of Chemical Physics*, *Journal of Physical Chemistry A*, and *Nano Letters*. The findings from the first and last reports are of particular relevance to the directions that were undertaken in FY 2015. In the first report, we combined atomic-force-

microscopy-based TERS point spectroscopy with the single molecule framework developed in FY 2013 and illustrated how time and frequency-dependent intensity variations in TERS spectroscopy can be uniquely assigned to molecular reorientation. In the last report, our findings identified gate-way molecular modes for mediating charge shuttling across a plasmonic gap.

In FY 2015, we further developed our TERS nanoscale chemical imaging capability and performed multi-modal imaging of novel plasmonic substrates using the suite of cutting-edge microscopy tools housed within EMSL at PNNL. Four key reports were published in the *Journal of Physical Chemistry C*, *Journal of Chemical Physics*, *Faraday Discussions*, and *Nano Letters*. In the first report, our results concluded that nanoscale TERS chemical images map the interaction between vibrational mode-dependent polarizability tensors of dimercaptostilbene (DMS) and enhanced electromagnetic fields at a plasmonic tip-surface nanojunction. The recorded TERS chemical images also provided insights into specific molecule-metal interactions; each pixel can potentially be used to broadcast intimate details about the local environments in which molecules reside.

In the second report, we investigated the plasmonic properties of a self-assembled 2D array of Ag nanospheres (average particle diameter/inter-particle separation distance of 9/3.7 nm). Combined with modest chemical enhancement factors, this study paved the way for reproducible, single molecule signals from this easily self-assembled SERS substrate. In a follow-up study, we used the same substrate for single molecule SERS spectroscopy. Taking observed chemical effects, as well as molecular orientation into account, we were able to estimate a single molecule enhancement factor of $\sim 10^{10}$ in our measurements.

In our last FY 2015 report, we reported tip-enhanced Raman imaging experiments in which information on sample topography and local electric fields was simultaneously obtained using our all-optical detection scheme. We demonstrated how a Raman-active DMS-coated gold tip of an atomic force microscope can be used to simultaneously map the topography and image the electric fields localized at nanometric (20 and 5 nm-wide) slits lithographically etched in silver.

In addition to the aforementioned publications, several collaborative works within the broader scope of this project resulted in peer-reviewed applications in FY 2015, in which partial support from PNNL LDRD was acknowledged. The principal investigator (PI) delivered a PNNL Physical Sciences Division Seminar; a talk at the Dynamics, Interactions & Electronic Transitions at Surfaces 2014 conference (and chaired the “Plasmonics & Nanoplasmonics” session); and two additional invited chemistry department seminars (University of Puget Sound and University of California Irvine).

Five peer reviewed publications resulted from research endeavors supported through this project in FY 2016 (through the end of May 2016). The first two (*Journal of Physical Chemistry B* and *Journal of Physical Chemistry A*) are theoretical in nature and were performed in collaboration with EMSL’s NWChem group. In the first report, we recognized that, although carbon-hydrogen (C-H) vibrational modes serve as key probes in the chemical identification of hydrocarbons and in vibrational sum-frequency generation spectroscopy of hydrocarbons at the liquid/gas interface, their assignments still pose a challenge from a theoretical viewpoint. As such, we reported a detailed study of the C-H stretching region of a model system using a new *ab initio* molecular dynamics (AIMD) module that was implemented in NWChem, and the code is now freely available to the global NWChem user community. Comprehensive anharmonic normal mode analysis of the C-H stretching region cast doubt upon previous experimental assignments of the shoulder on the symmetric C-H stretching peak.

In the second report, we simulated the spectroscopic properties of short-lived thermal and photochemical reaction intermediates and products. The latter is a challenging task, as these species often feature atypical molecular and electronic structures. Moreover, the complex environments in which such species typically reside in practice add further complexity. This problem was tackled *in silico* using the same AIMD module described above. We simulated the ultraviolet- (UV)-vis and infrared (IR) spectra of iso-CHBr₃ in the gas phase, as well as in a Ne cluster (64 atoms) and in a methylcyclohexane cage (14 solvent molecules) representative of the previously characterized matrix isolated and solvated iso-CHBr₃ species. We exclusively performed fully quantum mechanical static and dynamic simulations. By comparing our condensed phase simulations to their experimental analogues, we stressed the importance of 1) conformational sampling, even

at cryogenic temperatures; and 2) using a fully quantum mechanical description of both solute and bath to properly account for the experimental observables.

The PI also was invited to write two different review articles of works performed and supported through this project. The first review in *Advanced PhysX* considered greatly informative, yet significantly underrated, signatures of single molecules. Ultimately, this review article outlined the future research directions that will be undertaken by the PI throughout several years to come. The second invited review article in *Analyst* will be part of the 2016 *Analyst* themed issue on emerging investigators and highlighted different techniques used to visualize the enhanced electric fields associated with SPs. This review article illustrated how SPs can be visualized at the ultimate limits of space (sub-nm) and time (sub-fs).

In the last FY 2016 report, published in the *Journal of Physical Chemistry C*, we demonstrated some results obtained using a novel imaging capability currently under further development at PNNL. The arrangement consists of a multimodal (absorption/dark field scattering/fluorescence) near-UV-Vis-near-IR hyperspectral optical microscopy setup. We demonstrated how the centers and edges of individual particles can be localized in 2D to within a single pixel (85 nm²), using a statistical method for examining texture based on a co-occurrence matrix. Observations of the scattering spectra of 31 different nanospheres are suggestive of nanometric structural variations over length scales much finer than the spatial resolution attainable using the all-optical technique described. The target systems that can be potentially addressed using this all-optical technique span the realms of several disciplines. In this regard, nonconventional approaches to bioimaging are possible using this unique capability, and it is the concept behind two different proposals that are currently under development, *vide infra*.

The technologies and advances achieved through this project in FYs 2013 through 2015 were leveraged to tackle novel, high-risk, high-reward research directions in FY 2016 and provided the basis for several grant proposals.

Besides more than 20 publications (to date) and the development of novel imaging techniques at PNNL (TERS and hyperspectral optical microscopy), this project has led to several novel research directions that will be further pursued by the PI (at PNNL) for years to come.

Low-Scaling Electronic Structure Methods for Accurate Modeling Chemical Transformations in Complex Environments

Bo Peng

This research is focused on developing low-scaling approximations and parallel algorithms for the expensive, yet accurate, coupled-cluster (CC) method to achieve the microscopic, molecular level characterization and understanding of chemical transformations during the process of photocatalysis, photovoltaics, climate change, and metabolism.

Among all available theoretical methods, the CC model, as one of the wave-function-based *ab initio* methods, is capable of providing very accurate results by proper inclusion of many-body effects. In particular, the CC model with single and double excitations corrected by perturbative triples, a.k.a. CCSD(T), has been recognized as the “gold standard” for high-accuracy computational chemistry. However, this method always suffers from very high computational cost due to expensive tensor contractions (e.g., canonical formulation of the CCSD(T) approach scales as $O[N^7]$ with N representing system size). The expertise of PNNL in the development of quantum chemical methodologies and the cutting-edge computational resources provide a unique platform to tackle this challenge. Employing the local character of the correlation effects and applying recent advances in applied math with efficient parallelization strategies can significantly shift the system-size limit tractable by accurate CC methods. We envision that our CC simulations will be enabled for systems consisting of hundreds/thousands of light atoms and lay the foundations for extension of local CC approaches beyond the ground-state applications, including description of electronic excited states and properties using linear-response CC formulations.

The CC module in the parallel infrastructure of NWChem open-source software, when running on the Cascade computing cluster, has shown an almost linear parallel scaling for the most computationally expensive N^7 part of CCSD(T) calculations.

Recently, we applied the extensions of the equation-of-motion CC module to accurately calculate the ionization potential (IP) and electron affinity (EA) of a series of carbon nanotubes consisting of hundreds of light atoms. Based on the accurate IP and EA values, we demonstrated that the electronegativity of the more relativistic carbon nanotube systems remains, to a large extent, independent of nanotube length.

Secondly, by rewriting the CC equations in terms of low-rank tensors generated by employing pivoted Cholesky decomposition on integral tensors, we successfully obtained a new working equation of CC with doubles (CCD). This new working equation shows a natural compatibility, in comparison with the conventional equation, with the high parallelism, which will facilitate an efficient parallel coding in the near future.

Thirdly, we successfully derived the CC formulation of Dyson equation, or Green Function CC, as well as the analytic form of its derivatives, with respect to the frequency. Not only can this formulation accurately describe the propagation of a “particle” added to the ground state of the molecular system, but it also provides an efficient way to accurately compute the self-energy of the “particle” and its derivatives, with respect to the frequency, which is essentially related to the coupling between the “particle” and the system.

In the next year, we are going to implement the new working equation of CCD and extend this low-rank factorization to the more accurate CCSD model, as well as its equation-of-motion and linear response variations. In this level, we will then be able to calibrate other, more ambitious approximations. In the meantime, applications of this new implementation for the ground state and excited state of large molecules will be carried out to test the performance of the new factorization in comparison with the conventional procedure, and with high expectation, to obtain some meaningful scientific results.

Making, Measuring, and Modeling Materials for Quantum Computing

Nathan A. Baker

The goal of this project is develop a basic science understanding of quantum computing materials and the tools and techniques that can be applied to many of the most promising qubit architectures.

Materials science has a well-established approach for the design of new materials and improvement of existing materials represented as the “make, measure, model” mnemonic.

Make. To make, the existing PNNL materials synthesis capabilities demonstrate the preparation of quantum computing (QC) materials by preparing nanostructured QC materials through focused-ion beam methods; improving the fabrication of QC devices through post-processing to remove photoresist residue from delicate device features.

Measure. Existing PNNL chemical imaging capabilities will demonstrate the characterization of QC materials by developing novel sample preparation methods that enable analysis of unique and delicate structures for materials of interest to the QC field; developing new methods in z-contrast scanning transmission electron microscopy (STEM) to characterize structures of interest to QC; and using atom-probe tomography (APT) to understand the structure and chemical composition QC structures.

Model. Existing PNNL mesoscale materials modeling and mathematics capabilities will be developed in the simulation of QC materials by evaluating the impact of materials defects on the performance of topological QC paradigms.

From this broad continuum of technical focus areas, PNNL will develop not only a basic science understanding of the QC materials but also the tools and techniques that can be applied to many of the most promising qubit architectures (i.e., quantum dots, defects in solids, and superconducting Josephson junctions).

The task of developing a supercritical fluid-based wafer cleaning capability was divided into two parts. The first was to design, build, and test a modular supercritical fluid system that could be easily adapted to suit different cleaning procedures. The other part of this effort was to establish a cleanroom capability that would provide a means of patterning,

etching, and depositing aluminum features on silicon wafers. Samples could then be generated with a hard-baked photoresist in different ways and used to gauge the performance of the new cleaning system.

A high pressure system capable of generating conditions needed for supercritical fluid media was successfully built with a dual-line configuration of syringe pump, mixing vessel, and cleaning chamber. This parallel construction affords the ability to operate at two different pressures simultaneously. Samples could be soaked at lower pressure ranges and sprayed with higher pressures of supercritical fluid. The mixing vessels in line with both manifolds provided an effective dissolution of stripping agents in the supercritical fluids and acted as reservoirs for either filling or spraying fluid into the cleaning chamber. Digital readouts for pressure and temperature were installed across the system so that precise cleaning conditions could be logged over time for later replication with a commercially available system. Protocols for spin-coating and UV exposure of I-line resist were developed and tested for use with the evaporative deposition of micron-sized aluminum features. Masks that provided thin, interdigitated structures were chosen as a model template for cleaning studies. The closely connected features hindered subsequent lift-off and were found to contain residual resist, which are ideally suited for cleaning studies. These wafer samples are being actively used to evaluate and improve the performance of the supercritical fluid cleaning system for work in FY 2016.

For the measure aspect of the project, the majority of research transitioned to an externally funded follow-on project. The modeling research specifically focused on topological phases of matter, how defects affect these phases, and how these phases relate to quantum measurement and computing. Much of this work is ongoing and will be the focus of FY 2016 efforts.

Our team worked to develop new bounds for the Egyptian fraction problem with few prime factors, an important mathematical problem dating back thousands of years with applications in electrical engineering, geometry, and topological phases of matter. The previous best known bounds were exponential, whereas ours are linear. This dramatic improvement should allow us to classify integral modular categories

through rank 15, of broad interest to the mathematical community and vital to the study of topological quantum computation. Additionally, it is a necessary step for understanding how the disorder affects topological phases. This work will enable new studies related to disorder and computational power in FY 2016.

A collaboration was initiated to understand the effects of non-standard lattices in models for topological quantum computing. Preliminary studies showed that these non-standard lattices can improve the robustness of the system in the presence of noise and defects. It is believed that the lattices may also have implications in wave guides and meta-materials. We have also begun to investigate how the mathematics used to study topological phases of matter applies to nucleotide pseudo-knots. This work is ongoing and promises to have implications in DNA origami, nucleotide conformation, and the behavior of amino acid chains in thermal environments. The team has worked with external researchers to understand how fermions can be incorporated into certain models for topological phases of matter.

Additional team members have explored the implications of new combinatorial approaches to optimal quantum measurements. There has been investigating into the relationship

between symmetric informationally complete positive operator valued measures, mutually unbiased bases, and mutually unbiased subspaces. Combinatorial techniques have been brought to a classical algebraic/geometric problem. Another collaboration was formed to understand the algebraic building blocks of topological phases of matter that succeeded in understanding the algebraic properties of loop braid groups that have applications in $(3 + 1)$ -dimensional topological quantum field theory, classifying all topological phases of matter that have five particle species, classifying topological phases to understand how they behave in the presence of disorder, and showing that there are only finitely many pre-modular categories of fixed rank. In ongoing work, we are continuing to study classes of categories that describe gauged symmetry (and disorder) and how one can incorporate fermions back into a bosonized quantum field theory.

The above work has resulted in five papers currently under review, six papers in preparation and contingent on ongoing research, a special session at the Joint Mathematics Meetings, an accepted proposal for a special session at the 2016 AMS Spring Western Sectional Meeting, three new hires, and four conference/workshop invitations.

Molecular Fingerprint of ANFO Detonation

Carlos G. Fraga

Our goal is to discover novel chemical signatures that may exist after the detonation of an ammonium-nitrate(AN)-based explosive that may better identify the type and origin of explosive material used in an attack.

AN is an oxidizer widely used in improvised explosive devices (IEDs) and homemade explosives (HMEs). The chemical signatures in these materials may help investigators locate those responsible for IED and HME attacks. In this project, we are simulating the detonation of AN-based explosives through the thermal decomposition and combustion of both AN individually and AN fuel mixtures, followed by the characterization of their decomposition and combustion products in a laboratory environment.

Prior work demonstrated that numerous molecular species are released or produced from the thermal decomposition of blasting-grade AN with and without a hydrocarbon fuel. Most of these molecular species have not been previously reported and may be useful for determining the source of AN or for identifying an AN fuel explosion after detonation. The thermal decomposition and combustion of minute amounts of AN and AN fuel mixtures are being conducted using a pyrolysis unit capable of instantly heating a sample up to 1,000°C. Gas chromatography/mass spectrometry (GC/MS) is then used to identify and quantify the molecular products produced from AN samples. Our objective is to use signature discovery principles and tools to discover forensic molecular products from the thermal decomposition and combustion of AN and AN fuel mixtures.

Our previous work estimated that the thermal decomposition of milligram amounts of AN is an acceptable approximation for modeling AN-based detonations. Our work resulted in the detection of the main chemical products (e.g., N_2 , CO_2 , NO , N_2O) produced from actual AN-based detonations. Additionally, we tentatively detected previously unreported trace chemical compounds such as alkyl nitriles, nitrobenzene, chloroform, and chlorinated hydrocarbons.

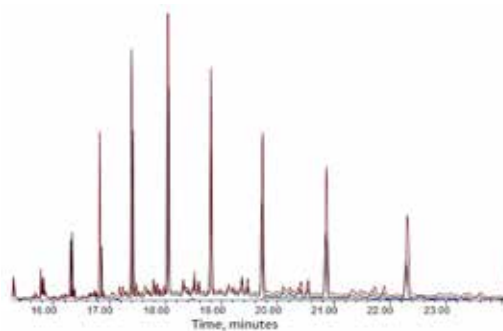
In FY 2016, we used the Signature Discovery Initiative approach “Fishing for Features” to analyze GC/MS data collected from the thermal decomposition of calcium ammonium nitrate (CAN) from two different CAN factories. CAN consists of 75 to 80% AN mixed with 22 to 25% dolomite or calcium carbonate. Less than 1% of the CAN is organic additives that act as anti-caking and anti-dusting agents. The heating of CAN to 250°C resulted in the decomposition of

AN and the detection of numerous organic compounds, presumably from the organic additives. For example, the GC/MS alkane band depicted in the included figure is attributable to paraffin oils commonly used as anti-dusting agents. Using classification trees and analysis of variance, the organic GC/MS signals for CAN were searched for any differences attributable to each factory; none were found. It is believed the same basic formulation is used by the two factories, given that they belong to the same company/manufacturer. In general, the GC/MS molecular signatures for CAN were consistent between the two factories.

In FY 2016, we also studied the effect decomposition temperature has on the molecular products detected from the thermal decomposition of AN. At 250°C, the main organic products detected in CAN were also detected in AN, pointing to a common manufacturing process used throughout the AN/CAN industry. For example, the alkane band depicted in the included figure was found in the analyzed CAN and AN fertilizers. The alkane band ended up being present at 350°C but absent at 450°C and 600°C. At temperatures above 450°C, the alkanes decompose to smaller components.

In addition to the alkane band, there were 200 organic compounds detected by the automated mass spectral deconvolution and identification system GC/MS software for AN heated to 250°C. One prominent group of detected compounds are siloxanes. The reason for their presence is yet to be determined.

In FY 2017, we intend to solicit external funding to look for molecular signatures released or produced at temperatures between 250°C and 350°C that may be useful for sourcing AN post detonation, for which there is a need. It is hypothesized that some of the 200 molecular compounds detected may prove useful for discriminating AN based on source after detonation, especially because actual AN detonations leave behind traces of non-combusted AN.



Paraffin oil GC/MS signature from the thermal decomposition of tannerite AN at 250°C using the pyrolysis GC/MS system. The depicted alkane band is present in all tested AN and CAN pills. The alkane band is not detected above 450°C.

Polymeric Ionic Liquids for Lubricants – A Breakthrough in Molecular Design

Lelia Cosimbescu

A new strategy is needed to reduce parasitic friction losses, resulting in greater fuel economy. By introducing multi-functionality in lubricant additives, this work will move us closer to an all-in-one additive that reduces lubricant complexity, negative effects associated with additive compatibility, and enables lubricant ease of use and disposal.

The project proposes a unique and innovative molecular design of a lubricant additive that will reduce the need and the amount of additives used in a formulated lubricant package, while further supporting the DOE mission of increasing fuel economy. The concept design combines polymer moieties, which are known to improve the viscosity index, with ionic liquid functionalities, which in turn, are known to be effective in reduction of friction and mechanical wear within an internal combustion engine (ICE). Viscosity index improvers (VIIs) are polymers that span various molecular designs and topologies. Conversely, anti-friction and anti-wear additives are small molecules generally comprised of soft metals with organic ligands or organic amphiphiles, which form dynamic interactions with contact surfaces promoting thin films. To our knowledge, no attempt has been reported to combine the benefits of ILs (ionic liquids) with VIIs to achieve a novel macromolecule with dual functionalities that address three major requirements in a lubricant: viscosity, friction, and wear (including scuffing).

In order to probe the design strategy, a co-polymer comprising two very different monomers had to be prepared. A polymethacrylate hybrid, combining a lipophilic monomer and an ionic monomer, with a well-defined architecture, is essentially unreported. Therefore, the main challenge of this project became synthesizing such a compound. Several synthetic strategies were tackled in an effort to prepare the desired diblock copolymer.

In the first, atom transfer radical polymerization (ATRP) and reversible addition-fragmentation chain transfer (RAFT) polymerization techniques were employed, in which the lipophilic monomer was added first, followed by the addition of the polar monomer after a few days. This approach led to a

low incorporation of the polar monomer (due to solubility differences of the reactants); post-modification of the amine group of the copolymer, to prepare the ionic liquid, was not successful. The second attempt, involving the direct copolymerization of the two monomers under ATRP or RAFT conditions, failed. And a third approach focused on free radical polymerization with azobisisobutyronitrile initiator present as the only successful route to preparing hybrid polymers. In this case, however, only random topologies were prepared.

In order to partly achieve our goal, the third methodology was implemented in synthesizing copolymers containing ionic liquid moieties. Three analogs were prepared via free radical polymerization: one containing 20% of an amine moiety (Analog 11), one containing 20% of ionic liquid moiety (Analog 29), and one containing only 10% ionic liquid (Analog 33). This small library of compounds allowed the preliminary assessment of the effect of the ionic liquid moiety on friction and wear. The compounds were dissolved in Yubase 4 oil at a concentration of 3.5% and their viscosity performance was evaluated. The viscosity indices (VIs) were lower than expected, which might have been in part due to a lower molecular weight (around 100 kDa). The fact that the polymer was soluble in a base oil, despite its high ionic liquid content is an accomplishment in itself.

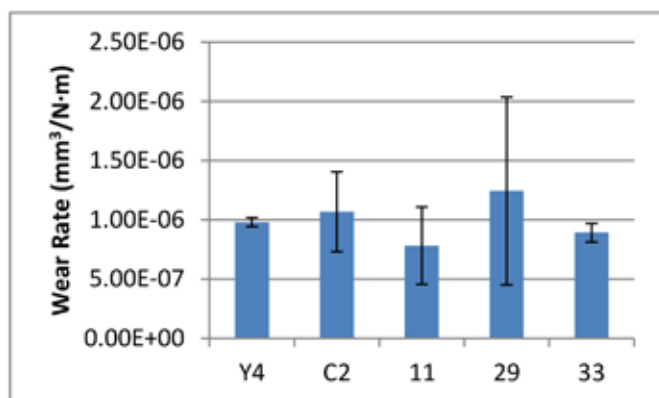
Viscosity data for all analogs tested (KV = kinematic viscosity at 40 and 100 °C, respectively; VI = Viscosity Index).

Analog	Composition	Type	KV 25 (cSt)	KV 40 (cSt)	KV 100 (cSt)	VI
11	20% amine	random	44.39	25.19	5.78	184
29	20% IL	random	40.90	23.66	5.29	166
33	10% IL	random	42.53	24.09	5.52	179

The analogs were evaluated by our collaborators at Oak Ridge National Laboratory, and the data revealed the following unexpected results: the wear rate is lowest for the amine copolymer, analog 11, followed by the copolymer with least IL content, Analog 33, then followed by Analog 29. The coefficient of friction follows the same trend. These are interesting findings that provide a glimpse of the chemistry that might happen at the surface in the friction boundary regime (100°C, load of 100N). It appears that the topology of the polymer provides an unfavorable interaction with the surface if a large polar segment (block) is not present. In addition, the

ammonium is too shielded by the large bis(trifluoromethane) sulfonamide (TFSI) counter anion. Another source of the enhanced wear might be the hydrolysis of the TFSI anion and subsequent generation of the acid form, HTFSI, which is very corrosive. One of the benchmarks in this study was a commercial polymer (C2), which was used to gauge the performance of our materials against industry standards. The data shows that our materials have comparable or better wear performance than the commercial control in a bench test.

The most significant accomplishment is that this is the first synthesis of a truly lipophilic polymer containing ionic liquid moieties, and it is its first application as a lubricant additive. Indeed, much work is needed to achieve a diblock topology and investigate other ion pairs, which are less sterically crowded, and other kinds of more reactive monomers. The ultimate goal is to achieve the diblock topology originally proposed, which enables a strong interaction of the IL with the metal surface and subsequent film formation.



Wear data at 100°C and 100N; Y4 blank base oil; C2 is a commercial benchmark.

Predictive Understanding of Self-Assembly: Particle-Mediated Growth

Kevin M. Rosso

We are developing a fundamental understanding of particle-mediated growth of crystalline materials from aqueous solution. The primary goal is to develop appropriate new theory that identifies and encompasses the relevant chemical physics and couples length and time scales through appropriate projection schemes.

Crystal growth by assembly of small particles or clusters, as opposed to ion-by-ion, is a recently recognized type of growth impacting our understanding of natural mineralization processes and holds promise for novel materials design. Oriented aggregation (OA) is an important case where larger crystals grow by the oriented self-assembly of smaller ones, typically nanoparticles. Observations show that approaching particles rotate to achieve co-alignment while separated by nm-scale solvent layers. Little is known about the forces that drive co-alignment, particularly in this “solvent-separated” regime.

This project will ultimately provide a foundation for predictive simulations on particle-mediated growth systems for comparison with self-assembly outcomes at various scales. An existing popular theory to describe energetics between two surfaces considers a simple balance between attractive and repulsive electrical double-layer forces, where parameters are strongly correlated with microscopic features like molecular structure and electron distribution. However, a complete understanding requires correlations over differing length scales in the energetics between two charged surfaces in aqueous electrolytes.

In FY 2016, we were successful in developing oriented ZnO(001) nanoprobe and in measuring rupture forces between co-planar ZnO(001)-aqueous solution-ZnO(001) ternary interfaces *in situ* using dynamic force spectroscopy (DFS). DFS using nanoengineered single-crystal, single-face force probes reveals an oscillatory attractive force with azimuthal co-alignment from 2.97×10^5 to 1.02×10^6 N m⁻² exhibiting 60° rotational periodicity. Classical molecular-dynamics-based potential of mean force simulations as a function of separation distance and azimuthal angle show several attractive free-energy wells distinguished by numbers of intervening water layers and that this number reaches a minimum at azimuthal alignment. The calculated activation energy to separate the attractively bound solvated interfaces shows the same 60° periodicity as in the force measurements, revealing

the key role of intervening water structuring as a basis to generate an interparticle torque when nearly co-aligned.

We were also successful in isolating and simulating the dispersion interaction between two model metal oxide nanocrystals in vacuum. Mutual lattice orientations dictate the types and magnitudes of forces between crystalline particles. When lattice polarizability is anisotropic, the van der Waals dispersion attraction can, in principle, contribute to this direction dependence. Using a combination of transmission electron microscopy and atomic-force microscopy technologies, we performed direct measurement of this attraction between rutile nanocrystals, as a function of their mutual orientation and surface hydration extent. At tens of nanometers of separation, the attraction is weak and shows no dependence on azimuthal alignment nor surface hydration. At separations of approximately one hydration layer, the attraction is strongly dependent on azimuthal alignment and systematically decreases as intervening water density increases. Measured forces were in close agreement with predictions from Lifshitz theory and show that dispersion forces are capable of generating a torque between particles interacting in solution and between grains in materials.

We also developed a molecular-to-mesoscale model to assess the role of ion correlation forces on OA. Ion correlation forces arise from dynamically interacting electrical double layers and can possibly explain face-selective attraction and particle rotation into lattice co-alignment from long range. Classical density functional theory simulations developed and performed for the archetype OA system of anatase TiO₂ nanoparticles in aqueous HCl solutions show that face-selective attraction from ion correlation forces outcompetes electrostatic repulsion at several nanometers apart, drawing particle face pairs into a metastable solvent-separated captured state. Analysis of electrolyte and pH dependence of interparticle interactions is in quantitative agreement with observed decreasing frequency of attachment between (112), (001), and (101) face pairs, with an adhesion barrier largely due to steric hydration forces from structured intervening solvent. More generally, they explain the driving forces for particle capture, the origin of facet selectivity and alignment forces in the solvent separated state, and barriers to attachment. In particular, the simulations revealed that the main attractive contributions to the interactions between nanoparticles are ion correlation forces with a typical range of 0.5–6 nm, static (zero frequency) van der Waals interactions between anatase particles across aqueous solution with the range of 0.5–10 nm for low concentration electrolyte and 0.5–2.5 nm for electrolyte concentrations above 1 mM, and short-range ion-surface dispersion interactions significant in the 0.75–1.5 nm range of interparticle distances.

Scalable Synthesis of Spinel Stabilized Metal Catalysts

Yong Wang

We are identifying, investigating, and developing new kinds of stable, active, supported metal catalysts that could be used by various industrial applications such as selective oxidation reactions and vehicle emission catalysts components.

Recently, we reported that MgAl_2O_4 support with controlled (111) facet can stabilize platinum catalysts under harsh reaction conditions. Conventional approaches via mechanical mixing of magnesia and alumina or co-precipitation of magnesium and alumina hydroxides typically lead to significant inhomogeneity in the amorphous mixture of magnesia and alumina, and consequently, in forming the spinel phase with inhomogeneity in local composition, structure, and morphology. We previously showed that MgAl_2O_4 , synthesized via a novel non-aqueous approach is capable of stabilizing platinum particles during severe aging at 800°C. We revealed that the superior stability is due to strong interactions between spinel surface oxygen and epitaxial platinum {111} facets. However, the approaches to synthesize MgAl_2O_4 spinel need more detailed investigation and understanding. For example, the solvent effect during synthesis and the surface properties of the formed products, as well as their relationship, need to be understood before scaling up the synthesis technology.

Our objective is to identify and develop a scalable synthesis approach of spinel MgAl_2O_4 via the novel non-aqueous approach and to understand the fundamental mechanism involved. Well-defined spinel MgAl_2O_4 can provide a base for ultra-stable metal catalysts. These catalysts have attractive hydrothermal stability and have potential applications in several important industrial reactions such as biomass conversion and vehicle emission abatement. Ultimately, we expect to have a synthesis pathway identified and developed that can produce homogenous, hydrothermal stable, and inexpensive MgAl_2O_4 catalyst support, as well as its supported metal catalysts.

In the past 2 years, we have identified that, thermodynamically, the most stable surface orientation of spinel MgAl_2O_4 is the {111} facet. Stabilization of precious metals, such as platinum, on the support is due to the following two main factors:

1) strong interaction between oxygen terminated spinel surface and epitaxial Pt {111} facets; and 2) spinel surface properties such as acidity/basicity, surface charge, and surface composition, etc. The first factor determines the stability of the supported metal particles, while the second affects the initial dispersion, as well. The ball milling technique of synthesis precursors in aqueous solution has been demonstrated as being able to reduce the synthesis (especially in large quantities) cost and achieve similar homogeneity as a novel non-aqueous synthesis approach based on various techniques like nuclear magnetic resonance, Brunauer-Emmett-Teller, X-ray powder diffraction (XRD), $\text{NH}_3(\text{CO}_2)$ -TPD, zeta-potential, thermogravimetric analysis, and discrete Fourier transform calculation.

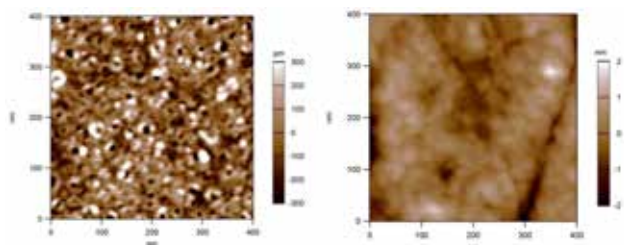
Recently, two single crystal MgAl_2O_4 {111} and {100} were loaded with platinum and treated under high temperature up to 800°C. It was verified that MgAl_2O_4 {111} is able to stabilize platinum particles well. Different solvents and precursor compositions away from Mg:Al stoichiometric were studied in order to optimize the ratio between {111} and {100} of the spinel support. A method for quantitatively measuring the preferred orientation toward the (100) and (111) planes by using XRD results has been introduced.

The search/match function in Jade (v.9.6, Materials Data Inc., Livermore, CA) XRD analysis software was utilized to determine which MgAl_2O_4 PDF pattern provided the best fit. The preferred orientation correction (G) was calculated using the March function (Eq. 1) from the Calculate Pattern function of Jade.

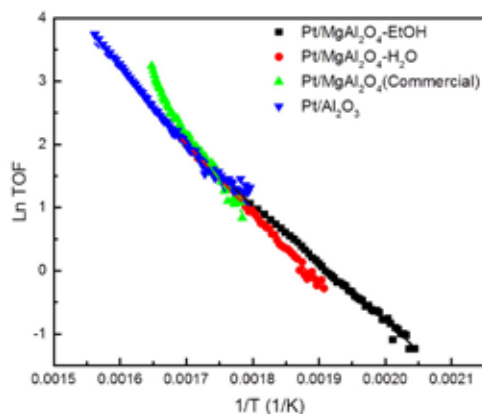
$$G = \left(C^2 \cos^2(\alpha) + \frac{\sin^2(\alpha)}{C} \right)^{-1.5}$$

Where C is the preferred orientation parameter. Complete randomness is represented by C=1 and decreasing C indicates increasing preferred orientation toward the plane. Based on XRD results, C was calculated. Our results show that for MgAl_2O_4 prepared using different solvents, samples prepared in H_2O exhibit more preferred orientation toward the (100) plane than samples prepared in isopropanol (IPA) and ethanol. On the other hand, when using IPA as solvent, higher Al/Mg ratio results in a lower degree of preferred orientation toward the (100) plane.

For FY 2017, ball milling technique, optimizing Mg/Al ratio in precursor, and solvent effect will be studied in parallel to finalizing the development of this low-cost process of large-scale spinel synthesis. During this finalization of the development, several scientific questions will be answered, as well, such as how to maximize the (111) facet of the spinel support, and how to load more precious metal onto the (111) plane instead of other facets during the initial impregnation step.



Pt sintering (~100nm particles) with (100) facet vs. stable small particles with (111) facets, after aging 24hr at 800°C.



Arrhenius plot of CO oxidation reaction of Pt catalysts. Pt/MgAl₂O₄ behaves the same as Pt/Al₂O₃ in CO oxidation reaction, which is an important reaction in vehicle emission abatement.

Sequence-Defined Polymers Based on a New Backbone Architecture

Jay W. Grate

This project is investigating a novel, bottom-up, self-assembly approach to achieve a controlled design and engineered polymer/nanostructure composites.

Sequence control in natural biopolymers, such as DNA and proteins, leads to tremendous structural diversity and functionality, such as material architectures, biocatalysis, molecular recognition, and transport across membranes. Realizing the same functionality in stable synthetic polymers will revolutionize materials science. Achieving this challenge will require both experimental synthesis and measurement of new polymer architectures with sequence control, as well as molecular simulations to predict and understand macromolecular behavior.

Epitomized in nature by polypeptides and poly(nucleic acids), sequence-defined polymers are composed of a multiplicity of monomers, each monomer distinguished from one another by having a different side chain, and the various monomers are sequenced into the polymer in a predetermined order. In nature, sequence-defined polymers create biomaterials, encode information, perform biocatalysis, participate in molecular recognition, and shuttle species across membranes. More stable sequence-defined polymers prepared by chemical synthesis likewise have the potential to create tremendous structural diversity and functionality based on monomer sequence. Such polymers will revolutionize materials science. The vast majority of synthetic sequence-defined polymers are, to date, either laboratory examples of the natural sequence-defined biopolymers, or close analogs based on similar structural units and bond-forming reactions. For example, pseudopeptides and peptoids both rely on amino acid structures and peptide bonds by analogy with polypeptides.

In this project, we have invented, patented, and published a new class of stable, synthetic, sequence-defined polymers. These new macromolecules are based on molecular precursors prepared from cyanuric chloride and assembled into polymers by a solid phase synthesis method. These new sequence-defined polymers have no precedent in nature and little precedent in chemistry literature; we call these triazine-based polymers (TZPs).

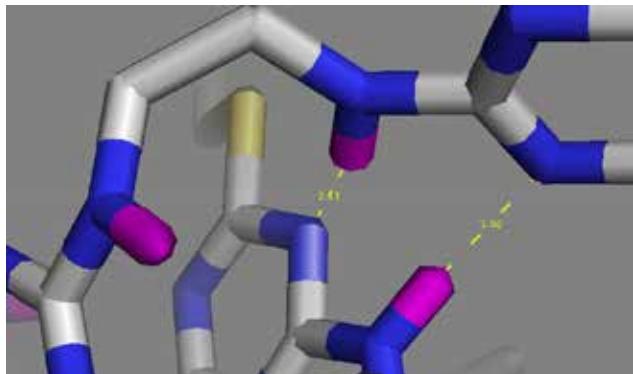
We successfully demonstrated the synthesis of molecular precursors that have a diversity of side chain structures. In addition to aliphatic and aromatic groups, we have created side chains with protected carboxylic acid and amine groups,

which lead to ionizable side chains after deprotection. In addition, monomers have been prepared with protected thiol groups, leading to polymers with thiol side chains. In these ways, the side chain diversity typical of peptides and proteins can be achieved in these new synthetic macromolecules. Molecular precursors with side chains have been sequenced into macromolecules using a submonomer solid phase synthesis technique that we developed for these TZP materials. The predetermined sequences range from arbitrary defined sequences to block copolymers, all of defined length.

Several methods were used to characterize the new polymers to prove that the sequences had been achieved. Proton nuclear magnetic resonance demonstrated that the side chain content was as planned in the synthesis, and tandem mass spectrometry methods proved that the monomers were, indeed, in the planned sequence.

In parallel with the experimental research, molecular dynamics simulations have been carried out that show intriguing patterns of noncovalent interactions between monomer units of the polymers. In single macromolecule studies, the molecules folded into nanorods stabilized by organized arrangements of hydrogen bonds and pi-pi interactions. These structures bear similarities to peptide beta sheets or helices and are, thus, biomimetic. In addition, simulations on pairs of molecules show similar patterns of hydrogen bonding, leading to nanorod structures.

Because these simulations demonstrate patterns of noncovalent interactions, they suggest that TZPs will lead to functional macromolecules and self-assembled materials. This work has demonstrated the following three key features in developing new sequence-defined polymers: 1) side chain diversity, 2) synthesis of macromolecules with monomer units in defined sequences, and 3) noncovalent interactions that can lead to intra- or intermolecular interactions and, hence, conformational behavior or self-assembly.



Paired hydrogen bonds between monomers in a triazine polymer conformation.

Spectrally Resolved Nanoscale Imaging of Single Molecules, Plasmons, and their Interaction

Patrick Z. El-Khoury

This project will enable the development of ultrasensitive multimodal (bio)chemical imaging technologies that may be used to interrogate molecules and metallic nanostructures, as well as to advance our understanding of molecule-plasmonic metal interactions. The potential applications of the envisioned technologies are far-reaching and span the realms of several scientific disciplines.

Recent advances in nanophotonics pave the way for advancing our fundamental understanding of processes of particular relevance to the energy sciences, including charge delocalization, transfer, and transport in hybrid molecular-metallic constructs. The need to develop a detailed understanding of these elementary processes is recognized in the 2015 DOE Basic Energy Sciences Advisory Committee report, titled, “Challenges at the Frontiers of Matter and Energy,” in the following excerpt: *The vision of this proposal is to understand, predict, and ultimately control energy flow within and across molecules interacting with plasmonic metal nanostructures. Prerequisite to fulfilling this vision is the development of novel multimodal imaging technologies that may be used to independently address molecules and plasmonic metals.* As we will illustrate, the technologies that will be developed through this proposal have applications in fields as diverse as ultrasensitive chemical detection and nanoscale chemical imaging, photocatalysis, and biological imaging, to name a few.

While major efforts in the field have been devoted to increasing the detection limits in linear and nonlinear optical microscopy, works addressing the fundamental mechanisms enabling ultrasensitive nanoscale optical spectroscopy are scarce. Molecule-plasmon interactions aside, it is almost surprising that the first report illustrating the modified optical absorption of molecules on plasmonic nanoparticles wasn’t published until 2016, in *Nature Photonics*, some 20 years after the first reports of molecular detection through surface-

enhanced Raman scattering (SERS). The recent work, albeit limited to colloidal solutions, questions the resonance conditions in several decades of (resonant) SERS measurements, knowledge of which is prerequisite not only to rationalizing molecular vibrational spectra, but also, for example, to understanding photochemical transformations near plasmonic nanoparticles. The need for novel imaging approaches that not only rely on vibrational contrast (e.g., Raman scattering or infrared absorption), but also on electronic contrast (e.g., ultraviolet-visible light absorption and scattering) presents itself in this context.

Although this project was only initiated on in June 2016, several accomplishments over the three-month period are noteworthy. The first is a technology-developed effort, enabling spectrally (377-1051 nm, $\Delta\lambda \approx 5$ nm) and spatially (diffraction-limited, sampled at 42 nm²/pixel) resolved fluorescence, dark field, as well as optical absorption microscopy measurements. A proof-of-principle report is currently under review, describing prototypical measurements targeting a variety of model systems, including 1) single fluorescent nanospheres, 2) single plasmonic nanoparticles residing in varying local environments, 3) a lipid (18:1 Liss Rhod PE) monolayer, and 4) an isolated, live *Tisochrysis lutea* cell. Whereas item 2 of this list is directly relevant to the science theme of this proposal, item 4 took advantage of hyperspectral optical absorption microscopy to identify and image algal pigments within a single live *Tisochrysis lutea* cell. Specifically, native fluorophores (carotenoids and chlorophylls) were addressed intracellularly by relying on the ground to excited electronic state transitions in these chromophores. The latter concept alone led to the development of several proposals in collaboration with our PNNL colleagues.

Additionally, we were able to acquire optical and optomechanical components required to build a custom and flexible optical microscope for correlated hyperspectral optical microscopy-SERS measurements. The capabilities of the envisioned setup (currently still under construction) will be demonstrated in FY 2017.

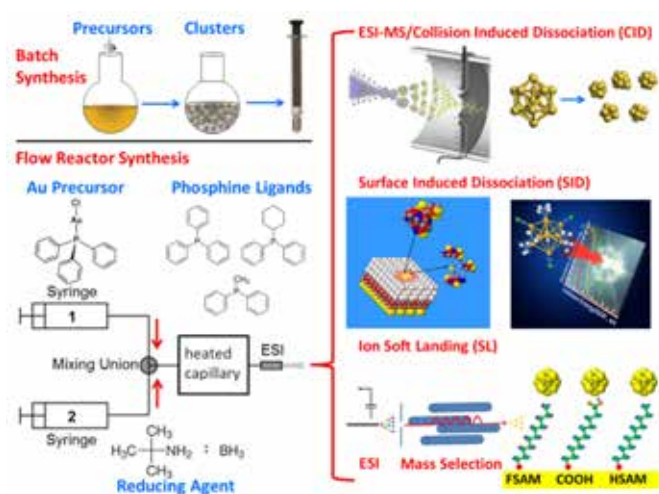
Understanding the Nucleation, Growth, and Deposition of Ligated Metal Clusters Using Mass Spectrometry

Grant E. Johnson

This project aims to develop novel instrumental approaches for understanding the nucleation and growth of ligated metal clusters using mass spectrometry (MS) and to utilize preparative MS as a versatile tool for the selective synthesis of well-defined clusters on surfaces. Metal clusters have highly size-tunable properties that give them broad potential applications in catalysis and energy storage technologies.

Sequence control in natural biopolymers such as DNA and We are focused on developing MS-based approaches for studying the nucleation and growth of metal clusters during solution-phase reduction synthesis. The scalable production of nanomaterials is important for potential commercial applications in catalysis, solar energy conversion, nanoelectronics, and biological sensing. Despite impressive advances in this field, the kinetics and mechanisms of cluster nucleation and growth in solution in the presence of organic ligands remain poorly understood. This presents a scientific challenge for the rational synthesis of clusters with predetermined size and properties. MS offers unique opportunities for studying the kinetics of the scalable synthesis of ligated metal clusters in solution. Furthermore, controlled preparation of individual clusters and their mesoscale assemblies on surfaces facilitates studies relevant to catalysis, chemical sensing, and energy storage. We are focused on understanding and controlling the size and charge reduction of soft-landed clusters through the use of differently functionalized phosphine ligands. We are also examining the effect of the physical and chemical properties of the surface and the surface coverage of clusters on their redox reactivity and stability.

We are employing an experimental approach that leverages three advanced MS techniques. Batch synthesis followed by characterization with electrospray ionization mass spectrometry (ESI-MS) in a commercial, quadrupole-ion trap or high-resolution Orbitrap instrument enables the exploration of how different combinations of reagents and reaction condi-



Schematic diagram illustrating the synthesis of ligated gold clusters by batch or flow reactor processes, which provides access to a range of intermediates and products for characterization by MS, SID, and SL.

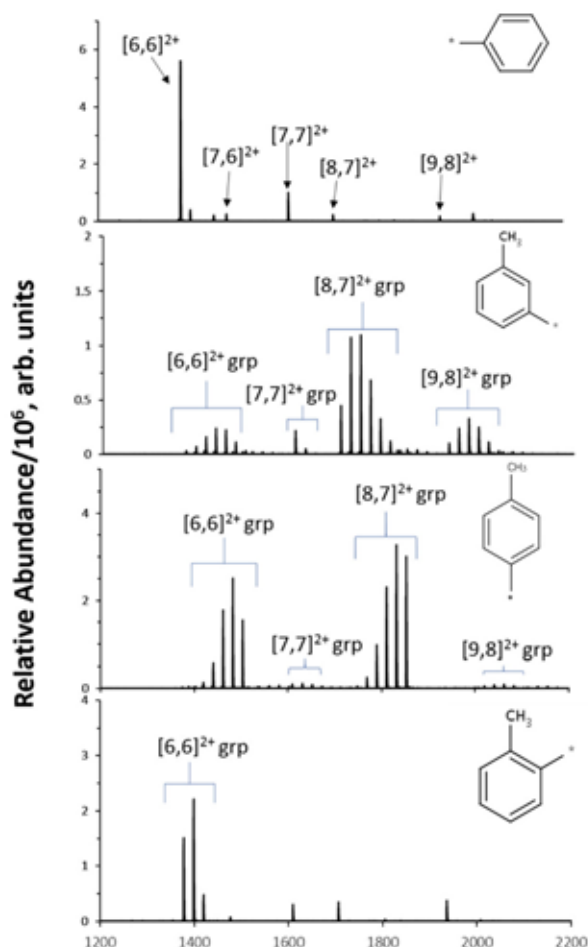
tions (e.g., type of ligand, relative concentration, time, and temperature) affect the final distribution of gold clusters formed in solution. For example, using ESI-MS, it is shown that substitution of cyclohexyl for phenyl groups in diphosphines may result in markedly different cluster distributions at otherwise equivalent synthesis conditions. ESI-MS also allows the investigation of ligand exchange reactions on pre-formed gold clusters. The mixed ligand species formed during exchange reactions are subjected to collision induced dissociation (CID) to gain insight into their structure and the relative binding energies of differently substituted phosphines to the metal cores. Compared to other characterization methods, ESI-MS and CID offer a versatile and straightforward way to determine the molecular formulae of small ligated cluster ions resulting from synthesis at different conditions. Quantitative ligand binding energies are extracted from fitting of the time- and energy-dependent fragmentation curves of the clusters obtained using surface induced dissociation (SID). The values serve to benchmark the results of high-level theoretical calculations performed by an external collaborator.

Novel clusters were produced by chemical reduction with a weak reducing agent, borane tert-butyl amine (BTBA). After initiating a batch reaction of AuPPh_3 (PPh_3 = triphenylphos-

phine) clusters, the solution was immediately introduced into the gas-phase by ESI followed by analysis with MS. Following acquisition of the PPh_3 spectra, tri(o-tolyl)phosphine (totp), tri(m-tolyl)phosphine (tmtp), and tri(p-tolyl)phosphine (tptp) ligands were introduced into the AuPPh_3 samples in equimolar amounts to initiate ligand exchange reactions resulting in formation of mixed-ligand gold clusters. The structure of totp is sterically hindered due to the methyl substituent being located two carbons away from the coordinating phosphorous atom.

Compared to totp, the structures of tmtp and tptp are less sterically crowded by the methyl substituent. Tmtp features a methyl group two carbon atoms away from the phosphorous atom, while tptp has a methyl group three carbon atoms away, or directly across from the phosphorous atom, making it the least sterically hindered ligand examined. Unlike totp, both tmtp and tptp exchange reactions produce a wide distribution of ligand exchange products. In the $[6,6]^{2+}$ range the cluster abundance increases with more tmtp ligand until the third tolyl ligand is exchanged, after which point the trend in abundance reverses. With tptp, a similar pattern is seen after the second tolyl ligand has exchanged.

In FY 2016, an invited article was published on this work in a special “emerging investigators” issue of *Analyst*. The accomplishments summarized in the preceding paragraphs were presented at an invited chemistry department seminar at Stanford University and at the 2016 Cluster-Surface Interactions Workshop held at Argonne National Laboratory. Parts of this work were also presented at the Noble Metal Nanoparticles Gordon Research Conference.



Representative ESI-MS spectra of a) AuPPh_3 clusters and ligand exchange initiated in b-d) solutions. Bracket notation indicates $[\text{Au}_x\text{L}_y]^{n+}$ (L = ligand), x and y indicate number of gold atoms and ligands present, and n+ is charge state of cluster.

Unveiling the Dynamic Microbial Biofilm and Plant Root Interface under Extreme Conditions

Xiao-Ying Yu

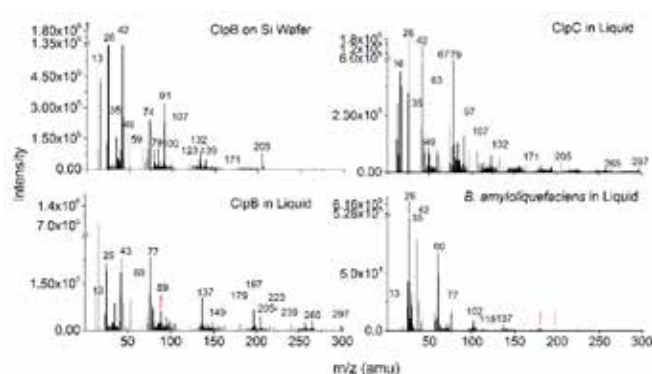
Biofilm formation on plant roots is known to be associated with biological control and pathogenic response; however, how plants regulate this association is not well known. We propose to enable unique PNNL multimodal chemical imaging capabilities to investigate the dynamic biofilm-plant root interface under the following two extreme conditions: 1) low relative humidity (RH), and 2) high salinity simulating drought to obtain measurements ranging from molecules to cells and associated communities.

Bacterial collagen like proteins (Clps) associated with promoting plant growth are a prerequisite for biofilm formation and adhesion to plant roots. Single cells play a complex functional role in bacterial biofilm surface components and extracellular factors in plant-bacterial association. Our knowledge of the identification and functions of extracellular proteins, DNA (eDNA), and lipids in the biofilm matrix of plant-associated bacteria remains limited. Cell-cell communication in bacteria-plant systems involves bacterial quorum sensing and the synthesis, release, and recognition of diffusible molecules such as indole. Production, perception, cellular sensitivity and response, as well as stability of signaling molecule(s), are key criteria to define how these molecules can be monitored dynamically.

This research aims to provide *in situ* dynamic multimodal chemical imaging of the interface between plant roots and microbial biofilms enabled by a versatile microfluidic reactor, namely System for Analysis at the Liquid Vacuum Interface (SALVI), which has resulted in a number of publications over the last several years. The following three tasks are proposed: 1) study the Clps and bacteria cell attachment at a surface, 2) obtain molecular distributions of key metabolic species in single bacterial cells at the biofilm-root interface, and 3) determine localized flux and mass transfer in biofilm cell-cell communications at different simulated extreme conditions.

In FY 2016, we mainly worked on Clps and bacteria cell attachment. Bacterial Clps associated with promotion of plant growth are a prerequisite for biofilm formation and adhesion to plant roots. We hypothesize that the localized water cluster microenvironment at the protein-root surface determines whether or not a bacteria cell attaches to the root surface. Our liquid secondary ion mass spectrometry (SIMS) positive m/z spectra can reveal discrete water cluster peaks (e.g., m/z 55, 73) that contain up to 40 water (H_2O)_n⁺ molecules. Water clus-

ters may also change corresponding to the oxidation state of the surface. For instance, more oxidized surface has many larger water clusters, thus making surface hydrophilic and potentially promoting attachment. In this task, Clps from plant growth promoting bacteria, such as *Bacillus amyloliquefaciens* FZB42, has been studied. Clps known to form triple-helical structures are of interest to obtain the characteristic water cluster distribution. Comparison with extracellular matrix proteins will allow us to elucidate the surface binding and aggregation of plant root promoting bacteria.



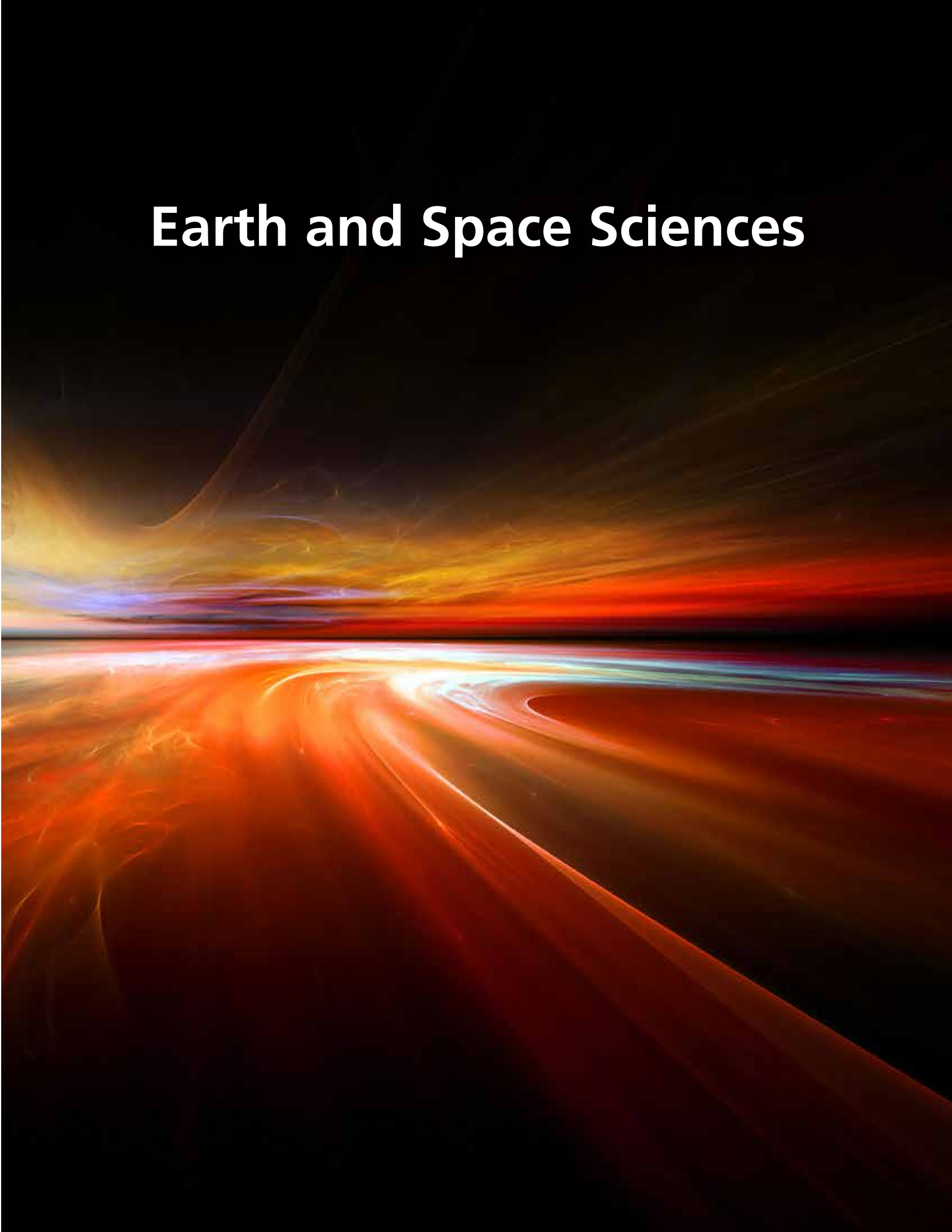
Negative SIMS mass spectra showing comparison of ClpB dried on Si wafer, ClpB in liquid, ClpC in liquid, and *B. amyloliquefaciens* in liquid. Red bars indicate the location of water clusters.

In addition, both aerobic and anaerobic biofilm culturing setups using our microfluidic reactors were developed to facilitate the next stage of experiments. *In situ* nuclear magnetic resonance biofilm imaging results using our SALVI were submitted for peer-reviewed publication.

In FY 2017, we aim at making a new microfluidic reactor that will accommodate the grass seeding in the microfluidic channel. *Pseudomonas putida*, a gram negative bacteria, may be used to compare its effect with *B. amyloliquefaciens*, a gram positive bacteria. We will focus on conducting the second and third tasks mentioned earlier, and obtain molecular-level understanding underpinning the principles of confident redesign of microbes and plants for sustainable biofuel production, improved carbon storage, or contaminant bioremediation.

It is anticipated that SALVI as a multifunctional platform for studying gene expression, regulation, and function in the spatiotemporal range of whole-cell environments and linking molecular-scale imaging to whole-cell, systems-level understanding can be illustrated in this project. Ultimately, we aim at providing 1) the state-of-the-art quantitative measurements of metabolomics analysis using unique PNNL capabilities, and 2) novel multiplexed imaging approaches to identify the relationship between external perturbations and microbial community responses under extreme conditions.

Earth and Space Sciences

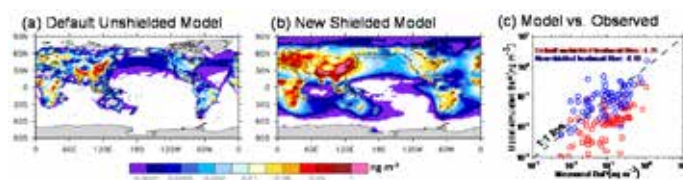


Assessing Climate and Human-Exposure Impacts of PAHs and SOA Particles

ManishKumar B. Shrivastava

Despite chemical kinetics measurements showing fast chemical degradation of particle-bound polycyclic aromatic hydrocarbons (PAHs), these PAHs persist in the atmosphere and travel thousands of miles. Based on latest advances in measurements, we developed novel modeling algorithms showing that health-relevant PAHs are protected in the atmosphere by climate-relevant secondary organic aerosols (SOAs).

Despite decades of research, the long-range transport of PAHs is not well understood, mainly due to incomplete knowledge of gas-particle partitioning and chemical loss rates of PAHs. Previous modeling studies have indicated that particle-bound PAHs like benzo(a)pyrene (BaP) need to undergo much slower heterogeneous loss to explain their observed atmospheric long-range transport. However, a mechanistic understanding of processes that cause this slow heterogeneous loss has been largely missing. Based on measurements that show that climate-relevant SOAs embed and shield PAHs from atmospheric chemical degradation, we developed new modeling paradigms in a global chemistry/climate model, wherein a shell of highly viscous (glassy) SOA forms around BaP that is adsorbed on the surface of soot/black carbon (BC) core. This shell effectively shields BaP from heterogeneous oxidation by ozone, at relatively cold/dry conditions.



This figure compares the default model treatment, where SOA does not shield BaP, to the new model treatment, which incorporates the effect of shielding of BaP by viscous organic aerosol coatings. The graph in (c) shows that the default unshielded treatment largely underpredicts BaP concentrations (red squares), while the new shielded treatment shows much better agreement (blue points are closer to the 1:1 line) at 71 background sites located in the United States, Canada, Western Europe, Africa, and Asia.

Using a global climate model, we demonstrate how this new model treatment largely increases BaP concentrations, lifetimes, and their long-range transport, globally.

Simulated BaP concentrations vary widely, with concentrations exceeding the PAH exposure limit of 0.25 ng m^{-3} over major source regions in Asia, Western Europe, Russia, and

Africa. At several locations globally the new shielded treatment predicts an order of magnitude larger BaP concentrations compared to the default unshielded. The new modeling treatment increases the long-range transport distance, remote BaP concentrations, and deposition fluxes of particle-bound BaP to both land and oceans, compared to the default unshielded treatment.

In addition, global population-weighted average incremental lifetime cancer risk due to exposure to BaP increases by a factor of four in the new shielded treatment, compared to the default unshielded treatment. The new treatment predicts that the global population weighted average lifetime cancer risk exceeds the acceptable levels of 1 in 100,000 persons, hence combustion-derived PAHs are predicted to be important for cancer risk. In contrast, the previous default model treatment predicts that cancer risk due to PAH exposure is within acceptable limits. This shows that the previous model will underestimate human lifetime cancer risk due to PAHs.

Due to its cross-cutting nature, the proposed research is expected to have multiple impacts affecting exposure-related human health and climate change. In addition, it would provide the necessary exposure data needed for connecting existing programs at PNNL including climate sciences, respiratory dosimetry and biological response to PAHs.

In FY 2016, this project team also has worked on a manuscript, titled, “Rethinking Long-Range Transport and Atmospheric Abundances of PAHs: Protective Effects of Highly Viscous Organic Shells,” which will be submitted to *Nature Geosciences*.

In the next fiscal year, we will continue the work described above to investigate which BaP sources (e.g., biomass burning, fossil-fuels) are most influential for global exposure and lifetime cancer risk. This will affect regulatory policy decisions for PAHs. Also, we will explore how future global warming will change human exposure to concentrations and types of PAHs (fresh versus oxidized), as well as which regions will be more susceptible to PAH exposure in the future.

Recent measurements also have shown that PAHs, in turn, affect climate-relevant SOA particles by increasing SOA mass and new particle formation. This increase in climate-relevant SOA particles affects cloud condensation nuclei and climate. We will develop new modeling paradigms that parameterize the synergistic effects of anthropogenic PAHs in increasing the number and mass of biogenic SOA particles.

We expect that this research will bridge the two disparate areas of health-impacts and climate change, and it will provide new modeling frameworks important for understanding how health-impacts and climate change are related to each other in the present day, as well as the future.

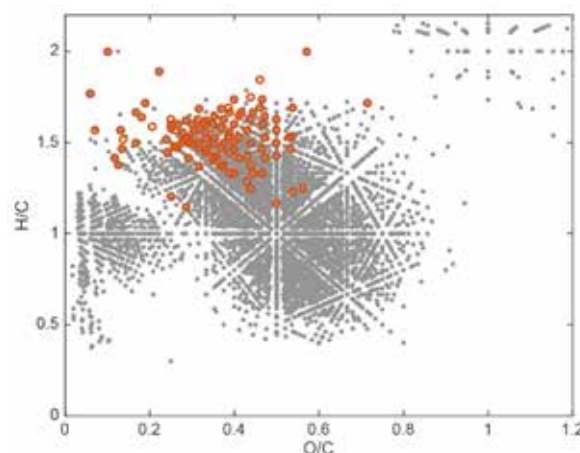
Characterizing Halogenated Organic Compounds in the Terrestrial-Aquatic Continuum

Li-Jung Kuo

This project aims to expand the formula assignment capability for ultrahigh resolution mass spectrometry (MS), which previously only considered compounds containing C, H, O, N, P, and S, to include halogenated organic compounds (HOCs). Such a capability expansion has enabled us to look into organic carbon that is outside traditional analyses windows and, thus, significantly enhance our abilities to better understand carbon sequestration and cycling mechanisms in different ecosystems across the terrestrial and aquatic environments.

Previous efforts in understanding the characteristics and environmental fates of HOCs were heavily concentrated in atmospheric chemistry, natural product discovery, and persistent organic pollutant analyses. Recent evidence shows that HOC may be an important fraction of natural organic matter (NOM) in soils and aquatic systems, in addition to their anthropogenic counterparts (e.g., pesticides, flame-retardants, and ozone depleting substances). However, the understanding of environmental fates of HOC and their impacts on carbon cycle is substantially hindered by the limited knowledge of their identities. Our project's goal is to provide means to confidently identify HOC in different ecosystems in a high throughput fashion using Fourier transform ion cyclotron resonance mass spectrometry (FTICR-MS), thereby allowing us to search for previously unknown HOC in nature.

An automated HOC assignment algorithm for FTICR-MS was developed and carefully tested. This algorithm uses isotopic pattern match to identify HOC in complex environmental samples. Identification confidence was measured with three independent, sophisticated scoring systems. The algorithm has successfully identified hundreds to thousands of HOC in each sample from different environmental matrices (i.e., soils, river and seawater, marine sediments, and algal external metabolites). The confidence of HOC assignment was benchmarked with 40 authentic HOC standards run multiple times as pure standards and matrix spikes. All standards observable with FTICR-MS were identified with high scores.

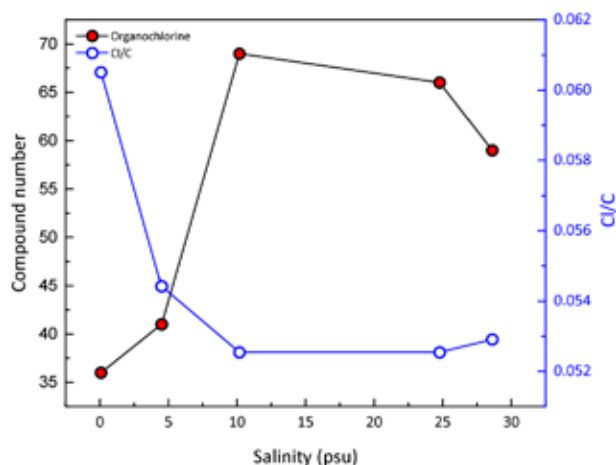


An example van Krevelen diagram depicted NOM identified in Suwannee River water. Grey dots are all compounds identified and orange circles highlighted HOC identified in the same sample.

A manuscript describing the HOC identification algorithm is in preparation to be submitted to *Analytical Chemistry*.

The HOC algorithm developed in our project has enabled us to study the distribution of halogenated compounds in natural samples from terrestrial-aquatic ecosystems (TAE). We have collaborated with Dr. Thomas Bianchi from the University of Florida to study HOC trends and speciation in water samples from river to the continental shelf. Samples were collected from Suwannee River along a steep salinity gradient (0 to 28 practical salinity units). Our results show that NOM within the river was dominated by terrestrial-derived materials, and these compounds were undergoing substantial remineralization into CO_2 . Chlorinated compounds, known to be present in plant matter decay, which were observed in the river samples. NOM on the continental shelf, outside the Suwannee River mouth, was dominated by marine primary production signatures. Organochlorine compound diversity increased significantly as salinity increased. However, organobromine and mix halogenated (chlorobromo) compounds were only observed in productive marine sites. In addition, chlorine to carbon ratio decreased substantially as salinity increased and as production regime switched to primary production, indicating potentially different HOC formation mechanisms along the TAE. Overall, results in this study clearly showed that HOC is widespread in TAE and can be used as a marker to distinguish terrestrial vs. marine NOM

sources. A manuscript to be submitted to a special issue in the *Journal of Oceanography and Limnology*, titled, “Headwaters to Oceans: Ecological and Biogeochemical Contrasts Across the Aquatic Continuum,” is under preparation. In addition, we have submitted an abstract for the 2016 American Geophysical Union Fall Meeting. An abstract also will be submitted to the Association for the Sciences of Limnology and Oceanography 2017 Aquatic Sciences Meeting.



Number of organochlorine and Cl/C along the salinity gradient.

In addition to aqueous samples, HOC were also identified in solid matrices like sediments. Samples were selected from an archived Puget Sound sediment core near Tacoma, WA. These sediment layers represent time frames with unique historical events (e.g., periods with substantial anthropogenic input), as indicated by polybrominated flame retardant and polycyclic aromatic hydrocarbons (PAHs) maxima, as well as pre-industrial era, which represented carbon signature only from natural input. The results reveal high HOC counts, particularly in sediment layers where flame retardant maximum and high coal input were observed.

The consistency between our results and targeted halogenated flame retardant analyses demonstrated that the algorithm developed in this project was able to robustly identify key historical events, during which high HOC counts were expected. In addition, previously unknown HOC were identified in the sediment layers, suggesting our algorithm can substantially improve our understanding of the diversity and fate of halogenated pollutants in the environment.

Determining Groundwater Residence Time through Ultra-Low Measurements of ^{39}Ar and Other Radiotracers

Jill M. Brandenberger

The goal of this project is to leverage the ultra-low background proportional counting (ULBPC) capability at PNNL to validate the utilization of the ultra-sensitive measurements of the noble gas radioisotope ^{39}Ar (half-life of 269 years) to constrain groundwater modeling in areas where groundwater depletion is drawing water recharged 50 to 1,000 years ago.

The need to refine the age distribution of groundwater is highlighted by the increasing number of groundwater resources that are being depleted faster than they are being recharged. Depletion contributes to water shortages, increased costs, land subsidence, and water quality deterioration.

Quantifying the Mean Residence Time (MRT) or age distributions of groundwater can be important for determining surface-groundwater interaction (recharge rates), evaluating connectivity of aquifers, verifying numerical groundwater models, and identifying depletion of groundwater aquifers critical for industrial/public use. Estimates of the MRT require multiple environmental tracers, covering age ranges of less than 50 years ($^3\text{H}/^3\text{He}$, ^{85}Kr) and more than 1,000 years (^{14}C). However, these established tracers do not adequately define the age distribution of groundwater aquifers recharging on intermediate time scales of 50 to 1,000 years.

Successfully building the ultra-sensitive ^{39}Ar measurement capability and attaining the scientific recognition required the following steps: 1) build efficient water degasification and Ar purification instruments (complete in FY 2014); 2) enhance the ULBPC measurements and increase gas transfer efficiency to ensure appropriate ^{39}Ar sensitivity for MRT (complete in FY 2015); 3) conduct verification sampling with the U.S. Geological Survey (USGS) to demonstrate the utility of adding ^{39}Ar to a suite of tracers used to determine age distributions (complete in FY 2015/FY 2016); and 4) build an enduring reputation through creating an ^{39}Ar reference sample and conducting an international intercalibration exercise (FY 2016).

The activity of ^{39}Ar is 1.01 Bq/kg Ar ($(8.0 \pm 0.6) \times 10^{-16}$ g/g), corresponding to an abundance of 0.808 parts-per-quadrillion. Assuming 1) a water temperature of 10°C, 2) that the water is (was) in equilibrium with atmospheric air, and 3) typical solubility of gases in water, the gas composition should be approximately 61.5% N_2 , 3.6% O_2 , and 2.5% Ar. Therefore, the target is to extract 1 L of Ar from approximately 3,000 gallons of groundwater by pumping through a membrane degasification system. The membranes are plastic hollow fibers that are hydrophobic. A vacuum is used to strip dissolved gases from the groundwater. The gas is collected in large, aluminum-coated, plastic sample bags and then pumped into compressed gas tanks (see included figure). Four verification sampling events with the USGS were conducted in the California Central Valley region. This region is in an extended drought and supports significant agricultural industries, which has led to water shortages and the need to understand the residence time of the groundwater to determine the rate at which it is being depleted.



Degasification unit connected to a Tedlar® gas intermediate sampling bag and yellow/red SCUBA tanks used to collect the extracted gas.

The collected gas from each groundwater well was compressed into SCUBA gas cylinders and shipped back to PNNL. Each sample was submitted for purification using PNNL's argon separation and purification process (as described in previous reports). All the samples have been counted in the

Shallow Underground Laboratory at PNNL. The included table shows the preliminary measurement results for these wells.

These international intercomparisons are critical to raising PNNL’s scientific visibility in the area of environmental applications of radiotracers. PNNL developed ³⁹Ar reference materials that could be analyzed by either low-level counting (LLC) or Atom Trap Trace Analysis (ATTA). The intercomparison would allow the direct comparison of ³⁹Ar determined by LLC measurements from the only two institutions capable of making this measurement: PNNL and the University of Bern (under Roland Purtschert). The same reference materials were also sent to Werner Aeschbach-Hertig and Markus Oberthaler at Heidelberg University to measure ³⁹Ar via ATTA. This provides a direct comparison of LLC and ATTA measurements of the reference material. The specific activity for the reference materials are 3x and 66x atmospheric background level (1.77E-6 Bq/cm³-Ar STP). PNNL is waiting for the partners to provide their results.

Estimated groundwater age determined from ULBPC ³⁹Ar measurements of California wells.

USGS Well ID	Date Sampled	³⁹ Ar Best Value, Years Old (Upper, Lower Bound) ¹
180-1	09/23/2014	523 (473,575)
180-1	09/25/2014	652 (590,720)
170	09/24/2014	565 (506,631)
180-1	09/23/2015	Sample not counted ²
183	09/24/2015	Sample not counted ²
180-2	09/25/2015	Sample not counted ²
TRLK-05	02/08/2016	363 (325, 401)
MOD-03	02/09/2016	In progress
MOD-02	02/10/2016	77.3 (40.5, 115)
MER-10*	02/11/2016	33.9 (0.54, 67.1)
CE-QPC-02	03/01/2016	39.0 (8.0, 69.3)
MER-11*	03/02/2016	95.8 (63.9, 127)

¹ 90% coverage interval.
² Unable to purify due to too much Nitrogen in the sample.
* Two wells contained an unusual decay trend. Upon further investigation, these two wells contained elevated levels of radon. Analysis is ongoing for these samples to determine if the rate of decay in the net count rate of the groundwater samples yields a half-life indicative of radon or some other short-lived isotope.

The FY 2016 activities focused on establishing scientific credibility in the area of ³⁹Ar to extend groundwater age distribution modeling. The principal investigator, as well as other members of this team, chaired the PNNL-hosted 2016 Low-Level Radiometric Measurement Techniques conference, September 26–30, 2016. Over 20 countries were in attendance, and it was the first time the conference was hosted in the United States.

The project provided two presentations and papers for publication in the Special Issue of Applied Radiation and Isotopes, which the team supported as guest editors. The list of submitted papers this year includes “Development of a Low-Level ³⁹Ar Calibration Standard – Analysis by Absolute Gas Counting Measurements Augmented with Simulation,” “Methods for Using Argon-39 to Age-Date Groundwater using Ultra-Low-Background Proportional Counting,” and “In Preparation with USGS, Ar-39 Fills the Gap: Age Distribution and Groundwater Recharge in Fresno, CA.” A team member also presented on “Application of ³⁹Ar Age Dating to Enhance Groundwater Age Distribution Estimation” at the National Groundwater Association Groundwater Summit in Denver, CO, April 24–27. The focus of the 2016 Summit was “Solving Groundwater Challenges through Research and Practice.”

A National Science Foundation proposal was also submitted in collaboration with Frank Loge at the University of California, Davis, and Tim Ginn at Washington State University titled, “Advanced Modeling of Groundwater-Agriculture-Energy under the Food-Energy-Water Nexus.” PNNL’s ³⁹Ar capability would augment the current data set (filling the “blind spot” of age interpretation between the half-lives of the common tracers tritium and ¹⁴C).

The project successfully demonstrated the first U.S. ULBPC ³⁹Ar measurement from several groundwater wells that have not been in contact with the atmosphere for 400 to 700 years. Key elements of this success included the development of a portable degasification unit and a higher efficiency dual reflux pressure swing adsorption that can provide up to 80% Ar recovery and is compatible with PNNL’s separation chemistry used to remove radon and other contaminating species. In addition, there are four ULBPCs characterized for both atmospheric and geologic ³⁹Ar to accurately determine the intermediate age distributions with errors less than 25%. Finally, the international intercomparison verified PNNL’s ³⁹Ar LLC capability, and these data are being used to address groundwater issues in the Central Valley of California.

Developing Multi-Criteria Assessment Methods under Uncertainty

Katherine V. Calvin

This research explores linkages between climate change mitigation and other societal goals (e.g., energy security, air pollution), enabling quantification of synergies and trade-offs among different objectives both regionally and globally.

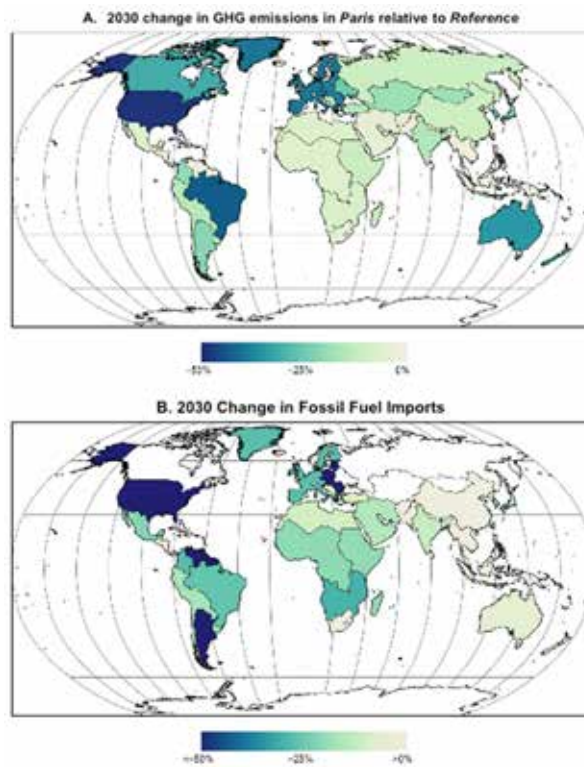
The integrated assessment modeling community has developed a broad literature over the last several decades, analyzing the effect of energy technology and climate policy on various aspects of the economy. A portion of that research has focused on comparing effort, focusing on a single objective (climate) and a small set of metrics (emissions and cost). More recent research has quantified the implication of addressing climate change on different societal objectives (e.g., energy security, air quality), providing insight regarding when potential synergies and conflicts may arise. This project combines these literature threads, assessing the effect of including multiple objectives on the evaluation of regional climate mitigation under different future scenarios.

This work uses an integrated assessment model, the Global Change Assessment Model (GCAM), to quantify the implications of different climate policies on multiple societal goals. GCAM couples together representations of energy, economy, agriculture, land, and climate. Using outputs from GCAM, we define one or more metrics for each of eight different societal goals, including climate change, energy security, energy access, food security, air pollution, and ocean health. We then compare the effect of the Nationally Determined Contributions (NDCs) of the recent Paris climate accord to these metrics.

We map the implications of NDCs on each of the defined metrics and contrast implications across regions. We find synergies between addressing climate change and addressing air pollution, as reducing greenhouse gas (GHG) emissions also leads to reductions in pollutant emissions. Efforts to reduce climate change, however, have differing effects on other goals. Furthermore, local effects may differ from international effects. For example, climate mitigation may improve the energy security in a fossil fuel importing region, but at the expense of

economic security in an exporting region. We note that there are differences in *contribution* and *consequence* of various efforts, as *direct* and *indirect* effects of actions, all of which are important to consider.

This study demonstrates that viewing mitigation through a broader sustainability lens could alter assessments of comparability. Including these factors requires detailed considerations of the interactions across societal goals, not only within domestic boundaries, but also internationally. Such a broad perspective would enable countries to gain a deeper understanding of the domestic impacts of NDC implementation on other societal goals, in a way that goes beyond the climate change mitigation space and also the international impacts of the activities implied by individual NDCs.



Changes in GHG emissions (A, top view) and fossil fuel imports (B, bottom view) when climate mitigation efforts, as proposed under the Paris Accord, are implemented. Emissions changes are a measure of climate mitigation effort, while fossil fuel imports are a measure of energy security.

Development of Optimal Visualization Techniques for Climate Model Performance Evaluation

Aritra Dasgupta

The goal of this project is to develop optimal visualization techniques for letting climate scientists understand similarities and differences among model performance, as well as to understand the trade-offs with respect to multiple metrics, which will ultimately lead to more accurate model calibration and, eventually, more accurate projections about climate change.

A key approach to climate modeling is to use multiple models or different configurations of a model for gaining confidence about the outputs, as well as for closely matching the observation data. For quantifying model performance, scientists use different performance metrics. The comparison of climate model performance across multiple metrics of skill with current tools is time-consuming and tedious. For example, the Atmosphere Model Working Group diagnostics package, which is used by the Accelerated Climate Modeling for Energy atmosphere model team for model evaluation and calibration, displays summary metrics either in printed tables or as Taylor diagrams. Both methods are ill-suited to the task of summarizing performance across many variables and model simulations (e.g., across 10 variables and 5 model simulations), which then requires tedious manual efforts by scientists. Taylor diagrams have an additional limitation of pre-determining the metrics that they are capable of displaying, prohibiting their use in displaying more statistically sophisticated measures of model performance; the habitual use of Taylor plots, therefore, potentially hinders the adoption of improved performance metrics.

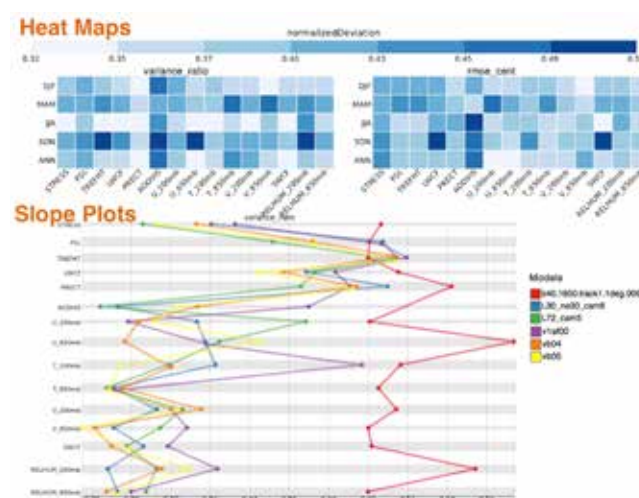
The techniques developed by this project will help speed up scientists' current analysis practices and also provide them with more effective tools for deriving insights out of their analysis. By using the interactive visualization interface, scientists can explore model performance data and also generate publication-quality images for presentation purposes.

We first designed and conducted a detailed, survey-based study for evaluating the relative effectiveness of different

model performance visualization techniques like heat maps, slope plots, Taylor plots, and bar charts. The results of the study show that, overall, climate scientists prefer heat maps and slope plots for performance comparison tasks, and their average accuracy across all tasks using the relatively unfamiliar slope plots is comparable to their accuracy when using more familiar visualizations like Taylor plots.

Motivated by the results of this study, we developed a visualization interface comprising heat maps and slope plots that enables climate scientists to explore model performance at different levels of detail and with respect to different combinations of models, variables, seasons, etc. We have submitted a manuscript to the Association for Computing Machinery Computer-Human Interaction 2017 conference, which includes the results of the user study.

We plan to leverage our collaboration with climate scientists to further develop the visualization tool. This will help fill a significant gap in model performance analysis techniques, as our initial results show that by using the developed techniques, scientists can more confidently and efficiently derive insights out of their model performance analysis processes than possible by using state-of-the-art analysis methods.



The visualization interface comprising heat maps and slope plots helps climate scientists interactively compare model performance with respect to multiple metrics, models, output variables, and seasons, at different levels of detail.

Development of Predictive Models on the Impacts of Heat Waves and Coincident Variability in Regional Water Availability on the Reliability of the Western Electric Grid

Nathalie Voisin

In this project, we demonstrate and quantify the stress of California heat waves on Western Interconnection operations under baseline water conditions and under selected critical drought conditions.

Droughts and heat waves are projected to be more frequent and more intense in the future. What is the vulnerability of the Western Interconnection to heat waves? How much do heat waves accentuate drought stress on Western Interconnection operations (reserve margin, unmet electricity demand, locational marginal price)? What are the opportunities for multi-scale joint water-energy management? These are all questions we seek to address.

In this project, we show that heat waves accentuate the stress from critical drought conditions by increasing the amount and duration of unserved energy. We further demonstrate that heat waves shift the system performance threshold (i.e., the chance of not meeting the summer energy demand raises from 11% to 14%). The outcomes of this project are an approach to mapping heat waves onto a building energy demand model and a quantification of heat wave stress in terms of the increase in peak and daily energy demand. A third outcome is the quantification of the compounded stress of heat wave and droughts on electrical grid performance (ability to meet the demand, reserve margin, locational marginal price). This work contributes to the emerging awareness of how a potential increase in frequency and intensity of heat waves combined with droughts could impact the performance of the whole Western Interconnection grid. It leads to further research opportunities for assessing adequate resources planning under future climate conditions and for increasing inter-regional coordination in joint water-energy management to meet the region's carbon emission objectives and to maximize the value of regional hydro resources.

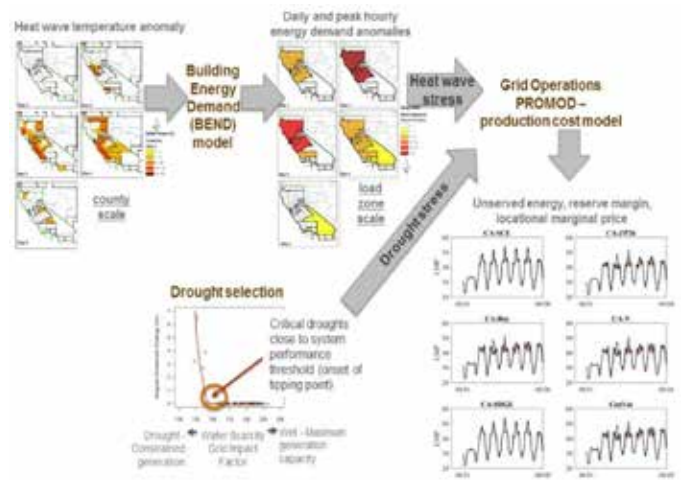
Heat waves and prolonged high-temperature periods stress the Western Interconnection by increasing the energy demand from air conditioning systems and exposure of infrastructure to wildfires. Utilities such as PG&E use daily probabilistic meteorological forecasts for the purpose of load forecasting over populated areas, especially for heat waves. Most energy demand models rely on degree days to represent the daily energy demand. Heat waves are characterized by locally extremely high temperatures, and the peaking building energy demand is not necessarily linearly related to the increase in temperature due to HVAC cooling system performance. Therefore, instead of using a linear regression between load centers and increase in temperature, we developed an approach to map (i.e., overlay) historical heat waves onto the operational grid modeling framework by simulating load anomalies using a building energy demand model. The approach allows for isolating the specific heat wave signal from the potential aggregation of other events in the historical sequence. This allows for characterizing the heat wave event with stress metrics (spatial extent, duration, intensity) to quantify its severity. The mapping is also consistent with meteorological forecasts provided daily for future applications in mitigation and response modes.

Experience from previous heat waves (2006) and prolonged high temperatures has led to incentives to install solar panels and demand conservation programs to mitigate the spur of energy demand at the source. The demand control facilitates the management of transmission and distribution lines that are extensively used to import energy from other regions, mostly from hydropower. Operating to supply adequate electricity resources for increased energy demand during heat waves, and in times of limited generation capacity due to extended and severe droughts, is challenging. The interdependency between the Western Interconnection operations and drought severity has been demonstrated, and a regional analysis revealed that drought severity in California defines the tipping point of the Interconnection operations beyond

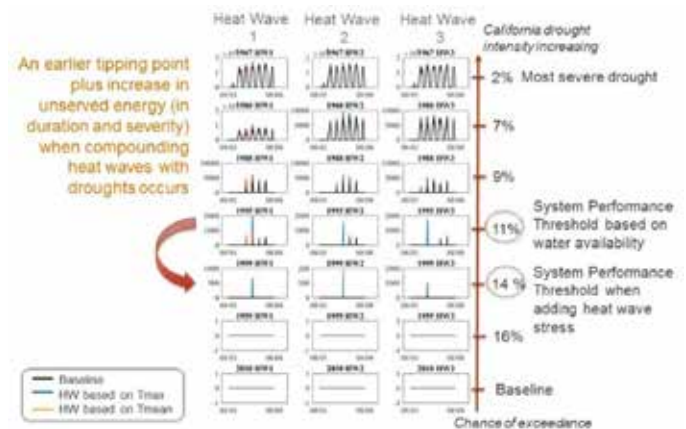
which the energy demand in August cannot be fully met due to constrained generation and transmission. Along with a baseline water year, a set of critical droughts at the onset and right after the tipping points were selected to be compounded with three heat waves chosen from a historical California heat wave catalog that we developed. Uncertainty around the definition of the heat wave is also quantified. Drought stress and heat wave stress both force a production cost model representing the 2010-level of grid operations over the Western United States.

Results indicate that, under average water availability, the heat wave stress on the grid operations will result in an increase in locational marginal price, which can spread over multiple load zones, even though the zones are not directly affected by the heat wave. Within historical inter-annual climate variability and normal grid operations, an earlier tipping point and an increase in unserved energy in duration and severity will occur when the heat wave is compounded with critical and near critical drought conditions. Under normal operations (prior to contingency plans for water use or generation portfolio requirements), there is a 3% chance that, under historical inter-annual climate variability, the grid as of 2010 will not be able to meet 6% or more of its August energy demand, with the failure to meet the demand located mostly in California. Based on drought severity, specifically in California, there is an 11% chance of not meeting the load in the Western grid. When compounded with a heat wave in California, this chance is raised to 14% for all three different heat waves considered in the analysis.

No resources adequacy studies presently look at the compounded effect of droughts and heat waves on electricity grid operations. Using the full distribution of drought characteristics and taking into account regional variability is highly recommended to evaluate and develop adaptation and mitigation studies under heat waves for resources adequacy analyses. This project is the first to explore operational grid impacts imposed by heat waves compounded by droughts in the western U.S. grid.



Experimental approach to quantifying the compounded effect of heat wave stress (increased energy demand) by drought conditions (constrained generation) on Western Interconnection performance (unserved energy demand, reserve margin, locational marginal price) using a production cost model.



Inter-dependency of Western Interconnection electrical grid performance and regional water availability, represented as a grid stress function; it represents the risk to the Western Interconnection, because it associates multiple levels of potential unmet energy demand with a range of probabilities of occurrence. The system performance threshold indicates the drought severity level (based on water availability across load regions), beyond which there is likely to be unmet energy demand if a contingency plan (change in energy management) is not in place.

Development of Surface-Modifying Tracers for Improved Geophysical Imaging of Fracture Networks

Vince R. Vermeul

This project seeks to evaluate the potential of using surface-modifying tracers for improved geophysical imaging of the subsurface, with the primary objective of developing methods for interrogating fracture surface area.

A major element of the SubTER New Sensors and Signals pillar is the development of methods for characterizing natural and stimulated fracture networks. The primary objectives of this element include determining the density and spatial distributions of fractures within the subsurface, interrogating fracture surface area, and providing for near-real-time monitoring/imaging of fluid flow within a fracture network. It is well recognized that the use of engineered tracers is likely to play a key role in new technologies developed to meet this goal. Consequently, research and development on the use of tracers for fracture network characterization is specifically called out as one of the pillar goals. Tracers that enhance geophysical signals can be used to both characterize fracture networks and monitor for changes that occur due to stimulation activities.

A series of laboratory experiments were conducted to evaluate the use of tracer suites with different surface-modifying characteristics as a means of interrogating surface area. Although the ultimate goal is to develop methodologies for measuring specific surface area (i.e., surface area to volume ratio) in fractured systems, this preliminary work was conducted using porous media admixtures of differing particle size and, thus, surface area conditions. Three separate granular basalt admixtures were used, including a coarse-grained mixture containing particle sizes ranging between 2.0 and 3.3 mm, a medium-grained mixture containing particle sizes ranging between 1.0 and 2.0 mm, and a fine-grained mixture containing particle sizes ranging between 0.5 and 1.0 mm. The experiments were conducted using a standard column test configuration. However, this work extends the standard column testing protocol to include electrical conductivity (EC) monitoring of the column materials as an indirect measure of tracer transport characteristics. Because retardation factors

(and associated distribution coefficients) for a given field test site are dependent on the types of rock(s) comprising the subsurface system and, therefore, are site-specific, laboratory-scale information on sorption characteristics is needed to develop field-scale test designs. Incorporating these results into forward simulations of tracer transport and associated electrical response can be used to develop appropriate operational and monitoring configurations for field-scale interrogation of fracture surface area. The tasks below were conducted.

Identify candidate tracers. This study focused on basalt as the target geologic setting, because it represents a relevant geology for both carbon capture and storage and enhanced geothermal systems applications. Two different surface-modifying tracers were identified, one applicable to only hot dry rock systems (changes in relative humidity) and one for water saturated systems (potassium fluoride adsorption/desorption).

Evaluate electrical properties of identified tracers. For each candidate surface-modifying tracer identified, a series of column experiments were conducted to determine whether EC measurements made directly on the column fill materials (i.e., granular basalt admixtures) could be used to distinguish between the tracer responses for varying surface area conditions.

An initial evaluation of relative humidity (RH) as a surface modifying tracer that could be monitored using electrical methods indicated that this approach was constrained by significant technical challenges. Brunauer–Emmett–Teller (BET) theory is a commonly used method to estimate the surface area of a sample from observations of water content over a range of RH. Increases in RH are expected to increase pore water film thickness (water content), leading to an increase in the EC of the bulk rock/fluid. For this project, bulk EC was used as a surrogate for water content, and a clear type II BET curve was observed; however, the overall change in bulk EC was quite small and would not be detectable under typical field scale conditions.

Results from an evaluation of potassium fluoride (KF) as a surface-modifying tracer indicate that, for the surface area conditions investigated, electrical properties can be used to

differentiate the sorption response for the various admixtures, and that increasing sorption was well correlated with independent measurements of surface area.

Initial efforts on the KF evaluation focused on demonstrating that column EC measurements could be used as a surrogate for tracer transport response and, thus, would be comparable to results obtained using standard effluent monitoring approaches. Column EC was measured over an integrated volume (1/3 of column volume) centered at the midpoint of the column. Column effluent EC was measured at the downstream end of the column with a microelectrode conductance meter (YSI Inc., model 32) and associated ion concentrations for selected experiments were measured by ion chromatography and inductively coupled plasma optical emission spectrometry. All column experiments were run with a throughput linear velocity of approximately 26 cm/hr (i.e., about 0.9 pore volumes/hr). Comparison of these EC data with associated anion and cation data (not shown) demonstrated that the observed EC responses resulted from transport of the injected tracer ions.

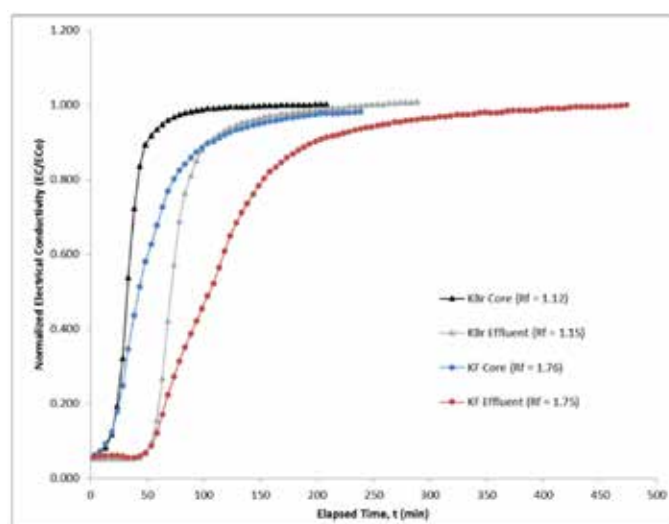
The retardation factor (R_f) for each tracer (and each EC measurement method) was calculated by integrating the area under the EC arrival curve to determine the arrival time for the centroid of the curve, and the resulting column velocity was calculated by dividing the distance to the point where arrival was measured by this arrival time. This velocity, relative to the volumetrically calculated velocity, was then used to calculate the R_f . As indicated, the calculated R_f for the conservative tracer (KBr) was approximately 1.1 (compared to 1.0 for an ideally conservative tracer), and the calculated R_f for the adsorbing tracer (KF) was approximately 1.8. Comparison of the R_f derived from these response curves shows good agreement between the core and standard effluent methods, and thus, the utility of using core EC to measure tracer transport response in these column experiments was demonstrated. The observed adsorption and desorption responses for KF were comparable, indicating a reversible sorption response.

Equivalent tracer experiments were conducted for the other two porous media admixtures, and EC response curves for each tracer were generated. The R_f determined, based on tracer adsorption response for these three different admixtures, each having a different specific surface area, were then plotted versus surface area (measured independently via N_2 gas BET). Results indicate that the relationship is strongly linear over the range of specific surface areas tested, with a method of least squares regression analysis resulting in an R^2 value of 0.999.

These data demonstrate the potential for using electrical methods to measure differences in tracer sorption characteristics *in situ* and in near-real-time, which in turn, can be used to interrogate fracture surface area. Although these results are encouraging, additional work is needed to demonstrate this capability for test configurations and specific surface area conditions more relevant to fractured rock systems. The next phase of testing would involve conducting a comparable set of column experiments, but the test configuration would be based on an engineered fracture system where surface area and aperture width (and thus specific surface area) could be varied in a controlled fashion. This work would also evaluate the use of more strongly sorbing tracers, which may be needed for geophysical interrogation in the lower specific surface area conditions that are characteristic of fractured rock systems.



Column assembly (30-cm-long) showing the inlet (bottom), outlet (top), EC electrodes (2 ports on left side of column), and relative humidity gauge (port on right side of column).



Observed tracer arrival responses, based on EC measurements, for the coarse-grained basalt admixture.

Ecosystem Transitions and Associated Greenhouse Gas Fluxes Following Salt-Water Intrusion from Relative Sea Level Rise

Heida L. Diefenderfer

The overarching goal of this project is to quantify, mechanistically understand, and predict shifts in CH_4 , CO_2 , and N_2O fluxes from an intact wetland community in response to salt water intrusion. Understanding the effects of these large-scale disturbances, predicted to occur at an increasing rate in coming decades, on greenhouse gas source and sink function is critical to improving process-rich models of climate change effects on coastal wetlands.

Coastal and fluvial ecosystems bordering the landward limits of salt water are seen as key depositional regions for terrestrial nitrogen (N) and carbon (C) bound for the ocean. These highly productive and globally distributed ecosystems are dominated by herbaceous, shrub, and/or forested vegetation. Long-term trends indicate that the zone of tidal freshwater will migrate substantially in the landward direction in coming decades. Soil chemistry is influenced by shifting hydrologic regimes at the intersection of river basin and coastal nearshore processes. The impacts of legacy material from disturbance-induced rapid succession on C and N cycling depends in large part on microbial community function. Understanding the controlling factors on these complex ecosystem processes is fundamental to DOE Biological and Environmental Research missions related to global elemental and water cycles. This project seeks to discern the sensitivity of soil microbiome molecular processes to altered salinity regimes.

Filling this knowledge gap is critical, as it currently limits our ability to robustly predict changes in ecosystem function as a consequence of altered salinity regimes. This project aims to fill this knowledge gap through *in situ* characterization coupled with hypothesis-driven experiments. Changes in porewater salinity will overwhelm the salt tolerance of plant and microbial communities. However, the time lags and magnitude of responses by microbial communities to saltwater intrusion, as measured by greenhouse gas flux function, currently remain unknown.

This research focuses on a tidally influenced wetland near the mouth of the Columbia River in Washington State. Climate change is altering the hydrologic regime in this, and similar, tidal freshwater regions such that historically river-dominated conditions are being increasingly affected by nearshore-ocean conditions, including saltwater intrusion. Aquatic-ter-

restrial transition zones like wetlands will experience the immediate consequences of changes in the magnitude and dynamics of hydrologic flows and associated fluxes. Coastal wetlands are particularly susceptible to storm events, salt-water intrusion, and increasing water levels. We initiated characterization of *in situ* conditions at the study site, with analysis of soils for total carbon, total organic carbon, total inorganic carbon, bulk density, particle size distribution, and total solids.

The effect of salinity on N_2O emission pathways has shown only variable results, and the impact of salinity on the microbial ecology of soil/sediment systems is unclear. Nitrifying and denitrifying soil bacteria are the sources of N_2O emissions in biological nitrogen reduction in soil/sediments, specifically autotrophic ammonia oxidizing bacteria, nitrite oxidizing bacteria and heterotrophic denitrifiers. The most important environmental factors identified to date that influence N_2O emissions through these processes are dissolved oxygen, nitrite, and organic carbon concentrations. Low dissolved oxygen and high nitrite accumulation trigger N_2O emission during nitrification. However, the quantitative relationships between N_2O production pathways have been reported sparsely.

We hypothesized that sudden changes in salinity regimes will strongly select for microbial taxa, likely including mortality events, and will, thereby, affect community functions, including the emission of greenhouse gases. We conducted an extended core-incubation study, subjecting freshwater-wetland soils to oligohaline-range diluted seawater, similar to expected model-predicted effects of climate change in tidal freshwater regions. The 3-month study is ongoing to determine the duration of the initial response, particularly the statistically significant difference in N_2O emissions in the treatment cores. Control cores functioned as sinks, while treatment cores functioned as sources of this gas (measured by hourly flux measurements, July through September, including night and day). The measured source effects are low in magnitude (i.e., less than $0.6 \text{ nmol/m}^2/\text{s}$) and near the detection limit of the automated gas analysis system employed for this study. However, this gas is notable for a disproportionately high contribution to the greenhouse effect, as compared to CH_4 and CO_2 , and few measurements of N_2O emission dynamics from coastal wetlands have been published due to the focus of prior research on the carbon cycle. During the core-incubation study, replicate cores were sacrificed for porewater analyses (pH, specific conductance, carbonate, chloride, sulfate, nitrate, total calcium, iron, manganese, potassium, and sodium) to support parameterization of a biochemistry model.

How Do Non-Linear Microbial Processes Lead to Linear Ecosystem Fluxes?

Katherine E.O. Todd-Brown

This project incorporates new measurements and understanding into soil carbon models, advancing predictive capabilities of biogeochemical models and bridging the gap between microbial processes and ecosystem observations.

Modeling soil carbon dynamics is critical to predicting how much carbon dioxide will be in the atmosphere to drive future climate change. Soil carbon models have remained essentially unchanged for decades. These models partition soil carbon into 1 to 9 pools; carbon leaves these pools at a rate proportional to the amount of carbon in the pool (i.e., first order linear decay); some of the carbon leaving these pools is diverted to other pools, and the rest leaves the system as carbon dioxide. Most research since the 1980s has focused on generating environmental sensitivity functions for the decay rates or physical/chemical characterization of the soil carbon pools. This model type tends to statistically fit observed soil carbon emissions using laboratory incubations. However, despite extensive attempts to characterize the environmental sensitivity, parameters must be re-calibrated for each location, making it difficult to confidently generalize these models to global projections.

In the past 15 years, new microbial characterization methods have led to a growing interest in the biogeochemistry community in microbially explicit decomposition models. In these models, soil carbon must first pass through a microbial pool before leaving the system as carbon dioxide. These models are appealing not only because they can describe a more fundamental understanding of soil decomposition, but also for their potential to incorporate new genetic and other 'omic measurements. However, there is an issue of scale: carbon decomposition occurs at the pore-scale (less than 1 mm³), and the fluxes of interest occur at the ecosystem-scale (more than 1 km²).

The objectives of this research are 1) to develop better soil carbon models in the short term via model-data integration of realistic field data into traditional decomposition models and 2) contribute to long-term improvements by capturing process-explicit representations of our evolving understanding of soil decomposition in new biogeochemical models.

In FY 2016, we have made progress in both advancing the understanding and parameterization of traditional soil decomposition models, as well as laying the ground work for a process explicit model representing microbial physiology,

mineral-organic interactions, extracellular enzyme kinetics, and microbial dormancy.

On the traditional modeling front, the principal investigator (PI) was a co-author on a *Journal of Mathematical Biology* manuscript with international authors from seven institutes. This paper explored the implications of using autonomous (constant inputs) analysis techniques for non-autonomous (varied inputs) systems in the context of first-order linear differential equation models used in traditional soil carbon simulations. In general, given the time scales of interest, this was an acceptable substitution, though care must be taken to distinguish between mean transit time and mean carbon age of the carbon pools.

The PI is also a co-author on a *Nature* manuscript that has been accepted for publication along with international authors from 41 institutes; this project provided statistical and modeling support for a study looking at novel ways to examine temperature sensitivity of soil carbon stocks across multiple field warmed manipulations. There is also a second manuscript being prepared, which the PI is leading, to place this information in a more traditional modeling context. This manuscript will be submitted as soon as the first manuscript is in press.

Soil carbon changes are generally considered to be too noisy to use for temperature sensitivity calculations; previous studies have instead relied on soil respiration changes. We have shown that it is possible to calculate a temperature sensitivity using soil carbon stocks directly, given a large enough sample size.

An additional manuscript examining the temperature sensitivity of soil respiration in a coastal wetland is also being prepared and is expected to be submitted early in FY 2017. This project is the result of a collaboration with researchers at the Marine Sciences Lab at PNNL and highlights the need to carefully consider the thermal effects of water intrusion in tidal systems, in order to accurately model biogeochemical cycling.

On the process explicit modeling front, the PI has developed and parameterized a new model from the literature, JAM (Just Another Microbial) model, which was presented at a Gordon Research Conference and selected for a talk at the associated Gordon Research Symposium. A manuscript is being prepared for submission in early FY 2017, outlining the general approach and highlighting the need for scale aware modeling. A second manuscript with the model code, parameter sensitivity analysis, and basic spatial heterogeneity effects on emergent system behavior is expected to be submitted by the end of FY 2017 and will be presented at the American Geophysical Union in FY 2017, as well.

Network Analysis of Soil Organic Matter Molecular Chemistry to Enable More Robust Modeling of C Cycle along Ecosystem Gradients

Nancy J. Hess

This project centers around rapid identification of molecular signatures of biologic or abiotic activity that can be used in natural organic matter obtained across ecosystem gradients.

Recent advances in soft solvent extraction protocols have enabled soil scientists to analyze and identify the thousands of organic molecules in a few milligrams of soil using ultra high-resolution mass spectrometry. However, existing visualization tools are only partially successful in allowing interpretation of this complex data set. The goal of this pilot project is to determine whether a network analysis approach of the neutral mass difference between the measured masses' organic molecules can be used to develop a semi-quantitative metric of the chemical state of the soil and identify biotic and abiotic processes that relate molecules in the resulting network.

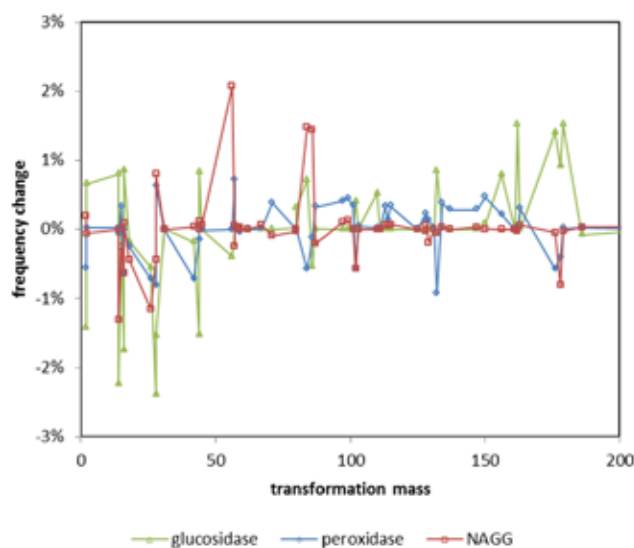
We conducted simplified “abiotic” enzymatic degradation experiments of filtered water extracts of peat soil to generate a dissolved organic matter (DOM) of suitable chemical complexity. The DOM was degraded using commercially available glucosidase, peroxidase, and N-acetylglutamate (NAG) synthase enzymes. The unreacted DOM, as a control, and the enzyme degraded DOM samples were analyzed using electrospray ionization Fourier transform ion cyclotron resonance mass spectrometry (MS) to measure the changes in the DOM molecular chemistry prior to and following enzymatic degradation.

We followed the changes in molecular chemistry in two ways. A standard approach using the O:C and H:C ratios of assigned molecular formula was used to classify the molecular compounds into aggregate molecular classes such as lipid, lignin, and tannin. We also used a novel network analysis approach based on the mass difference between MS peaks (network edges) to reveal changes in the connectivity of all MS peaks (network nodes). We hypothesized that network analysis would show a change in the connectivity of the network and frequency of transformations that reflects the specific enzyme activity.

The reacted DOM displayed an increase of between 10 to 20% mass spectral peaks, suggesting greater chemical diversity in

the reacted samples. For the glucosidase enzyme, the impact of enzymatic activity was evident using a standard aggregate compositional analysis. For example, a 20% decrease in CHO compounds and corresponding increase in CHON was observed. Insignificant shifts in composition were noted for the peroxidase and N-acetyl-glutamyl-glutamate (NAGG) degraded DOM.

An advanced network analysis approach, Edit Distance, was then used to compare nodes whose specific connectivity, considering both number and identity of the chemical transformation (edges), either remained unchanged or changed significantly following enzymatic activity. For nodes whose connectivity changed significantly, all three enzymes produced distinctive differences in chemical connectivity. The glucosidase reacted DOM showed an elevated number of chemical transformations involving amino acids; the peroxidase reacted DOM displayed a shift to more oxygenated organic compounds; whereas, NAGG reacted DOM displayed a repression of these transformations and an increase in transformations involving pentose, glucose, and glucuronic acid subunits. While the results are still preliminary, Edit Distance is a promising approach to understanding the chemical state of DOM.



Each of the three enzymes studied shows a characteristic change in the frequency of different chemical transformations between unreacted and reacted DOM, plotted as a function of transformation mass.

Non-Stationary Climate Considerations – Floods and Consequences

David Judi

The objective of this research is to develop modeling and simulation tools and methodologies to help stakeholders (including policy makers, planners, and engineers) understand vulnerabilities and consequences related to the non-stationarity of precipitation events and urban development.

Flooding has been observed to become more prevalent and is expected to worsen with a changing climate, including potentially increased precipitation and snowmelt runoff. For example, some coastal areas frequently experience flooding resulting from relatively minor precipitation and tidal events because of local changes in sea level. Inland areas have also become more vulnerable to flooding. The majority of the United States has seen an increase in short-duration, high-intensity precipitation events, which are expected to further increase in frequency and intensity. The observed and projected changes in precipitation are commonly referred to as a non-stationary climate.

In many cases, the infrastructure designed under stationary climate assumptions (e.g., historical 100-year return period) is largely inadequate to handle changes in flood events. To this end, recent policy changes in floodplain management guidelines call for the inclusion of climate-informed science. However, actionable climate information or guidelines for its inclusion at a local-scale are not widely available.

Meanwhile, urban areas become further susceptible to flooding risks accompanied with an urbanization process, including urban infills and sprawls. Population migration, redevelopment, and market economics add more uncertainty to the magnitude of the consequences of flooding risks to urban areas. Urbanization, therefore, can be regarded as another non-stationary variable in future flood prediction.

The objective of this research is to develop modeling and simulation tools and methodologies to help stakeholders (including policy makers, planners, and engineers) understand vulnerabilities and consequences related to the non-stationarity of precipitation events and urban development. This includes the translation of global, long-term climate projections and regional socioeconomic data into local-scale, actionable information. These tools will provide a means to

develop mitigation and adaptation strategies to ensure resilient designs for critical infrastructure systems.

The focus of this project was the development and utilization of flood frequency distributions of flow rates, historical and future, combined with historical socioeconomic datasets to derive estimates of flooding and consequences for the Snohomish River located near Monroe, WA. The Snohomish River watershed was selected as a demonstration region, given relative contributions of rain, snow, and rain on snow to potential flooding. The frequency distributions were developed using regional climate model forcings for representative concentration pathway (RCP) 4.5 for a period up to year 2100. For comparison, frequency distributions using historic U.S. Geological Survey (USGS) data were also developed.

The crux of translation of climate data to local-scale runoff distributions is the accurate simulation of the hydrological processes forced by regional climate data. The Distributed Hydrology Soil Vegetation Model was used to simulate the complex dynamics of precipitation, infiltration, snow accumulation/melt, and runoff at an hourly time-scale. We calibrated this hydrologic model with a distinct emphasis on capturing extreme runoff events using observations from USGS stream gages, observed meteorological data from local weather stations, and historical data from the North American Land Data Assimilation System. This model was specifically calibrated for high flows to ensure that extreme events are properly accounted for in future time period simulations.

The future hydrologic periods were forced by regional climate model data developed at PNNL. These include hourly observations of air temperature, wind speed, relative humidity, incoming shortwave radiation, long wave radiation, and precipitation at 1/8 degree resolution. We used these meteorological forcings for one scenario, RCP 4.5. These data cover a time period from 2005 to 2100.

Two frequency analysis methods were used to characterize the runoff from the simulations: Bulletin 17C and Generalized Extreme Value. Using the frequency distributions, we were able to quantify changes in intensity and frequency of extreme runoff events (including rain, snow, and rain on snow) over future periods and scenarios. For example, RCP 4.5 showed a consistent 15% increase in runoff over all return periods. In addition, a 100-year flood event today was projected to become approximately a 48-year event in the future.

The historic and future flood frequency distributions were used to develop spatial and temporal distributions of flood depth and velocity. This was done using modeling and simulation based on the shallow water equations and available topographic data. We used Monte Carlo methods to develop 135 flow forcings for each scenario (historic and RCP 4.5). From each simulation, spatial distributions of peak depth and velocity were developed and stored for use in consequence estimation.

A primary objective of this research was to quantify the relative contribution of future flood consequences from both climate change and urban development. Urban development includes new development and changes in market conditions based on property value. In this research, we specifically focus on flood consequences within the residential and commercial sectors. To represent the urban development component, we constructed spatial datasets of the urban environment using Census, parcel, and tax valuation data from 2001, 2006, and 2011. We also developed fragility models, a relationship between damage and flood characteristics, for each sector. For each simulation, the flood hazard, built environment, and fragility were combined to assess population at risk and direct economic impact from structural damage. These results were used to estimate the frequency of total damage (\$), annual risk (\$/year), and the expected total annual loss (\$/year) relative to the probability of the event.

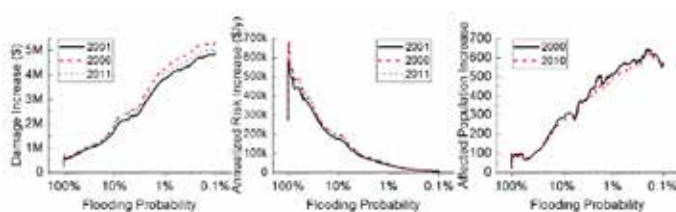
Regardless of year, relative to the representation of the built environment, the change river flow rates from the historic climate to the future climate under the RCP 4.5 scenario results in significant increases in expected flood damage, especially for the high magnitude, low probability events. (Figure 1). To separate the relative contributions of climate and development impacts, the total annualized loss of 2006 and 2011 development conditions with the future climate forcing was compared to the 2001 development condition forced by the historic climate data (Figure 2). Thirty-four percent (34%) and 29% total annualized loss increases were estimated for 2006 and 2011, respectively, with the highest changes in both years occurring in the residential sector. Within each sector, the contribution from climate change accounts for slightly more than half of the total increase in structural damage. In total, the climate and development variables contribute to 18% and 15% of the total change of annualized loss for 2006 and 18% and 11% of the total change for 2011.

Interestingly, the annualized loss in 2011 decreased compared to 2006, consistent with observed market changes in the commercial sector for the region.

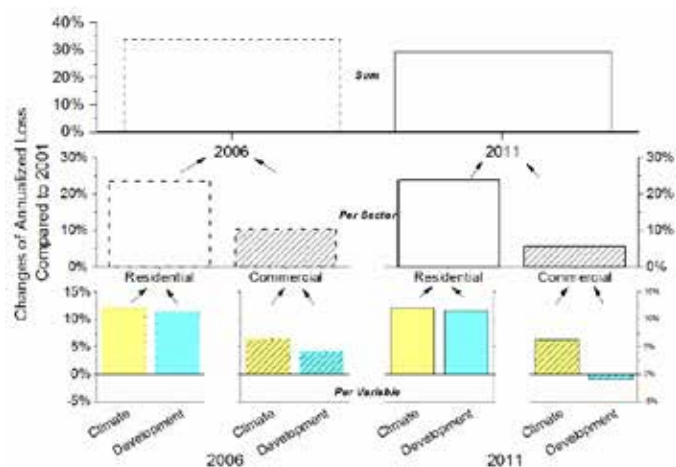
The outcomes of this research highlight the importance of not only considering non-stationary patterns in climate when estimating future flood consequences, but that flood control

planning should also include non-stationary patterns in the urban environment, which include not only new development, but market dynamics, as well. While this study only utilized urban development data over a 10-year period, it is shown that dynamics in the urban environment can contribute to the increase of flood loss by the same order of magnitude as the contribution from climate change.

There are obvious uncertainties associated both with quantifying future climate conditions and with estimating conditions within the built environment, including market conditions. The objective of this study was not to absolutely quantify future conditions, but rather to develop tools to help understand the relative contributions of conditions, given best available knowledge. The toolset and datasets developed in this study also provide the ability to incorporate mitigation and adaptation strategies and quantify the impact strategies have on minimizing damage and the population at risk, which will be targeted in the future work.



Increase in damage, annualized risk, and affected population from the current to the future climate scenarios (all of the costs were normalized by 2011 U.S. dollars).



Percent changes of annualized loss attributed by sectors and variables (all of the changes were calculated as the absolute percentages compared to the 2001 development condition forced by the historic climate data; the first row of the plot represents the sum of changes for 2006 and 2011 development conditions with the future climate forcing, the second row represents the percent changes per sector for each year, and the last row represents the separate changes caused by climate variation and urban development within each sector for each year).

Quantifying Carbon Fluxes and Underlying Mechanisms Using Multiple Data Sets with a Joint Land-Atmosphere Ensemble Kalman Filter Data Assimilation System

Ghassem R. Asrar

The purpose of this project has been to utilize and further develop a coupled land-atmosphere carbon data assimilation system that will simultaneously handle multiple modeling components and multiple streams of data to assess the spatial and temporal variations in carbon sources and sinks.

The coupled land-atmosphere carbon data assimilation system this project is developing was evaluated specifically with observations from Orbiting Carbon Observatory-2 and GOSAT satellites, together with other datasets such as CO₂ measurements from flask sampling network, remotely sensed vegetation indices and fraction of Absorbed Photosynthetically Active Radiation, FLUXNET CO₂ and water and heat flux measurements, vegetation and soil biometrics, and land cover/use history. The working hypothesis has been that using a combination of these measurements in the proposed coupled land-atmosphere data assimilation system will enable us to characterize the spatial and temporal variability, hence the uncertainties, associated with the major global/regional carbon sources and sinks. The scientific knowledge gained and data sets developed are important for 1) bounding the uncertainties associated with climate sensitivity to CO₂ build up in the atmosphere, 2) determining the capacity of terrestrial ecosystems in sequestering the excess CO₂ from the atmosphere, and 3) developing climate- and carbon-related policies and management practices for future.

In FY 2016, we carried out several tasks over different phases of this project, including those that follow.

Development of a joint land-atmosphere CO₂ data assimilation system. The main focus was on the testing and validation of coupling between the VEGAS and GEOSchem models via the observation operator and the use of both surface-based and space-based CO₂ measurements.

Analyses of Tier 1 datasets. We examined GOSAT XCO₂, flask CO₂, FLUXNET, and NDVI/EVI/fAPAR.

Assembling and processing Tier 2 datasets. These datasets included soil and vegetation biometrics, flux tower footprint,

and land use and disturbance history, as a complement to Tier 1 datasets.

Conducting experiments. We conducted coupled land-atmosphere experiments in forward mode; compared transported CO₂ at representative stations, as well as direct comparison of land fluxes/vegetation with FLUXNET; and derived phenology and physiology information from remotely sensed observations and conventional data sets. We also assimilated flask CO₂ measurements with atmosphere models and compared them against traditional atmospheric inversions as a benchmark.

During the current period, we continued to assemble key data sets, including historical remotely sensed observations and high-resolution photographs for key eddy flux observation sites and major agro-economic zones in the United States.

We participated in an ISI-MIP project that is focused on inter-model comparisons to compare VEGAS vegetation-atmosphere model with other comparable models for better characterizing their uncertainties in simulating the carbon cycle, which is essential for uncertainty analyses of coupled carbon data assimilation experiments later on. ISI-MIP consists of two sets of experiments: historical and future. There are four historical experiments, primarily based on different climate data presented in table below. ISI-MIP also provides land cover type and land use data based on HYDE3.1 crop land and pasture areas. The ISI-MIP project requires spatial resolution of simulations to be at 0.125 degree. In addition, we run the VEGAS model at 0.5 and aggregated to 2.5 degrees spatial resolution for further analyses.

Based on these simulations, we observed 7 to 10% variation in net primary productivity (NPP) and 6 to 8% in soil carbon under different historical climate conditions. We also observed some differences in carbon allocation to plant roots and woody biomass. Other than these variables, we have not seen appreciable differences among other VEGAS model outputs. We presented these findings at the Ecological Society of America in Spring 2015, Baltimore, Maryland.

Next, we focused on the ISI-MIP future scenarios experiments. There were a total of 5 experiments and 13 different simulations for the major Representative Concentration Pathways;

climate forcing of 2.6, 4.5, 6.0, and 8.5 W/m²; and corresponding CO₂ concentrations.

Other research activities conducted in FY 2016 included a series of coupled carbon data assimilation experiments to understand and quantify 1) the complementary features of CO₂ measurements by different methods; 2) explore the limits to quantifying seasonal, inter-annual, and decadal variability in CO₂ exchange between atmosphere and terrestrial ecosystems; and 3) carry out coupled CO₂ assimilation experiments to understand and quantify the spatial variability in sources and sinks of CO₂ for major biomes and eco-regions, globally.

We developed indicators of terrestrial ecosystems phenology and eco-physiology changes based on Landsat and MODIS remotely sensed observations to be used in the vegetation-atmosphere models such as VEGAS. The initial focus of this task was the United States, but we are testing the applicability of this approach for some major select eco-regions (e.g., tropical and boreal systems) worldwide.

The main components of the data assimilation system when applying *in situ* and satellite CO₂ data consisted of observations from surface stations flask and OCO-2, atmospheric carbon data assimilation uses LETKF-Carbon with GEOSChem model for atmospheric transport, and flux (Fnet) assimilated as model parameter (space-time varying) together with CO₂ concentration.

We have successfully conducted several data assimilation experiments for the period of 2000 to 2016. The seasonal cycle of the global land-atmosphere CO₂ fluxes are consistent with expectation, based on comparison with the VEGAS model. The VEGAS model has been compared for its performance and comparability with several other models in the international TRENDY and ISI-MIP, as well as nationally in MSTMIP model inter-comparison projects. The results are also comparable to many atmospheric inversion results that have used GLOBALVIEW-CO₂ network data in the past 20 years. During the OCO-2 period (2015), using satellite data alone, we were able to obtain the seasonal cycle, albeit with some noise, suggesting the limitation in temporal coverage and/or bias in the data.

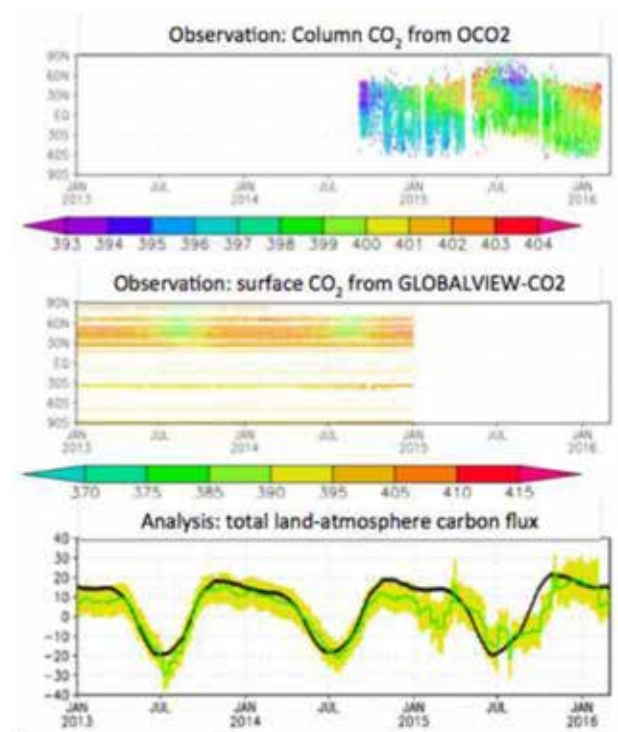
Relative to the large seasonal carbon cycle (global total of more than 30Gt/y) due to the Northern Hemisphere biospheric growth and decay, the long-term mean carbon sources and sinks (global total 1-2 Gt/y) were more challenging to capture. The data assimilation system succeeded in capturing the general patterns (Fig.4) such as the NH carbon sink. Comparison with other inversion products shows that the detailed spatial pattern may differ, because there is limited observational constraint at model grid resolution. The information at that scale comes from either prior biosphere model (CarbonTracker) or other assumed large-scale correlation (Jena). The LETKF system uses very limited prior information, so it is quite remarkable that the system is able to

obtain reasonable grid-scale information without observations at that scale. Nevertheless, major biases are seen in subtropical regions such as India and Sahel.

When OCO-2 data is injected starting from September 2014, such biases are significantly reduced (Fig. 4 lower-left panel). We hypothesize that even with infrequent sampling, OCO-2's tropical-subtropical coverage results in this improvement, as a complement to the GLOBALVIEW-CO₂ *in situ* network (Fig.2). The OCO-2 data was pivotal in constraining the assimilation in those regions where GLOBALVIEW-CO₂ data is inadequate.

The assimilation system now produces some products as proposed by this project, including 3-D fields of analyzed CO₂ as an optimal combination of model and observations, as well as surface carbon fluxes from data assimilation analysis, constrained by a combination of OCO-2 and *in situ* data. These assimilated data products are at 6-hour and monthly resolution for 2000 to 2016 period.

In FY 2017, we plan to engage in joint land-atmosphere assimilation with multiple datasets and experiment with XCO₂ and SIF data from OCO-2 as soon as available. We also will quantify uncertainties in fluxes and model parameters/processes, and prepare a final set of papers for publication in open literature.



Hovmuller diagram (time-latitude plot) of zonal mean column CO₂ (XCO₂) from OCO-2 satellite (a), *in situ* GLOBALVIEW-CO₂ network (b), global total land-atmosphere carbon fluxes I ppm (c), and carbon flux Gt/y from the data assimilation using GEOSChem atmospheric transport model and the observations described above. Note the higher noise in 2015 when only OCO-2 data is available. In (c), black: VEGAS modeled flux is presented for comparison.

³²Si Geochronology Gap Analysis

Gary A. Gill

This project will produce new and enhanced capabilities that will have significant impact on environmental geochronology and other low-background radiation measurement applications, significantly advancing PNNL as a national leader in these areas.

Two previous projects, (Enhanced Sediment Geochronology Achieved Using Ultra-Low Background Materials and Ultra-Sensitive Detection Capability – GeoSed) were to create methodology (chemistry) and an analysis platform (BIDS Detector), with the ultimate goal of utilizing ³²Si as a sediment chronometer. This project carried that plan forward with the challenge of finalizing the detector requirements and processing the existing sediment samples for analysis in the shallow underground laboratory (SUGL).

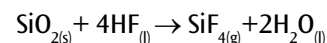
Ultra-sensitive beta detector development. The background performance of prototype low-background beta particle detectors was improved in FY 2016. The completion of a new shield system in the PNNL SUGL allowed the detector backgrounds to be reduced from about 40 counts per day (cpd) per counter in 2015 to 23±1 cpd for a set of 2 detectors in 2016 (about 11.5 cpd per detector), as seen in Figure 1. This background is significantly lower than typical beta counter backgrounds of approximately 1,500 to 3,000 cpd per counter and represents over an order of magnitude sensitivity improvement. A new detector housing was also assembled and tested, eliminating high-frequency noise pickup and providing robust sample integrity to protect the clean room housing the counting system.

A demonstration measurement of the reference fission product isotope ⁹⁹Mo showed that a sample with a starting activity of 1 mBq (86 decays per day) could be quantified with 15% uncertainty. Measurement of the first ³²P sample separated from a Puget Sound sediment core (Carr core) showed that the sample was not sufficiently radiopure to meet project ³²Si age dating goals. This result inspired the project team to develop a path forward to improve radiopurity of the final ³²P samples for ³²Si age dating.

Alternative Si chemistry. The chemistry for silica separation was taken from the initial project GeoSed, and the initial

cores were utilized from this project. During this year, the chemistry supported the measurement of the uppermost section of the Carr Core after radiochemistry processing showed an excess background rate as high as approximately 1,600 cpd, equating to roughly 1.1 cpm or 18.3 mBq. This sample-associated background level prevents us from taking advantage of the beta detector's potential for low level ³²P measurements. During development of sample process chemistry in parallel with the detector development, samples were measured on a low background germanium spectrometer; measured activities were less than the gamma minimum detectable activity for all isotopes, but not sufficiently low to ensure reaching the desired background for beta particle measurement.

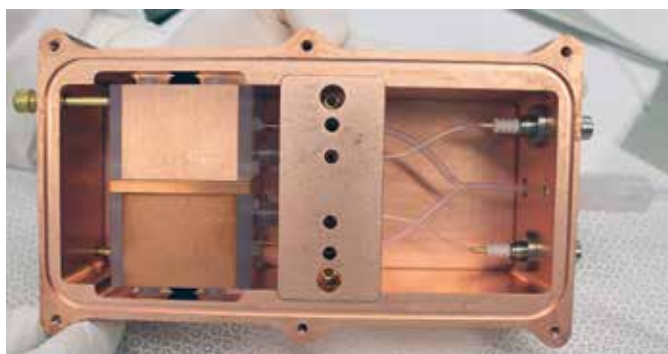
An alternative radiochemical purification method has been identified for future ³²Si age dating research. The off-gassing of SiF₄ during dissolution of silica with hydrofluoric acid can be engineered into a distillation process useful for purification of silicon, using the following chemical reaction.



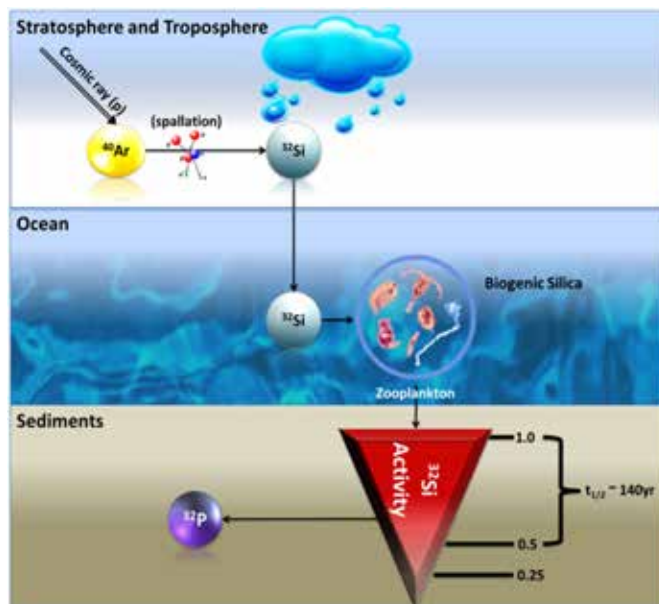
A distillation process of this type may be effective in isolating the distillate from radio-impurities in the original sample. In this process, a key step is recovery of the gaseous SiF_{4(g)}. A literature search revealed that a byproduct of phosphorus-based fertilizer production is SiF₄, a highly reactive and a toxic gas, which, therefore, must be treated before it is released into the atmosphere. Industrial-scale wet scrubbing units are employed where the gas is bubbled through water to capture the gas and convert the silicon into SiO₂. On a smaller scale, this method can be used in the purification of silicon recovered from sediment samples.

In summary, this project has successfully demonstrated a low background beta detector with more than an order of magnitude improvement in sensitivity relative to the most sensitive beta detectors used to support national security missions (e.g., National Technical Nuclear Forensics and radiochemistry supporting Nuclear Test Monitoring). One of the original goals of the project, to establish a new ³²Si sediment age dating capability, is not fully in place due to sample-associated backgrounds that do not allow full utilization of the radiation measurement capability. After this issue was identified, a novel radiochemical separation approach was developed; ini-

tial research into this method indicate it has significant potential to meet the project sediment age dating goals. It is important to note that, although not all of the goals were achieved in terms of the chemistry, that the detectors built on this project will be able to help develop and optimize the low-level chemical separations for future projects, since the sensitivity is now below what was possible with the low level gamma spectrometers.



Set of low background beta detectors assembled into copper carrier.



^{32}Si is produced in the troposphere by cosmic radiation; it is removed from the atmosphere in wet and dry deposition, incorporated into plankton, and removed to sediments (biogenic silica). ^{32}Si activity decreases with depth (half-life ~ 140 years) in sediments, providing a means to age date sediment layers. The daughter of ^{32}Si , ^{32}P , is quantified, as it has a higher activity and is in secular equilibrium with ^{32}Si .

Simultaneous ^{14}C and T Dating of Environmental Organic Matter

Jim Moran

We are developing high-sensitivity methods for tritium (T) age dating of organic compounds. Once developed, this advanced technique can be coupled with other approaches (including 14-carbon [^{14}C] analysis) to provide a powerful tool for detailed analysis of carbon cycling through environmental systems.

A well-established method, ^{14}C age dating of organic samples, enables research investigations in areas as varied as microbial substrate tracking, carbon cycling, dating anthropological samples, atmospheric chemistry, and biomarker sourcing. By definition, however, organic molecules also contain hydrogen that can be independently age dated using T analysis. Tritium has a much shorter half-life than ^{14}C , thus it can be used to track different processes and target different age ranges than ^{14}C .

While T has widespread applications in hydrologic, limnologic, oceanographic, and other investigations, current measurement approaches make it difficult to apply to organic molecules due to the large sample size requirements for analysis. Furthermore, T content in the biosphere spiked during the 1960s at approximately 1,000 times natural levels due to anthropogenic inputs, but these levels have since largely subsided, making measurement of T to track a number of biological and physical processes more challenging.

The primary technological goal of this project is to improve the measurement sensitivity of T to enable more widespread use of T measurements in environmental investigations. One potential application space is in exploring organic bound T (OBT) and processes involving these molecules. Once developed, our approach will provide a platform for enhancing understanding of molecular cycling and stability of environmental organic compounds and refine our understanding of chemical recalcitrance with respect to the C cycle.

Our efforts have focused on 1) improving measurement sensitivity for T and 2) developing methods for converting small samples into a suitable form for measurement (i.e., conversion into gaseous methane). Together, successfully accom-

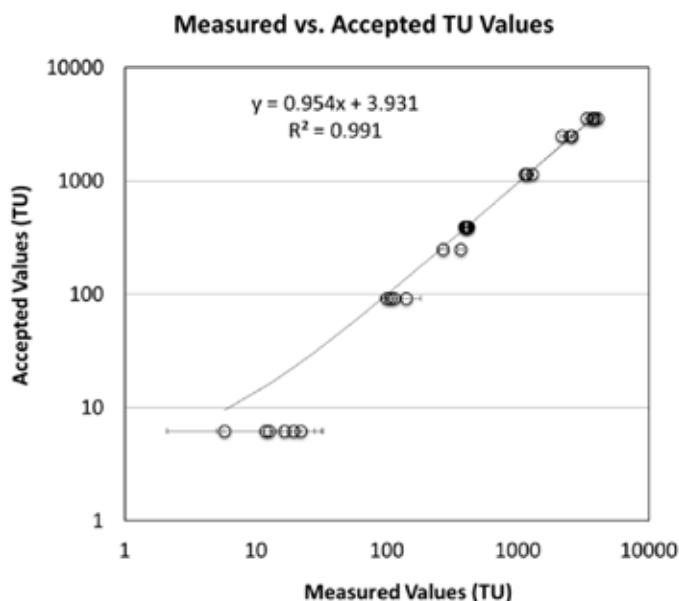
plishing these goals will link small environmental samples to accurate T measurement, and the capability can be applied to help address a wide range of environmental science questions.

Multiple ultra-low-background proportional counters (ULBPCs) were fully characterized for this project using 7-atm background composed of a 90% argon/10% methane (P10) gas blend and measured in PNNL's Shallow Underground Laboratory. This laboratory offers significantly reduced interferences from background activity to help enable improved measurement sensitivity and, thus, smaller sample sizes. Furthermore, materials developed in this underground lab were incorporated into the detectors to reduce inherent background signals and help improve overall sensitivity. Backgrounds and efficiencies were characterized on a suite of ULBPCs. Further, we developed the stand-alone hardware needed to fill these counters with a known volume of sample-derived methane and then introduce the proper amount of argon fill gas.

We demonstrated simultaneous ^{14}C and T measurement using samples of environmental methane collected from a landfill and wastewater treatment plant site. Comparison of simultaneous measurements performed at PNNL to individual T and ^{14}C quantification made by a commercial vendor (with much higher sample volumes) demonstrated the dual isotope approach's vastly improved sensitivity (requiring orders of magnitude less sample) with highest precision and accuracy for samples with elevated T, compared to ^{14}C content.

Central to the project's success is the ability to convert organic samples into methane, which is an ideal gas for dual isotopic measurement by proportional counting. We have developed the hardware needed to combust organic samples and quantitatively collect the resulting product water and carbon dioxide. We recently constructed the hardware needed to convert the resulting combustion water into methane to permit T measurement. The general approach utilizes a heated zone with zinc reagent to thermally crack water and trap the byproduct oxygen (immobilized as either a zinc or manganese oxide) resulting in net molecular hydrogen production. A heated ruthenium catalyst catalyzes methane formation from the hydrogen and carbon dioxide. Finally, a series of cryogenic steps are performed to assure methane purity and to facilitate sample loading into the ULBPC for measurement.

We used a series calibrated water standards containing different amounts of T to test the accuracy of the method (inclusive of methane synthesis, purification, and T measurement). We observed high chemical conversion yields to methane, efficient cleanup of the product methane using multiple cryogenic steps, high transfer efficiency from the reaction chamber to the ULBPCs, and a strong correlation between our measurements and the accepted T content of the starting water standards. Importantly, however, was that this was accomplished using one to two orders of magnitude less starting water volume (approximately 120 μ L) than for traditional approaches. The added sensitivity of these methods over alternative approaches offers a valuable technological capability for expanding T measurements into a wide range of environmental applications.



Accuracy of the developed method is demonstrated by making measurements of water standards having known T content. There was high agreement between the measured results and accepted values ($R^2=0.991$). A point of distinction, however, was that our measurements were performed using one to two orders of magnitude smaller sample sizes than those used to calibrate the standards.

The Role of Hurricanes in the Carbon and Oxygen Dynamics of the Coastal Zone and its Global Significance

Laura Bianucci

Extreme events (e.g., hurricanes, storms) are expected to increase with climate change, but their role in modulating the carbon and oxygen cycles in the coastal ocean remains uncertain. By using different datasets (i.e., satellite, in situ, and reanalysis), we established the important role of hurricane-induced sediment resuspension relative to the transport by river plumes and will evaluate ways of including this process in earth system models to improve climate predictions.

Carbon burial in the ocean is dominated by the transport and burial of terrestrial material from the continents to the coastal zone. This is due not only to the refractory nature of molecules formed in terrestrial vegetation and soils, but also to the rapid sedimentation rates in coastal zones that quickly bury organic carbon below the shallow oxic layer of most sediments. These sediments can be resuspended by the strong shear generated by hurricane-induced currents at the bottom boundary of the shallow continental shelf, re-exposing them to oxygen and enhancing the remineralization of previously buried organic carbon (both terrestrial and marine).

This process produces carbon dioxide and consumes oxygen; however, it is not well represented in current global models, which tend to oversimplify (or ignore) sedimentary processes and, typically, cannot explicitly resolve hurricanes. Moreover, hurricanes can increase the transport of organic material to the coastal ocean, increasing the pool of organic carbon to be remineralized. In this project, we aim to evaluate the importance of sediment resuspension and transport by hurricanes, with a focus on the carbon and oxygen cycles. Furthermore, we plan to parameterize these processes such that they can be included in regional and global models.

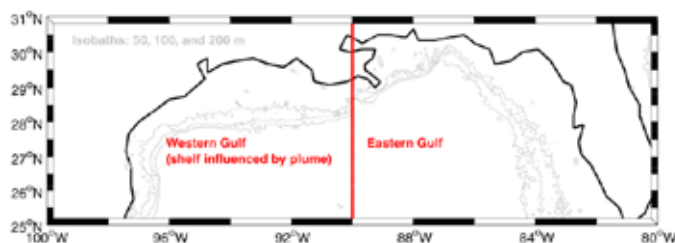
We use the northern Gulf of Mexico as the test bed to investigate the potential impacts of hurricanes and strong storms on dissolved oxygen (DO) in the coastal ocean, with a suite of *in situ*, remotely sensed, reanalysis, and published data sets.

Using sea surface salinity from model reanalysis and satellite data, we first demonstrate a time lag of approximately 5 to 7 days from the time a hurricane makes landfall near the Texas-Louisiana shelf to the time when the Mississippi River plume induced by the hurricane arrives at the mouth of the delta. This time lag allows the local impacts of hurricane-induced mixing on DO at the continental shelf to be isolated from the delayed response to the plume. Furthermore, we use satellite-derived observations of total suspended matter and particulate organic carbon, as well as spatially sub-sampled data from the eastern Gulf (with less river plume influence) and a western Gulf (dominated by plume dynamics) to estimate the local and delayed response of DO to hurricanes. Our analysis agrees with literature estimates of the total resuspension of sediments by hurricanes in the region (about 10 times the annual deposition by the Mississippi/Atchafalaya River system).

Finally, using ship cruise measurements of DO before and after hurricanes, we analyze the two primary processes that occur during the hurricane and before the arrival of the plume: 1) vertical mixing that homogenizes the water column and decreases the hypoxic area, 2) sediment resuspension and subsequent remineralization that tend to increase the hypoxic area. We find that the consumption of DO from sediment resuspension partly compensates the effects of vertical mixing and is a significant sink of DO (about 10 to 15% of the effect of remineralization of terrestrial organic matter brought by the river plume). Furthermore, most shelf regions subjected to hurricanes are not influenced by large river plumes, while we can expect that sediment resuspension will be an ubiquitous process in mostly every continental shelf under extreme weather events. Therefore, our results point to the important role of sediment resuspension on hurricane-induced oxygen response, not only in the Northern Gulf of Mexico, but also in other regions, with potential implications for the carbon cycle.

This work will be presented in January 2017 at the American Geophysical Union (AGU) Chapman Conference and will be submitted as a journal article before the end of 2016. Furthermore, in FY 2017, we will use a 1-D ocean-modeling framework (which includes sediment deposition and resus-

pension) to further test and quantify the role of sediment resuspension and transport of terrestrial organic matter due to hurricanes in oxygen and carbon cycles. The goal will be to develop a parameterization that allows for including these effects in regional and global climate models. Most of these models do not have enough resolution to explicitly resolve hurricanes and their role in enhancing transport of organic matter from the watershed to the coastal zone; moreover, climate models tend to underestimate the magnitude of extreme precipitation events, misrepresenting the transport of terrestrial material. Climate models also usually ignore or oversimplify the sediment-water boundary, which prevents the proper representation of sediment resuspension. The proposed parameterization would allow climate models to represent the hurricane-intensified carbon export and remineralization by using variables readily available in global models (e.g., wind speed, bathymetry, river discharge and organic carbon loading, and local primary production).



Map of Gulf of Mexico. Red line at 90°W divides the western Gulf (plume-influenced region) and the eastern Gulf (not strongly influenced by the plume).

Understanding Polar Climate Sensitivity

Hansi A.K. Singh

The polar regions play a key role in moderating global climate, suggesting that climate change in the Arctic and Antarctic will have major consequences for Earth's climate system. This project aims to pinpoint the relative roles of atmosphere and ocean dynamic processes in determining the sensitivity of polar climates to anthropogenic forcings.

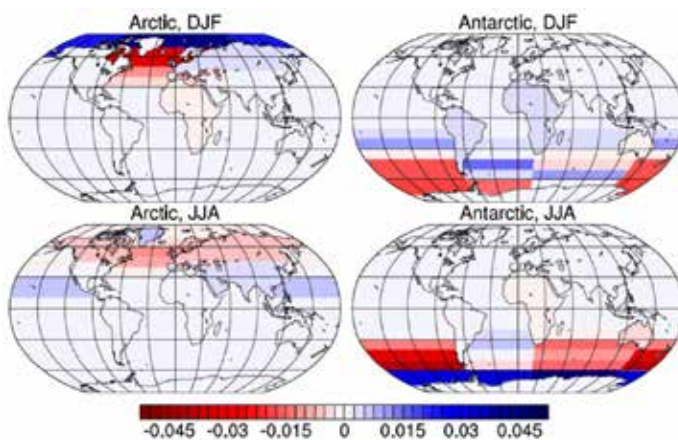
Previous studies have identified many local factors that enhance polar climate sensitivity, such as ice-albedo feedback processes, breakdown of winter temperature inversions, increased atmospheric precipitable water, and positive cloud feedbacks. However, the impact of large-scale dynamics on polar climate sensitivity, particularly changes in ocean and atmosphere energy flux convergence, remains an open question. The aim of this project is to isolate the role of ocean and atmospheric dynamics on the perturbation response of polar climates to forcings. Such an approach will highlight how differences in the general circulation in the Arctic and Antarctic affect the local climate sensitivity of each region.

This project started late in FY 2016. In the short time it was active, the Principal Investigator (PI) participated in writing a proposal for computing time at the National Energy Research Scientific Computing Center and prepared a manuscript on moisture transport into the polar regions using model runs from global climate model experiments that include numerical water tracers. This paper will be submitted for peer review in early FY 2017. In this work, we find that, from a Lagrangian perspective, the hydrologic cycle perturbation response to CO₂-induced warming is very similar in both the Antarctic and Arctic. In particular, we find that moisture source regions shift equatorward in summer but become strongly focused over regions of sea ice retreat in winter.

The project continues to assemble and develop the appropriate models needed for this research and plans to use a range of varying-complexity models to study the many dynamic mechanisms that mediate polar climate sensitivity, including single-column radiative transfer models, dry dynamical cores, moist dynamical cores, atmosphere-only models with gray

radiation, atmosphere-only models with full radiation, atmosphere models coupled to a slab ocean without continents (aquaplanet) and with continents, and atmosphere-ocean models (fully coupled) with different configurations of polar orography.

In FY 2017, we will begin new modeling work assessing the sensitivity of polar climates to changes in ocean energy flux convergence and heat uptake. We will conduct a series of model experiments with the Community Earth System Model in a slab ocean configuration, in which both the ocean heat transport and deep ocean heat uptake are adjusted. Comparing these slab ocean experiments to the fully coupled atmosphere-ocean case permits the role of ocean dynamics to be isolated. By comparing a fully coupled CO₂-doubling experiment with several of these slab ocean simulations, the relative sensitivity of Arctic and Antarctic climates to changes in ocean energy transport and ocean heat uptake can be ascertained. Given that ocean heat uptake is a transient process, while ocean heat transport is expected to settle at an equilibrium state, such studies can also be used to constrain the transient and equilibrium climate sensitivities of the polar regions.



Change in the fraction of Arctic (left panels) and Antarctic (right panels) precipitation sourced from different regions due to CO₂-doubling. DJF refers to the December-January-February climatology, and JJA refers to the June-July-August climatology. Experiments show that local regions where sea ice retreats become a crucial source for polar precipitation in winter, while remote regions become more important in summer.

Understanding the Role of Coastal Wetlands in Carbon Cycling – An Integrated Modeling-Observation Approach to Improve Regional Earth System Modeling

Zhaoqing Yang

Carbon fluxes among land-coastal and wetland-ocean interfaces are substantial components of the global carbon budget, yet remain poorly understood and quantified, largely due to the highly heterogeneous and dynamic nature of these interfaces. This project aims to quantify carbon fluxes and transport across the interfaces of wetland-estuary, air-sea, and water-sediment and to better understand the role of wetlands in coastal carbon cycling using an integrated modeling-observation approach.

Carbon movement from terrestrial systems to the oceans, through coastal wetlands, is a critical pathway of the global carbon cycle. There is growing research showing that these ecosystems may be relatively large carbon sinks, globally. Coastal wetlands, in particular, have been persistent atmospheric CO₂ sinks over recent millennia, but are significantly understudied and pose the greatest uncertainty and variability with regard to carbon cycling. Coastal wetlands are absent in Earth System Models (ESMs) at global and regional scales. Therefore, there is an urgent need to develop a coupled wetland-ocean model that simulates the biogeochemical processes in the coastal wetland carbon pool, as well as fate and transport processes at wetland-estuary, wetland-atmosphere, and water-sediment interfaces.

The proposed study establishes a new capability of integrated tools with *in situ* measurements and coupled ocean-wetland models for simulating the fluxes and transport processes of carbon cycling in the coastal zone. The wetland model simulates carbon transport and transformation at the interface of terrestrial and ocean ecosystems and, thus, fills the current gap between land and ocean components of ESMs. The model addresses key physical transport and biogeochemical processes, as well as the interactions and feedbacks between them.

An integrated modeling and observation approach was used to develop predictive and *in situ* measurement tools to understand the carbon cycle in coastal wetlands. Model development largely leverages existing research conducted at PNNL. Sequim Bay, WA, was used as the study site for field measurements of key carbon parameters in coastal waters and greenhouse gas exchanges in marshes and model testing.

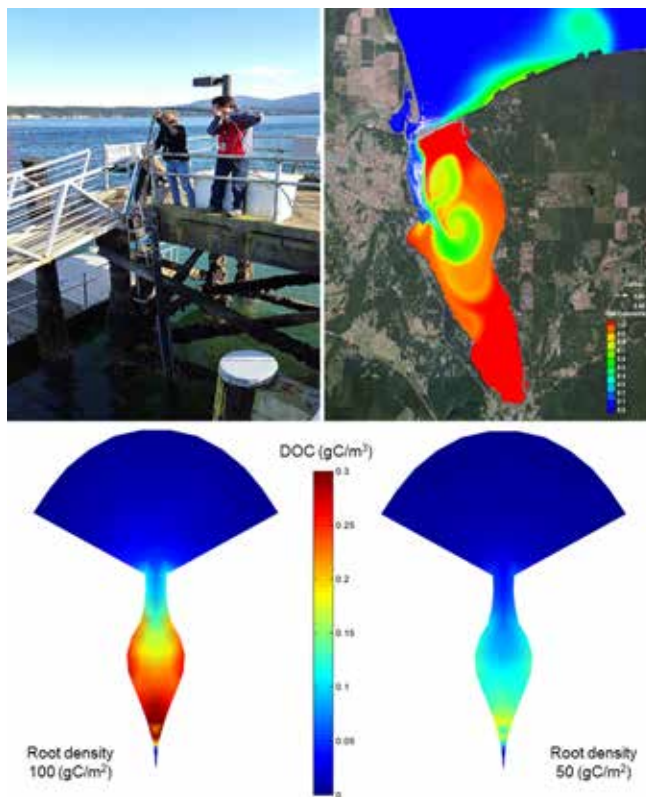
In situ measurements of key parameters were conducted in the water column at the dock of the Marine Sciences Laboratory in Sequim Bay and in a channel connecting the bay to a tidal salt marsh lagoon. High-frequency data for temperature (T), salinity (S), dissolved oxygen (DO), colored dissolved organic matter (CDOM), dissolved organic carbon (DOC), total suspended solids (TSS), and nitrate (NO₃) were periodically collected from spring to summer. Greenhouse gas emissions, including CO₂, CH₄, N₂O, and NH₃ in the marshland were simultaneously collected as the water column sampling. Collected data showed strong tidal signals, indicating the carbon flux and cycling in coastal wetlands is influenced by the physical processes. Data collected are used to parameterize the coastal wetland model and support model validation in Sequim Bay.

A coastal wetland model called FVWETLAND has been developed within the framework of the Finite Volume Community Ocean Model (FVCOM), which includes a nutrient, phytoplankton, zooplankton, detritus module; sediment diagenesis module with two layers of organic-matter-enriched sediments; and dissolved organic matter (DOM) module in the water column and the sediments. The wetland model is directly coupled with the coastal hydrodynamic model to simultaneously simulate coastal circulation and biogeochemical processes.

The coupled hydrodynamic-wetland model was tested to evaluate the effect of marsh root exudation on DOM flux from sediment bed into the water column in an idealized model domain, which mimics the spatial scale and physical-biogeochemical processes in Sequim Bay with marsh root mass placed along the coastal line nodes of the embayment.

Preliminary model results demonstrated the coupled hydrodynamic-wetland model is able to simulate DOC production contributed from marsh root, sediment diagenesis, and interaction with water column under different marsh root density conditions.

With continued funding in FY 2017, a submerged marsh growth module, which includes leaves, shoots, stem, and root, will be developed to simulate the dynamics of biomass and organic carbon cycling and flux across the terrestrial-aquatic interface.



Understanding the of coastal wetlands in carbon cycling: a) field operation; b) hydrodynamic modeling of Sequim Bay; c) simulated dissolved organic carbon in an idealized estuary and coastal bay.

vPASS 2.0: Mechanistic Process Components of a Virtual Plant Simulator

Yilin Fang

There is a critical need to improve the accuracy of plant process contributions to the carbon cycle in climate model projections. A systematic, mechanistic, and quantitative understanding of how plants invest and allocate carbon in the context of their immediate environment will be key to addressing this need. This research project will develop an integrated plant ecosystem modeling capability for plant performance as a function of genotype and environmental conditions.

Brachypodium has been well-studied for investigations of genomic potential for plant performance; however, no computational process models of *Brachypodium* structure and function have been published. This project will develop a novel prognostic Virtual Plant-Atmosphere-Soil System (vPASS) model that integrates a comprehensively detailed, mechanistic, single plant model with microbial, atmospheric, and soil system processes in its immediate environment. This development will focus on *Brachypodium sp.* as a model for bioenergy grasses. There are three broad areas of process module development under this project, including 1) incorporating models for root growth and function, rhizosphere interactions with bacteria and other organisms, litter decomposition and soil respiration into established porous media flow, and reactive transport models; 2) incorporating root/shoot transport, growth, photosynthesis, and carbon allocation process models into an integrated plant physiology model; and 3) incorporating transpiration, volatile organic compound emission, particulate deposition, and local atmospheric processes into a coupled plant/atmosphere model. Partnering with other research organizations, the developed model will be released as a community open source model.

The project started late in FY 2016. Principal efforts have focused on planning and coordination, and the identification, acquisition, and testing of plant modeling software. The vPASS team has had ongoing discussions on data needs with the other focus areas involved in *Brachypodium* experiments. A sound experimental design is key to the successful parameterization and testing of the vPASS simulation capability. To date, the following models have been acquired for testing.

Multiscale Framework Model for Arabidopsis developed by the Plant System Biology Modeling group in University of Edinburgh, UK.

ROOTBOX model developed by Daniel Leitner, University of Vienna, and Andrea Schnepf, Forschungszentrum Jülich GmbH (Germany). It can easily create time dependent branched geometries of growing plant root systems. The resulting root systems can be coupled to arbitrary soil models such as nutrient and water uptake models.

SimRoot model from Pennsylvania State University. This 3D architectural root model also calculates nutrient uptake as the roots grow and receive photosynthate from the shoot.

WIMOVAC/BioCro model from University of Illinois at Urbana-Champaign. It simulates system scales from leaf-level photosynthesis to ecosystem-level carbon and water balance.

We have reviewed papers and code documentation for these models and successfully installed and tested them. We acquired code documentation of Root-Soil Water Movement and Solute transport, a numerical model for simulating solute transport and water flow within the soil-plant system from Forschungszentrum Jülich GmbH. We also had model development and collaboration discussions with plant modelers in the Plant Systems Biology Group at the Chinese Academy of Sciences-Max-Planck-Gesellschaft Partner Institute for Computational Biology and the Icahn School of Medicine at Mount Sinai.

The team presented the vPASS concept to the 2016 Multiscale Plant Modeling workshop, August 25–26, and participated in the 2016 EMSL Multiscale Ecosystems Analysis and Design Workshop, September 12–13. An abstract was submitted to the 2016 American Geophysical Union Fall Meeting, December 12–16.

In FY 2017, we will continue the root modeling development effort, as well as the *Arabidopsis* model testing with existing data sets. As the *Brachypodium* experimental data become available, *Brachypodium*-specific process module development will accelerate. The goal is to have a preliminary whole plant *Brachypodium* model, BP1.0, with basic functionality, ready for testing at the end of the fiscal year.

Energy Supply and Use



Agent-Based Test Bed for Complex Building Control Systems

Siddharth Goyal

This project is developing a test bed to expedite the application of advanced control theories and tools in buildings, which will benefit energy efficiency, occupant comfort, and reliable operation of the power grid.

Multi-agent systems for distributed controls have been successfully applied to robotic systems, air traffic management, and other uses. However, only small, fragmented exploratory studies have been performed so far for agent-based building controls. Partly because of the lack of realistic and flexible test beds for distributed building controls, those studies are limited to either ad-hoc simulations, or they only tackle simple problems for a specific building. To fill the gap, this project was initiated with the following two major objectives: 1) to develop an agent-based test bed for distributed control of building systems and equipment and for interactions between buildings and the grid, and 2) to demonstrate use of the test bed in investigating agent-based control approaches for representative building systems.

The project is designing and deploying an advanced building controls test bed in real buildings on PNNL's campus. The test bed is expected to have the following characteristics: 1) it supports interactions among heterogeneous components and systems (e.g., pumps, air-handling units, lighting systems, water heaters, and occupant behavior); 2) it can easily be reconfigured to test/validate/demonstrate different control methodologies and tools under realistic but controllable conditions; and 3) it is an integrated part of a larger test bed that includes renewable generation, energy storage, grid, and peer buildings. These characteristics will enable the building controls test bed to support testing and demonstration of new control solutions and technologies developed from other related projects.

In FY 2014, we identified the test bed requirements from different aspects such as test bed core characteristics and capabilities, data collection, monitoring, communication, experimental setup and management. Based on these requirements, we proposed a building test bed infrastructure.

The building tested supports both physical and virtual devices. To achieve distributed controls in real buildings, each application-specific controller is associated with a single-board computer that has VOLTTRON™ installed, which is an agent execution platform developed at PNNL. Different control algorithms can be implemented as different agents in the single-board computer. When testing, the applicable control logic implemented in VOLTTRON™ will override the original control logic in the corresponding application-specific controller. The multi-node communication capability of VOLTTRON™ enables a controller to get data from related controllers, if necessary. A high-level computer is an interface for communication with external entities. The infrastructure was implemented at the Building Diagnostics Laboratory (BDL), which is a test facility on PNNL's campus. A small test case on the single-duct variable-air-volume system at BDL was created to try out the key VOLTTRON™ functionalities to be used in the test bed.

The test bed infrastructure can support different control topologies, depending on the location of control functionalities. For example, centralized controls are achieved if all single-board computers at the bottom level provide no control capabilities except passing data while the high-level computer has all control functionalities. Similarly, hierarchical controls can be achieved by introducing middle-level single-board computers and allocating control functionalities accordingly.

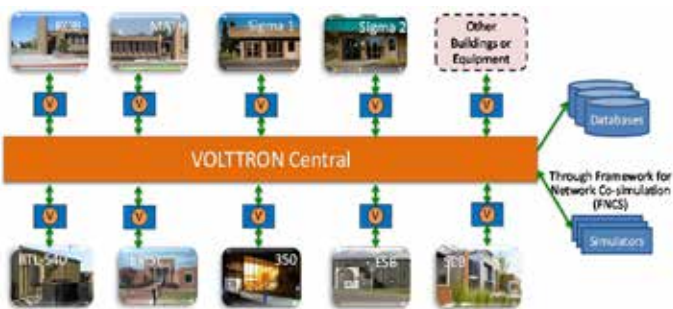
In FY 2015, the building test bed module infrastructure was enhanced to allow switching between original control systems and new controls. The building test bed was expanded to include the System Engineering Building (SEB). A couple of use cases were implemented to verify the ability of the initial test bed to support local data storage, data collection at different intervals, remote access and configuration, different control topologies, and communications among heterogeneous devices. A number of Modelica models were developed for different building components such as variable-air-volume terminal boxes, air-handling units, chillers, and cooling towers. A solution to hardware-in-loop based on VOLTTRON™ and Modelica models was designed and implemented.

In FY 2016, this project’s final year, we achieved several notable outcomes. In coordination with another related project, we have expanded the building test bed to connect to nine real buildings. Building operation data have been continuously collected from those buildings at 1-minute intervals for a few months. Moreover, the building test bed structure enables easy expansion to incorporate additional buildings and equipment in the future.

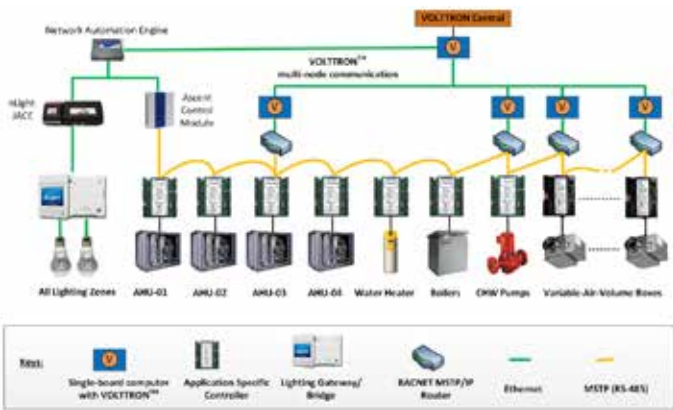
We added the capability of lighting controls to the building test bed. Power consumption of all LED lighting fixtures in SEB can be controlled to support experiments using buildings to provide ancillary services. The SEB building can now support interactions among heterogeneous components and systems (e.g., lighting and HVAC). The infrastructure was built to demonstrate the capability of interfacing between VOLTRON™ and physical devices at two different levels: the supervisory control level for the whole building and the local control level at each device.

We have managed to gradually build the trust of PNNL’s Facilities and Operations team. A number of documents, such as the rules of engagement and experimental plan templates, were developed to govern future experiments using laboratory facilities.

A preliminary version of advanced control theories has been applied on real buildings of the test bed. All major functionalities expected from the building test bed were successfully tested.



Connected Buildings



Building Test Bed Implementation in SEB

Aggregate Load Modeling and Control for Power Grid Regulation Services

Jacob Hansen

This project is developing aggregated load models that can be utilized for the design and analysis of control strategies that support provision of demand-based regulation services for the grid.

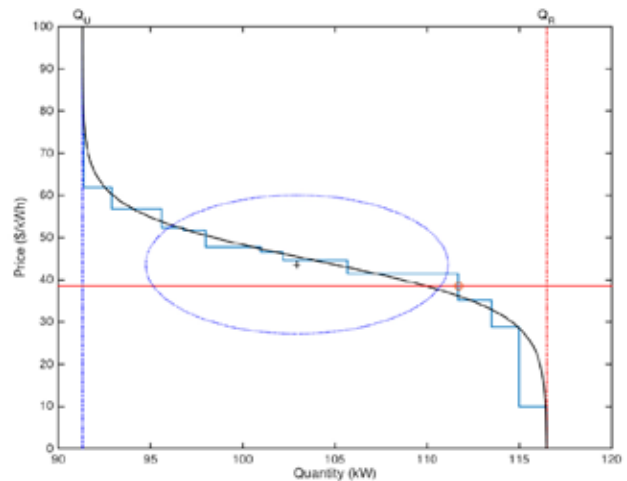
Modeling controllable load is the first step toward reliable integration of renewable resources in power grid operation. In this project, appropriately chosen and calibrated load models were used in the design of control systems that enable utilization of smart energy consuming load resources and renewable energy generation resources in bulk-power system operations. The aggregate load modeling problem has been a long-standing challenge to the design of scalable, fast-acting, demand-based control solutions that use electricity consuming systems to enhance and support both planning and the real-time operations of the bulk power system. This project provides aggregate demand models toward comprehensive use of loads in the larger problem of optimizing energy, power, and allocation of ramping resources in bulk power systems.

Prior literature on the topic of modeling electric loads for demand response captured the load variability power spectrum as a function of a set of independent load components. The PNNL project team modeled the aggregate load as a composition of underlying models that could be directly measured or indirectly disaggregated from existing data. The periodic electricity consumption components of controlled loads were captured using the mathematical formalisms of modern control theory.

In FY 2015, PNNL collaborated with the Electrical Engineering Department at University of Victoria to develop a model of aggregate load control at the utility scale, based on a new discrete-time zero-deadband residential thermostat. The outcome was a linear time-invariant load control model that enables design of demand dispatch strategies. The model was leveraged with a variety of standard control design techniques for the development of load control systems. The new aggregate load control model is simpler than previously developed models and enables utility engineers to implement load utilization in power grid operation in a manner similar to the controlled use of generation for operations.

In collaboration with the Economics Department at University of Victoria, PNNL investigated the short-term elasticity

of demand response using data collected from the Pacific Northwest GridWise Testbed's Olympic Peninsula study and the Northeast Columbus GridSmart Demonstration projects. The team applied a random utility model and found that demand response bids in transactive systems could be modeled using a logistic demand curve. Using this function, we have devised a simple formula to estimate the short-term demand elasticity of loads participating in a transactive control system. In collaboration with the Mechanical Engineering Department at University of Victoria, PNNL developed an interconnection-scale model of transactive control systems to enable the design and validation of market-based demand response control systems that use aggregate load models to deliver power grid services.



Random utility model of short-term demand response applied to a single transactive market clearing event in Columbus, OH. Blue stair-step line = true demand bid curve; black line = logistic demand curve fit; dotted ellipse = average and one sigma range for the expected price and quantity; solid red line = clearing price, with the blue circle showing clearing quantity and marginal demand units; dot-dashed blue and red lines = demand response lower and upper limits, respectively.

In FY 2016, the project team finalized the implementation of an interconnection-scale model of demand response control in the GridLAB-D™ software for aggregate load control systems and used the model to demonstrate a method for economic dispatch of grid-friendly appliance resources from an aggregation of controllable building loads. A retail, market-based control strategy was developed to engage these grid-friendly appliance resources. This could form the basis for planning and operation of load-based primary, secondary, and tertiary regulation services required by grid operators.

Analysis of UrbanSim and SoundCast for Parallelization

Kevin A. Glass

The goal of this project is to improve the performance of open source simulation packages used by the Puget Sound Regional Council (PSRC) to understand and reduce specific carbon emission sources within their sphere of influence.

Metropolitan areas are a major source of greenhouse gases, so finding sustainable ways to reduce the carbon footprint in these areas will help meet the U.S. DOE carbon emissions goals.

The PSRC uses UrbanSim and SoundCast software packages for their simulation, and the compute-intensive part of their workflow is an open source package called Daysim. This currently takes 6 hours to run on their existing hardware, making their simulation cycle for understanding the effects of policy modification on carbon production too slow. We hope to improve the performance of the Daysim workflow by a factor of four, without sacrificing its accuracy.

We ported the Daysim code to a local PNNL machine and acquired non-disclosure-agreement data from PSRC to run a real-size problem for benchmarking, and then we ran the code as configured and used by PSRC. The runtime for this benchmark was about 5 hours. By modifying the run configuration parameters, we were able to reduce that runtime to 3.5 hours. We did some coarse-grain profiling and identified areas of the code to focus our future code optimization efforts on. We also have identified some areas for improving scaling when running on larger hardware configurations.

While we have made some progress in reducing the runtime for this step of the workflow, we have more to do to reach our goal. We will continue to narrow the focus of code to be modified by further profiling, and we hope to dramatically further improve execution time by improving data access patterns and optimizing execution of the software.

Climate-Informed Decision Support System for Identifying Sustainable Hydropower Development Opportunities

Kyle Larson

Climate change may affect future sustainability of water resources, including our nation's largest source of renewable energy: hydropower. While the science of assessing possible hydro-climatological futures continues to improve, so must methods and tools that allow decision-makers and stakeholders to assess uncertainty at multiple scales. This project focused on using geo-information technology to fuse hydro-climatological science and several other factors affecting sustainability and project siting to better support complex decision-making processes encountered in hydropower and water resource planning.

Future sustainability of hydropower is intrinsically linked to changes in the hydrologic cycle and the cascade of environmental and societal effects that stem from often competing uses of water. Understanding the role climate change may play in this complex equation is becoming increasingly important, but reconciling the state-of-the-science with decision support needs of regulatory agencies, water managers, and affected stakeholders remains a challenge.

This project developed a geoinformatic approach and demonstration web application for integrating climate-informed analyses of future water availability and water-dependent generation stress with national hydropower resource assessments and other relevant planning information. A key goal was to illustrate how geospatial attribution can be used to transform and unify independent datasets or analyses for a common purpose and, thereby, help bridge information gaps between science and resource management communities. The project also emphasizes the importance of distilling scientific models and analyses into intuitive metrics for non-technical audiences.

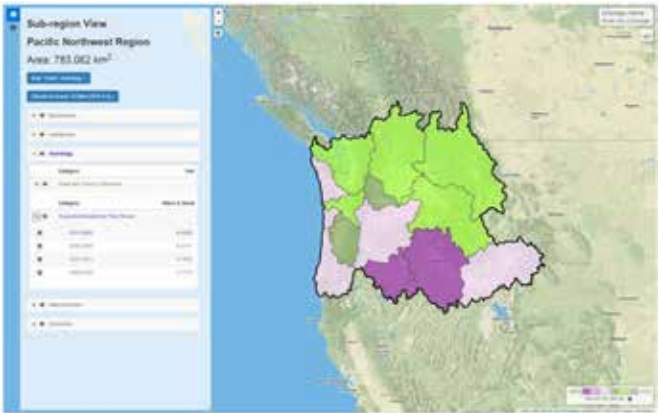
DOE's national-scale assessments of undeveloped hydropower potential were used as a basis for illustrating how

future hydropower potential could be examined in multiple contexts affecting sustainability and feasibility, including future water availability and energy economics under future climate scenarios, environmental concerns, and estimated interconnection cost. Components of PNNL's Platform for Regional Integrated Modeling and Analysis framework were leveraged to simulate future water availability and water-related stress on the western U.S. electricity grid under two greenhouse gas concentration scenarios. Three example metrics were derived from these analyses to illustrate their use in decision support, including timing of change in annual and seasonal hydrographs for river outlets of sub-regional drainages, deviation of streamflow from historic conditions at sub-regional to watershed drainage scales, and percent change in locational marginal price of electricity.

Non-climate-related information relevant to hydropower siting was also included in this study, including screening-level estimates of interconnection cost and a suite of environmental concerns data. Our method to estimate interconnection cost was consistent with an industry method for transmission infrastructure, but was modified to better represent connection of distributed small hydropower (less than 30 MW capacity) to local distribution systems. The method was implemented for the entire geographic domain of the western U.S. electricity grid.

Geospatial transformation and relation techniques were utilized to disaggregate, quantify, and visualize sustainability and feasibility factors by a hydrologic drainage framework that is commonly used in water resource planning. A demonstration web application was built to showcase how our approach could be implemented in tool form. The application follows the hierarchically nested design of the hydrologic framework, allowing users the ability to drill in/out of drainages while maintaining proper hydrologic context of the issues being examined. Drainages are dynamically color-shaded to quickly visualize relative differences among those displayed. Underlying data also can be accessed and downloaded from within the application.

This project demonstrates how geoinformatic approaches and technology can be used to un-silo disparate data and analyses to enhance integrated approaches to assessing sustainability and feasibility of future hydropower. These capabilities are especially useful for integrating climate-related impacts into decision-making realms, from local to national scales, and helping bridge the gap between climate science and public understanding. Future work will focus on building new collaborations to expand and apply our approach and demonstration web application for specific use cases.



View of the web application illustrating relative differences in projected hydrologic deviation (measured in number of standard deviations above/below the historic period of 1985–2010) during the 2015–2045 time period for the selected climate model and scenario (CCSM4: RCP 4.5). Drainages are shaded based on the relative position of their individual values within the range of values for all drainages shown. In this example, the table indicates the mean deviation for all drainages shown is -0.4250 (below normal), with drainages in the northern portion having lower deviation (more negative) than drainages in the south.

Complex Systems Control Testbed

Rick M. Pratt

This project provides an experimental and testing infrastructure in which distributed control methodologies and tools can be validated and verified.

Physical testbed demonstrations of advanced theoretical control system methodologies can validate the theoretical development and evaluation processes and provide confidence in extending the theoretical methods to larger scales, heterogeneous systems, and devices with non-ideal behavior. Verified theoretical control approaches are critical to effective integration of intermittent and decentralized renewable resource electrical generation and controllable loads (e.g., hot water heaters, energy storage, electric mobility) to utility portfolios. Distributed control has the potential to cost-effectively integrate decentralized resources by leveraging the extensive, network-based communications infrastructure. The network-based communications and control system uses software modules called agents to enable the testbed to be flexible, reconfigurable, and provide scalable operational capability.

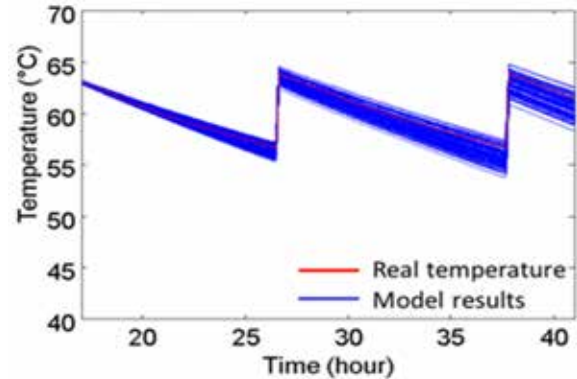
Agents were developed on the PNNL's VOLTTRON™ platform to enable drivers to interface with existing campus and regional load and generation resource controllers at Lab Homes, electric vehicle charging stations, and energy storage systems. Agents were also developed to obtain Environmental Molecular Sciences Laboratory photovoltaic array, Bonneville Power Administration load, wind and hydro generation, California Independent System Operator market data, and weather data. Physical device populations were then scaled by implementing emulated devices into the system.

In FY 2016, it was recognized that a new VOLTTRON™ driver class was needed for a low-cost and scalable hardware and software control platform.

This significant effort included upgrading the entire testbed software infrastructure to the current VOLTTRON™ version. These upgrades enabled collected data to be stored using the VOLTTRON™ historian for post-experiment processing and evaluation, while maintaining the capabilities needed in the event of a communication fault or transitioning between experimental mode and normal device operation.

The limited scalability of physical testbeds was addressed by developing emulated devices with characteristics that

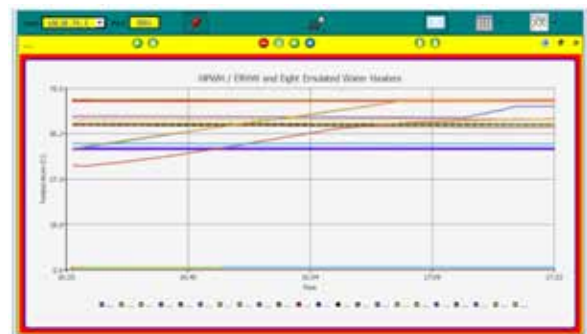
approximate physical devices. For example, the water heater has four states: heater ON, heater OFF, water valve ON, and water valve OFF.



The physical water heater thermal performance can be measured in each state and be used to define a large heterogeneous population of water heaters with characteristics based on physical devices.

This approach was tested and demonstrated to provide 64 water heaters based on the measured characteristics of two physical water heaters. Additional testing is needed to determine the upper limit of emulated water heaters.

The theoretical development team was provided a new capability that allowed MATLAB-based agents to test the control system performance without creating the python agents needed by VOLTTRON™.



Graphic display of selected control and system data.

Network-based, distributed control systems utilize digital communications between devices that can be difficult to troubleshoot. A capability was added to graphically display selected control and system data, as needed, to troubleshoot device/algorithm operation. This display was enabled to operate on the common hardware platform.

Construction and Testing of a Mesocosm Seawater Wave Tank for Conducting Environmental Oil and Gas R&D under Simulated Arctic Conditions

Michael Huesemann

A unique, temperature-controlled mesocosm wave tank was constructed and tested for use in future oil spill mitigation research, in order to better understand the fate and effects of petroleum hydrocarbons with and without dispersants following a potential oil spill under Arctic conditions such as seawater with floating ice under breaking and non-breaking wave conditions.

Due to scaling problems associated with interdependent physical and biological processes, it is impossible to obtain meaningful data on the simultaneous dispersion, weathering, volatilization, and biodegradation of crude oil with and without dispersant under different wave conditions in small-scale bench-top experiments. As a result, only a few reliable published studies on crude oil dispersion have been conducted in indoor or outdoor wave tanks. A particular challenge is the performance of oil dispersion experiments in the presence of floating ice, and the only published study on this topic was carried out in a very large outdoor wave tank at the Ohmsett Facility in New Jersey in the middle of winter (to induce ice formation). However, there are currently no temperature-controlled, small-scale, seawater wave tanks in the United States that enable the performance of crude oil dispersion experiments under simulated Arctic Ocean conditions. To enable future research on crude oil spill mitigation with dispersants under Arctic conditions, a temperature-controlled, mesoscale seawater wave tank was constructed, installed, and tested at the PNNL Marine Sciences Laboratory in Sequim, WA.

A mesoscale circular wave tank, as shown below, was constructed out of 0.25-inch, highly corrosion resistant stainless 316L steel. The tank has the following dimensions: 216" (548 cm) length x 79" (200 cm) width x 60" (152 cm) height. The maximum water volume is approximately 1,500 gal (5,678 L).

Waves with selected frequency (0–60 Hz) and stroke (0–8 inches) are generated at one end of the circular tank with a computer controlled wave-board. The end opposite of the wave generator contains a beach module with two large observational windows to study crude oil-sediment-water interactions, with and without dispersant, under different wave conditions. The middle of the wave tank has two large, opposing windows that allow for optical analysis of dispersed oil via photography- or laser-based methods. The wave tank is also equipped with six sampling ports with waterproof septa, equally spaced 7.4" (18.8 cm) apart.

Clean, filtered seawater is supplied to the wave tank from Sequim Bay. The inside oval of the tank contains a large heat exchanger, which is connected via hydraulic lines to a heat pump, enabling the rapid cooling of the seawater in the wave tank to below freezing temperatures (-2°C) to induce ice formation and simulate Arctic Ocean conditions. The seawater can also be heated to simulate ocean conditions in temperate or tropical/subtropical climates, such as in the Gulf of Mexico where oil exploration activities take place.

In order to ensure that operations are safe from an environmental and health perspective, the tank is covered with semi-transparent, twinwall polycarbonate sheets to avoid leakage of hydrocarbon vapors into the surrounding laboratory space. During future oil dispersion experiments, emanating hydrocarbon vapors will be removed from the tank headspace through a vent in the cover by an activated-carbon-operated fume extractor. Following completion of an oil dispersion experiment, the crude oil contaminated seawater will be pumped through an activated carbon drum to remove all hydrocarbons prior to discharge into the adjacent wastewater treatment system. To avoid accidental environmental contamination by oil-laden seawater leakage from the wave tank, the entire experimental area will be surrounded by oil booms. PNNL health and environmental safety permits are currently under review.

This unique research platform will enhance PNNL's fundamental capability in conducting oil fate and effects research—including biodegradation, volatilization,

weathering, and migration—under simulated Arctic Ocean conditions.



(a)



(b)

Side (a) and top (b) view of wave tank in operation.

Control Framework for Large-Scale Complex Systems

He Hao

This project is developing a framework for decomposing and engineering distributed control in next-generation power transmission and distribution systems.

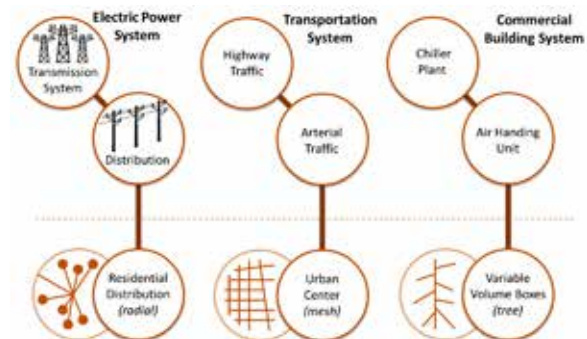
With rapid technological advancement in communication, computing, and control, dynamical systems become increasingly complex. The complexity of dynamical systems (e.g., large-scale, dynamic coupling, uncertainty) presents challenges to the current engineering paradigm.

The goal of this project is to investigate and develop analysis and synthesis methods to systematically architect and design control hierarchies for large-scale complex systems. The outcome of this investigation is a set of requirements that will inform control architecture definition and algorithm selection for a given system size and physical shape, as well as communication and computation architectures. As an example, the performance tradeoff between solution convergence rate and system size will determine the number of decision layers in the system and/or the number of agents in each decision layer.

In FY 2016, we investigated control algorithm scalability to characterize tradeoffs between system scale and information exchange rates between control nodes. The project team conducted a preliminary literature review on the definitions of

scalability, contexts where scalability is being studied, and the metrics for scalability evaluation. We then investigated layered (dual) decomposition approaches and their application to architecting control decision layers for a distributed energy resource coordination problem, with the aim of improving the convergence rate of a distributed coordination algorithm. Numerical results validate the advantages of a distributed approach to coordination.

In FY 2017, we will investigate theoretical approaches to system decomposition. We will address the research in the following three stages: 1) representing next-generation grid models as stochastic hybrid systems, 2) decomposing stochastic hybrid systems into subsystems based on fundamental representations of input-output properties, and 3) translating this decomposition to a coordination and control framework.



Side (a) and top (b) view of wave tank in operation.

Decision Theory for Incentive-Compatible Mechanism Design

Abhishek Somani

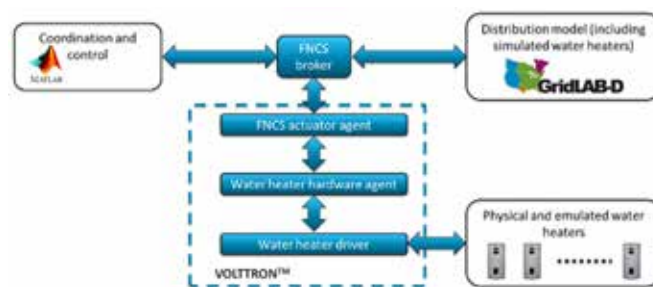
Distributed control of a complex system may be viewed as a problem of incentive-compatible mechanism design, so that the emergent outcomes, due to actions taken by agents acting out of self-interest, are consistent with the overarching control objective. Mechanism design theory—a field of micro-economics and game theory—provides a framework for solving such problems.

In FY 2014, we used agent-learning methods to derive smart HVAC agents' bidding behavior double auction retail markets, like those used to manage feeder constraints in the recently concluded American Electric Power gridSMART® smart grid demonstration project. The bidding strategy in double auction markets is derived without assuming a model for the agents' economic behaviors using a reinforcement learning algorithm. The results from comparison of the model-based and model-free approaches demonstrated that the model-free bidding strategy gave similar performance to the model-based bidding strategy.

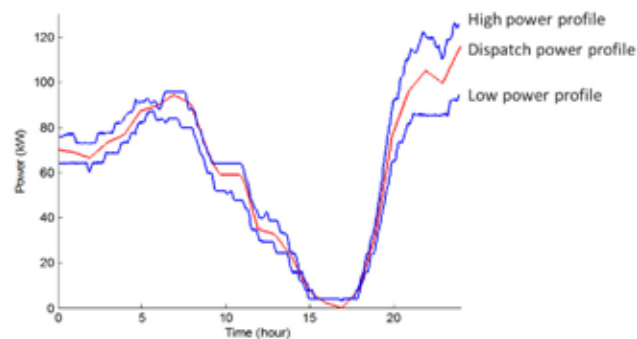
In FY 2015 we proposed and formulated a cooperative power allocation strategy for a population of commercial buildings within a campus using the theory of Nash bargaining. Numerical simulations showed that our allocation strategy was effective in achieving a power allocation for buildings while respecting their own preference and flexibility. The model was also extended to include multi-period negotiation to enable power allocation in a smart residential community. The concept was tested in a simulation setting only and was later demonstrated using physical water heaters in FY 2016.

Also in FY 2016, we collaborated with other principal investigators to devise an integrated demonstration of the capabilities developed under the various projects. The demonstration team included over 10 staff members with diverse backgrounds spanning 5 different projects. The integrated demonstration focused on distributed coordination of electric water heaters for the purpose of load management.

The testing of the control system was performed in a phased manner, with increasingly higher-fidelity test environments, including: 1) simulation of water heaters and the control system in Matlab only, 2) simulation of water heaters in GridLab-D™ and the control system in Matlab, and 3) hardware-in-the-loop testing. The simulated water heater models were calibrated based on real data from the physical water heaters, and the variance in their performance relative to physical water heaters was captured using an uncertainty quantification method developed in another project.



The testing and validation of the control system was done in a test environment that included water heater control signals coming from the solution of a Nash-bargaining problem in Matlab, with water heaters and the distribution system simulated in GridLab-D™. The test environment also consisted of two physical electric water heaters with local controls. The information exchange with the physical water heaters was facilitated by the use of a VOLTTRON™ agent, while a framework for network co-simulators broker enabled the information exchange between Matlab and GridLab-D™.



The results from simulation of 1,000 water heaters, along with the responses from physical water heaters.

Demonstration of Advanced Bi-Additive Vanadium Sulfate Electrolyte

Wei Wang

PNNL patented new, advanced additive technology that significantly improves energy density and temperature stability of the vanadium redox flow battery electrolyte. This project seeks to successfully demonstrate this technology, which will provide the energy storage industry with a revolutionary candidate for large-scale energy storage.

The traditional vanadium sulfate acid electrolyte precipitates whenever the temperature is over 35°C or below 10°C. In the new additive technology, we identify and introduce unique foreign ions to change the vanadium ions solvation chemistry and, therefore, to frustrate its precipitation mechanism.

The new electrolyte is demonstrated at the stack level with a 30% increase of energy density and 80% increase of temperature stability window compared with traditional vanadium

sulfate electrolyte. The demo unit with its test results serves as pivotal proof of technology for the on-going commercialization licensee and research and development contract negotiations.

Eighteen liters of the vanadium sulfuric acid electrolyte were made with the addition of the additives, and the performance of the additive electrolyte was tested on the 3-cell stack designed in 2015 under the current densities of 160, 240, and 320mA/cm² under the nominal flow rate of 400cc/m/cell. At each current density, the tests were carried out at 30, 40, and 50°C.

The additive electrolyte demonstrated comparable performance with the traditional electrolyte. More importantly, no precipitation was observed, even when the operating temperature was elevated to 50°C. The performance data thus proves the superior temperature stability of the additive electrolyte compared with the traditional sulfuric acid electrolyte.

Performance of the additive electrolyte.

3 Cell 1.5M bi-additive 2 layer ELAT Nafion 212	Columbic	Voltage	Energy	Discharge	Pressure (psi)	Pressure (psi)	Flow Rate/cell	Discharge
Current Density (A/cm ²)	Efficiency (%)	Efficiency (%)	Efficiency (%)	Capacity (Ah)	negative	positive	(cc/min)	Average Temp (°C)
160	97.0%	84.9%	82.4%	56.8	9.0	6.8	400	27.4
160	97.3%	84.5%	82.2%	57.5	8.5	5.9	400	38.6
160	96.6%	86.1%	83.2%	59.5	7.2	5.2	400	50.0
240	97.2%	76.8%	74.7%	43.1	9.8	6.9	400	26.1
240	97.6%	78.8%	76.9%	48.8	8.1	5.8	400	37.1
240	97.5%	79.7%	77.7%	49.4	7.6	5.1	400	46.8
320	97.8%	70.3%	68.8%	36.7	10.8	7.0	400	24.6
320	98.2%	72.3%	71.0%	38.0	9.1	5.9	400	35.6
320	96.6%	74.7%	72.2%	37.7	7.1	5.1	400	46.5

Development of Lithium-Ion Battery Electrolytes for Low-Temperature Applications

Wu Xu

Commercialization of electric vehicles (EVs) demands that the battery systems have high safety, low cost, high energy and power density, long cycle/calendar life, and a wide working temperature range over the state-of-the-art (SOA) lithium (Li)-ion batteries. Utilization of the optimized electrolytes developed in this project will enable the SOA Li-ion batteries to meet the above requirements for large-scale applications of plug-in hybrid EVs and pure EVs.

In FY 2015, we developed an electrolyte technology by using CsPF₆ as an electrolyte additive to protect graphite anode in propylene-carbonate- (PC)-containing electrolytes under the Advanced Battery Materials Research Program within the DOE Vehicle Technologies Office. The electrolytes with CsPF₆ additive can form an ultrathin, uniform, and stable solid electrolyte interphase (SEI) layer on the graphite anode surface so that the following performance indicators can be achieved: increased rate capability, enhanced cycle life at room temperature and elevated temperatures, and greatly improved low-temperature discharge capacity. However, the above results were obtained in small capacity (about 4 mAh) coin cells, and the electrolyte formulations have not been optimized.

In this project, the electrolyte formulations will first be optimized in coin cells and the optimized electrolyte formulations will then be evaluated and compared with the conventional Li-ion electrolyte in 4 mAh coin cells and 1,000 mAh pouch cells. The large-size pouch cell batteries should have the same or similar results as in small-size coin cells. The battery performance results from the large-size batteries are more reliable to the battery manufacturers.

Eight electrolytes were studied in coin cells of synthetic graphite anode and $\text{LiNi}_{0.80}\text{Co}_{0.15}\text{Al}_{0.05}\text{O}_2$ (NCA) cathode, and they are based on the formulation of 1.0 M LiPF₆ in EC-PC-EMC with 0.05 M CsPF₆ and with different EC and PC contents, where EC and EMC are ethylene carbonate and ethyl methyl carbonate, respectively. The test results show that the EC content in the electrolyte significantly affects the discharge per-

formance of graphite | NCA full cells at low temperatures, especially below -20°C. The increase in EC content in the EC-PC-EMC solvent mixture does not decrease much of the ionic conductivities of the electrolytes at low temperatures, but it does greatly reduce the discharge capacity of Li-ion cells, especially at -40°C. This is because more EC content in the electrolyte will result in a relatively higher solidification temperature of the electrolyte, a higher viscosity, and a lower ionic conductivity, thus a lower discharge capacity at low temperatures. However, these eight studied electrolytes show very similar cycling stability in graphite | NCA full cells at room temperature, as well as at elevated temperature (60°C), except for the electrolyte with 20 wt% PC and 10 wt% EC in the solvent mixture; this is probably due to the lower EC content leading to poor SEI layer on graphite anode surface.

The optimized electrolyte was then compared with a conventional Li-ion electrolyte in graphite | NCA coin cells. The test results demonstrate that the optimized electrolyte developed in this project has nearly the same long-term cycling performance as the conventional electrolyte at room temperature and elevated temperatures (45 and 60°C), but it does show much better low-temperature discharge performance than the control electrolyte, especially at -40°C.

When tested in 1,000 mAh pouch cells with natural graphite anode and $\text{LiNi}_{1/3}\text{Mn}_{1/3}\text{Co}_{1/3}\text{O}_2$ cathode, the optimized electrolyte developed in this project also shows similar results (i.e., much better low-temperature discharge performance at -40°C) and similar rate capability and cycling stability at room temperature to the conventional Li-ion electrolyte.

The above results demonstrate that the electrolyte technology developed in the project does greatly improve the low-temperature performance of Li-ion batteries and maintain the battery performances at room temperature and elevated temperatures. The optimized electrolyte shows promise for replacing the SOA Li-ion electrolyte in EV applications for a wide temperature range.

The next step is to approach the interested electrolyte manufacturers and battery manufacturers to license this electrolyte technology.

Distributed Control of Large-Scale Complex Systems

Jianming Lian

This project investigated distributed control to support future developments in the areas of electricity infrastructure and building technology. The resulting theory was applied to establish the theoretical foundation for a newly proposed concept of transactive control in the domains of grid and building, and to enable the distributed, optimal control of smart grid assets for demand response.

Modern systems, such as electric power systems and building systems, are becoming more interconnected and complex. In contrast to centralized control, distributed control is emerging as a promising paradigm for the control of large-scale interconnected systems.

Transactive coordination and control is emerging as a new approach for integrating various distributed energy resources (DERs) into the power distribution system, where individual resources are automated and engaged through market interaction. In transactive energy systems, the economic signals, such as prices or incentives, are used to engage the self-interest of the assets and to coordinate their operation with the power grids. One example of transactive energy systems is the one designed in the American Electric Power (AEP) gridSMART® demonstration project. In this project, we investigated the underlying characteristics of transactive approaches and established a general theoretical framework for the analysis and design of existing and future transactive energy systems.

In any transactive energy system, there are two levels of decision-making. At the device level, each type of controllable resource (e.g., water heaters, residential air-conditioning units) was modeled as an automated agent that can communicate with other agents in the system and decide on their own actions. The agents are self-interested with private local information and individual local objectives. The control decision process at the resource level was modeled as an optimization problem for surplus maximization. The coordinator pursues global, system-level objectives when coordinating various DERs. For example, the coordinator objective could be to minimize the overall operation cost, while engaging DERs to balance supply and demand and provide ancillary services. The objective function is then defined as the difference between total utility of all the DERs, which represents the value of power consumption, and total cost of energy. In welfare economics, a branch of economics that uses micro-economic theory to evaluate well-being at economy-wide

level, this type of system-level objective is referred to as social welfare. The system-level constraints could be constraints imposed on power balance, line flow, voltage magnitude, and system reserve. The local constraints of individual resources are capacity and ramping limits. The coordinator aims to solve this optimization problem in order to obtain the optimal energy allocation.

With the mathematical analysis, we identified two challenges in designing and deploying any transactive control approaches for power system applications. The first challenge is related to device modeling. We have to model not only device dynamics, but also economic preferences. The second challenge is associated with the computational issues. Although many concepts are very clear from the economic point of view, it may be very difficult to achieve them from the computational point of view. For example, it has been known as a non-deterministic polynomial-time hard problem to compute the Nash equilibrium of a game.

In FY 2014, we applied the proposed theoretical framework to analyze the transactive energy system in the AEP gridSMART® demonstration project. We explained the underlying principles of system operation and identified the issue of the original bidding strategy. We found that the original bidding strategy cannot guarantee the global optimality of the system outcome and the feeder capacity limit can still be violated during system operation. Hence, we proposed a new bidding strategy to address this issue.

In FY 2015, we revisited the bidding strategy we had previously proposed. Because the implementation of this bidding strategy requires the knowledge of some device parameters that are not measurements, we proposed a learning-based bidding strategy whose implementation will only be based on the available measurements. In addition, we also proposed a new distributed-control strategy for multi-zone commercial buildings. This new control strategy enables commercial buildings to be a great flexible resource for providing various grid services through building-to-grid integration. We first developed a distributed method of characterizing the flexibility and then applied a distributed method to allocate the air flow among different zones.

In FY 2016, we collaborated with other projects to demonstrate the proposed distributed-control strategy on a building testbed facility. We first performed a proof-of-concept testing of the proposed strategies. This proof-of-concept testing was performed by utilizing building models of increased fidelity developed by the testbeds team and simulation and co-simulation tools developed by the tools team. First we tested the proposed transactive strategy using MATLAB and leveraged the EnergyPlus model of a campus building at PNNL.

Electrolytes Enabling Low-Temperature Battery Operation

Wesley A. Henderson

The objective of this project is to develop electrolyte formulations based upon classes of previously unutilized ketone and nitrile solvents to optimize low temperature Li-ion battery cell performance with comparable or superior room-temperature performance relative to commercial electrolytes.

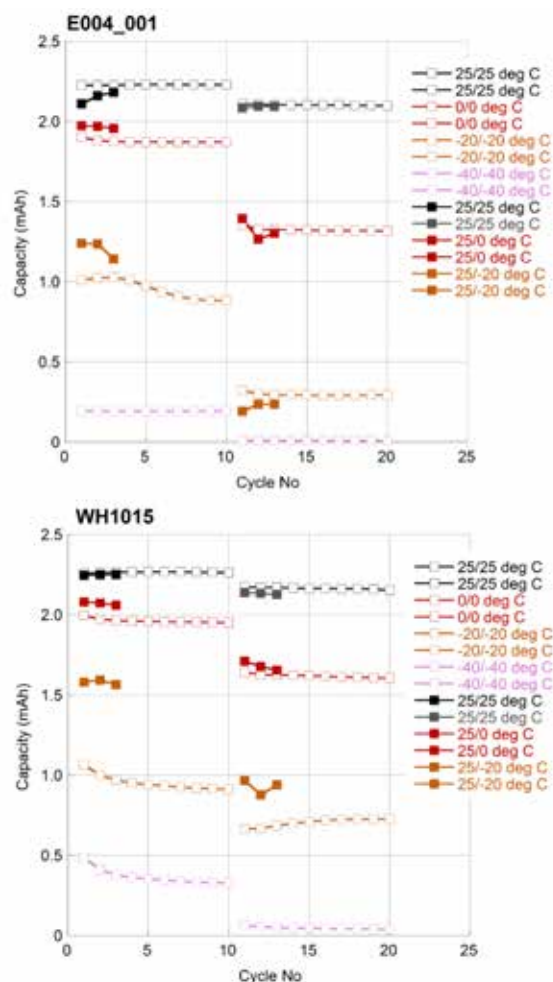
State-of-the-art Li-ion batteries contain electrolytes based on LiPF_6 and a mixture of carbonate solvents (i.e., ethylene carbonate [EC] and an acyclic solvent such as ethyl methyl carbonate [EMC]). Although providing excellent cycling behavior at room temperature, such electrolytes result in poor performance at low temperatures, with a significant decrease in capacity/power, in large part due to the increasingly sluggish Li^+ cation transport within the electrolyte as the temperature is decreased. Batteries with electrolytes also containing ester solvents (e.g., mixing solvents such as ethyl butyrate or ethyl acetate with the carbonate solvents) have improved low-temperature performance and may operate down to -40°C , but generally only at very low discharge rates (having been charged at room temperature).

Significantly improved low-temperature battery operation is particularly relevant for increasing the driving range of electric vehicles and for 12V Start-Stop applications. Li-ion batteries, which provide a similar energy and power density in the -30°C to 25°C temperature range, may dramatically revolutionize the cost (due to a reduction in battery pack oversizing) and public acceptability of electrified vehicles, as well as greatly facilitate the market accessibility of such batteries for other applications. Commercial battery producers are highly conservative with regard to altering the materials within batteries, but the poor low-temperature performance of Li-ion batteries remains a critical impediment for their widespread adoption for numerous uses, particularly for vehicle propulsion. A solution without detrimental effects on the room-temperature characteristics and lifetime of the batteries could be widely adopted by the industry.

Low-temperature electrolytes were developed for the Li-ion cell chemistry utilizing lithium titanate ($\text{Li}_4\text{Ti}_5\text{O}_{12}$ or LTO) as the anode active material and lithium iron phosphate (LiFePO_4 or LFP) as the cathode active material. This anode does not require the formation of a solid electrolyte interface from the electrolyte components, as is the case for the graph-

ite anode used in standard Li-ion batteries, which adds substantially to the cell's impedance. The operating voltage of the LFP cathode also enables a wider selection of solvents and salts to be employed than is the case for other cathode active materials.

Noncarbonate electrolyte solvents were selected with the goal of eliminating excess electron lone-pairs from the electrolyte components, thereby reducing the high-energy barriers to solvent and ion rotational/translational motion at low temperature. The new electrolyte formulation [WH1015] showed an almost 2X improvement in capacity retention when charging and discharging at -20°C .



Comparison of LFP/LTO coin cells with 1M LiPF_6 -EC/EMC electrolyte [E004_001] and modified electrolyte formulation [WH1015]. The open symbols are for charging/discharging at the same temperature, while the filled symbols are for charging at 25°C and discharging at the indicated temperature. The charge rate was C/2. Cycles 1-10 used a C/2 discharge rate, while cycles 11-20 used a 2C discharge rate.

Experimental Management for Controls of Complex Systems Test Bed

Mark J. Rice

Through the creation of this project's test bed, we are reducing researcher experimental overhead and improving the overall experiment process so that additional time and energy can be focused on the research while simultaneously delivering a more concrete product.

In complex systems, development and deployment of large-scale physical and virtual experiments may encompass the description and configuration of up to millions of devices. Researchers often devote significant time and project resources to experimental setup. A core element of our project is a powerful, flexible test bed that enables researchers who have developed new control schemes to examine performance and behavior in an experimental environment to capture the macroscopic behavior of increasingly complex interacting systems with an adequate degree of fidelity.

The experiment management system (XMS) is designed to provide functions such as experimental design assistance and automation, instrumentation guidance, data collection and curation, model cataloging and sharing, and experiment execution. XMS will manage experiments performed in the test bed, and visual analytics capabilities and graphical interfaces will support the management and monitoring of the test bed setup, experiment orchestration, instrumentation, and data analysis. We are defining the requirements and specification needed to allow tools to be integrated into the test bed and experiments to be run to test and validate control theory and methods developed. The XMS will be built in a way that allows changes in modules (e.g., simulators) to be implemented without the need for updates to other modules.

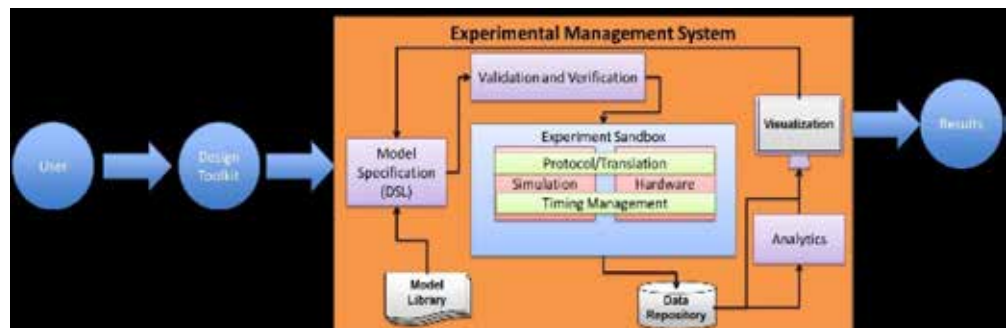
In FY 2015, we built the framework to deploy control system experiments using multiple simulators from a web portal. The project used OpenStack, a cloud computing management platform, to enable the deployment of multiple computers into an isolated network, a tenant, to perform experiments with. The platform allows XMS to replicate an experimental setup for other research, to start a new study with the same infrastructure.

Within XMS, we built virtual machines containing the

common simulation software, including GridLAB-D™ (an electrical distribution system simulator), ns-3 (a communication simulator), matpower (a wholesale market simulator), and framework for network co-simulators (FNCS), a PNNL-developed software that enables multiple independent simulators to work in a coordinated manner. Using these images of the simulator Arion, a related project that is developing a domain-specific language, we deployed a demonstration experiment to showcase the XMS and other tools for an annual review. The experimental setup included multiple GridLAB-D™ virtual machines, an ns-3 virtual machine, and the FNCS virtual machine. This demonstration showed that we can create the environment (tenant) in less than 10 minutes to perform the experiment.

In FY 2016, we integrated the agent-based distributed buildings controls and laboratory homes controls project to work with the XMS with the capabilities built by the projects at the communication/networking. We have continued to work on PNNL agent platform VOLTTRON™ virtual machines in both the XMS and central sever to interact with VOLTTRON™ installations used as part of the test bed, which allows for application-level support between various parts of the test bed. The central server deploys new agents that are different control schemes to VOLTTRON™ installations at laboratory homes and for building a control laboratory. In addition, we created a repository for the data collected during experiment and experiment descriptions hosted in the XMS and provided as a service to other projects that would like to mine collected data.

In FY 2017, we will continue to add features and work with control researchers to perform experiments using the XMS as portal into the test bed. New features to the XMS for FY 2017 will include the use of the XMS to deploy an experiment on a combined system that includes both physical hardware (hardware-in-loop) and integration of the Controls Modeling Language. Also in FY 2017, we will have beta testers of the XMS, which will help improve the usefulness of the environment.



The workflow of performing an experiment in the test bed.

Geochemical Sensors of Fracturing in the Subsurface

Eirik J. Krogstad

This project seeks to evaluate the potential of using rare earth elements (REEs) as chemical tracers to elucidate key subsurface processes associated with hydraulic fracturing, with the primary objective of developing a simple, low cost method for interrogating fracture surface area.

Global energy demands continue to increase at a dramatic pace, with an increase in total energy consumption of approximately 350% over the last 50 years. Worldwide, nearly all of this energy is supplied from the burning of fossil fuels. Much of this is being driven by technological advances in hydraulic fracturing or “fracking,” which have enabled unconventional resources of natural gas and oil trapped inside low-permeability geologic formations, such as tight sandstones and shales, to be economically produced. Today, nearly all natural gas wells drilled in the United States use this approach. Hydraulic fracturing is also a key step in developing enhanced geothermal reservoirs (EGS) and could serve as a mechanism to establish EGS as a carbon-free, sustainable energy alternative to fossil fuels.

The use of tracers to understand the development of hydraulically induced fractures has been a focus of research efforts for more than 4 decades. Due to the recent boon in shale gas development afforded by the ubiquitous deployment of horizontal drilling and multi-stage hydraulic fracturing, there has been a renewed focus on the use of tracers for fracture characterization during or after stimulation treatment. Further advances are coupling pulsed tracer injections with other technologies, including electrical resistivity tomography, microseismic monitoring, cross-polarized ground penetrating radar, and acoustic emission monitoring, to perform near real-time analyses of fracture. While these combinatory approaches hold promise, they require a level of field infrastructure and operation that will not be practical for widespread deployment.

Therefore, we hypothesize that REEs can be used as simple, yet novel chemical sensors to elucidate the surface area of fractures around the zone of drilling and stimulation. In order to test this hypothesis, we developed surface area cor-

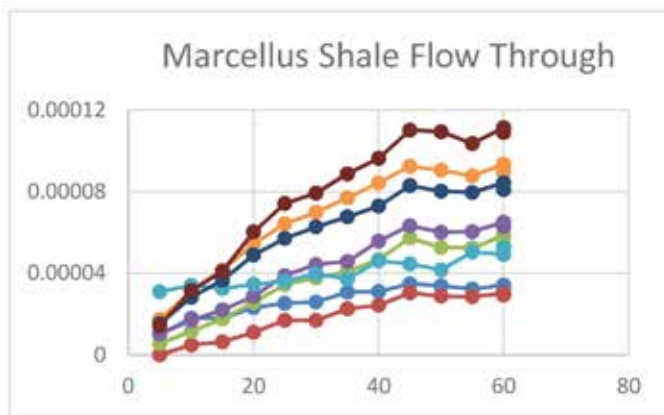
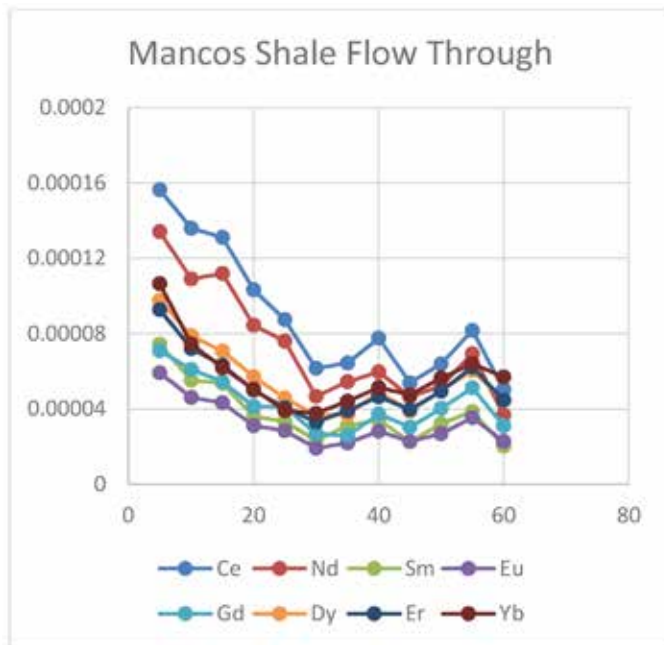
relations across multiple sample configurations (ranging from crushed rock to whole-rock cores). Four distinct lithologies were selected for use in this study. Three of the lithologies (i.e., basalt, andesite, and granite) are relevant rock types observed or anticipated at current or proposed enhanced geothermal system reservoirs; while the fourth lithology, shale, was selected due to its importance in unconventional resource development.

In our studies of these lithologies, we found that the REEs in the tracer solution were strongly adsorbed. Typically, the observed loss of tracer was greater than 99%, despite the relatively low pH in the tracer solutions that were used. This loss of the REE was due to interaction of the rock and the tracer to consume hydrogen ions: the pH change made the REE incompatible in the solution. Nonetheless, the high specificity of the isotope dilution mass spectrometric approach, combined with the high sensitivity of the multicollector mass spectrometers, allowed clear patterns to be observed among the various REEs.

Fine grain sizes led to higher degree of fractionation of the REE ratios, with the heavy REE (HREE) being more strongly adsorbed, probably due largely to their higher surface area. This effect is greater than the effect of time on fractionating the light REE (LREE) from the HREE (e.g., Nd/Er).

The fractionation of the LREE from the HREE (e.g., Nd/Er) was also a function of the rock type. For example, the Mancos and Marcellus shales, which were tested in a triaxial core holder, had notable differences in their fractionation of the REEs; in the Mancos shale, the REEs were progressively removed, with the LREE lost later than the HREE. In the Marcellus shale, by contrast, the REEs initially were strongly adsorbed, with the LREE being more compatible, and the HREEs were the earliest to increase in concentration. The differences on the behavior of the REEs in these two shales imply that mineralogical controls are a factor.

The loss of the REEs in these simple solutions would limit the usefulness of this type of tracer. However, more complex and realistic solutions, with alkali concentrations in the 10s to 100s of millimolar range, may be more capable of maintaining the low pH required to stabilize the REEs in solution.



Flow-through experiments in the triaxial core holder along a simple fracture in two representative shales. Concentration of the REE, in parts per million, is plotted on the y-axis; the number of pore volumes of fluid passing through the fracture is plotted on the x-axis.

Hardware Integration Platform for the CCSI Test Bed

Jereme N. Haack

This project is developing an integrated framework for simulation, hardware, and experimental control by adding functionality to the PNNL-developed VOLTTRON™ framework that is specific to supporting other related PNNL projects.

This project works closely with other related projects to integrate them all into a seamless platform. Projects to be integrated include those focused on simulation, experiment control and data storage, and a hardware test bed for realistic test deployments. This project will develop enhancements to the VOLTTRON™ platform needed to facilitate easy implementation and execution of experiments that include physical devices and systems, integration of a greater variety of physical devices/systems than possible today, providing an easy-to-use environment for building new control modules (agents), and integrating physical devices/systems with the simulation environment and the experiment management system.

VOLTTRON™ serves as integration point for the physical (and emulated) devices in the test bed. Together with the co-simulation environment, it will establish communication between the physical test bed environment and the simulation environment. It will also support communication for experiment configuration and data storage with the experiment management system. Test bed developers will use VOLTTRON™ to communicate with the hardware devices, build and deploy modules that implement their control methods for the hardware, and set up topologies required to implement their control methods for experiments.

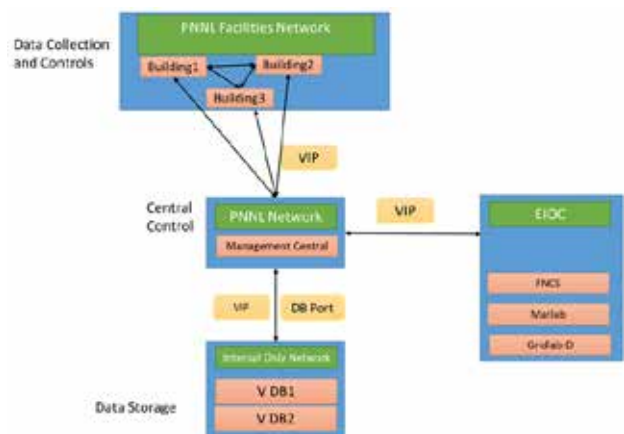
In 2016, the project integrated custom drivers developed for water heaters to bring them into VOLTTRON™-platform compliance. This allows them to be used more generally by the VOLTTRON™ community and increases their maintainability.

The MATLAB bridge developed this year is a major addition to the VOLTTRON™ platform that allows for agents to interact with simulated buildings and the testing of MATLAB-based control algorithms. Several projects used this capability to support annual review demonstrations. Additionally, this capability generated interest from the greater VOLTTRON™

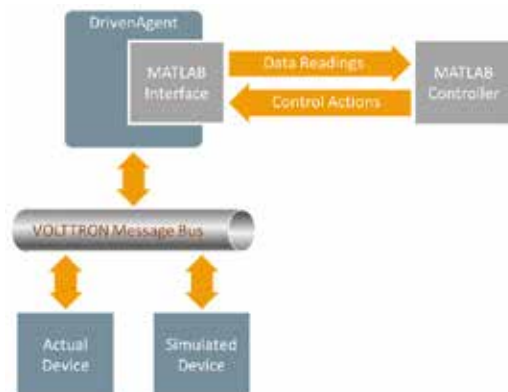
community and was quickly picked up and incorporated into at least two external research projects.

Integration with simulations was further enabled by building an integration point with a PNNL simulation project. This powerful combination allows large-scale simulations to interact with hardware being controlled by the VOLTTRON™ platform.

Cross-network data and control streams were also demonstrated in 2016. Data collected on the isolated facilities network was directed to a protected network via the VOLTTRON™ platform. Control actions inside the protected network were then sent out to the PNNL network to update agents being deployed in the test bed. This capability will be further developed in FY 2017 to provide flexible control and collection capabilities across multiple networks.



This project used VOLTTRON™ to cross network and firewall boundaries to integrate hardware, simulations, and control.



The MATLAB bridge allows applications to be tested against simulated buildings before actual deployment. It also allows researchers to develop and verify controls in MATLAB before paying the transition cost for the deployable Python version.

Impacts of Communication Network on Distributed Control

Di Wu

We are studying the effects of communication networks on distributed control where system components are not physically co-located and information needs to be exchanged through imperfect communication channels. This project analyzes and evaluates the impacts of communication network effects, such as time delay and packet drops on the performance of distributed control systems, and develops distributed control algorithms that can best accommodate various network effects.

In FY 2014, we reviewed literature of distributed control for building and grid applications and evaluated the impacts of communication network effects such as time delay, packet drops, and asynchronous effects on the performance and convergence of existing distributed control algorithms. We found that the existing distributed algorithms that designed without considering imperfect communication could fail the control system.

In FY 2015, we developed the following two distributed control algorithms that are robust for communication network effects: 1) A ratio consensus algorithm for distributed load shedding in power systems. In this method, each energy resource agent maintains two internal states and updates their values only using the information received from neighbors. 2) Minimum-time consensus-based distributed algorithms for load shedding and economic dispatch problems. The proposed algorithms are capable of solving these problems in a minimum number of time steps rather than asymptotically, as in most existing studies, and therefore, accelerate the convergence speed and alleviate the communication burden.

In FY 2016, we developed a hierarchical control method to incorporate thermostatically controlled loads (TCLs) into distributed energy resources (DERs) coordination that considers the dynamics of individual TCL devices. The proposed method is a combination of economic and control techniques, as it synthetically captures the underlying economics from demand response, as well as detailed dynamics in device-level control.

Algorithm 1 Multiple storage devices

```

1: Initialize  $k = 0$  and  $\lambda_i(0) = 0 \in \mathbb{R}^T$ .
2: repeat
3:   procedure LOCAL OPTIMIZATION
4:     for each node  $i = 1, \dots, N$  do
5:        $p_i(k) = \arg \min_{p_i \in \Omega_{N,i}} \sum_{t=1}^T C_i(p_{i,t}) - \lambda_i(k)' p_i$ 
6:     end for
7:     for each node  $i = N+1, \dots, N+M$  do
8:        $\{p_i^+(k), p_i^-(k)\} = \arg \min_{\{p_i^+, p_i^-\} \in \Omega_{M,i}} \lambda_i(k)' (p_i^- - p_i^+)$ 
9:        $p_i(k) = p_i^+(k) - p_i^-(k)$ 
10:    end for
11:  end procedure
12:  procedure CONSENSUS AND GRADIENT
13:    for each node  $i = 1, \dots, N+M$  do
14:       $\lambda_i(k+1) = \lambda_i(k) - \underbrace{\beta \sum_{v=1}^{N+M} \ell_{iv} \lambda_v(k)}_{\text{consensus part}} - \underbrace{\alpha_k (p_i(k) - D^i)}_{\text{gradient part}}$ 
15:    end for
16:  end procedure
17:   $k = k + 1$ 
18: until Error small enough
19: for each node  $i = 1, \dots, N$  do
20:    $p_i^{\text{sol}} = p_i(k-1)$ 
21: end for
  
```

This project used VOLTTRON™ to cross network and firewall boundaries to integrate hardware, simulations, and control.

Additionally, we extended DER coordination to include energy storage over multiple time periods subject to constraints at both system and device levels, such as power balance constraint, power/energy limits from each device, storage dynamics, and charging/discharging efficiencies. Fully distributed algorithms are proposed to dynamically and automatically coordinate distributed generators with multiple/single storages. With the proposed algorithms, the coordination agent at each DER only maintains a set of variables and updates them through information exchange with a few neighbors. We showed that the proposed algorithms, with properly chosen parameters, solve the DER coordination problem, as long as the underlying communication network is connected.

Moreover, we proposed a distributed algorithm based on the gradient push-sum method to solve an optimal coordination problem over communication networks, potentially with time-varying topologies and communication delays. We showed that the proposed algorithm is guaranteed to solve the problem if the time-varying directed communication network is uniformly, jointly, and strongly connected. Moreover,

the proposed algorithm is also able to handle arbitrarily large but bounded time-varying delays on communication links.

Finally, we have developed the framework and scripts for evaluating and comparing distributed coordination algorithms using test beds designed for other projects within the same initiative. The scripts are required for the interaction between different software, including GridLAB-D™, Matlab, and ns-3. With the scripts and the capability of Framework

for Network Co-Simulation (the chosen platform), we are able to simulate the power distribution system and controllers, while capturing the communication network effects. In this way, we better represent the interdependency of network effects and coordination algorithms and study the characteristics of networks effects such probability distribution of time delays and packet drop rates.

Integration and Demonstration of Scalable Power System Simulation for CCSI Test Bed

Mallikarjuna R. Vallem

This project has enabled researchers to assess the behavior of a bulk power system to several control components in simulation and real-time experimentation, demonstrating the value of Opal-RT to the Control of Complex Systems Initiative (CCSI) test bed.

The CCSI test bed envisions an integrated experimentation platform for distributed buildings and buildings to electrical system control components. A robust, commercial, real-time digital simulator that performs grid simulation, Opal-RT is integrated into the CCSI test bed to provide bulk-grid behavior to several controls components.

The focus of the project during FY 2016 has been on seamless operation of Opal-RT integrated with the CCSI test bed to enable non-real-time bulk-grid simulation. This work primarily involved the tasks described below.

Demonstrating the value of Opal-RT to the CCSI test bed.

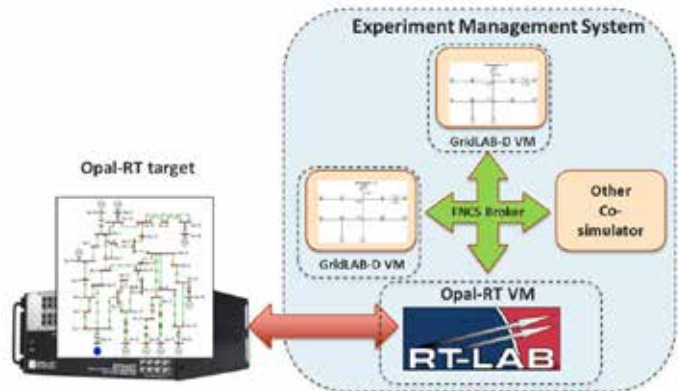
The project team has developed eight use cases that resulted from brainstorming sessions out of CCSI leadership. The use cases developed have been presented, and a path to implement them with Opal-RT integrated with CCSI test bed has been contemplated. This task demonstrated the unique capabilities of Opal-RT in the support of control experiments by testing the integration of Opal-RT with other CCSI test bed capabilities. To validate the mitigation developed, several specific features of a successful Opal-XMS [-extended memory specification] integration were identified and assessed.

Seamless integration with XMS. The existing Opal host is built on a Red Hat image with a custom Linux kernel. Being part of an older version of Red Hat Kernel, XEN extensions

cannot be installed on the Opal-RT virtual machine. The project will identify suitable mitigation to overcome this hurdle. When a grid simulation is performed on the Opal virtual machine, it will consume the majority of the computational resources. To ensure better performance, the simulation will be performed on a target (field-programmable gate array [FPGA] computer box that can perform Hardware-in-the-Loop [HIL] simulations). The FPGA computer is specialty hardware that cannot be virtualized. It is required to exist as an independent physical machine outside of the XMS environment. The team will work with XMS to ensure connectivity to Opal-RT Linux HIL simulator.

Use case demonstration and support for experimentation.

The objective of this task is to collaborate with relevant projects and support experimentation. Through preliminary interactions, it was determined which projects would benefit the most from collaboration. Through this effort, we will establish the need for control methodology and demonstrate the impact of implemented control.



Opal-RT integration for bulk-grid simulation.

Measurement and Verification in Controlled Complex Systems

Jim Follum

As revolutionary control architectures are implemented in complex systems, equally advanced methods of measurement and verification (M&V) are needed to support deployment. In this project, methods to confirm and quantify the impact of control at each level of the architecture are being developed to reduce the costs of verification, provide ongoing monitoring capabilities, and better link incentives with measured performance.

Effective control of large-scale infrastructures, such as energy, building, and transportation systems, can improve efficiency and reliability, but associated methods of ensuring that the desired improvement is realized are largely unexplored. Traditional M&V methods applied to energy- and water-saving projects require detailed, manual, and at times subjective analysis of datasets collected over long periods of time. This project will significantly advance the state of the art for M&V by applying methods from the fields of statistical decision theory and machine learning. These methods, which can meet the needs for scalability and noise tolerance inherent to this problem space, can be automated to allow ongoing performance monitoring and quantification at each level of the control architecture. By allowing system and device participation to be verified and quantified, the development of M&V tools will facilitate development and deployment of novel distributed control schemes at lower costs and shorter timescales.

Methods were developed in the following three areas: distributed M&V, advanced statistical data mining, and financial analysis. Each of these areas was explored while considering a distributed control method applied to a feeder serving homes with air conditioners regulated by smart thermostats. The developed methods have broader applicability that will be explored in the future.

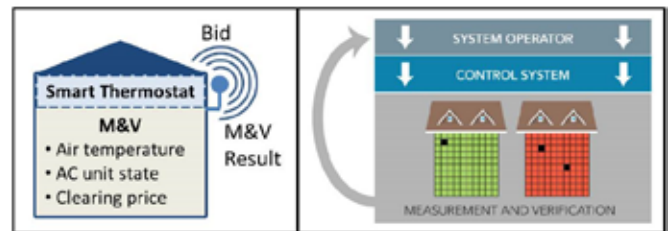
The distributed M&V focus area involves the dispersion of M&V tasks out to the device-level controllers. In the test case,

correlations between local measurements and system variables were examined within each home's smart thermostat. Based solely on values needed to implement the control, each thermostat was able to report back to the central controller whether or not the control was effective at the local level.

Within the area of advanced statistical data mining, work focused on the application of machine learning methods to categorize system behavior, despite highly complex relationships between system variables and significant noise levels. By examining training data from test cases, the transfer entropy between feeder power and the base price for electricity was used to verify the distributed controller's impact at the system level.

Finally, the area of financial analysis was explored by examining the controller's architecture. This type of analysis will aid system operators as they compare the costs, such as incentivizing participants, and benefits, such as deferred infrastructure investments, of implementing control.

In all three areas, results indicate that the developed methods can be implemented with minimal costs, while allowing ongoing monitoring at system and device levels. In the following year, the project will focus on testing with measured data, developing the capability to predict system behavior, and extension of the methods to a broader set of control architectures and complex systems. As the methods are refined, they will be integrated as a suite of tools for performing M&V with complex systems.



Shown here is conceptualization of the distributed M&V approach, along with results from simulations demonstrating the high accuracy rate in categorizing the participation of individual home air conditioners in a control architecture.

Multidimensional Membrane Theory to Predict Power System Oscillations

Laurie E. Miller

This research applies concepts of multidimensional oscillating membranes, similar to that used in the “theory of everything” (M-theory) in modern physics, to formulate a new model of electric power grid dynamics and oscillations, forming a topology-based, dynamic characterization. We extend this approach to find and predict singularities in the conventionally established power system stability regions.

Low-frequency oscillations can and do occur during interchange of power between neighboring regions or areas of operation. These inter-area oscillations remain a challenge for grid analysts, despite being a problem of long standing. With increasing power system size and complexity, designing for a robust and resilient system becomes a larger and more complex problem, as well. Inter-area oscillations add a difficult extra layer of complexity in power system planning, since they are a phenomenon that 1) can be spontaneous, 2) are not well-understood, and 3) lack predictive and actionable mitigation. Another important aspect of power system stability is the ability to support stable voltages in the system (voltage stability). The link between voltage and small signal stability is very important for connecting different types of stability into a single framework (like in the “theory of everything,” where all aspects of matter are explained in their interaction).

The core idea behind our proposed oscillating membrane theory of a power grid is that, in an electrical and electromechanical sense, the grid has a different topology compared to its two-dimensional layout on the earth’s surface. By allowing geographically distant points to be brought close by high-voltage transmission lines, the resulting image of the grid in the electrical sense can be represented as a multidimensional membrane, which oscillates over time. Our hypothesis confirmed in this research is that this membrane has certain natural frequencies while oscillating in a multidimensional space. The natural frequencies depend on the system size and topology. The actually observed free oscillations are



A conceptual view of a multidimensional electromechanical power system model.

excited by small or large perturbations of system parameters and can appear or disappear with changes in system configuration.

We have extended the idea to trace the connection between oscillations and voltage stability and pursue evidence of singularities (unstable “holes”) within the voltage stability region of power systems; this is a phenomenon not yet known in the published literature for real-life power system models. Power grid stability regions are fundamental to both the real-time operation of power grids and to the long-term design and planning of power grid systems and system enhancements. Conventionally, the outer boundary of the presumed region of stable operation is computed, and as long as the operation of the power grid stays within that region during operation, it would be assumed to be in a stable condition. The newly discovered inside-region “holes” change this traditional perception: the “holes” can cause system instabilities of a new kind, previously unknown to system analysts and engineers. This indicates the necessity of investigation of thin structures of power stability regions.

In this project, we developed a new approach to power system stability analysis, pathing a way for new engineering techniques and tools; demonstrated that power system oscillations can be a function of system size and topology; and connected oscillations with voltage stability, pathing a way to a single power system stability model.

Optimal Management of Building Loads and Energy Storage for Grid and End-User Services

He Hao

In this project, we aim to develop a unified modeling and coordination framework for flexible building loads and energy storage to optimally provide power grid and end user services, which are essential for renewable integration and energy efficiency.

Optimal coordination and management of building loads and energy storage are extremely important in order to fully exploit their flexibility and harvest their benefits to the grid. In the power system, power balance between supply and demand must be maintained at a second-to-second basis. However, deep penetration of variable generations into the supply side makes maintaining the power balance very challenging.

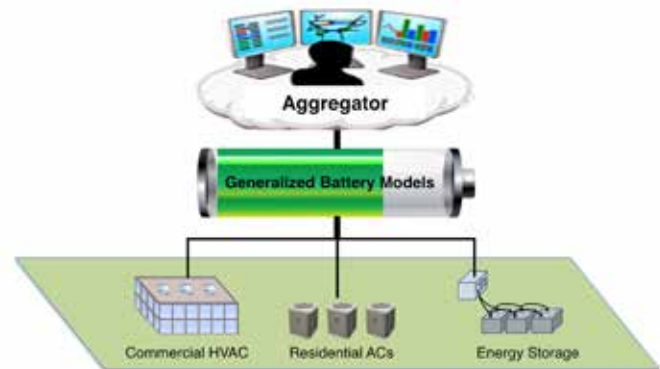
Renewable energy, such as wind or solar, has a “free fuel” component, but at the cost of variability. Currently, the uncertainty of renewables is handled through supply side reserves. This approach has many drawbacks, since it decreases generation efficiency, reduces net-carbon benefit from renewables, and is economically untenable.

Demand response and energy storage present a great potential to absorb the uncertainty of these variable generations. Demand-side control presents a novel way to manage the power balance; among various demand-side resources, buildings in which we live and work have significant impacts on energy consumption. More specifically, buildings consume about 40% of energy and 75% of electricity in the United States. Their massive power consumption and enormous thermal capacity present a great potential for providing various power grid and end user services. Moreover, recent developments and advances in energy storage system technologies provide another viable solution for providing flexibility to both the power grid and the end users.

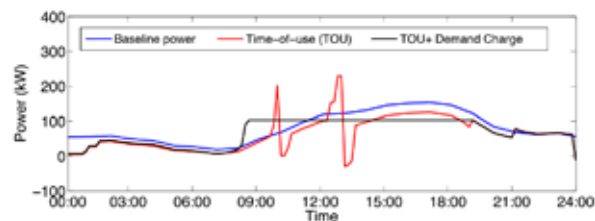
However, most of the existing studies are limited to modeling, aggregation, and control of a single type of resources and ignore the coordination and cooperation between multiple types of smart grid assets. In this project, we first propose a unified modeling method to capture the flexibility from various resources considering their physical characteristics, environmental parameters, and human behaviors, etc. Additionally, we characterize their aggregate flexibility simply using a set of standard battery parameters. Based on the proposed unified model, optimal coordination and dispatch strategies are then developed for power grid and end-user services.

We present a Generalized Battery Model (GBM) to describe the flexibility of building loads and energy storage. An optimization-based approach is proposed to characterize the parameters (power and energy limits) of the GBM for flexible building loads. We then develop optimal coordination algorithms to provide power grid and end user services such as energy arbitrage, frequency regulation, and spinning reserve, as well as energy cost and demand charge reduction.

Several case studies have been performed to demonstrate the efficacy of the GBM and coordination algorithms and to evaluate the benefits of using their flexibility for power grid and end user services. We show that optimal coordination yields significant cost savings and revenue. In particular, the revenue of providing grid services or the savings from providing end user services from coordinating 20 residential air-conditioners and 1 commercial HVAC with 17 zones is greater than twice the revenue or savings of using a 50 kW/100 kWh battery. Moreover, we find that the best option for power grid services is to provide energy arbitrage and frequency regulation. Furthermore, when coordinating flexible loads with energy storage to provide end user services, it is recommended to consider demand charge in addition to time-of-use price in order to flatten the aggregate power profile.



Schematic of the proposed modeling and coordination framework.



Comparison of aggregate power profiles of residential and commercial HVACs among baseline, considering TOU only, and considering both TOU and demand charge.

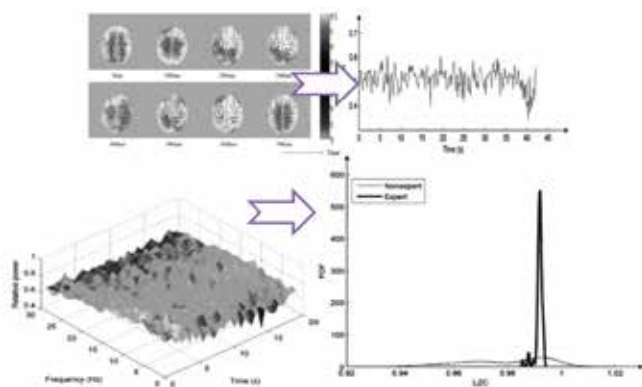
Preliminary Study on Knowledge Automation for Complex Systems – Uncertainties Impact in Decision-Making

Hong Wang

This project investigates the impact of the uncertainties of human decision-making on the optimization of the process operation.

Human operators are extensively involved in decision-making and plant operation of complex systems. The decision-making phase of human operators contains uncertainties in the time-horizon in terms of observation, use of knowledge, and decision realization. Since these uncertainties will affect the result of the decision-making, there is a challenging issue here on how the negative effect of these uncertainties on the optimization can be minimized.

A preliminary study in 2013 has shown that the uncertainties of human operators' decision-making can be measured through EEG (brain) signals, where the brain signals of inexperienced operators would exhibit a large portion of uncertainties and randomness, which lead to a widely distributed probability density function (PDF). It is, therefore, imperative that such a measure be used by well-defined process variables in the minimization of the impact of uncertainties embedded in human operators' decision-making. This forms the main objective of the proposed study.



PDFs of EEG signals for experienced and non-experienced operators.

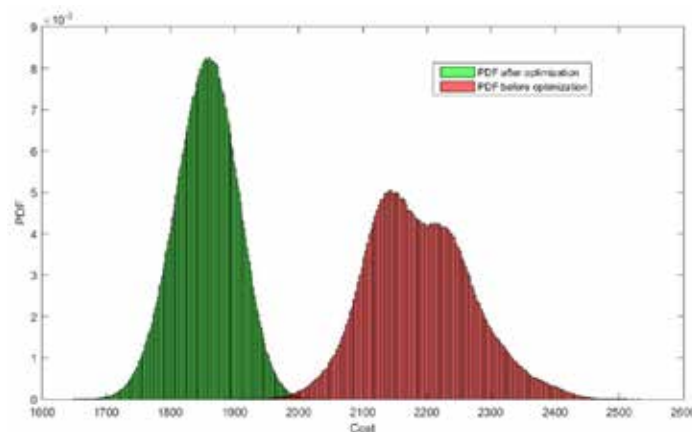
In FY 2016, we investigated the ways to quantify knowledge quality and randomness using signals collected from the brain via EEG. Relevant context and defined use cases are

selected for brain-computer-interface-based knowledge automation. We have also developed a method that can be used to quantify quality and randomness of knowledge extracted from the human brain. This has been achieved through a framework that represents the randomness of the knowledge extraction in the optimization phase and how it can be used to design optimization algorithms.

We have proved that any optimization problem can be treated as a feedback control design problem, where rather rich control theory can be directly applied to deal with optimization issues. The process performance is subjected to two types of uncertainties: one to the “controller” to represent human operators' uncertainties and the other to the “plant.”

The performance index J is, therefore, a random process, and its optimization should be performed so that the mean value of J is minimized, while the randomness of the optimized J is minimized. This is a PDF shaping problem, where the stochastic distribution control theory originated by the principle investigator can be applied so that the PDF of J is moved as close as possible to the left, and its spread area is made as narrow as possible.

We have applied the PDF shaping method to solve the feedback control problem. In a real-world scenario, millions of dollars could be saved.



PDF shaping results on stochastic economic dispatch.

Preparation for Tidal Turbine Deployment at the Marine Sciences Laboratory

Genevra EL Harker-Klimeš

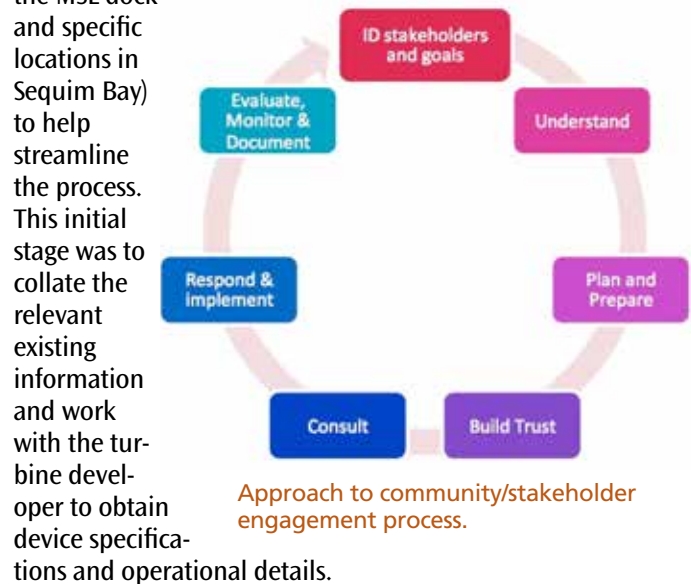
Clean energy development, such as marine renewable energy (MRE), is critical in the effort to reduce our dependency on fossil fuels and to reduce the impacts of climate change. In order to make MRE a viable energy alternative, we must remove the current barriers to development and deployment. This project provides the opportunity for a working tidal turbine to be deployed in Sequim Bay, adjacent to the Marine Sciences Laboratory (MSL), so researchers can study aspects beyond the device performance metrics, such as environmental effects, materials performance, and power integration studies.

MRE is an important energy source for the future, both for increasing energy security and tackling climate change. Although there are many MRE device designs advancing through technology maturity, to date, there are no commercial deployments of marine energy devices in the United States. The growth of the tidal turbine industry depends on performance testing, as well as opportunities to study the environmental effects and conduct tests in well-characterized locations in nearshore marine waters to help remove regulatory barriers that are currently preventing the commercialization of this clean, renewable energy generation alternative. The objective of this project is to characterize tidal hydrodynamics in Sequim Bay to determine optimal tidal turbine device placement and to conduct preliminary community engagement and permitting preparation work to streamline the process for the eventual deployment of the tidal turbine at MSL.

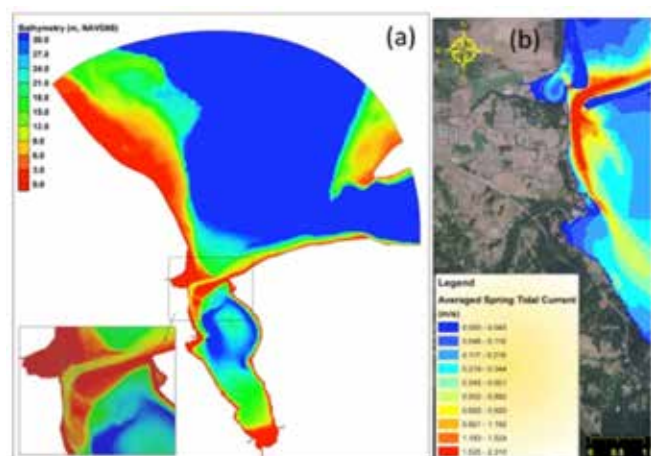
We developed a high-resolution, standalone hydrodynamic model to simulate tidal circulation for Sequim Bay that is based on the unstructured-grid, Finite-Volume Community Ocean Model and covers the entire Sequim Bay and part of Strait of Juan de Fuca with a semi-circle open boundary. The model was applied to characterize detailed velocity distributions in the bay, which were used to determine the most suitable locations for testing tidal turbines. Both Surface Modeling System and ArcGIS software packages were used to visualize model results. The resulting plots will enable PNNL, in conjunction with the developer, to determine appropriate locations for the deployment that allow research to be undertaken in a practical way.

A community engagement strategy was developed and executed to inform key local community members of the proposed project and to gather feedback prior to moving forward with the permitting process. Several successful meetings were held with key local stakeholders and the project will move forward without opposition.

Permitting the deployment/installation and operation of a multi-turbine unit and necessary infrastructure is likely to take 6 months to 1 year. Our approach is to leverage information that was gathered for the existing 5-year permit (includes the MSL dock



The completion of these tasks has illustrated that moving forward with this multi-turbine unit proposal is feasible, and the work has informed the next steps that need to be taken to provide research opportunities around clean energy development in the marine environment.



Quantifying Hazardous Turbulence Conditions and Dose-Response for Fish Passage through Hydropower Turbines

Marshall C. Richmond

A unique laboratory testing capability has been developed to measure fish responses to turbulence conditions that are representative of those occurring in hydropower projects. Such a facility allows the biological impacts of turbulence to be characterized and safe levels to be identified.

Turbulence is currently the most uncertain and inadequately quantified of hydro-turbine injury mechanisms for fish. Existing studies of turbulence effects on fish are of limited use for design and evaluation of turbine systems, because they use qualitative descriptors that cannot be directly related to variables the engineering design process can compute or measure. By recreating turbulent conditions in a laboratory environment, repeatable studies of fish response to turbulence can be conducted in isolation from the other stressors of fish passage. A unique feature of the research was the use of highly resolved computational fluid dynamics (CFD) models of complete hydro-turbine systems to estimate realistic values of turbulence metrics that are representative of operating full-scale turbines. These metrics mined from the CFD results were used as the basis for designing a novel turbulence test tank that can produce realistic exposure conditions for live fish testing.

A laboratory facility was constructed at the Aquatic Research Laboratory at PNNL in Richland to generate highly turbulent flow conditions within a test volume. Turbulence conditions were controlled using two randomly actuated synthetic jet arrays of 25 independent submersible pumps in a 5x5 grid. This technique successfully created turbulence velocities up to an order of magnitude greater than those of previous applications.

A total of 200 spring Chinook salmon were used in turbulence stressor tests following approved Institutional Animal Care and Use Committee protocols. All fish from all treatments immediately regained equilibrium after the treatment period ended, and there were no significant differences in survival

of fish exposed to the control and turbulence treatments in the current study. The turbulence conditions that were created in this experiment are below the threshold for significant behavioral or survival effects on juvenile Chinook salmon. This information can be used to evaluate CFD models of new turbine designs to establish a lower threshold.

The fish survivability observed in these tests will contribute to the understanding and assessment of biological response to hydroturbines and other submerged structures. These tests have demonstrated that turbulent velocities of 24 cm/s are below the threshold for significant behavioral or survival effects on juvenile Chinook salmon.

Two randomly actuated synthetic jet arrays of 25 independent submersible pumps in 5x5 grid units in the flume tank, showing the fish test volume defined by the net pen.



Test fish in the net pen between the two jet pump arrays.

Resilience in Large-Scale Distributed Control Systems

Wei Zhang

This project is developing approaches to analyze, quantify, and improve resilience of distributed control systems. The research is expected to yield new design tools for distributed control of the electric power grid.

In distributed and physically interconnected systems, the effects of local physical faults or cyber attacks propagate through the physical and communication networks and impact the rest of the system, presenting a challenge for the resilient operation of the overall system.

This project will develop methods to evaluate when a distributed control system can still satisfy its operational and performance requirements after abrupt contingency-driven changes to the system structure. While there are many studies that provide specific insights for particular resilient control scenarios, the main research thrust of this project lies in the development of a mathematical framework that can be used to study resilient distributed control, as well as methodology for scenario-independent resilience quantification for power system applications.

In FY 2015, we reviewed the literature on resilience of distributed control systems applied to power systems. We also developed a framework using graph-based models to analyze resilience of distributed control systems. In this framework, the cyber and physical coupling between different subsystems are modeled by a graph, and contingencies (attacks or failures) are described as perturbations to the system graph. Distributed control design problems are described as distributed optimization problems, and resilience is quantified by the change in the value of the objective of the optimization problem driven by a contingency event.

We analyzed the resilience of optimal power dispatch operation strategy using the Institute of Electrical and Electronics Engineers (IEEE) 30-bus system. We simulated the impact of about 1,600 attacks/failures contingencies that cause loss of one or two transmission lines in the system. Most contingencies resulted in cascading failures, which led to up to 40%

load shedding. We proposed a re-dispatch method and showed that on average it reduces load shedding by more than 20%.

In FY 2016, we focused on quantifying resilience for wide area control (WAC) of the bulk power systems. WAC is designed to improve the stability of the grid with increased penetration of renewables and distributed energy assets. All types of WAC rely on real-time Phasor Measurement Unit (PMU) measurements sent over communication networks. We studied WAC resilience under a class of cyber attacks that may cause loss of communications from the PMU sensors.

As WAC is a small signal stability problem, we used a linearized network-reduced power system model to study its resilience. A WAC is defined by a system-wide state feedback gain matrix K , whose (i,j) element represents how the state of bus j affects the control decision at bus i . The (i,i) block is zero if bus i control action does not rely on the PMU measurement of bus j state.

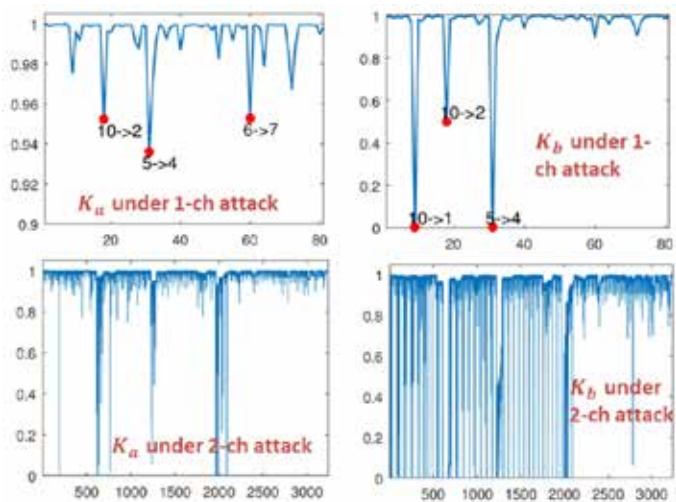
One of the results was a new way to quantify the resilience of a WAC controller as the performance degradation under the communication loss attacks. The resulting resilience index takes values between 0 and 1, where 0 means “not resilient” (can be destabilized by some attacks) and 1 means “completely resilient” (will not be affected by any attacks).

A second result was a computationally efficient approach to quantify resilience (a problem with combinatorial complexity) using a convex relaxation that provides sufficient conditions for resilience. We also derived sufficient conditions under which the solution of the relaxed problem is exact.

We validated our resilience quantification method on the IEEE 39 bus system, which includes 10 buses and 81 direct communication links. The computed resilience indices of two WAC controllers, K_a and K_b , under 1-channel and 2-channel attacks on the communication links, show that K_b is less resilient than K_a , and its corresponding closed-loop system can be destabilized by attacking one communication link (bus 10 to bus 1 or bus 5 to bus 4). In addition, when attacking two communication links, there are many more opportunities to destabilize the system under K_b than that under K_a , as shown in the bottom two figures.

This provides grid operators with a formal way to compare different WAC, from both a performance and resilience perspective, and helps identify the most critical communication links to be protected from cyber attacks.

In FY 2017, we will further our work on resilience analysis for WAC systems by providing a link between the resilience metric and dynamical systems analysis concepts, and we will extend our investigation to market-based control systems.



Resilience index for two WAC controllers under 1- and 2-channel attack scenarios.

Structural Framework for a Holistic Transportation System Model

George G. Muntean

This project will document relevant data sets and assess the potential structural requirements for a simulation platform that is data driven, self-populating, and self-calibrating.

Transportation systems simulations are on the brink of revolutionary change that will allow us unprecedented situational understanding. With this understanding will come transformational innovations that will lead to significant efficiency improvements, as well as improvements in safety and convenience. This change is being enabled by the confluence of megatrends in Big Data, communications, sensing, data analytics, and computations. When implemented, we will, for the first time, have simulations that are data driven, self-populating, and self-calibrating. As a first, limited step to the realization of this vision, this project will document relevant data sets and assess the potential structural requirements for such a simulation platform.

Numerous categories of transportation simulation models have been developed to answer various needs within the community such as traffic control, congestion management, planning, and forecasting. These models range from macro to micro scale and can be either static or dynamic. The earliest efforts (1960s) were static, trip-based models. At this time, infrastructure development was in full swing, and the fidelity of these models was sufficient for the task. As pressure began to mount in the 1990s to better utilize existing infrastructure, activity-based models were developed to help improve model fidelity in the increasingly congested traffic environment.

The current era in transportation is seeing radical new changes. New technologies (connected and autonomous vehicles), new business models (ride sharing and mobility as a service), and new attitudes (reduced car ownership and increased use of public transport) are creating an increasingly complex landscape. Incorporation of person choice, multiple modes, and human behaviors are becoming increasingly critical in mobility simulations.

Simultaneously, we are experiencing an explosive growth in connectedness and data availability. This confluence is usher-

ing in the next transformational evolution in traffic simulations. The grand challenge for the research community is to access these data resources and synthesize them to provide actionable understanding and control of the traffic system. To accomplish this will require sophistication and advancements in numerous areas such as data acquisition, extraction, cleaning, integration, aggregation, fusion, analysis, modeling, and visualization. Tools such as machine learning (and deep learning), gaming, and agent-based modeling will be required to automate the creation of this situational understanding.

In this vision, a transportation planning area will be modeled through a highly automated process that brings in the hard infrastructure overlay through geographic information system databases and creates a set of smart agents through access to local information resources. The population set and “intellect” of the individual agents is predicated on the availability of data within the study area. In regions with high levels of data access, numerous classes of agents can be created with unique behavioral characteristics. These characteristics may be codified using synthesized data from social media, communication systems, traffic management, and transit information, etc. These smart agents will then decide on frequency of travel, mode of travel, routing, and destinations. Their behaviors during travel will be simulated as well. This tool can be utilized not only for operations management but for planning and situational preparedness.

This project is a scoping study that proposes a conceptual construct for this new class of simulation and investigates the range of data sets that are currently available for utilization within this construct.

A critical enabling aspect to the emergence of a data-driven simulation platform is the ability to access and utilize relevant data sources. The primary outcome from this project was a thorough and extensive review of the data landscape. The data traditionally used for transportation modeling was procured either by contracting companies who specialize in traffic count data, procuring local data (e.g., pneumatic traffic counters), and/or accessing federal data sources (e.g., census and registration data). These data were highly specific and tended to focus primarily on traffic flow and congestion simulations. Examples of these data streams include the follow-

ing: traffic volume counts (mainline and intersections), traffic speed data, bicycle/pedestrian counts, origin-destination surveys, transportation network inventory, travel time data, vehicle classification counts, attitude and opinion surveys, safety data analysis, regulatory compliance inventory (ADA, etc.), video data collection, parking occupancy.

More recently, commercial entities such as AirSage, HERE, INRIX, and TomTom have been offering data services through licenses and agreements. For example, AirSage obtains cellular data from two national carriers and has specialized in creating origin-destination matrices for specialized localities. These services offer good data on what trips may be taken, but they do not offer much insight into decision-making and why trips are made. These data fall into the larger category of Location-Based Services and are a growing part of the “data as a commodity” trend. Of particular interest in the last decade has been the advent of “Open Data” and the increasing number of “Open Cities” giving the public access to

numerous data sets. This is a promising trend but suffers from the classic “Big Data” challenges around quality, reliability, and homogeneity.

The primary conclusions from this work are that data and the way it is viewed are evolving quickly. While it is true that ever-increasing quantities of data are being generated and shared, it is also the case that it is being viewed more and more as a commodity to be monetized. While the monetization of the data ensures entities will provide the data analytic services to make the data useful (aka “liquid data”) it also complicates the access and utilization of the data for public use. Finally, an exciting, but complicated, frontier in data is the use of social data streams to investigate behaviors, choices, and decision-making. To date, little has been done to formalize these data streams into larger micro-simulators for traffic modeling.

Visual Analytics Environment for the Smart Grid

George Chin, Jr.

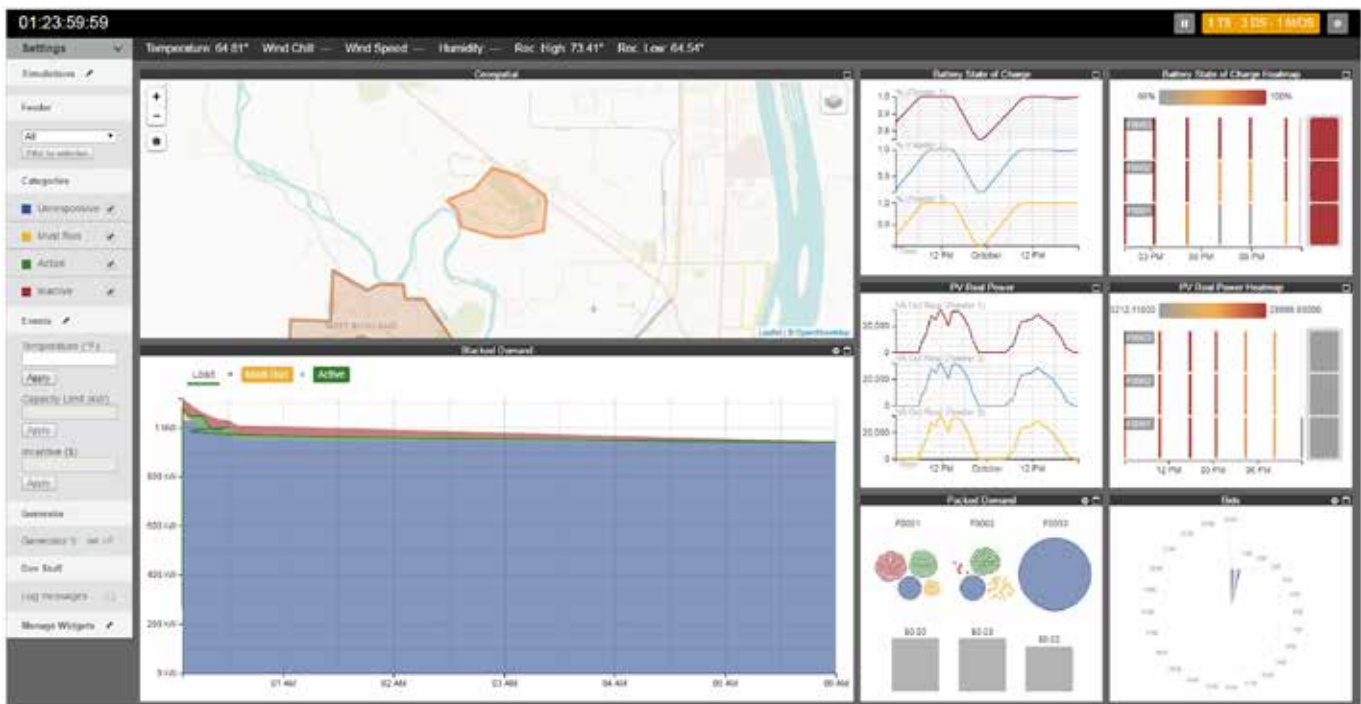
This project is developing a more comprehensive visual analytics platform for smart grid operations and analysis by extending the Visual Steering and Modeling Environment (VSME) to combine the additional two essential smart grid areas of photovoltaic generation/integration and distributed energy storage with demand-response.

Today, a modernization effort is underway, whereby new digital technologies are allowing system planners and operators to better monitor and measure the status and performance of transmission and distribution lines, as well as to interact more directly with consumers to effectively address real-time conditions and needs. The electric power grid is rapidly evolving into the “smart grid”—a term summarizing the contributions of new digital technologies, advanced sensors and devices, and tighter relationships and communications between suppliers and consumers. With the rapid growth and evolution of the smart grid, new visual analytics capabilities

and tools are needed to make sense of the huge bounty of streaming data and to support new and emerging operating paradigms.

Visualization and visual analytics tools are available for conventional transmission system side analysis through select software systems such as PowerWorld. For distribution systems, however, visual analytics capabilities are scarcely available or applied. As for smart grid models and systems, these are evolving today. More and more smart grid systems are being deployed and tested across the country by various utilities and smart grid demonstration projects. Advanced visual analytics tools would be extremely useful in the analysis, evaluation, and operation of these rapidly emerging systems.

This project identified the following three critical, emerging smart grid areas in need of visual and data analytics support: 1) demand-response – smart meters and appliances allow consumers to adjust demand based on price signals; 2) photovoltaic (PV) generation/integration – it is becoming much more common for homes to be outfitted with solar power generation systems; and 3) distributed energy storage – utilities are deploying systems to locally store energy.



The enhanced VSME dashboard integrates and displays demand-response, photovoltaic generation/integration, and distributed energy storage information across multiple visualizations.

VSME was originally developed as a visual analytics environment to enable the monitoring and analysis of dynamic smart grid data. The concept of visual analytics focuses not only on interactive visual interfaces, but also on the analytic reasoning that such interfaces may facilitate. Thus, VSME provided advanced interactive visualization techniques and tools to allow users to understand, comprehend, and interpret dynamic smart grid data, combined with analytics capabilities that allow users to navigate, explore, and mine the dynamic data to gain insights, discover knowledge, validate hypothesis, and make informed decisions.

To ground development on a concrete smart grid use case, VSME initially concentrated prototype development on supporting transactive control and demand-response operations and analysis, which was the focus of several other simulation and modeling projects. As a result, the dynamic data that was steered and visualized through visualization tools consisted of information such as load categories, bid prices, bid quantities, clearing prices, demand curves, and incentive amounts.

To make VSME more compelling and attractive to utilities, DOE, and other smart grid stakeholders, the project expanded VSME's limited demand-response analytics functionality to incorporate the other emerging smart grid areas of PV generation/integration and distributed energy storage. Combined with demand-response, three smart grid areas in total are now supported in VSME. For each of these areas, the project expanded the data stream (and its schema) to pass

more information, selected and adapted information visualization tools to best convey the new dynamic data, and instantiated and integrated new coherent views of the data and visualizations. The streaming data from which the visual analytics capabilities were developed came from the distribution system simulator GridLAB-D™, which has explicit computational models for each of the three essential smart grid areas.

The outcome of the project is an enhanced, comprehensive, integrated VSME for monitoring and analyzing dynamic demand-response, PV generation/integration, and distributed energy storage information. VSME may be connected to data streams from physical devices, simulations, or a mixture of both to enable different types of usages and applications.

A distribution system operator may use the enhanced VSME in the control room to monitor and operate the smart grid (devices) or train on certain scenarios or events (simulations). A distribution system planner may use VSME to conduct near- and intermediate-term planning by running models against current and emerging conditions (devices plus simulations). An analyst might replay collected data (devices) in VSME to interpret and analyze grid, consumer, and operator behaviors. For simulation and model developers (e.g., DOE and laboratory researchers), VSME may provide an integrated testbed environment for evaluating and analyzing smart grid paradigms, models, and simulations

Engineering and Manufacturing Processes



Bulk Thermally Stable Nanocomposite Processing

Scott A. Whalen

In this project we are developing the underlying science and engineering processes for fabricating bulk tubing and bar with thermally stable nanocomposite microstructures. Our research in far-from-equilibrium shear processing is envisioned to enable fabrication of industrial-scale products, with game-changing material properties that have heretofore only been achieved in powder, flake, thin-film, or very small specimens.

The goal of this project is to use shear-assisted processing and extrusion (ShAPE) to consolidate and extrude, in a single step, nanoscale Mg-X and Al-X precursors (powder or flake) without any loss of microstructure in the resulting bulk form. Thereby, it will maintain property improvements derived from nanoscale features in the precursor. Current Assisted Pressure Activated Densification (CAPAD) is also being used for the same purpose in Fe-Si.

Although a large body of research exists for using various high shear processes to refine microstructures, only the ShAPE process offers promise for fabrication of bulk nanocomposite materials at an industrial scale. We expect that the processes developed and material properties achieved will lead to publication in high-impact journals.

AZ91 ribbons, fabricated by melt spinning at PNNL, were consolidated and extruded using ShAPE to form tubing with an outer diameter of 7.5 mm, wall thickness of 0.75 mm, and length up to 15 cm. Microstructural analysis showed that the 5 μm grain size and nanoscale second-phase particles within the precursor ribbons were preserved in the bulk tubing. It was also discovered that the ShAPE process is effective at breaking up millimeter-scale segregated phases and dispersing them as micron-size particulates in the bulk tubing.

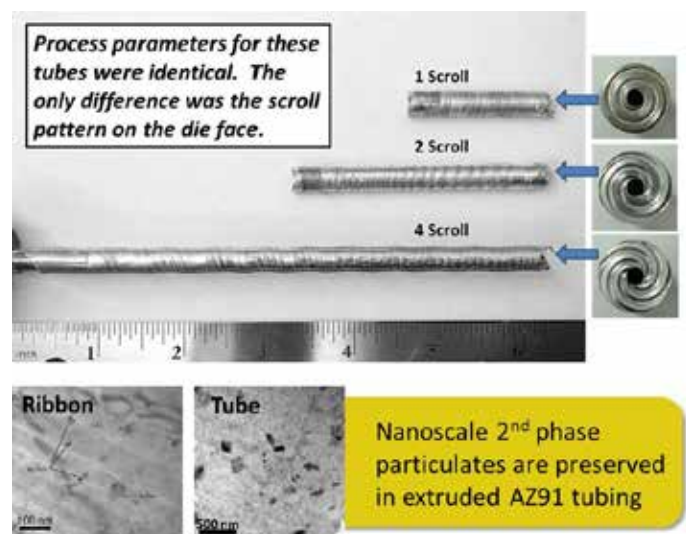
The simultaneous linear and rotational shear inherent to ShAPE, controlled by feed rate and tool rpm, respectively, has been found to impact textural alignment in the extrudate. For example, the (0001) basal planes in AZ91 tube walls are highly aligned perpendicular to the extrusion direction. Varying the feed rate and tool rpm can result in a degree of textural control that is advantageous from a material property anisotropy standpoint. Based on this discovery, we also

investigated the impact of tool face scroll features on textural alignment. Microstructural analysis from these studies is still forthcoming, but it is clear from visual observations of the tubes, that differing scroll patterns significantly affect flow in the extrudate.

Early in FY 2017, we plan to complete installation and calibration of the custom ShAPE machine at PNNL and new CAPAD machine at the University of California, Riverside. These one-of-a-kind machines will be used to consolidate (CAPAD) and extrude (ShAPE) rapidly solidified flake and/or mechanically alloyed Mg-X, Al-X, and Fe-Si precursors. Processed materials will be characterized to determine the extent to which the preferred precursor microstructures are retained in the bulk form. Relevant material properties will also be measured and material specific processes developed.



Schematic of ShAPE tooling and new state-of-the-art machine.



Extrusion of AZ91 melt spun ribbon using the ShAPE process with three different scroll patterns. Transmission electron microscopy images showing preservation of nanoscale particles in extruded AZ91 tubing.

PN1602212799

Graphene-Oxide-Based Structured Laminar Membranes

David Gotthold

Structured laminar membranes, consisting of hierarchically stacked, overlapping layers of 2D graphene oxide (GO), are a fascinating and promising new class of materials with the potential for radically improved water separation with excellent selectivity and high permeability.

This work was based on published results that showed GO has significant potential as a highly selective membrane for water vapor, with selectivity above 10^{10} .

In order to enable practical applications for this new class of structured materials, key fundamental questions must be answered. What is the mechanism responsible for water transport in laminar membranes (such as GO) and what are the influences of structure and surface chemistry on the transport properties? How does the laminar structure assemble and how do we develop a scalable synthesis route to make commercially viable separation membranes? By developing the fundamental understanding of how these new laminar membranes function and assemble, we will enable the development of practical water separation membranes.

In the first year (FY 2014), this program focused on evaluating basic material properties such as graphene flake size, membrane structure, and permeability. In FY 2015, we built on that basic understanding to develop a scalable process for synthesizing large membranes, achieving membranes up to 600 cm^2 with a path toward continuous production. During that work, we identified new challenges in materials stability, especially when the potential application involved direct liquid contact. Addressing those challenges was the primary focus in FY 2016. We evaluated the physical and chemical stability of GO membranes under a range of conditions and evaluated the ability of using both chemical modifications and structural processing to improve stability in liquid environments.

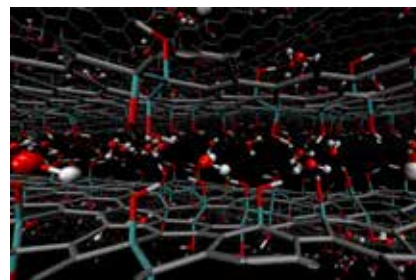
In the first year, we determined that larger flakes produced more physically robust membranes with higher selectivity and permeability. In FY 2015, we developed an improved gel preparation method that maintained large GO flakes, which are key for scalable GO membrane formation. Casting of thin and large GO membranes was demonstrated ($2\text{ }\mu\text{m}$ thick and greater than 20 cm long) using this graphene oxide gel.

In earlier phases of the project, the application work was focused on gas separation, such as dehumidification, but in

FY 2016, the focus shifted to liquid separation such as biofuel dewatering. We prepared multiple membranes using both traditional vacuum filtration and the new gel-based coating method. They were tested for stability in liquids by immersing them in different solvents, including water, ethanol, dimethylformamide, and acetone. The GO remained stable in all solvents except water, in which the membranes disintegrated, typically within a few hours. The time to completely re-suspend the GO in water depended on both the preparation process and the amount of time that had elapsed between membrane fabrication and testing, with vacuum filtered and older membranes lasting significantly longer: days to weeks. We evaluated multiple paths for improving the stability, including accelerated aging with heat, short-wavelength light exposure to drive reactive chemistry on the graphene surface, and vacuum drying with a combination of vacuum and heat producing the best stability and performance.

We performed separations of ethanol-water mixtures with the treated membranes at a range of temperatures and concentrations. The membranes were stable over the test period of 5 days, with a maximum water/ethanol selectivity of 874 and a concentration-dependent water flux of $0.33\text{--}1.32\text{ kg m}^{-2}\text{ h}^{-1}$ at 90°C .

In previous years, a sequence of quantum and classical simulations were carried out to probe the behavior of the water confined between the layers, as well as the interaction of the water with the functional groups in the membranes. In FY 2016, we performed molecular dynamics simulations of water interactions in GO membranes at several hydration levels using a model system with hydroxyl groups only and a C/O ratio of 4. Our results show that water diffusion in GO is an order of magnitude slower than in bulk water due to strong hydrogen-bonding interactions between H_2O molecules and the OH group. The optimum distance for the hydrogen bond in our simulation is 0.17 nm and the oxygen-oxygen distance is typically 0.27 nm . Even at the highest hydration level of $23.3\text{ wt.}\%\text{ H}_2\text{O}$, only about 21% of the H_2O molecules were free or bulk-like. We observed large water clusters comprising 10 to 30% of the water molecules present in the system. Such clusters can span across oxidized regions, graphitic regions, and defects or holes that have been experimentally observed, thereby contributing to rapid water transport.



In Situ Fastening of Metals to Plastics

Piyush Upadhyay

Mainstream use of multi-material components in assembly has the potential to reduce waste, save energy, and lower the cost of manufacturing in transportation, infrastructure, and consumer products. We will develop a novel manufacturing method for joining of metals to carbon fiber reinforced polymer (CFRP) composites using an in situ mechanical interlock between materials during the assembly process.

Multi-material components that include CFRP composites and metals are increasingly used in aerospace, automotive, and consumer products to meet local mechanical property needs and to attain effective weight savings. Enabling efficient use of metal and composite vehicle components through development of new, solid-state joining techniques is a critical path to meeting the 30% weight reduction targeted in the *EV Everywhere Grand Challenge*. This requires effective joining methods that can save costs and reduce assembly weight. Current state of the art methods in CFRP-metal joining, including bolting, riveting, and adhesive bonding, add complexity and increase assembly time.

The lack of fast, reliable, and robust manufacturing processes for joining dissimilar materials has been identified as a key technology gap for both light- and heavy-duty vehicle systems in recent DOE Vehicle Technology Office workshop reports. Maximizing structure efficiency through advanced joining is also necessary to realize the full potential of advanced composites in lightweight aircraft and aerospace structures.

The objective of this project is to enable multi-material design and efficient material use in manufacturing through development of the Friction Stir Scribe Technology (FSST) to join metals to fiber reinforced polymers. This results in an *in situ*, rivet-like fastening between metal and polymer produced by simultaneously inducing local flow of the polymer matrix and mechanically disrupting the surface of the adjoining metal into the polymer interface. Development targets of the method include optimization of joint strength for specific combinations of metals and composites. This work aims to increase understanding of process challenges, generate property datasets, and produce demonstration samples for joints between aluminum and carbon reinforced thermoplastics for the benefit of prospective end users of the technology.

Friction stir scribe tools and associated stationary shoulder parts tools were designed and fabricated to join 3-mm-thick carbon fiber reinforced polyamide-6,6 plaques to 3-mm-thick AA6061 aluminum sheet.

The quality of the produced joints was initially tested by drop tests, peel tests, and monitoring of the crown surface. Identified parameters for defect-free welds were used to produce lap joints for testing, characterization, and demonstration purposes. Water jet-cut specimens were lap shear tested and joint cross-sections were examined to understand effects of welding parameters. Lap shear fracture modes observed include interfacial failure and fracture via CFRP weld nugget with initiation near the hook feature and traveling near the crown. The importance of tool wear in friction stirring of CFRPs became apparent in this work, emphasizing the need to match tool materials with FSST material system characteristics.

This project has increased our understanding of the FSST process as it relates to joining industry-relevant carbon fiber reinforced thermoplastic composites to metals. Development experience has elucidated challenges and potential for the technology. FSST, which may be used in combination with adhesives, may be used to reduce assembly time requirements for adhesive cure. Creation of mechanical fastening between CFRP and metal components using FSST during assembly may prove advantageous to the use of conventional fasteners in terms of lighter weight assemblies with fewer parts and faster assembly time.



A representative FSST joint between carbon fiber reinforced nylon and aluminum sheet.

PN1604712824

Novel Heat-Treatment for Automotive Castings

Aashish Rohatgi

The goal of this project is to develop a novel, energy-efficient heat-treatment technique for metallic alloys. We anticipate that this technique will enable development of high-strength, light-weight, and low-cost aluminum and magnesium automotive castings that will lead to better fuel economy and reduced green-house gas emissions.

Heat-treatment of metallic alloys is essential to obtain desired functional performance such as strength, toughness, fatigue life, and corrosion resistance. Heating a material activates atomic-scale phenomena that produce the microstructures responsible for the material's functional performance. These microstructural changes often occur very slowly, which therefore, requires many hours of heating. Such extended heating is energy intensive and increases the product cost. For example, the required time in a traditional oven/furnace for heat-treating aluminum (Al) castings can be quite long (up to 24 hours). Therefore, some metal castings, despite their potential to reduce a car's weight, may be uneconomical for the mass-market automotive industry. In this project, we propose to develop a novel, energy-efficient heat-treatment approach to accelerate the heat-treatment process and, thus, lower the associated energy requirements, time, and cost.

An aluminum casting alloy, referred to as a 319 alloy in the aluminum industry, was used as the test material in this work. Commercial software from Ansys was used to develop a multi-physics model to simulate the specimen heating. Thermal and other physical properties of the Al alloy used in the model were obtained from the technical literature. Finite element solvers in the Ansys model were used to predict the sample temperature distribution as a function of the process parameters.

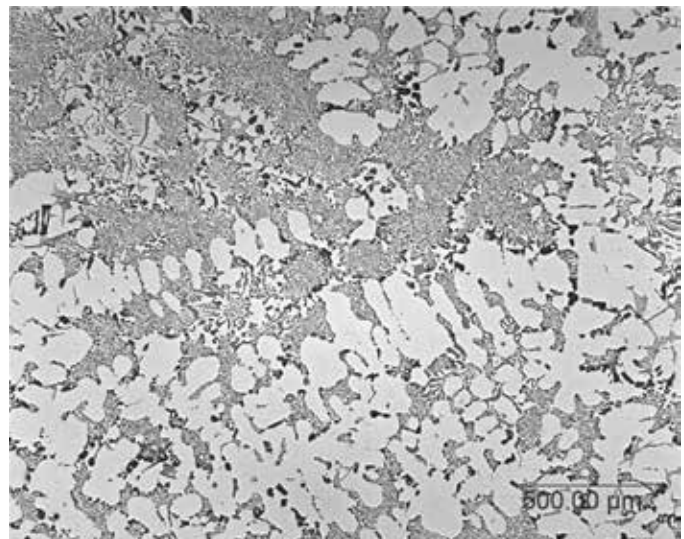
The as-received alloy material was machined into test samples and subjected to different types of heat-treatment using both conventional furnace heat-treatment and the new method. The heat-treated samples were analyzed using metallography, scanning electron microscopy, hardness testing, and differential scanning calorimetry (DSC).

The heat-treatment model demonstrated that iterative feedback between the solvers allowed the simulation results to converge within several iterations. Good correlation between

the measured sample temperature and the model predictions were obtained with a heat-loss coefficient of $5\text{--}10\text{ W/m}^2\cdot^\circ\text{C}$ to account for convective heat losses from the sample surface.

The microstructure of the as-received material was a typical as-cast microstructure with aluminum dendrites, Al-Si eutectic, and various intermetallics comprising Al in different proportions with copper, iron, manganese, and silicon. The DSC experiments of as-received and solution-treated samples showed several endothermic peaks between approximately $500\text{--}550^\circ\text{C}$ and between about $550\text{--}625^\circ\text{C}$ corresponding to the dissolution and melting, respectively, of different microstructural phases. The hardness of the as-received, as-cast material was measured to be approximately 45 RB (Rockwell B scale), while the hardness of the solutionized (495°C for 8 hours) plus aged material was approximately 73 RB. By comparison, initial experiments suggested a sample that was solution-heated by the new method, followed by aging, could achieve a similar high hardness with only 1 hour of solution heating, supporting our hypothesis of the advantage of the new approach.

In FY 2017, additional experiments will be performed to confirm the applicability and advantages of the new heat-treatment method on other alloys and to understand the underlying mechanisms responsible for improved performance. Additionally, the multi-physics model developed in FY 2016 will be extended to enable temperature prediction in samples with arbitrary geometry.



Optical micrograph of the as-received aluminum alloy sample showing the as-cast microstructure that typically needs to be heat-treated to improve its mechanical properties.

Scalable Processing of Nanostructured Materials

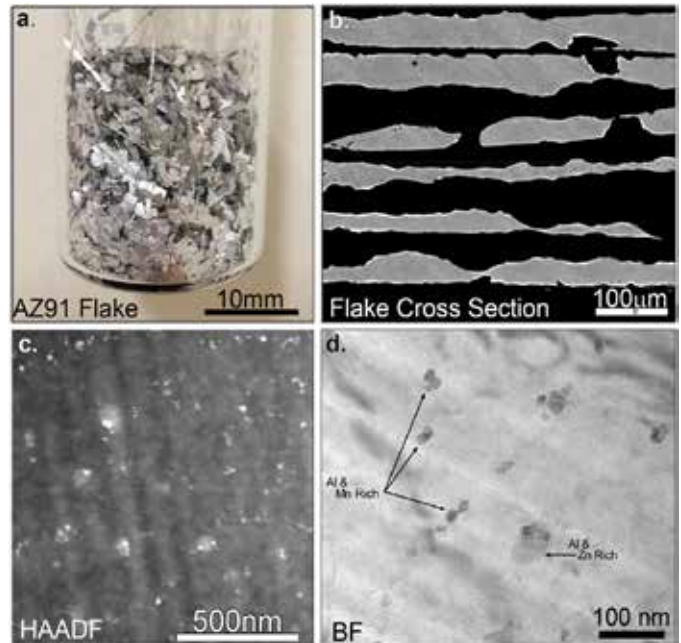
Nicole R. Overman

We are developing novel and scalable processing methods to fabricate unique structural and magnetic nanocomposites. This work is being performed in an effort to advance high-throughput manufacturing technologies and design new lightweight structural alloys and soft magnetic materials.

Prior research on far-from-equilibrium alloy development has revealed several key factors that can have profound effects on improving material properties such as strength, toughness, corrosion resistance, and in some cases, magnetic properties. The basis for these advances has relied heavily on microstructural control and refinement. The underlying science behind them boils down to a materials engineer's ability to impart maximum disorder into the alloy at a microscopic level. The difficulty with this approach is in stabilizing the disordered or far-from-equilibrium material enough so that it can be retained despite extreme environments encountered during processing and in service.

The current research is part of a strategic thrust area focused on production and stabilization of unique nanocomposite materials that can only be produced using specialized processing methodologies. This work bridges the research efforts of two sister projects under the nanocomposites thrust area, focused on 1) modeling thermally stable far-from-equilibrium alloy compositions and 2) severe shear deformation consolidation approaches through the production of far-from-equilibrium precursor materials.

The key goals of this project are first to provide rapidly solidified (RS) material for consolidation testing and, secondly, to understand and identify key processing parameters that can be exploited to produce and homogenize these novel precursor materials on the nano-scale level. Processing regimes under investigation target two primary fabrication approaches, melt spinning and ball milling. Using this multifaceted approach, we are able to take advantage of both thermodynamic and kinetic effects to produce precursor materials for consolidation that exhibit novel alloy compositions with enhanced homogeneity, processability, and thermal stability.



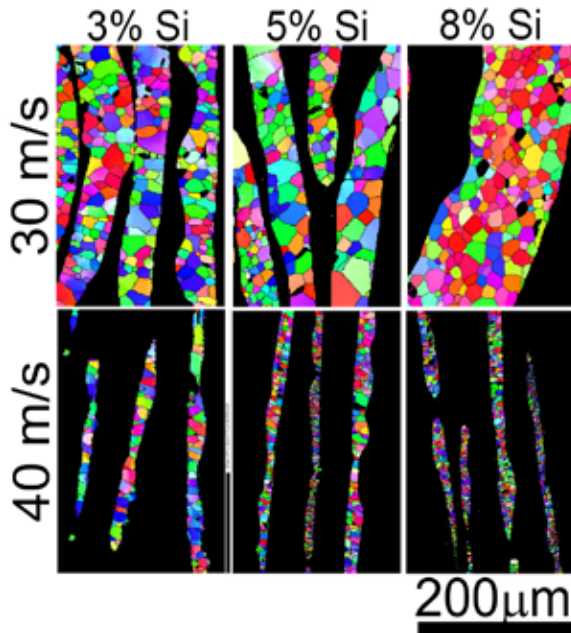
As produced AZ91 RS flake material is shown in (a), along with a cross-sectional view of the flake via scanning electron microscope backscatter imaging (b); transmission electron microscopy images (c,d) show the presence of nanoscale second phases.

Our approach in year one has focused on optimizing structural refinement through systematic evaluation of processing parameters for both of these methods.

Over the past year, significant progress has been made in the key material systems targeted. Demonstrator alloys were identified and fabricated based on their ability to serve as game-changing material systems. AZ91, a widely used magnesium-based alloy, was chosen for preliminary process development, as well as Fe-Si (3–9wt %). Both alloys were fabricated using melt spinning. Evaluation of the melt spun precursor material has revealed key fabrication insights, showing fine distributions of nano-scale second phases. Ball milling research has resulted in the development of generalized curves defining optimal milling time for grain refinement and magnetic properties optimization.

A unique outcome of the work performed in year one has been an increased understanding of the effect wheel speed can have on the as-produced microstructure of melt spun ribbon. We have shown that the grain size and structure of Fe-Si based ribbons can be significantly refined when the through thickness width of the ribbon is decreased. We

have also shown that the ribbon thickness and, therefore, grain structure can be reduced by increasing the wheel speed. This result was found to be highly dependent on the material system under evaluation; the Mg-based ribbons did not show the same behavior.



Electron Backscatter Diffraction results of ribbon cross sections show grain orientation in AZ91 RS Fe-Si ribbons. Increasing the wheel speed was found to dramatically refine both the flake thickness and grain structure over a wide variety of compositions.

Preliminary studies on the thermal stability of the melt spun ribbons using differential scanning calorimetry have shown the absence of equilibrium phases common to as-cast AZ91. Following a melting event, repeat analysis of the same specimen shows dramatically different behavior and the presence of equilibrium phases. This result is significant because it validates the ability of melt spinning to produce far-from-equilibrium precursor materials. Unique second phases, potentially exhibiting metastable chemistries have also been identified in these novel precursor materials and were evaluated using transmission electron microscopy.

During FY 2017, we plan to extend our knowledge of processing parameters to alloy compositions via thermodynamic and atomistic modeling studies. We will also evaluate the effect of melt spinning and high energy ball milling on the final consolidation microstructure when these processing methodologies are used in series. Finally, we plan to further develop our analysis capability to streamline thermal stability assessment of RS flake precursors.

Materials Science and Technology



3D Printing of Electrical Sensors for Biological and Chemical Detection

Rebecca L. Erikson

Three-dimensional (3D) printers have captured the imagination of Americans both young and old with the opportunity to create anything imaginable while in the comfort of your own home. This project provides a vision and a pathway to demonstrate 3D-printed parts capable of detecting chemicals that may someday be used as glucose monitors for diabetes monitoring or for vapor detection of hazardous chemicals such as gas leaks.

To date, 3D printers have been used primarily to create structural components of various systems, but limited efforts have been concentrated in the area of a functional sensing component. This project focuses on the development of methods in materials science, physics, and engineering to enable a fully functional, electrically active, biological and/or chemical detector produced solely from a 3D printer. A large focus of this project is around custom formulations of materials that can be used with standard fused deposition modeling 3D printers.

Our multidisciplinary team has demonstrated significant science and engineering outcomes with our research this year. We have developed and demonstrated techniques to produce both physical and electrical changes in material during a 3D print by altering the magnetic field around the extrusion nozzle during fabrication, which led to filing a formal patent application. By altering the magnetic field, we have shown that a commercially available material can be made to change its conductive properties by threefold and that the surface area can also be increased or decreased.

We have focused our efforts on the development of polymers and polymer hybrids that can be used as feedstock to our printers with very specific chemical properties of interest. These materials have included polymers that have very high conductivity (20 Ohms/cm), polymers that remain flexible

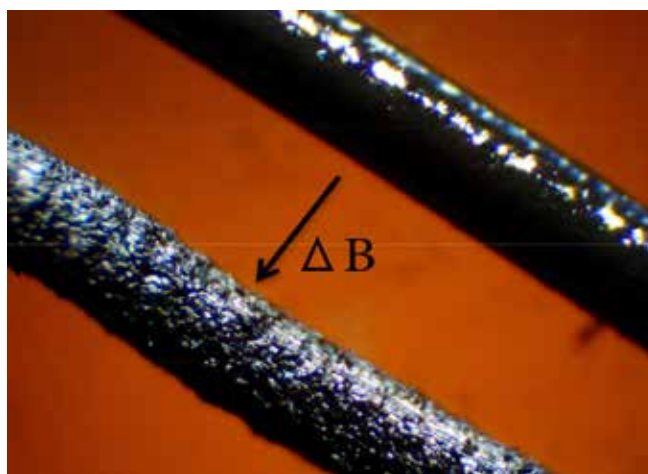
after printing while maintaining conductivity, polymers that demonstrate piezoelectric properties after printing, and polymers that predictively change their conductivity when exposed to chemicals of interest.

We have investigated the relationship between mechanical form and performance for polymer electronics and have optimized the geometry for sensitivity to a handful of chemicals.

We have demonstrated the ability to merge our polymer based chemical sensors into traditional electronic circuits and microprocessors, creating robust sensing systems capable of analyzing vapor and making a chemical assessment of that vapor within 30 seconds. Using this platform, we have demonstrated the ability to speciate between volatile chemicals based on response profiles over time.

Our results are being drafted into a paper for journal publication, and we expect publication to occur early next year.

Our research for the next fiscal year includes focusing on increasing the surface area through changes in material properties to enhance our sensitivity thresholds, functionalizing polymers to react to very specific chemicals of biological interest, and focusing on additional publications and securing follow-on funding.



A change in the magnetic field (ΔB) significantly changes physical and electrical properties in this commercial polymer.

Atomistic View of Solid-Liquid Interfaces Using *In situ* X-ray Probes

Vijay Murugesan

This project is developing a unique imaging capability based on X-ray microscopy that can provide unprecedented chemical imaging of complex interfaces.

Molecular interactions at the heterogeneous interfaces, such as solid-liquid interface, are pivotal to energy storage processes; however, predictive understanding about the impact of solvation phenomena, electrode surface chemistry, and kinetic process on their interfacial interactions is seriously limited. More specifically, a lack of comprehensive understanding about interactions between polysulfide and lithium-(Li-)anode prevents the straightforward tailoring of optimal materials for Li-sulfur (Li-S) battery technology. In this work, we analyzed the interaction between polysulfide and Li-anode during the charge/discharge process using combined theoretical and experimental X-ray photoelectron spectroscopic (XPS) methods.

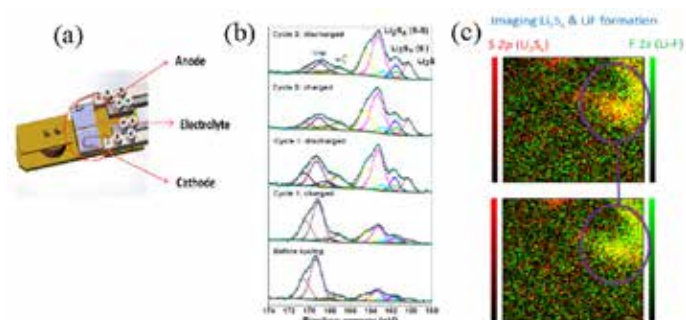
We developed a unique *in situ* XPS and spatial imaging probe that can simultaneously gain spatially resolved chemical imaging, as well as core-level spectra of critical elements. The *in situ* technique is applied to a model Li-S battery system to gain fundamental understanding about the evolution of solid electrolyte interface (SEI) layers on Li-anode during the battery operation.

The *in situ* XPS results show direct evidence of polysulfide shuttling and subsequent reaction with Li-anode during the battery cycling process. This *in situ* technique uniquely reveals the chemical identity and distribution of active participants of parasitic reactions between Li-anode and polysulfide species leading to the fouling process as part of SEI layer formation.

XPS spectra also reveals vital information about the decomposition of counter anion from electrolyte at the anode surface, leading to LiF formation as part of SEI layer

and suggesting reaction between Li^+ cations and decomposed F^- from counter anions. The Li metal anode surface of Li-S batteries suggested the decomposition of TFSI anion (i.e., $[\text{N}(\text{SO}_2\text{CF}_3)_2]^-$) in the liquid electrolytes when it is in contact with Li metal and produces stable lithium fluoride phase as part of SEI layer. The high-resolution XPS also revealed that the TFSI anion can easily decompose to CF_3^- and $\text{CF}_3\text{SO}_2\text{NSO}_2^{2-}$ constituents by breaking the C-S bond, even when electrolyte salt was placed far from the Li metal surface. The $\text{CF}_3\text{SO}_2\text{N}^{2-}$ reduced further by breaking all C-F bonds, followed by the removal of two oxygen atoms, which left CSN , F^- , C^+ , and O^{2-} anions on the Li metal surface completing the TFSI anion decomposition process. The decomposition process can also start with a rupture of the S-N bond, but will result in the same end products on the surface after the salt is completely decomposed.

This *in situ* XPS analysis with spatial chemical imaging capability provides a quantitative view of SEI layer evolution through the polysulfide fouling process and anion decomposition on Li-anode. These new molecular-level insights about SEI layer evolution on Li-anode are crucial to delineating effective strategies for the development of the Li-S battery.



(a) Schematic representation of *in situ* XPS probe, (b) *in situ* high-resolution S2p core-level XPS spectra during Li-S charge/discharge cycles, (c) *in situ* chemical imaging of polysulfide and lithium fluoride fouling process as part of SEI layer formation.

Bridging Length Scales in Complex Oxides: From Point Defects to Defect Superstructures

Peter V. Sushko

This project provides fundamental understanding of defect formation and interconversion mechanisms in multicomponent materials in order to advance their functionality by means of controlled synthesis and processing. Our aim is to reveal how cooperative effects that underpin materials functionalities emerge from the properties of individual building blocks.

Development of fundamental understanding of the roles of heterogeneity, interfaces, and disorder in materials systems represents a transformative opportunity to move from ideal materials to the complexity of real systems under realistic. The overall aim of this project is to advance fundamental understanding of how local inhomogeneity (i.e., deviation from an ideal composition and structure) gives rise to cooperative behaviors that underpin materials functions. In particular, we seek to 1) reveal coupling between zero-dimensional (0D) defects and 1D, 2D, and 3D structures; 2) investigate the effects of defect concentration and spatial distribution using theoretical and computational methods; and 3) identify control parameters that enable utilizing this cross-scale coupling in energy-storage and conversion processes.

For the purpose of this project, we selected materials systems and associated phenomena of ever-increasing complexity. Our results provide mechanistic insights into the origin of their functionalities, optimal synthesis, and processing needed to harness their properties and functions in practical applications.

The high electron mobility of SrTiO_3 (STO) makes it an intriguing candidate for solar applications. However, its usefulness is limited by its large optical band gap (3.2 eV) and propensity to form oxygen vacancies that undermine transport properties. To break through these limitations, we designed a new material: $\text{Sr}_{1-x}\text{La}_x\text{Ti}_{1-x}\text{Cr}_x\text{O}_3$. Our results demonstrated that Cr^{3+} ions at the Ti^{4+} sites lower the band gap by approximately 0.9 eV, while La^{3+} ions at the Sr^{2+} site provide electrostatic charge compensation, thus suppressing oxygen vacancies formation. Published in the American Chemical Society (ACS) journal *Chemistry of Materials*, this work reveals

association mechanisms of intentional defects and suggests a path for imprinting quasi-1D wires in thin films.

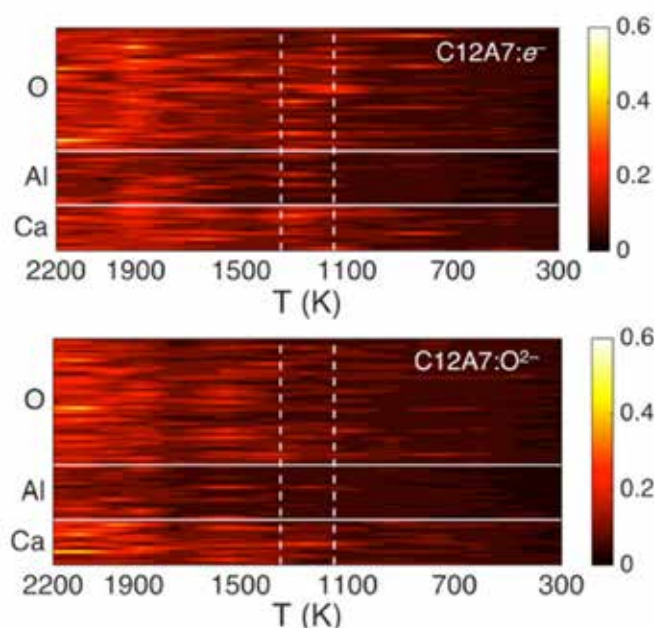
Replacing Li^+ with a bivalent or trivalent ion is a promising route to improving storage capacity and energy density. Mg is an especially attractive alternative to Li due to its large specific capacity, small ionic radius, and high abundance. Transitioning from Li^+ to Mg^{2+} requires new electrode and electrolyte materials. We identified $\beta\text{-SnSb}$ as a promising electrode material and investigated the mechanisms of Mg incorporation and extraction using *ab initio* methods.

Together with the experimental data obtained separately, these results allowed us to create a model for $\beta\text{-SnSb}$ electrode evolution and predict optimal electrode structure. These results, published in ACS's *Nano Letters*, point to nanostructured Sn electrodes in the form of about 30-nm particles as a potentially optimal Sn electrode for Mg-ion batteries.

Novel approaches to efficient NH_3 synthesis at an ambient pressure are actively sought in order to enable production at compact facilities. The key is the development of a high-performance catalyst that enhances dissociation of the $\text{N}\equiv\text{N}$ bond. Prior research demonstrated that nanostructured oxide $[\text{Ca}_{12}\text{Al}_{14}\text{O}_{32}]^{2+} \cdot (2e^-)$, denoted as C12A7:e^- , has a low work-function and can be used as an efficient electron donor. We examined how this property of C12A7:e^- enhances catalytic activity of nanoscale Ru. Using *ab initio* simulations, we established atomic-scale mechanism for how C12A7:e^- promotes electron transfer to N_2 molecules and significantly reduces the dissociation energy of N_2 . Further experimental studies demonstrated that with Ru/ C12A7:e^- catalyst, the rate controlling step of NH_3 synthesis is not N_2 dissociation but the subsequent formation of N-H_n species. Thus, our results, which were published in *Nature Communications*, not only reveal the factors responsible for enhancement of catalytic activity but also redefine the bottleneck step in NH_3 synthesis.

We extended the concept of *excess electron* for catalytic applications to another class of materials: metal hydrides. In particular, in the case of CaH_2 , we found that hydrogen vacancies are readily formed at catalytically relevant temperatures. Importantly, in contrast to other ionic materials, Ca^{2+} ions remain bound at the surface, even at high concentrations of anion vacancies, thus providing confinement potential to the trapped electrons and promoting catalytic function. Using

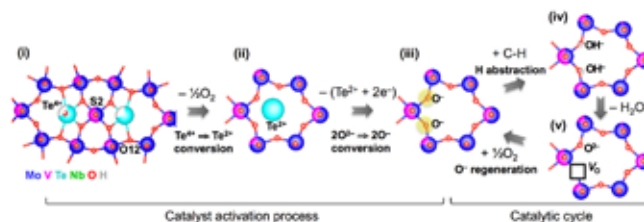
these data, we proposed a class of materials in which excess electrons can be formed without undermining the overall stability of the lattice.



Atomic speed ($\text{\AA}/\text{fs}$) distributions of individual atoms for C12A7: e^- (top) and C12A7: O^{2-} (bottom) during quenching indicates lower T_g in the electron-rich systems. The mobility of Al atoms ceases by $T \sim 1900$ K; Ca and O atoms remain mobile until a substantially lower temperature.

One of the key challenges in materials design is identification of degrees of freedom that ensure robust control of the properties of interest. Small changes in composition of crystalline materials can lead to large changes in their electronic properties such as optical absorption and electrical conductivity. Similarly, small concentration of (co)dopants in amorphous materials can be used to control their optical properties. However, delicate control of viscosity and glass transition temperature in multicomponent materials (T_g) has remained elusive due to the collective origin of these properties. Substitution of atomic anions with electron anions in materials to form electrides introduces an additional degree of freedom. We demonstrate that electrons in $[\text{Ca}_{12}\text{Al}_{14}\text{O}_{32}]^{2+}$ ($2e^-$) dramatically change dynamics of atoms in an inorganic amorphous material, which strongly affects its T_g . Extending the concept of such electron anions to other amorphous materials would provide a powerful instrument for the design of glasses with finely tuned characteristics.

Catalytic function emerges from a synergistic interplay between the catalytic material and its reactive environment. Our modeling of MoVTeNbO $_x$ catalyst revealed that its function emerges from a cooperative process, in which zero-dimensional defects (oxygen vacancies) form under catalytic conditions and trigger a dramatic change of the electronic structure within 1D lattice channels which, in turn, promotes the formation of chemically active O $^-$ species at the 2D material's surface. This realization of a 0D-1D-2D cooperative lattice effect demonstrates the inter-related nature of processes in complex oxide and opens new strategies for designing highly active oxidation catalysts.



Schematic of the cooperative activation step and catalytic function in MoVTeNbO $_x$. (i) Thermal treatment in a reducing environment induces Te^{4+} to Te^{2+} conversion. (ii) Emission of Te^0 results in the formation of chemically active O^- species. (iii) In the presence of alkane molecules, these O^- species abstract H atoms and form surface OH^- . (iv) The OH^- groups recombine and leave the surface. (v) Interaction of the resulting vacancies with O_2 regenerates O^- , providing continuous activation.

In FY 2017, we propose to build on our experience with complex oxides and hydrides and consider phenomena at the interfaces of these two classes of materials. We anticipate that the ability of hydrogen species to switch reversibly between H^- and H^+ states will give rise to new physical and chemical phenomena. This direction of research is consistent with PNNL's broader focus on the science of complex interfaces; it will also help us to mount research efforts on atomic-scale interactions at hetero-interfaces and fundamental origins of functionality.

Bulk Nanostructured Alloy Optimization: Designing for Processing and Thermal Stability

Aashish Rohatgi

This project will help develop the next generation of advanced materials for fuel-efficient transportation and high-efficiency power generation systems. These advanced materials possess a specially designed nanostructure to counter undesirable microstructural changes that otherwise limit the use of traditional metals and alloys at high temperatures.

Nano-sized microstructural features can produce significant improvement in structural and functional properties of materials. For example, nano-sized precipitates, present within a matrix of micron-scale grains, act as obstacles to dislocation motion and form a key basis of strengthening in a wide range of engineering materials such as steels, superalloys, aluminum (Al) and magnesium (Mg) alloys, etc. Even greater strengthening is predicted by the Hall-Petch relation if the grain size itself is also reduced to within a certain nanometer (nm) size range.

In soft magnet materials, both theory and experiments show that a composite of soft iron grains (about 10 nm) embedded in a ferromagnetic amorphous matrix with approximately 2 to 3 nm separation will give the best magnetic performance. Unfortunately, nanostructures are inherently unstable and coarsen rapidly when heated. This coarsening is traditionally countered by a “kinetic” approach, whereby nano-sized precipitates are used to pin the grain boundaries. However, such pinning can be eventually overcome at higher temperatures and/or longer time. Therefore, several research groups have proposed a “thermodynamic” approach that is expected to be more effective than the kinetic approach in preventing coarsening of the nanostructure. This thermodynamic approach relies upon solute atoms to segregate to the grain boundaries and lower their energy, thus reducing/eliminating the driving force behind grain coarsening.

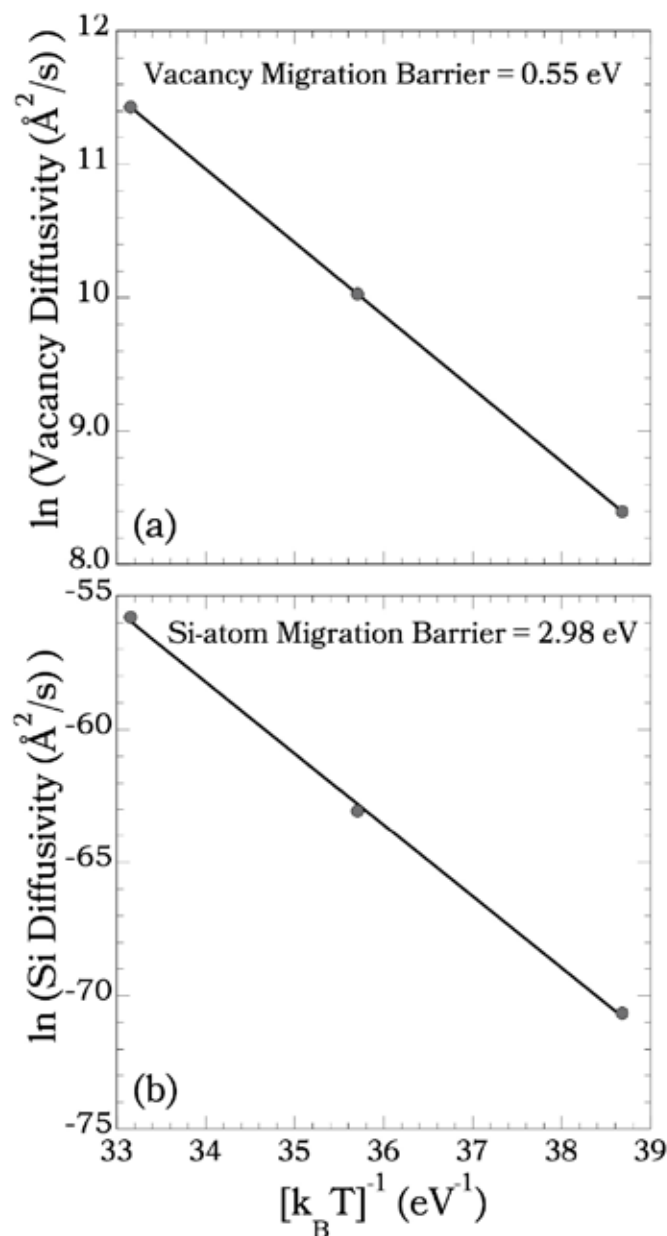
In this project, we will follow the thermodynamic approach and use existing analytical models to identify thermally stable Al-, Mg-, and iron-silicon (Fe-Si) based alloys. We will develop experimental techniques to evaluate the thermal stability of

these alloys and use atomistic modeling techniques for a fundamental understanding of how solute atom-grain boundary interactions lead to thermal stability.

This research will follow an integrated experimental and modeling approach in order to achieve its objectives. We will use existing thermodynamic stabilization models to down-select a list of potential nanostructured alloy candidates that are predicted to be thermally stable based on criteria such as reduction in grain boundary energy, enthalpy of mixing, enthalpy of segregation, etc. Experimentally, we will use magnetron sputtering to create nanostructured thin films of model-predicted Mg-, Al-, and Fe-based compositions. Thermal stability of these nanostructured compositions will be evaluated by subjecting them to elevated temperatures and characterizing the resulting grain growth, solute distribution and diffusion, and precipitate formation. The experiments will be accomplished using analytical techniques such as transmission electron microscopy, atom probe tomography (APT), X-ray diffraction, etc. Finally, we will use atomistic and molecular dynamics simulations to understand interactions between solute atoms and grain boundaries.

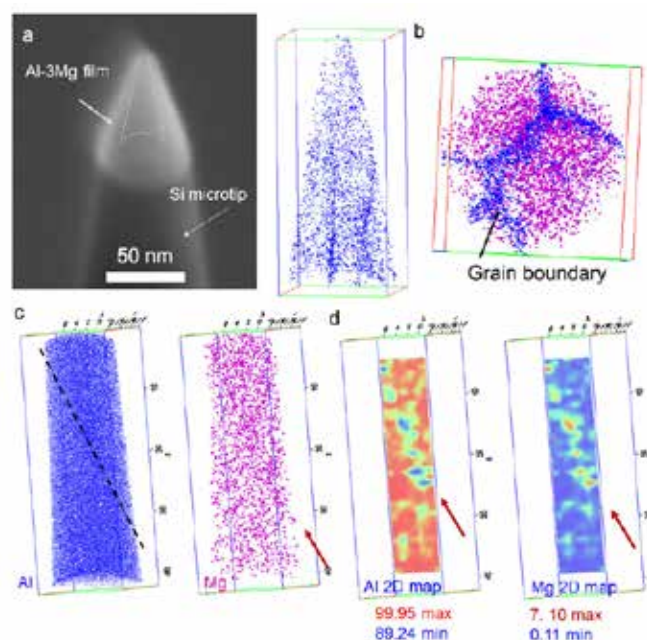
In FY 2016, a lattice Monte Carlo method was used to evaluate the thermal stability of various nanostructured Al- and Mg-based alloys. Software tools were developed to calculate the enthalpy of mixing, enthalpy of segregation, and internal energy of selected alloys, focusing on systems with positive heat-of-mixing. The alloys were plotted on thermodynamic stability maps based on which Al alloys containing Mg solute were predicted to be thermally stable. Guided by these predictions, nanostructured Al-Mg thin films with different Mg contents were fabricated. Initial APT analysis of nanostructured Al-Mg films showed that Mg atoms tend to segregate in small clusters near the grain boundaries, although continuous segregation of Mg atoms along the Al grain boundaries was not observed. We used density-functional-theory- (DFT)-calculated formation energies to derive pairwise interaction energies of Fe-Fe, Si-Si, Si-Fe, Fe-vacancy, and Si-vacancy pairs. These pairwise interaction energies were then used in an atomistic model to calculate activation energies for various vacancy-atom exchange processes and, hence, allowed us to determine the diffusivities of vacancy and Si atom in a Fe lattice.

In FY 2017, the thermodynamic modeling approach from FY 2016 will be modified and extended to investigate thermal stability in Al- and Mg-based alloys with negative heat-of-mixing. Since thermal stabilization ideally requires the solute atoms to be distributed uniformly along grain boundaries, the potential of low-temperature annealing to enable Mg atom clusters to diffuse and distribute uniformly along grain boundaries in Al-Mg films will be evaluated. Grain growth kinetics in Al-Mg films will be determined and compared to baseline pure Al to validate the model predictions that Mg segregation can, indeed, stabilize Al against grain growth.



Arrhenius plot of diffusivities of (a) vacancy (b) Si atom in a Fe lattice, where k_B is the Boltzmann's constant and T is the absolute temperature in Kelvin.

Analogous to Al-based systems, Mg-based alloy films (e.g., Mg-yttrium (Y)) will be fabricated and analyzed for solute segregation and grain growth kinetics to evaluate the thermal stabilization potential of such solutes. Considering that grain boundaries in polycrystalline Al-, Mg-, and Fe-based alloys comprise a distribution of energy configurations, atomistic modeling will study how the grain boundary character influences the interactions between solute atoms and the grain boundary. Finally, the diffusion of Si in Fe matrix at various solute (i.e., Si) concentrations will be determined to understand temperature-driven microstructural phenomena in high-Si Fe-Si alloys



Analysis of grain boundary segregation in Al-Mg thin film experimentally using the APT technique. The 3D elemental maps generated by APT along (b and c) with 2D concentration maps (d) are uniquely capable of identifying and quantitatively analyzing the extent of segregation of elements (e.g., Mg in Al matrix), which can then be used to validate the computational modeling results.

Carbon Rods with Unexpected Humidity-Driven Water Expulsion

David J. Heldebrant

This project focused on the predictive science of synthesis, with the aim of understanding the structure and interaction of water at interfaces of nanomaterials to control the adsorption/desorption and transport of water.

Studies were carried out on recently discovered, iron-rich, carbon-based nanorods that adsorb water at low humidity and spontaneously expel half of the adsorbed water at high relative humidity (RH).

During FY 2016, we strived to gain mechanistic insight into this unique water expulsion at high humidity by studying the rod's geometry and surface chemistry to enable larger scale synthesis of the rods or develop a template for the synthesis of other nanomaterials. We studied the surface and internal composition of the rods and how they affect the interaction of the rods with water and impact the onset of evaporation (desorption). We used advanced microscopy and spectroscopic techniques to characterize the interfacial and surface chemistry before and after exposure to water.

During FY 2016, we also investigated the effect of rod synthesis temperature and amount of catalyst on the onset of evaporation and studied different synthetic strategies to increase water adsorption/expulsion capacity by achieving control over and understanding surface chemistry.

We discovered that there is potential for these rods to collect water from a lower RH gas and then desorb it into a higher RH

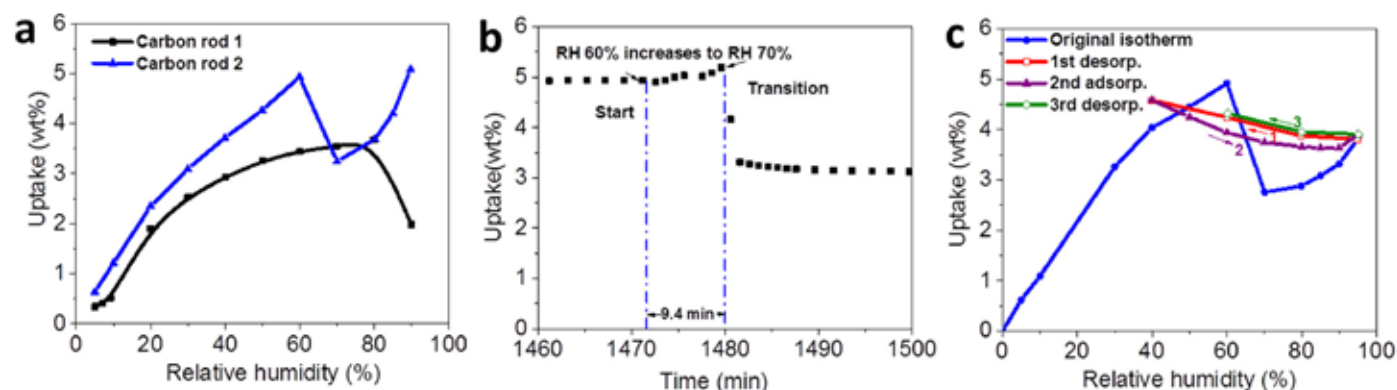
gas with little or no temperature swing, which opens the door for unique water harvesting or dehumidification devices.

Our current understanding is that the response to a change in RH shifts the equilibrium water pressure primarily through a change in the heat of adsorption. Exposure to high RH is predicted to result in an endothermic shift in the material, while exposure to low RH will result in an exothermic rearrangement. These shifts will increase the heat dissipation required during adsorption and heat adsorption required during desorption. Temperature swings beyond that required to achieve acceptable rates of heat transfer do not appear to be necessary.

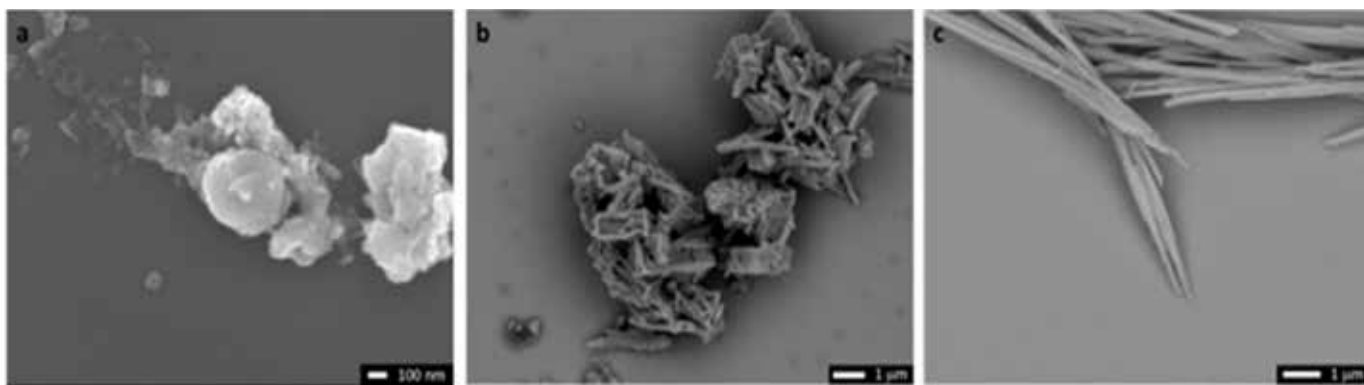
Under these auspices, water appears to be moving against its chemical potential (from low RH to high RH), so the additional heat flow or other features in the cycle must offset the Gibbs energy of mixing.

This preliminary thermodynamic analysis provides clues into how the material may behave but still does not get at the nature of the cost to drive this process in the observed direction. Ultimately, an improved understanding of the nature of the energetics of the inter-particle interactions that result in the nanorods changing their spacing distance is needed to design viable devices for water harvesting. Collecting this data is the focus of next year's work.

During FY 2016, a patent application was filed, and four manuscripts were published in peer-reviewed journals; a fifth manuscript is being drafted, as well.



(a) Water adsorption isotherm at 25°C. (b) Water adsorption kinetic data during transition process for the carbon nanorod obtained using 0.2 M FeCl₃ solution.



Iron-rich, carbon-based materials synthesized at different temperatures: (a) at 180°C, (b) 200°C, and (c) 230°C.

Combining Isotopic Measurements Using *In Situ* Liquid SIMS and *In Situ* TEM to Determine the Mechanism and Kinetics of Li Ion Mobility in SEI Layers

Zihua Zhu

We are using the unique capabilities developed at PNNL, including in situ liquid secondary ion mass spectrometry (SIMS) and in situ transmission electron microscope (TEM), to resolve critical questions regarding how Lithium-ion (Li-ion) migrates through the solid-electrolyte interphase (SEI) under electric field, as well as to provide a quantitative measure of its mobility.

Li-ion batteries are now indispensably used as energy storage devices for portable electronics and electric vehicles and are starting to enter the market of the renewable energies. The rechargeable capacity and the battery life depend critically on the structural stability of the electrodes themselves, the electrolyte degradation rate, and the electrode-electrolyte interaction layer: the SEI. Processes that occur at the SEI in batteries are critical to battery lifetime and performance, but their details remain elusive because of the difficulty of examining the interface during battery operation. Although electron microscopy and magnetic resonance approaches have provided new insights, isotopic and time-resolved measurements using our novel *in situ* SIMS in combination with *in situ* TEM offer the potential to provide an unprecedented level of detailed information about mechanistic and dynamic processes. Finding answers to these critical questions will enable further advancement on the designing of rechargeable Li-ion batteries for enhanced performance.

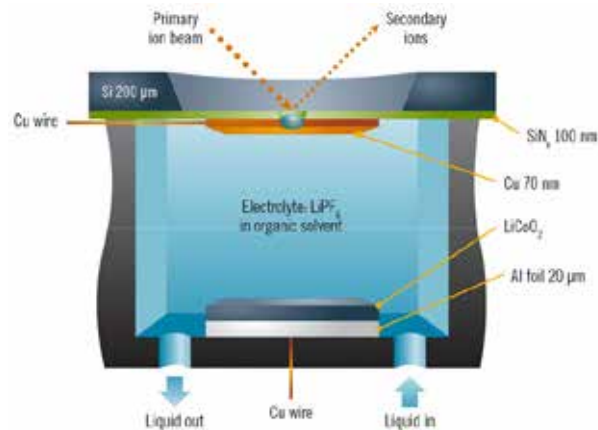
In this research, our effort will focus on using *in situ* SIMS and *in situ* TEM to resolve the following two important issues of SEI: 1) How does Li^+ move through SEI? 2) What is Li^+ diffusivity and conductivity through SEI?

To answer these two questions, we need to elucidate the formation mechanism of the SEI layer first. Actually, so far, many basic questions about SEI layer *in operando* are still not clear, despite previous numerous efforts. Critical questions include, but are not limited to: when does an SEI layer start to form?

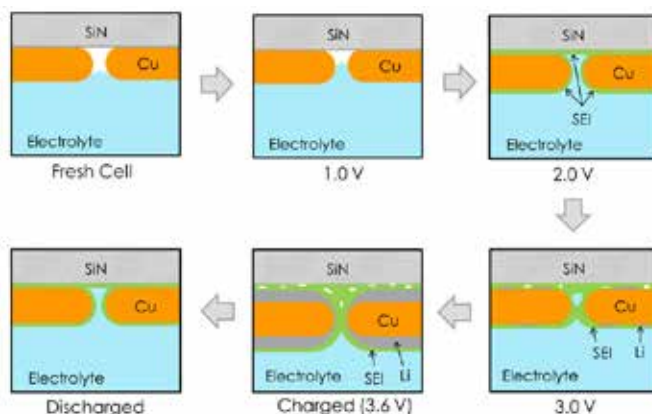
When does Li metal start to deposit on the anode surface? What is the thickness of the SEI layer at operational condition? What are major chemical components in the SEI layer?

During FY 2016 (the first year of this project), we began by improving the design of our model battery and optimized the SIMS measurement conditions, leading to filing a U.S. patent and publication of two papers (*Chemical Communications* and *Journal of American Society of Mass Spectrometry*). More encouragingly, the DOE Office of Science highlighted our *in situ* liquid SIMS-SEI work.

Thereafter, we focused on searching answers to the above



A schematic illustration of the design of the model battery used for *in situ* liquid SIMS study of Li^+ ion transport mechanism in the SEI layer.



An illustration of the SEI formation mechanism based on our *in situ* liquid SIMS data.

four questions related to the formation mechanism of the SEI layer. The model system is composed of a Cu metal anode with lithium bis(fluorosulfonyl)imide-Dimethoxyethane (LiFSI-DME) as an electrolyte, which is a very promising candidate in the next generation of Li-ion batteries.

Our results show that the SEI layer starts to form at about 1.5 V and almost fully forms at 2.0 V. At 3.0 V, a small amount of Li metal already deposits on the Cu electrode surface. Upon charging to 3.6 V, the Li metal layer continuously forms on the Cu electrode surface. After discharging, the Li metal is stripped from the electrode surface, but the SEI layer still stays on the Cu surface. A very interesting observation is that the components and thickness of SEI after discharging are very similar to those of the SEI layer upon charging to 2.0 V, indicating the SEI layer almost fully forms at 2.0 V.

In addition, the thickness of the SEI layer is about 10 to 15 nm *in operando*. More interestingly, Li oxide and Li hydroxide are found to be the major components in the SEI layer, but the concentration of Li fluoride is unexpectedly low. Our

computer simulation results show that the low concentration of fluorine in the SEI layer can be attributed to the formation of an electrical double layer at the electrode-electrolyte interface. The above findings provide key insights for designing the next generation of Li-ion batteries.

In FY 2017 (the second year of this project), we plan to organize the above data and publish them in top-level journals. At the same time, we will combine isotopic labelling techniques and *in situ* liquid SIMS to study the Li⁺ transport mechanism in the SEI layer. In brief, an SEI layer will be prepared with a natural abundant LiClO₄-DME electrolyte. Following a stable SEI layer formation, the natural abundance LiClO₄-DME electrolyte will be replaced by a ⁶Li-enriched electrolyte (⁶LiClO₄). After one or more charging-discharging cycles, *in situ* liquid SIMS will be used to examine the change of the Li isotopic ratio in the SEI layer to determine the mechanism of Li⁺ ion transport through SEI and, therefore, the Li⁺ diffusivity and conductivity through SEI.

Commercially Viable Slow-Release Solid Nitride Fertilizer

Josef Matyáš

A low-cost, slow-release, solid nitride fertilizer is being developed that would improve fertilizer-use efficiency and significantly decrease the greenhouse gas footprint of nitrogen (N) fertilizers. The specific goal of this project was to develop and demonstrate a steady N-release rate for solid nitrides synthesized from realistic feedstocks such as fly ash and kaolin.

A variety of slow-release fertilizers, typically ammonium salts or urea encased in porous polymer film or sulfur-coated, have been employed to deliver nitrogen to growing plants and to extend or delay release of this nitrogen. To be commercially viable, slow-release fertilizers must release more than 80% of their nutrients at a steady rate over the course of a crop growing season, which is typically 12 to 16 weeks in length. Our previous work with aluminum nitride (AlN) mixed with N-release additives successfully demonstrated steady near-linear release of more than 80% of the N during a 16-week period. Since AlN is also a major solid nitride in the mixture synthesized by carbothermal nitridation of fly ash or kaolin, the same approach was extended to consider nitrides prepared from these realistic industrial feedstocks. The synthesized nitrides were blended with additives that facilitate the desired release rate of the N, with a goal to demonstrate rates and extents of N-release that are similar to those obtained for AlN.

A raw, low-grade kaolin from a leading producer in Georgia and a Class-C fly ash from a midwestern power station that burned Powder River Basin coal were nitrided for 12 hours at 1,400°C under 1.5 L min⁻¹ of N₂ at atmospheric pressure before being mixed with additives designed to control the rate of N-release and then transformed to mechanically robust solid nitride fertilizer granules for an efficient release of N. These granules were immersed in water and tested for the rates of N-release at 18.5 and 28°C. Initially, two preliminary short-term (2-week) N-release tests were conducted to optimize the concentration of N-release additives. This was followed by a long-term (16-week) test to investigate the effect of milling the synthesized nitrides

to increase reactive surface area before mixing with N-release additives and making the fertilizer granules.

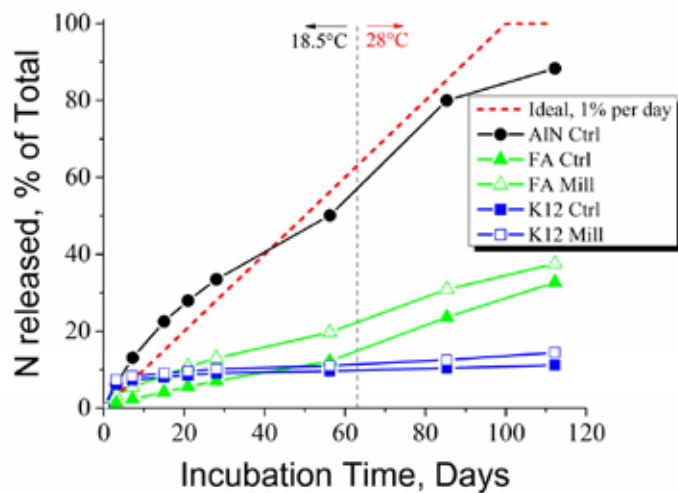
The total N content of the nitrided kaolin and fly ash samples ranged from 16 to 26 mass percent and averaged 21 mass percent. After mixing with additives, the N content in the fertilizer granules averaged 11 mass percent.

The two most important criteria for slow-release fertilizers are the rate and degree of N-release. An ideal fertilizer would release at a steady, near-linear rate of about 1% of the initial total N per day until full depletion. In our long-term experiment, a near-ideal rate (about 0.9% per day over 56 days) was achieved with AlN when mixed with the N-release additives. N-release rates for unmilled nitrided kaolin and fly ash fertilizer were about 0.17 and 0.22% per day, respectively, over 56 days. Milling significantly increased the N-release rate of the fly ash fertilizer to 0.35% per day. This strong response to milling highlights the importance of high reactive surface area in improving overall N-release rate and suggests that N-release rate can be adjusted as needed during the fertilizer-formulation process. An equally strong response to milling was not seen for kaolin fertilizer, however, with N-release rate increasing only to about 0.20% per day.

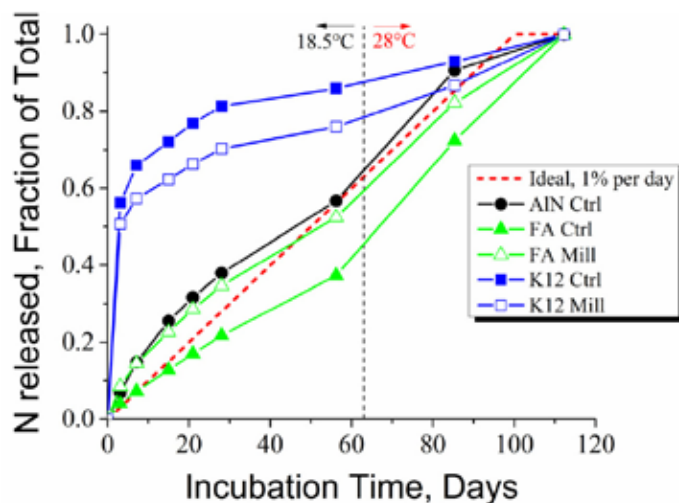
The linearity of N-release is also a critical parameter. In the long-term experiment, the fly ash and AlN fertilizers showed highly linear N-release during the first 56 days. The fertilizer produced from kaolin, however, showed a rapid initial release during the first few days and then much slower, albeit linear, release thereafter. It is hypothesized that boron (present as an impurity in kaolin) precipitated on the nitride surfaces thus decreasing the rate of N-release.

In the long-term experiment, the temperature in the incubator was increased from 18.5°C to 28°C to simulate the rise in temperature of the soil during a crop season. This temperature increase further improved the overall linearity of the N-release and increased the N-release rate by about 60%.

The project achieved its primary objective of demonstrating a steady linear N-release by a solid nitride fertilizer prepared from a realistic feedstock (fly ash) over the length of a typical growing season. The results for the nitrided kaolin suggest that further study to determine the cause of the nonlinear release is warranted.



Nitrogen-release curves for fertilizers prepared from nitrided fly ash (FA) and kaolin (K12). Nitrided FA and K12 milled prior to making fertilizer granules are indicated by "Mill." Unmilled FA, K12, and AIN are indicated by "Ctrl." The dashed line shows the calculated ideal N-release rate. N-release values are % of total N initially present.



Normalized N-release curves for nitrided FA and K12 fertilizers and calculated ideal N-release rate. N-release values are fraction of total N released during experiment.

Development of Aluminum Alloys with Improved Strength at Elevated Temperatures

Saumyadeep Jana

This project aims to develop Al alloys with improved elevated temperature strength through L_{12} phase precipitation. Al alloys that can withstand high temperatures are of particular interest to the automotive industry, as it means improved fuel efficiency through weight reduction, implementation of newer combustion techniques, and more.

Over the last three decades, research has been primarily focused on development of dispersion-strengthened Al alloys for improved elevated temperature strength. However, the dispersion strengthening intermetallic phases have incoherent interface with α -Al matrix. Design of an Al-alloy with cubic L_{12} precipitates can push the high temperature limit of Al alloys ($0.45 T_m$) to that observed in Ni-base superalloys ($0.75 T_m$) through 1) presence of a coherent interface with the Al-matrix, and 2) resistance to coarsening at elevated temperatures.

The main aim of this project is to demonstrate formation of metastable L_{12} structured Al_3M precipitates in an Al matrix through careful selection of alloy chemistry.

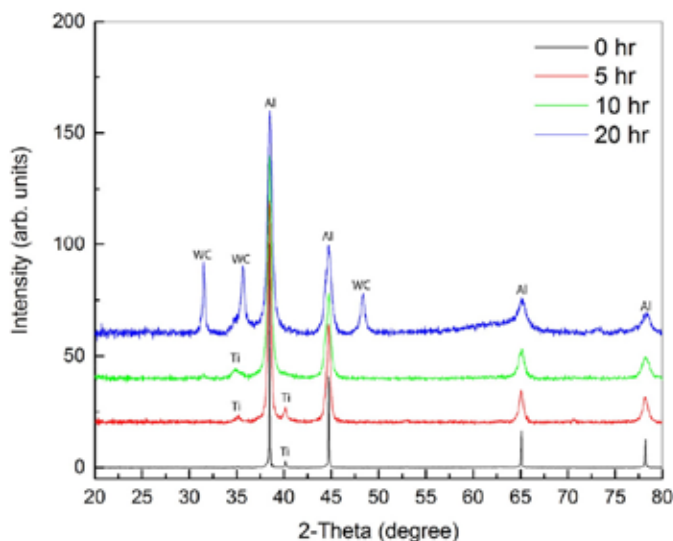
The current study focuses on Al-Ti system. Al_3Ti has a stable DO_{22} structure. However, addition of minor transition series alloying elements is known to improve the metastability of cubic L_{12} structured Al_3Ti phase. Therefore, we have selected Cr as a minor alloying element for stabilization of L_{12} structure. Ball milling has been used for alloy synthesis. Mechanical alloying in Al-Ti-Cr system has been attempted using 1) SPEX mill, and 2) planetary mill. Ball milled powder has been extensively characterized using X-ray powder diffraction (XRD) and scanning electron microscopy (SEM). Differential scanning calorimetry (DSC) analysis has been carried out to understand the thermal behavior. Crystal structure of the precipitating phase has been determined through transmission electron microscopy (TEM) imaging and XRD.

XRD analysis of the ball milled powder shows that mechanical alloying is occurring in Al-Ti-Cr system when using SPEX

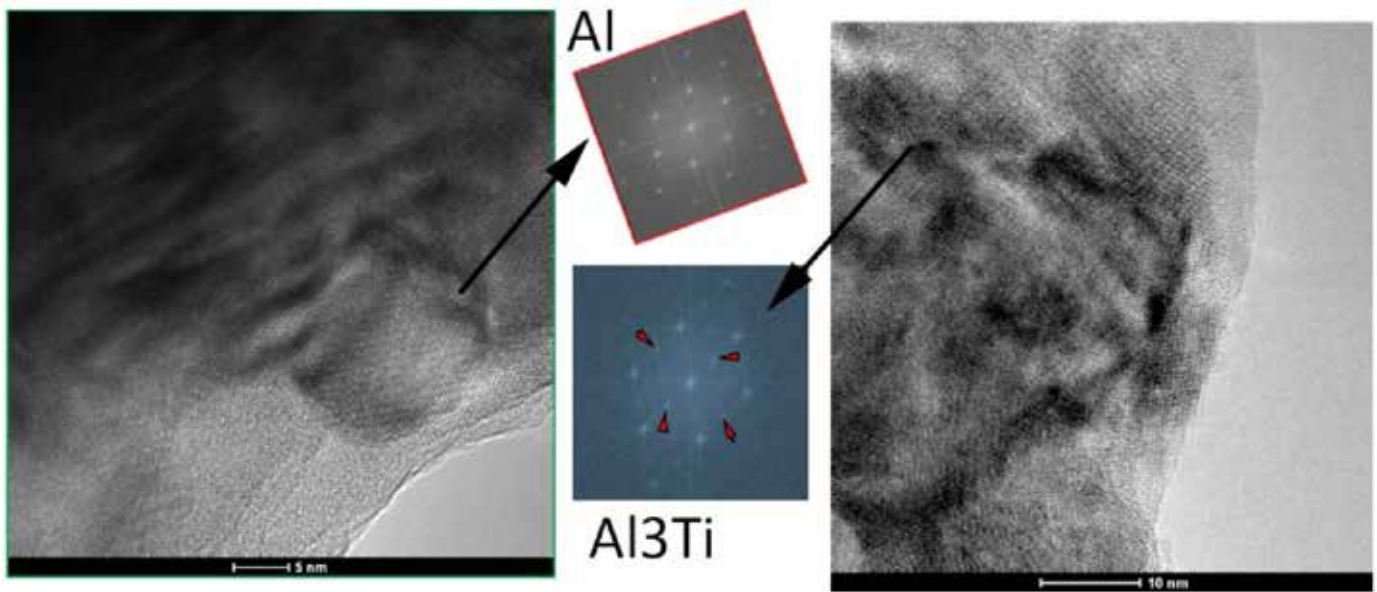
mill equipment. Characteristic Ti peak intensity reduced as a function of milling time and was completely absent after 20 hours of milling, which has been confirmed for three Al-Ti-Cr compositions. However, mechanical alloying did not take place even after 80 hours of planetary ball milling. SEM images show a homogeneous Al-matrix enriched with Ti from SPEX mill runs, whereas planetary mill run results in significant refinement of Ti particles that remained homogeneously distributed as numerous bright particles within Al-matrix. WC has been used as milling media in SPEX mill runs, and leads to powder contamination. WC contamination has been controlled by stopping the milling process at an intermediate stage and loading fresh WC balls. The effect of lower ball to powder ratio has also been studied.

Mechanically alloyed Al-Ti-Cr powder has been heat treated inside DSC to learn about the thermal behavior. A broad exothermic peak at 380°C has been noticed for all three mechanically alloyed Al-Ti-Cr compositions that indicates precipitation event.

In conclusion, this study shows that it is possible to generate cubic L_{12} Al_3Ti precipitates in mechanically alloyed Al-Ti-Cr systems. Further heat treatment studies are required at this stage to learn more about various microstructural features.



Progress of mechanical alloying in Al-Ti-Cr system as a function of milling time using SPEX mill.



HR-TEM image of L1₂ Al₃Ti precipitates in Al-matrix. Short red arrows indicate superlattice reflections associated with ordered L1₂ structure. Nanocomposite Al-Al₃Ti microstructure is forming.

Development of Hierarchical Porous Structured Materials for Energy Storage Applications

Luis Estevez

In this project, we are developing novel porous materials with tailorable properties. This ability to tune the structure/property relationship in these materials will allow us to target the particular prerequisites of certain electrodes for energy storage applications—specifically lithium ion battery anodes and supercapacitor electrodes.

The United States transportation sector is overwhelmingly powered by the internal combustion engine (ICE). This results in roughly a quarter of our total energy use being in the form of petroleum. Not only does the use of this fossil fuel have detrimental environmental effects, but it is, coincidentally, similar to the amount of petroleum we import as a country every year. Thus, there has been a great demand for a move towards an electric vehicle (EV) powered fleet, particularly as petroleum becomes scarcer. A major impediment to this goal is the fact that the current energy storage technology of choice for EVs, the lithium ion battery (LIB), is roughly an order of magnitude away in terms of energy density, and at least that in terms of power density.

In order to bridge these gaps, this project will advance two distinct energy storage technologies that can be utilized in an EV (potentially even together in a hybrid EV): supercapacitors and silicon-based LIBs.

Supercapacitors (SCs) are one of the few energy storage technologies that can compete with the very high power densities present in an ICE (typically about 1 to 10 kW/kg for either system). Unfortunately, the most common SCs, electric double layer capacitors (EDLCs), can usually only muster energy densities at least two orders of magnitude below ICEs (typically 1 to 10 Wh/kg for SCs and more than 1,000 Wh/kg for an ICE). Since the mechanism for energy storage in an EDLC is the adsorption of the ions present in the electrolyte onto the electrode surface, high surface area electrodes are typically employed with specific surface area (SSA) values typically greater than 1,500 m²/g. The most common SC electrodes are usually activated carbons that are electrically conductive and meet the large surface area prerequisite through an extensive network of small, nanometer-sized

(typically less than 5 nm) pores. The problem is that these commercially activated carbons typically have a ramified porous structure that can give rise to kinetic barriers for faster moving ions, reducing the power density of the SC in order to increase the energy density.

In our project we have synthesized a hierarchical porous carbon (HPC) materials platform with controllable porosity in the three porosity length scales (as designated by International Union of Pure and Applied Chemistry): micropores (less than 2 nm), mesopores (2–50 nm) and macropores (greater than 50 nm), by utilizing three different templating mechanisms: CO₂ activation, hard colloidal templating and ice templating (respectively). This has enabled our HPC materials to have an interconnected vascular-like network of larger macro/mesopores leading into smaller micropores. This open structure will enable the HPCs to be maximized for both power density and energy density by using the interconnected mesoporous network to mitigate kinetic barriers for the conduction of the ions to the high surface area sorption sites. Additionally, this project has exploited the ability to synthesize both extensive micropores and larger meso/macropores in order to fabricate HPCs with both high SSA and large amounts of void space—typically measured as pore volume. These large pore volumes contain the void space necessary for the loading of transition metal oxides into the larger mesopores, which add additional pseudocapacitance to the EDLC based capacitance from the high SSA micropores. Finally, this project has also explored alternative HPC materials that are more graphitic, but still have the same tunable morphology, combined with large SSA and pore volume characteristics.

By utilizing the triple templating synthesis strategy described previously, we have synthesized HPC materials with varying surface areas, pore volumes, and pore size distributions. Highlights include HPC materials with a very large SSA simultaneously combined with colossally large pore volumes—textural characteristics that are usually exclusive to each other, due to the different pore size requirements for each. For large SSA, smaller pores (less than 5 nm) are usually required, whereas for large pore volume values, larger mesopores—on the order of tens of nm—are typically employed. For mesoporous carbons, pore volumes of over 5 cm³/g are considered a high value and quite rare. Rarer still are porous

carbons that combine these large pore volumes (greater than $5 \text{ cm}^3/\text{g}$) with large SSA values (typically greater than $2,000 \text{ m}^2/\text{g}$). In the synthesis of our HPC materials, we can control the morphology to include both porous regimes, and thus, we have synthesized HPCs with high SSA of greater than $2,800 \text{ m}^2/\text{g}$ (as measured via Brunauer–Emmett–Teller) and pore volumes of about $10 \text{ cm}^3/\text{g}$ altogether in one material. As of this time, we cannot find any nanoporous carbons in the literature with the combination of these values. Attempts to fabricate these extremely porous HPCs into electrodes for EDLCs have proven difficult due to their outstanding textural properties, but samples have been sent to collaborators at Florida State University for testing as lithium ion capacitors (LICs). Our HPC material showed promising initial results, and the cyclic performance was excellent, with very little capacity fading.

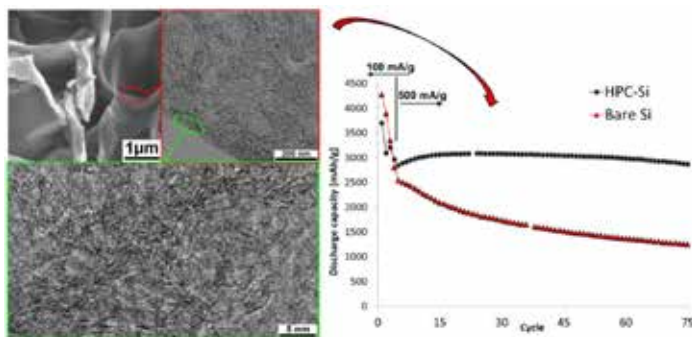
Currently, we have also been optimizing the synthesis of graphitic HPCs (denoted as HPC-Gs). We have synthesized HPC-G materials with a superior graphitic structure that was verified via high resolution transmission electron microscopy (TEM), as well as x-ray powder diffraction.

While SCs can help bridge the ICE gap in terms of specific power, employing a silicon-based anode in an LIB is one way to bridge the gap in terms of energy density. Silicon has over 10 times the theoretical capacity of graphite (about $4,200$ and 350 mAh/g , respectively), but the major impediment to its use as a battery electrode is the fact that the Si anode pulverizes and breaks apart during charge/discharge cycling.

Initial results in attaching the HPSiO_2 and core/shell nanoparticles (CSNPs) to the graphene oxide nanoplatelets look promising, but we have focused this last fiscal year on the work utilizing a functionalized HPC-Si composite anode. Our results consistently demonstrate the HPC-Si composite anode maintains more of its initial discharge capacity compared to the Si nanoparticle-based cells (without the functionalized HPC added) as the cells cycle. Typical results are approximately 75% capacity retention for the HPC-Si cells versus about 35% capacity retention for the bare Si cells after 100 cycles.

For this project, we have achieved unprecedented control of the HPC morphology, resulting in an excellent material for supercapacitor electrodes. We are currently preparing a manuscript based on the HPC supercapacitor project and anticipate submission near the start of the new fiscal year. For the HPC-Si composite LIB anode work, we have successfully shown how a small amount of HPC can be utilized to mitigate the capacity loss present in electrodes fabricated with bare silicon. We are currently finishing up our work with the Si LIB anode project and expect to start preparing a manuscript based on this work.

Additionally, our expertise in the functionalization of porous materials led to a collaboration involving vanadium redox flow batteries that were being finished at the end of the last fiscal year. This resulted in the work being published in the journal *ChemSusChem*. Additionally, we have leveraged our ability to tune our HPC materials into other potential applications via collaborators both internally and externally. We anticipate working within these collaborations into the next fiscal year while using the LIB anode effort and supercapacitor work as an excellent platform for concentrating on LICs.



Results showing the HPC-Si composite anode maintaining more of its initial discharge capacity as the cells cycle, compared to the Si nanoparticle-based cells without the functionalized HPC added.

Durable and Stretchable Materials for Protective Equipment and Coatings

Curtis J. Larimer

This project developed flexible and stretchable liquid-repellent coatings for applications in healthcare and emergency response. Their implementation has the potential to drastically reduce the incidence of infection in healthcare settings.

Personal protective equipment (PPE) is essential for the safety of healthcare workers and first responders. However, PPE can serve as a vector for disease transmission when infectious material sticks to the surface and is transferred from one ill patient to another or to other surfaces in the environment. Coatings can be used to improve performance and comfort of PPE by imbuing the materials with antimicrobial and nonstick properties. Biomimetic materials based on the structure and chemistry of the lotus leaf have extreme water repellent properties, but to date, these materials have proven too fragile for many practical applications.

With this project, we aimed to develop and demonstrate a durable, flexible, and stretchable lotus-inspired superhydrophobic coating that was inexpensive, nontoxic, and easy to apply. When applied to PPE, the coating could prevent the adhesion and transfer of infectious liquids such as blood and other bodily fluids. Instead of spreading over the surface, these liquids have the tendency to form ball-shaped droplets that have the tendency to roll off the surface.

It is the micro and nanostructure of the coating that causes air to be trapped under surface liquids and reduces the contact area of the liquid with the material by two to three orders of magnitude. In previous studies, microstructured materials were limited by a low durability to mechanical abrasion and by an inability to yield to mechanical deformation (i.e., flexion and stretching). We developed a novel, nontoxic coating that integrated a super-repellant surface with a pliable and stretchable base (e.g., latex and nitrile). This had the synergistic effect of also improving the coating's durability to abrasion.

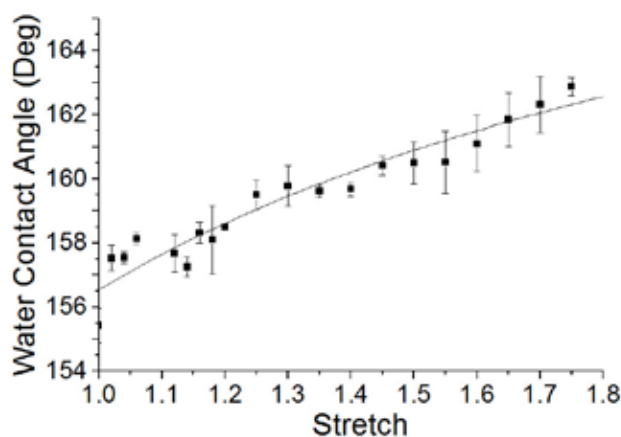
This project undertook a systematic study to characterize and better understand the physical wetting behavior of stretching superhydrophobic materials. We discovered a non-intuitive property of these coatings: the material became more hydrophobic as it was stretched. We studied the microstructure of the stretched coating with electron microscopy and interferometric imaging and observed a transition from nanostructure

to a hierarchical micro and nanostructure. We then developed a theoretical model of this phenomenon. The model predicts a wetting state that was previously undiscovered.

We then validated this theoretical model with experimental data. The combination of novel material design and processing with a newly discovered physical phenomenon led to disclosure of an invention and filing of a provisional patent. Potential commercial applications in healthcare, industry, and emergency response have spurred interest from external parties.

Finally, we conducted a study to demonstrate the ability to prevent contamination by an infectious liquid. We exposed test and control materials to cultures of bacteria and spores and compared the amount of residual that adhered to each sample. Results indicated a significant reduction in bacterial adhesion. The results suggest that the material would effectively reduce transfer in a hospital setting.

Future work on this coating technology could include continued exploration of the unique wetting states that were observed. We also aim to explore concepts for new coating methods that will be easier to control, more cost effective, and produce materials with better performance. Additionally, we aim to continue to optimize the formulation of the coating. Finally, thorough biological testing with a broader range of relevant infectious microbes will demonstrate the advantages using this new coating for PPE and other commercial applications.



This project developed a new material with extreme liquid repellency. The material becomes more repellent (higher water contact angle) as it is stretched to nearly double the original length. The material could be used as a durable coating that minimizes transfer of infectious liquids.

Free Space Transistors for Advancing the Art of Software-Defined Radio

Ethan D. Farquhar

The objective of this proposed work is to collaboratively acquire expertise in the design, fabrication, and use of Free-Space Field-Effect Transistors (FS-FET) for the dual purposes of advancing the science of new nano-technological devices and applying these advances to various national security application spaces such as the further miniaturization of software-defined radio systems, as well as harsh-environment sensing systems.

The invention of the transistor and the subsequent miniaturization and integration of these devices has driven revolutionary changes to society over the course of the past generation. Quite possibly, the invention of this device has yielded the largest creation of wealth ever. However, these devices have speed limits that cannot be exceeded. Because of these limitations, researchers have been looking to technologies other than traditional silicon semiconductor processes to develop ever-faster and lower-power processing.

In standard complementary metal-oxide-semiconductor (CMOS) transistors, electrons are able to flow from the source terminal to the drain terminal, with the current being modulated by the field from the gate terminal. The electrons, though, are forced to conduct through the material between the source and the drain. Interacting with the crystal lattice along this path impedes the progress of this charge carrier, thus slowing it down and limiting the speed (frequency) at which this transistor can operate. This interaction limits conduction velocities in standard silicon CMOS to the order of 1×10^7 cm/s.

In contrast to standard CMOS, free-space transistors show the promise to run possibly as much as three orders of magnitude faster than traditional CMOS systems. This is due to the fact that electrons are ballistically transported through the channel instead of through a resistive channel. Also, due to this configuration, there is a potential for greater power gain than with a standard MOSFET.

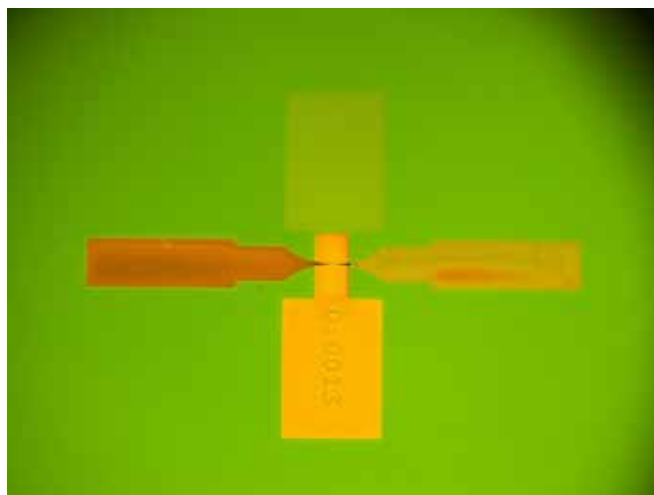
Early devices fabricated and described by Meyyappan have demonstrated cutoff frequencies for the FS-FETs as high as 450 GHz. This frequency is high enough to give coverage over much of the radio frequency spectrum and is even of relevance in the millimeter wave band (30–300 GHz).

While progress on this particular project has been challenged by pushing the capabilities of our facilities, there have been several accomplishments and prospective application spaces that make this work more and more exciting.

One of the accomplishments is the ongoing collaboration with Dr. Meyya Meyyappan from NASA Ames Research Center, who has written many books on nanotechnology and carbon nanotubes and is considered a leader in the field.

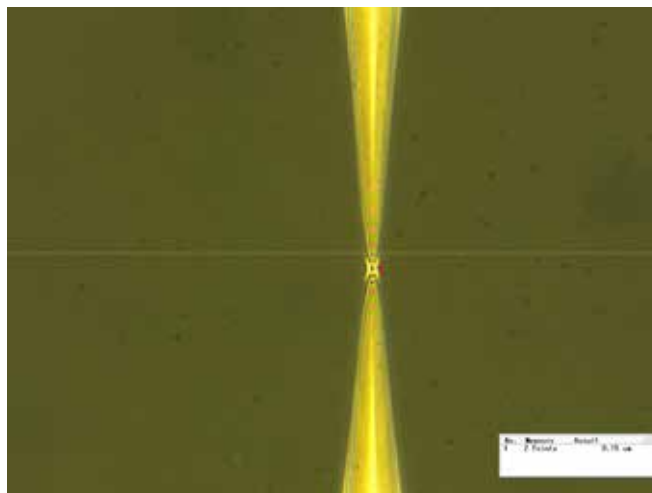
Also, we have pushed the science of what was previously seen to be the boundaries of what is possible in the EMSL micro-fluidic fabrication facility. For example, we are the first to ever sputter coat such things as tungsten and silicon-nitride within our facility.

In addition, we have fabricated our first devices with gaps between them of approximately 150 nm. These devices are incredibly new, and we are not awaiting bonding to these devices to begin measurements. We expect these measurements to begin with the new fiscal year. Most excitingly, new ideas are being generated that specifically relate to the national security mission of our directorate.



The two points comprising the collector-emitter region of the VFET. This magnification appears to show a 150 nm gap.

Lastly, we have new and innovative thoughts regarding ways to generate that gap in the first place, and we expect to begin that work early in the new fiscal year. This fresh approach promises even greater control over the size of the gaps we are building, and if successful, we expect to produce new areas for publication.



Our first full device; this shows the collector-emitter terminals surrounded by both a top gate and a bottom gate.

Fundamental Insights into Gamma-Radiation Effects at Complex Oxide-Water Interfaces from First-Principles Simulations

Sebastien N. Kerisit

This project will utilize state-of-the-art computational techniques to provide molecular-scale insights into the effects of radiation fields on the reactivity of mineral phases relevant to the processing of radioactive waste.

Aluminum is a major component of high-level waste (HLW) sludge stored in underground tanks at the Hanford Site and is principally present as aluminum oxyhydroxide phases, with gibbsite and boehmite as the most abundant phases. Aluminum is an undesirable component of the waste stream, as large concentrations of aluminum are detrimental to glass formation and, thus, HLW immobilization. Therefore, gibbsite and boehmite will be dissolved by caustic leaching prior to vitrification.

An important unknown is the effect of the radiation field present in the tank wastes on the dissolution behavior of aluminum-bearing solid phases during HLW processing. Can the presence of a radiation field explain the unexpected behavior of aluminum oxyhydroxide phases in tank waste? Indeed, the abundance and properties of boehmite in actual tank waste samples fall outside simple estimates based on knowledge from the aluminum mining industry. Further, boehmite from tank waste has been found to be more resistant to dissolution in caustic solutions than commercially available material, hinting at radiation hardening of this phase.

Finally, enhancement of radiolytic production of dihydrogen gas (relative to pure water) has been observed at boehmite surfaces but not at chemically similar gibbsite, indicating significant differences in the response of these solvated surfaces to the presence of a radiation field. This project, therefore, focuses on boehmite and gibbsite as model systems and aims to develop a fundamental understanding, from first-principles simulations, of the reactivity of these phases in the presence of a γ -ray field to elucidate the elementary interfacial processes that give rise to observed differences in surface reactivity.

This fiscal year, the initial focus of this project was to determine the structure and energetics of boehmite and gibbsite surfaces in aqueous conditions as a baseline for studying the effects of the presence of a radiation field. Extensive first-principles simulations were performed using density functional theory (DFT) to characterize the structure and energetics of low-index boehmite and gibbsite surfaces, both in the absence and presence of water. A survey of exchange-correlation functionals was undertaken to identify the best-suited level of approximation and quantify the associated uncertainties. The simulations yielded predictions for surface energies that enabled the determination of morphologies for boehmite and gibbsite particles. The calculated morphologies were in good agreement with those observed experimentally using electron microscopy. The approach was also validated by comparing the predicted surface energies against experimental calorimetric data published in the literature.

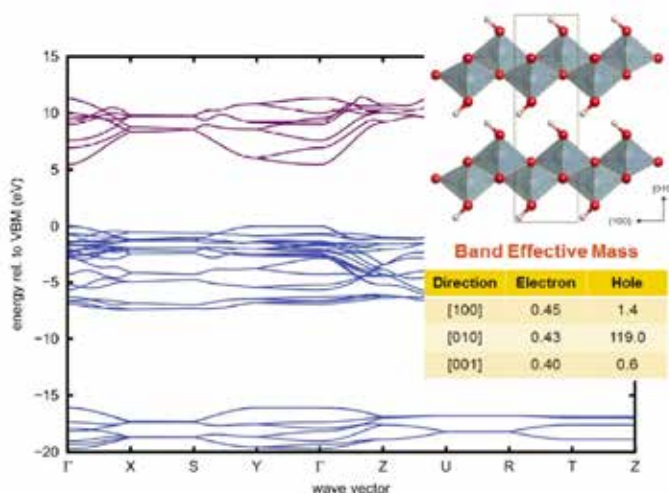
In addition, the ionic strength can be particularly high in Hanford tanks, and traditional aluminum oxyhydroxide hydrothermal synthesis routes used in this project to generate new materials usually also involve high ionic strength solutions. It is, therefore, important to understand what role electrolyte ions might play in determining interfacial structure and energetics (e.g., surface energies and particle morphologies).

In FY 2016, we performed a series of simulations to understand the interactions of concentrated NaNO_3 solutions with boehmite surfaces. Published classical force field parameters were validated against DFT calculations for Na^+ adsorbed as inner-sphere and outer-sphere complexes at the boehmite (010) surface, which dominates boehmite morphology. The validated force fields were then used in large-scale classical molecular dynamics (MD) simulations of 2 M NaNO_3 solutions in contact with several boehmite surfaces. The MD simulations revealed extensive Na^+ adsorption at the surfaces, which significantly affected the surface energies relative to pure water, showing that high ionic strength may indeed affect particle morphology.

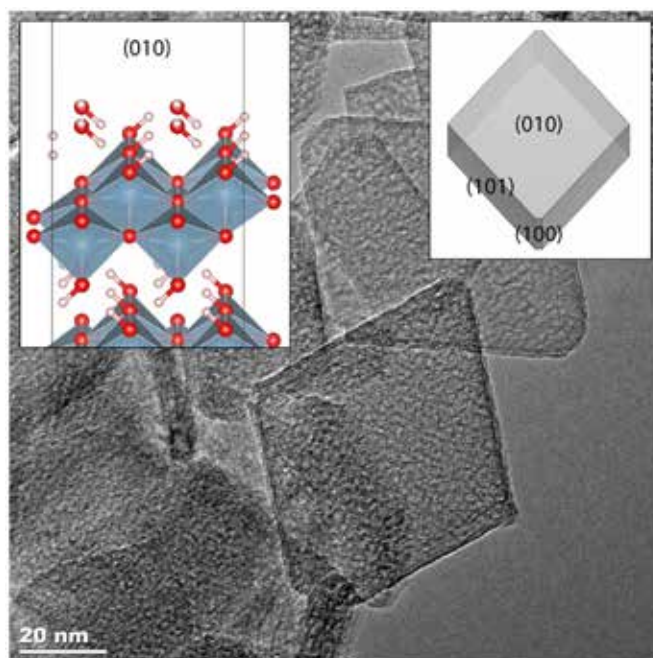
This fiscal year, we also synthesized boehmite particles, aiming to measure their bandgap energies with electron energy-loss spectroscopy. Initial results on a small number of particles showed bandgap energies in the vicinity of 7 eV, in closer agreement with our calculations. Further experiments are being planned to refine these measurements and look at the effect of particle size. We speculate that the published bandgap energies obtained from UV measurements might have been probing gap states due to the smaller energy range used in these experiments.

Previous work showed that radiolytic activity of oxide particles appeared to peak at 5 eV when plotting H₂ yield as a function of bandgap energy. However, these measurements were carried out uniquely with oxides and not oxyhydroxide or hydroxide phases. Additionally, some oxide phases with a bandgap energy in the vicinity of 5 eV did not show radiolytic enhancement. We conclude that radiolytic activity is likely not a simple function of the bandgap energy. Indeed, the bandgap energy of gibbsite is very similar to that of boehmite for all levels of approximation considered; yet, the two mineral phases show very different radiolytic activities experimentally. Analysis of the electronic band structures obtained from our first-principles calculations shows that, for both mineral phases, the band mobility of electrons is direction independent, whereas holes transport much more slowly across hydrogen-bonded layers (i.e., along stacking direction). This means that holes generated in the bulk phase by γ -rays might concentrate at edge surfaces. We will, therefore, explore whether the nature and/or the proportion of edge surfaces can account for the known differences in radiolytic activity between the two aluminum phases.

In addition to the planned activities already mentioned, we will focus our efforts in FY 2017 on exploring an alternate reaction pathway for radiation enhancement of interfacial reactivity, which involves radiolysis of interfacial water molecules, followed by reaction of radiolysis products with surface functional groups.



Boehmite electronic band structure. Inset shows the boehmite unit cell and band effective masses of electrons and holes in the three directions derived from the band structure. Holes are much less mobile along stacking direction, whereas electron mobility is direction independent.



Transmission electron microscopy image of boehmite particle synthesized via hydrothermal method. Left inset shows water adsorption at (010) surface from first-principles calculation and right inset shows the predicted particle morphology based on first-principles surface energy calculations.

Fundamental Investigations of Photoelectrochemical Water Splitting of Model Oxide Electrode Surfaces

Kelsey A. Stoerzinger

We are developing oxide-based heterostructures that effectively absorb the solar spectrum and efficiently utilize solar-generated carriers to generate chemical fuels, such as hydrogen and oxygen gas, from water splitting.

Harvesting renewable solar energy is a tantalizing solution to addressing concerns of energy security and global warming. However, storage of this intermittent energy source poses a significant challenge to grid integration and even use in remote applications.

One approach is utilizing the sun's energy to form chemical fuels through water splitting. The efficiency of this process exists in driving the hydrogen and oxygen evolution reactions via electron-hole pair generation, propagation to the electrode surface (without recombination), and electrochemical conversion. This can only be done through specially designed and synthesized solids with optical and electronic properties that facilitate these processes with a high degree of efficiency. Metal oxides constitute ideal aqueous interface layers because they are much more stable in solution than traditional semiconductors. Our goal is to fabricate oxide-based heterostructures that absorb well in the visible portion of the solar spectrum, have low defect densities (to minimize recombination and carrier trapping), and exhibit band bending and band alignments that facilitate carrier transport to and through the electrode surface to the aqueous medium. The outcome will be an important new set of design principles, along with useful materials, for photoelectrochemical water splitting.

This project employs oxide molecular beam epitaxy (MBE) systems and a suite of characterization tools available in the Environmental Molecular Sciences Laboratory to design and fabricate well-defined oxide heterostructures. We will measure functional properties, including photoconductivity and photoelectrochemistry, to assess suitability for solar water splitting applications. Mechanistic investigations of the interaction of these surfaces with aqueous solutions are carried out via ambient pressure X-ray photoelectron spectroscopy (APXPS) at the Lawrence Berkeley National Lab Advanced Light Source (ALS).

Work is ongoing to establish a testing methodology of oxide thin films and heterostructures thereof in an inert atmosphere, providing unprecedented control over surface preparation and its characterization following photoelectrolysis. A custom photoelectrochemical cell has been designed and fabricated to electrically contact and chemically isolate oxide MBE films for measurement. After measurement in a nitrogen box, the sample can then be directly transferred back into vacuum for characterization of the surface species present, eliminating the potential for contamination from ambient atmosphere. In the next fiscal year, this testing methodology will be validated and employed to measure the photoelectrochemical activity of novel oxide heterostructures for water splitting.

Initial work has also begun on mechanistic investigations of the water/oxide surface interaction. Using APXPS at the ALS, we have probed the speciation resulting from interaction of oxides with water and electronic band bending at the surface. The absolute core-level binding energies measured with the surface saturated in water vapor yield valuable information of the effect that the aqueous environment and the associated adsorbates have on the presence and strength of built-in potentials in the near-surface region of the oxide. Likewise, the detailed line shapes, particular those for oxygen, reveal

which species are present at the surface. Future combination of this information with photoelectrochemical reaction rates in the upcoming fiscal year will facilitate realistic models of light-induced electrolytic activity at these surfaces.



Photograph of custom-designed photoelectrochemical cell for measurement of oxide heterostructures grown by MBE.

PN1610512882

Fundamental Mechanisms of Nucleation and Growth of Particles in Solution

Marcel D. Baer

This work will provide the necessary theoretical framework and simulation tools that enable an understanding of the microscopic principles of nucleation, growth, and self-assembly.

Inorganic polymers form a large class of materials ranging from sol-gel networks to metastable zeolites and hybrid metal-organic framework materials. The synthesis and use of these highly engineered systems has evolved over the past 100 years from “black magic” into a technical art. The next step—the one that approaches modern science—requires the continued development and application of a gamut of synthetic methods, illustrative model systems, characterization techniques, and interpretation via computation and simulation that can bridge from quantum mechanics through fluid and structural mechanics.

We want to understand the growth kinetics of size-selective molecular clusters that form the secondary building blocks of polymeric inorganic networks in a joint experimental and theoretical effort. Thus, our focus in this project is on the use of molecular models to mimic and understand the underlying principles of how surface assembly of inorganic oxide supports their related surface-grafted organometallic fragments. To this end, this project is developing new research capabilities for the simulation of reactivity and structure in complex heterogeneous and homogeneous environments, establishing a theoretical framework, and using simulation tools to understand the microscopic principles of nucleation.

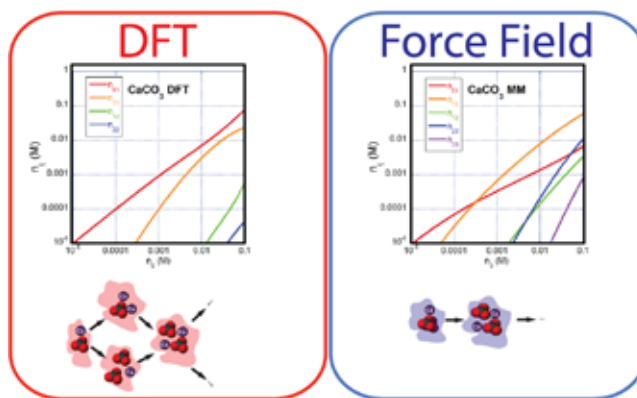
In FY 2015, we continued with the experimental focus of understanding how silica particles nucleate and grow from monomeric silane species. We used a solution-based acid catalyzed hydrolysis reaction to grow octameric polyhedral oligomeric silsesquioxane starting from a monomeric $R^+Si(OR)_3$. We monitored the reaction progress as a function of temperature and pH during assembly, utilizing mass spectroscopy and solution phase ^{29}Si nuclear magnetic resonance to identify intermediate oligomers that were formed during the process. Our results showed that we formed significant amounts of tetrameric species with small amounts of octameric, decameric, and dodecameric species; however, almost no dimeric species were seen during the growth process.

A new project was started to test the hypothesis that, according to the theories of nucleation, a population of transient

alkali halide clusters may exist on the mica surface prior to nucleation. We used fast-scanning atomic-force microscopy to investigate the existence and behavior of such small alkali halide clusters, ideally characterizing both their populations and lifetimes. Because the concentration of such clusters is directly related to their free energy of formation, this approach can provide an alternative route establishing the free energy landscape for nucleation. Thus far, the controlled growth of microscopic crystals was shown for RbI on mica.

The theoretical effort in FY 2015 was focused on ion-pairing as the first step of nucleation and growth for calcium carbonate in the high dilution limit. We used state-of-the-art *ab initio* density functional theory (DFT) and classical force field representations of interaction to obtain condensed-phase potential of mean forces (PMFs). This process allowed us to coarse-grain the ion-ion interaction to study the nucleation of small clusters and their free-energetics using dynamical nucleation theory.

In FY 2016, we developed the capability to image the precursors to nucleation using *in situ* atomic-force microscopy with molecular resolution, demonstrating the ability to image the formation of aluminum hydroxide films from molecular scale clusters on mica. Preliminary results suggest that non-classical nucleation pathways, such as barrier-free coalescence may occur. Results using the Aggregation Volume Bias Monte Carlo (AVBMC) method show significant differences for the coarse grained models based on classical force fields and DFT for the early stages of nucleation of $CaCO_3$. Differences in the PMFs lead to very different pathways for the formation of small clusters. The classical force field shows $CaCO_3$ as a “building block,” even for low concentrations, whereas the DFT interaction potentials show the free ions as dominant species.



AVBMC using coarse grained potentials to effectively sample free-energies of clusters formation in the condensed phase and show pronounced differences in nucleation pathways, depending on the concentration condensed phase.

High Aspect Ratio Functional Composites for Thermal Optical Applications

David M. Reed

We are developing novel composite structures that will reversibly change from transparent (or any underlying color) to opaque in a targeted temperature range. Innovative design strategies are employed to fabricate inexpensive functional materials for applications in the energy management of buildings, vehicles, and consumer markets.

Over the past few decades, a great amount of effort has been made to improve the energy management of buildings, vehicles, and products in consumer markets. We are developing and demonstrating composite coatings, when applied to a product, can change its reflectivity in the visible and near infrared (IR) spectra as the temperature changes, thus enabling it to act as an active cooling or heating component with environmental changes. Composite membranes have been fabricated with durable and low-cost materials that change their reflectivity by utilizing the temperature-dependent, intrinsic properties of at least two phases in the composite.

Composite materials have found numerous applications in various technology markets, because their collective properties are often unique and not found in single-phase materials. The diameter, morphology, volume fractions, and connectivity of phases within the composite materials are often used to tailor the desired properties. Transparent structural composites (i.e., glass fiber loaded polymers) have found limited applications because of their tendency to have a very narrow temperature window in which they are transparent. With changes in temperature, these composites become translucent due to the refractive index (n) mismatch that is accompanied by interfacial light scattering. The refractive index variation with temperature (dn/dT) of polymeric materials is often one to two orders of magnitude higher than inorganic glasses. This difference in the dn/dT determines the rate of change with temperature of the mismatch in the refractive indices and controls the temperature-dependent transmission of the composite.

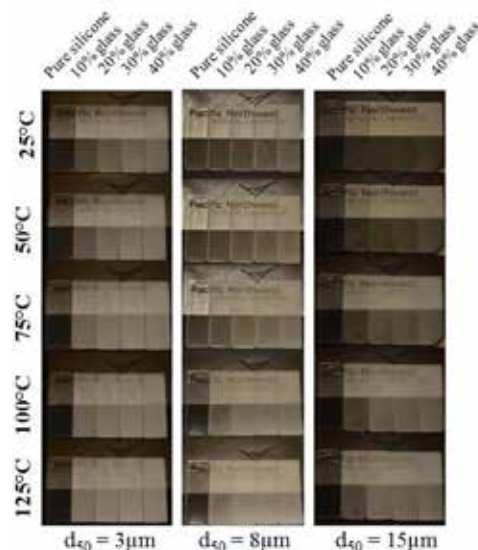
We have developed composite materials that transform from a transparent region (or any underlying color) to opaque over a desired temperature range by careful selection of the refractive index (n) and temperature dependence (dn/dT) of the filler and matrix materials, as outlined above. Proper selection of material chemistry, refractive index, dn/dT , filler particle size, and loading were initially chosen via modeling, followed by experimental verification. As a first approximation, modeling efforts focused on using Rayleigh-Gans light scattering theory for a two component system with small differences in refractive indices. The refractive indices of a number of poly-

mer systems (i.e., epoxies, thermoplastics, silicones, etc.) and inorganic glasses were measured as a function of temperature.

Silicones were the focus in FY 2016, because they have the greatest dn/dT , have superior stability and durability, are easy to process, and have excellent weather-resistant properties. In general, the silicones studied had dn/dT values in the range of -5×10^{-4} , whereas the inorganic glasses had dn/dT values of about $+10^{-6}$. A number of silicones with various glass powder fillers were prepared into 1-mm-thick, free standing films. The silicone and glass powders were combined in a centrifugal mill to ensure proper dispersion and mixing, followed by de-airing to remove trapped air. The samples were then cast and cured into 1-mm-thick films, and the composite films were characterized using UV-vis-IR spectroscopy to understand the influence of particle size, volume fraction, dn/dT of constituents, etc., on the final transmission of the composites as a function of temperature.

Samples were placed on an aluminum block, on which the top half has the PNNL logo inscribed in black and the bottom half is painted solid black. The samples were exposed to various temperatures, at which point a photo was taken. All pure silicone samples remained transparent over all temperature ranges, whereas samples loaded with glass filler showed different responses depending on the particle size and volume fraction loading.

The ability of these smart membranes to change their reflectivity with temperature can render the composite either an active cooling or heating component due to environmental changes to minimize the energy load. The reversible transition from transparent to opaque can be tailored to meet a specific application by utilizing the material properties of the matrix and filler such as the refractive index (n), thermo-optic coefficient (dn/dT), filler particle size, filler volume fraction, and thickness of the composite film.



This is an example of a composite consisting of a silicone and a glass powder with different volume fractions and particles sizes exposed to various temperatures.

High-Information Content Polymers and Their Assembly into Structural Motifs

Chun-Long Chen

We are developing high-information content polymers to understand their self-assembly into higher order structural motifs, an ability that will ultimately enable biomimetic functional materials.

High-information content polymers will mimic the ability of proteins and peptides to exhibit secondary and tertiary structure, as well as self-assemble into oligomeric complexes and extended ordered matrices. The self-assembled materials developed in this project can perform functions beyond protein- and peptide-based materials. This work will lead to an understanding of the assembly and thermodynamic and kinetic controls on those pathways, the rates at which assembly occurs, and the degree of order in the final structures. In short, we seek to create a scalable, robust approach to synthetic high-information content polymers with sophisticated functions; use powerful methods of investigating self-assembly to develop a fundamental understanding of assembly in the synthetic systems; and integrate polymer synthesis, characterization, and theoretical modeling to create a true predictive science of synthesis for high-information content polymeric materials.

In FY 2014, we designed and synthesized two types of sequence-defined polymer sequences for self-assembly of highly ordered materials. We tuned the peptoid self-assembly chemical characteristics rationally, and we observed that a slight change of sequence design caused dramatic differences in polymer self-assembly. The first designed class of synthetic polymers contained an architecture that did not involve peptide bonds. Using R-group substituted monomers, we performed batch synthesis of homopolymers, solution-phase synthesis by addition of one monomer at a time using deprotection steps, and a solid-phase method using a submonomer approach to create polymers by extending the chain a fraction of a monomer at time. Our other modeled class of synthetic sequence-defined polymers informed experimental designs structurally. In the simulations, it was determined that imine sidechains increased helical propensity and chiral linkers increased conformational order.

In FY 2015, we synthesized over 10 three-fold peptoids and investigated their self-assembly into porous networks. Adding reversible chemical bonds into the design, we demonstrated that self-assembled peptoid materials exhibited reversible structural changes in responding to pH. We also synthesized over 20 membrane-forming peptoids and demonstrated that attaching functional groups at arbitrary locations in the peptoid sequence leaves the basic membrane structure intact. Under dry conditions, atomic force microscopy (AFM) results show that these free-standing peptoid membranes exhibit a thickness of 3.5–4.0 nm, and analyses via X-ray diffraction (XRD) determined that they are highly crystalline. Transmission electron microscopy (TEM) results showed that they also contain well-aligned strips of flexible carboxylate groups. Based on these AFM, XRD, and TEM results, we generated a proposed structural model for peptoid membranes. Molecular dynamics were used to confirm the model and understand the interactions leading to a robust peptoid assembly. Simulated structures showed extensive π -stacking in the hydrophobic region and disorder in the carboxyl tails. In addition, simulated XRD data is in good agreement with the experimental data, and the formation of well-aligned strips of disordered hydrophilic domains agree well with TEM data. Molecular simulation further confirmed that peptoids packed anisotropically to form membranes.

By exposing peptoid membranes to a range of solvents and high temperature, we demonstrated that they are highly stable. For example, *in situ* AFM studies showed that no significant disruption was observed when they were placed in a mixture of water and organic solvents, or even in pure CH_3CN and EtOH for over 6 hours. Similar to lipid-bilayer membranes, *in situ* AFM results demonstrated that peptoid membrane exhibited salt-induced thickness changes. By exposing the materials to NaCl solution or phosphate buffered saline buffers of increasing concentration, we observed thickness changes from about 4.2–5.4 nm. We also demonstrated that peptoid membranes could coat surfaces in single-layers through a two-step heterogeneous nucleation process. Moreover, they exhibited anisotropic self-repair.

During FY 2016, we designed and synthesized membrane-forming peptoids containing functional groups that include fluorescent dansyl (DNS) dye and cyclodextrin. We further used *in situ* AFM to systematically investigate and characterize the self-repair and patterning of two-dimensional membranes like peptoid materials, we demonstrated that functional groups could be introduced and patterned within peptoid membranes through membrane repairing process and by co-crystallization approach. By changing the side-chain chemistry of hydrophobic domains of membrane-forming peptoids, and varying the charge, length, and chemistry of polar domains of membrane-forming peptoids,

both experimental and simulation results showed that the interactions between peptoid hydrophobic domains drove the formation and stabilization of peptoid membranes. Based on that conclusion, we further designed over 30 amphiphilic peptoids that assemble into nanotubes and demonstrated that simple variation of peptoid side chains can be used to tune the structure of peptoid assemblies. We further used AFM, TEM, and XRD to demonstrate that peptoid assembly pathway and the thermodynamic and kinetic parameters could be tuned by changing peptoid side-chain chemistry. Based on the work mainly supported by this project, we published several manuscripts in FY 2016.

High-Resolution and 3D Imaging of Nanomaterials

Ilke Arslan

Advances in technology are often linked to advances in methods for characterization. This project aims to advance characterization methods to provide input for the rational design of next-generation materials.

We are advancing and combining nanoscale techniques to further complex materials examination. This year, we have developed two new, exciting methods in scanning transmission electron microscopy (STEM) to gain new understanding of materials and synthesis that has not been observed before: 1) *in situ* synthesis in the liquid phase at elevated temperatures (up to 100°C) applied to zeolite synthesis, and 2) 3D chemical imaging by combining electron tomography with energy dispersive x-ray (EDX) spectroscopy in the aberration-corrected STEM to understand the 3D composition of a complex set of materials called interplanetary dust particles (IDPs), which are collected in the upper atmosphere.

Catalysis with zeolites is a multi-billion dollar global investment in the petrochemical industry every year. For decades, zeolites have been synthesized with “secret recipes” by oil companies with minimal understanding of the fundamental formation mechanisms. The initial stages of formation are, of course, on the atomic to nanoscale and not easily accessible at the temperatures and pressures of standard synthesis. Now, through a recent release of heating capability in liquid up to 100°C by the holder company Protochips, we have been able to do the first experiments on *in situ* zeolite synthesis.

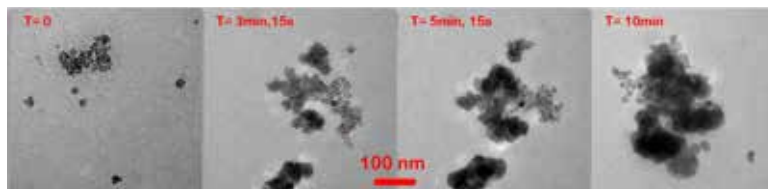
Zeolite Y was chosen as the first experiment for the following reasons: the lower temperature and pressure of zeolite formation compared to other zeolites (70°C), fast formation, no organics involved in the synthesis, and the ability to correlate with other *in situ* methods such as *in situ* nuclear magnetic resonance and x-ray absorption fine structure. The zeolite solution, or “gel,” is composed of an aqueous solution with approximately 10% NaOH, making it a very challenging experiment, as the solution will become more corrosive at higher temperatures.

There are two hypotheses as to how the zeolite can form. The first is that there are amorphous but solid nanoparticles that undergo a process of dissolution and reformation into the crystalline phase to form a zeolite, with the Al coming from solution. The second hypothesis is that the zeolite particles form out of solution directly into a crystalline phase. Our results

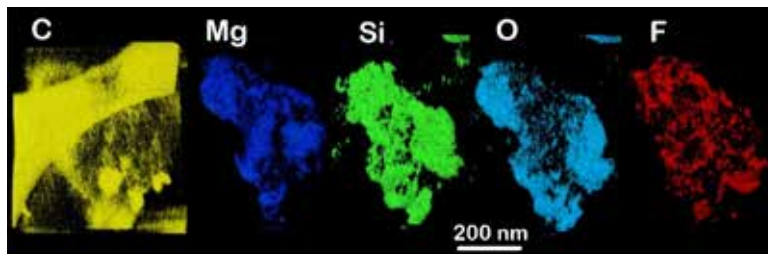
indicate that the zeolite formation follows the latter process in which the particles grow out of solution. The silica solid particles maintain their size, shape, and distance from each other. These results have implications for the future of zeolite synthesis and the potential to impact many technologies.

The second method that was developed is EDX tomography to obtain 3D chemical information on the nanoscale. Several recent and planned space missions have returned or will return samples of extraterrestrial bodies that include a fine-grained fraction. The fine-grained fraction often is the most sensitive recorder of environment and processing, and thus is critical to a full understanding of the formation and evolution of the parent body, which in this case can provide information on the formation of the solar system itself. Our initial results show that these materials are very complex with many elements present. The distribution is unexpected and nanoscale, making EDX tomography the only method capable of extracting this information. The 3D spatial resolution with chemical sensitivity is currently about 10 nm. With our continued work on the acquisition parameters and reconstruction algorithms, we expect to reach 5 nm and below. This method will be applicable to any complex material system.

The results of both sets of work are under analysis for publication. These technique developments, together with the developments in 3D reconstruction algorithms made in FY 2015, are moving us towards our goal of realizing *in situ* 3D chemical imaging, an essential need for the characterization and rational design of next-generation energy materials.



Time lapse of *in situ* zeolite synthesis at 70°C in NaOH/H₂O solution. The zeolite synthesis occurs through growth out of solution, rather than dissolution of amorphous silica particles that then recrystallize with Al. The scale bar is the same for all images.



First results of an IDP using EDX tomography. The 3D distributions of five different elements have been reconstructed and are shown separately for clarity. The scale bar is the same for all elements.

Ion Implantation and Characterization of Epsilon Metal Phase Formation in Ceria

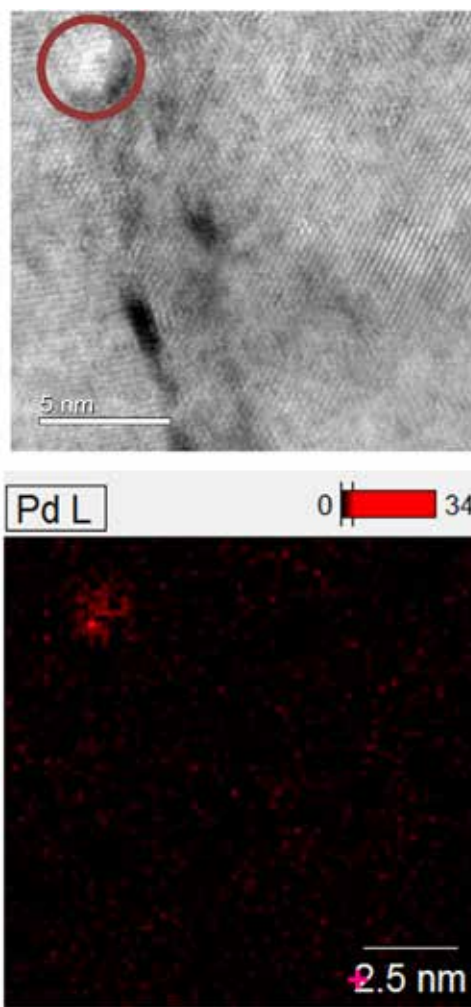
Ram Devanathan

This project integrates synthesis, ion irradiation, microscopy, and modeling to develop fundamental understanding of fission products in irradiated nuclear fuel. This research will improve the economics of nuclear power by enabling high burnup of fuel and identify signatures of nuclear material processing.

The mechanism of formation of metallic phases and their morphological evolution are key knowledge gaps in our understanding of nuclear fuels. The phase diagram of this metallic system is five-dimensional, and the reactor conditions under which the phases form are far from equilibrium. However, our understanding is limited to a small region of phase space under equilibrium conditions. Efforts are currently underway at PNNL and at external institutions to perform careful irradiation of nuclear fuel under controlled conditions in a test reactor, recover metallic phases that may be present, and characterize their structure and chemistry. Due to the long duration, cost, and radiological concerns associated with fuel irradiation, as well as the complex physical processes in nuclear fuel, targeted ion bombardment studies of a surrogate material can add tremendous value to reactor-based studies. This project will test the following hypotheses: 1) defects produced by fission fragment damage enhance the migration of ϵ -phase metals; and 2) metals diffuse to grain boundaries, cracks, and pores from surrounding regions. Our work will reveal the conditions under which metallic phases form in irradiated fuel.

We performed pulsed laser deposition of CeO_2 (surrogate for nuclear fuel UO_2) doped with Mo, Re, Ru, Rh, and Pd on a polycrystalline yttria-stabilized zirconia substrate at 550°C . The ceria film was polycrystalline with randomly oriented grains. We irradiated these thin films with 90 keV He^+ ions at 400°C to a fluence of 4×10^{17} ions/ cm^2 to accumulate a peak damage level of 13 displacements per atom. We performed the ion irradiation at Texas A&M University through a collaborative agreement. The irradiation is intended to represent fission fragment damage.

We have performed initial characterization of the as-grown and ion-irradiated samples using transmission electron microscopy. We observed the formation of Pd-rich particles having a size of about 3 nm in the ion-irradiated sample. We did not see these particles in the as-grown film. In an effort to determine the nature of radiation damage that is a driving force for the phase formation, we have initiated molecular dynamics simulations of fission track damage in CeO_2 and UO_2 . Our integrated effort using synthesis, ion irradiation, chemical imaging, and atomistic simulation is providing valuable insights into metallic phase formation in irradiated nuclear fuel surrogate.



Scanning transmission electron microscopy image and energy dispersive spectroscopy map reveal the formation of Pd nanoparticle in irradiated CeO_2 .

Modeling the Interfacial Effects, Partitioning, and Production Routes of Epsilon Particles in Uranium Oxide

Richard A. Clark

The goal of this work is to elucidate the physical and chemical mechanisms that govern the formation of these metallic inclusions in irradiated fuel to understand the radiological, physical, and chemical conditions leading to their formation and unique properties.

The chemical fractionation of five-metal “epsilon phases” (composed primarily of the elements Mo, Tc, Ru, Rh, and Pd in a reduced state) from nuclear fuel under irradiation is not well understood. The development of these phases within uranium oxide matrices over time in fuel under irradiation has been linked to many important phenomena over the life cycle of nuclear fuel, from in-reactor operations to long-term spent fuel disposition. It has been hypothesized that epsilon phase formation can affect fuel performance, control cladding erosion, and cause stress corrosion through both the physical disruption of the fuel matrix caused by growth and the alteration of the fuel’s electrochemical behavior caused by presence as a unique chemical species.

This effort aims to understand the formation mechanisms of the epsilon metal phase during nuclear fuel irradiation and to understand the solid-solid-gas interfacial equilibrium effects that lead to variations in the properties of this unique phase. Developing a fundamental understanding of the chemical, radiological, and material structure conditions that led to precipitation of these phases will significantly improve our knowledge of nuclear fuel under irradiation conditions.

The expected outcomes of this research project are to characterize the epsilon metal phase and reveal the driving factors that produce epsilon metal precipitates in uranium oxide (UO_2). Some of the challenges we will tackle and queries we intend to resolve include the following:

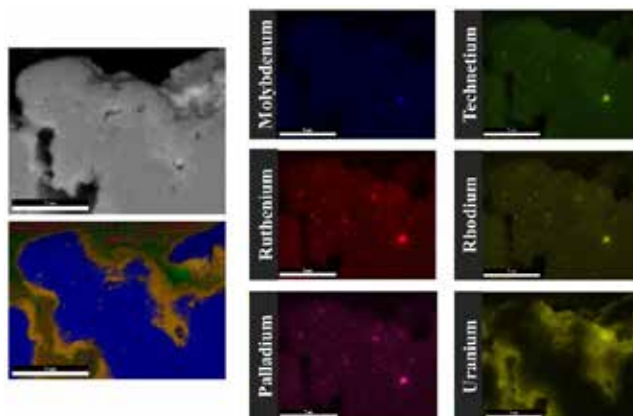
- Are epsilon metals in used nuclear fuel a single phase or a mixture of several discreet phases?

- Are the elements in these phases a result of fission products, decay products, activation products, or a mixture of all of these?
- At what concentration or key burn-up do we expect to see epsilon phases form?
- Does the radiation environment during reactor operations play a role in epsilon product formation?
- What mechanism controls the transport of Mo and Ru?
- Do the other metals follow a similar transport mechanism or are they produced *in situ*?

A very late FY 2015 start, this project had only a few accomplishments planned or achieved for FY 2015, with many of the milestones tied to instrument purchases, including a scanning transmission electron microscope detector for the Radiochemical Processing Laboratory’s focused ion beam/scanning electron microscope (FIB-SEM) and a Vienna Ab-initio Simulation Package license. Other less tangible accomplishments included using this project (and others) to support staff conversion and supporting several new postdoctoral fellows. Technical work during FY 2015 was focused on setting up the modeling frameworks that this project will use over its lifespan. Competing physical mechanisms for epsilon phase formation were developed based on dominant compositions of fission or activation product elements, and schemes for evaluating these mechanisms using density functional theory (DFT) were created.

For fission product elements, mobility of individual elements to grain interstices during irradiation is expected, and calculations of the energetics of element impurities on the bulk UO_2 lattice are anticipated to provide information about the relative driving force associated with this mobility. For a primarily activation element composition, reactive gas transport of MoO_x and RuO_4 is expected to be the primary formation mechanism. Characterization of epsilon particles and formation mechanism were a focus of research in FY 2016 and are separated into three key focus areas: Dissolution and Isotopic Analysis, *In situ* Analysis, and Modeling. Results were recently published in the *Journal of Alloys and Compounds*.

Future work is focused on extending the model binary system to include additional components (particularly Tc, Rh, and Pd), as well as interactions at interfaces. The UO_2 surface is the most likely candidate for the site of particle nucleation. DFT simulations will involve generating a surface of UO_2 and calculating the energy of an absorbed MoO_3 or RuO_4 at varying geometries and positions, with respect to the UO_2 . These results will provide a framework in which to test proposed mechanisms of particle formation.



Scanning transmission electron microscopy image and energy dispersive spectroscopy map reveal the formation of Pd nanoparticle in irradiated CeO_2 .

Observing and Quantification of the Initial Stages of Nucleation and Growth in Liquids

Nigel D. Browning

We are examining the dynamic processes behind the synthesis and operation of energy materials that provide unique insights into structure-function relationships needed to develop and implement new energy technologies.

Key processes important to the synthesis and operation of next-generation energy technologies involve atomic movement on the microsecond timescale or faster. Typically, the movement occurs at specific interaction locations that can be sub-nanometer in spatial extent and in complex gas or liquid environments (e.g., the electrode-electrolyte interface in energy storage technologies). We will make direct dynamic observations about these length and time scales using unique, state-of-the-art microscopy methods and use these observations to develop a fundamental understanding about how the atomistic mechanisms define the all-critical material/device performance on the mesoscale (i.e., the length and/or time scale over which a particular structure “locks in” the desired functionality).

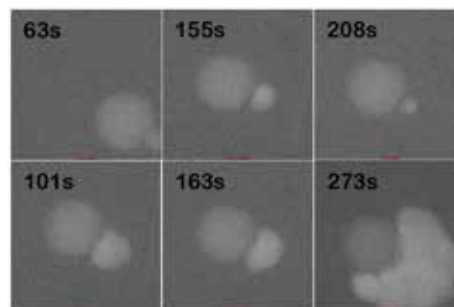
Our research will focus on a systematic analysis of a representative set of systems that exhibit specific processes important to energy technologies. A fundamental atomistic understanding of precipitation phenomena, such as the nucleation and growth of nanostructures from an aqueous solution, can have far-reaching implications on new material synthesis and the design of cheap, stable, efficient batteries, which are research themes that align well with DOE goals. As part of our work, the unprecedented sub- μ s temporal resolution of the dynamic transmission electron microscope (DTEM) will be developed to obtain a quantitative observation about the evolution of structures from individual atoms to mesoscale structures and complexes under varied environmental conditions that will be correlated with first principles modeling/simulations. These observations, including the fast transient intermediate states in each process, will provide unique insights to optimize material synthesis routes and operating parameters for advanced energy technologies.

Our work in FY 2015 focused on establishing the DTEM as a working instrument capable of time-resolved imaging. In addition, preparations have been made for nanoscale experiments that can be performed when the DTEM becomes operational. These preparations involve two sets of experiments:

one, a synthesis of nanoparticles involving bimetallic core shell architectures; the other, the identification of active catalytic sites in an oxide catalyst. A key result was the demonstration that the DTEM could work as a pulsed microscope.

Work in FY 2016 focused on the optimization of the laser pulses and the alignment of the microscope column to achieve high spatial and temporal resolution images. Alignment improvements during the course of the year both identified several areas of the optical beam path that needed to be modified and improved the alignment of the column to push the spatial resolution of the pulsed images towards approximately 10 nanometers. During the course of this work, many of the critical lifetime limitations of the emission process were determined, and an optimal approach to increasing the operation time of the microscope was determined.

Work to prepare samples for dynamic imaging in the DTEM has focused on materials that can be imaged both in liquids and gases. Results were obtained from two high-resolution preparation experiments. In the first test, a set of iron oxide nanoparticles were prepared with different surface functional layers. The test experiments in conventional microscopes demonstrated that the dynamics of the ions in the Stern layer around the nanoparticle could be observed directly. Such experiments demonstrate that the higher time resolution of the DTEM will be able to map dynamic functionality at solid-liquid interfaces with extremely high spatial and temporal resolution. In the second experiment, the atomic resolution images of an MoVTeO catalyst sample under reaction conditions showed that a novel diffusion of Te mechanism appeared to have a dominating effect on the activation of the catalyst. The future DTEM experiments will be able to directly observe the individual diffusion events leading to an atomic-scale understanding of the catalyst functionality.



Sequence of images showing the growth at the surface of L-cysteine functionalized nanoparticle. This growth is reversed by reducing electron dose rate and magnification. At a higher dose rate, ions accumulate at the particle surface, forming an amorphous, iron-rich phase. When the dose is reduced, iron dissolves back into the solution.

Optical Properties Modification in Complex Oxide Epitaxial Films via Alloy Formation

Ryan B. Comes

We are engineering the optical and electronic properties of complex oxide thin films by doping the materials to reduce the band gap and interface formation to promote separation of optically excited charge carriers.

There is great interest in finding new materials that can harvest sunlight effectively for converting photovoltaics and powering photochemical reactions. Because the solar spectrum is more abundant in visible than in UV light, finding photoactive materials that absorb in the visible is of particular importance. Many recent popular photovoltaic materials use elements that are either toxic or rare; moreover, their surfaces form oxides in the atmosphere, which can alter their otherwise useful properties. An ideal class of materials for future photovoltaic applications is complex metal oxides, which are already stable in air and afford flexibility in designing and synthesizing tailored materials. Complex oxides also exhibit a wide range of optical and electronic properties. SrTiO₃ (STO), for example, exhibits the highest reported electron mobility of any perovskite oxide. When epitaxial strain is applied, it can be made ferroelectric, producing a built-in electric polarization to separate electron-hole pairs in photovoltaic material. However, STO has a wide optical band gap of 3.2 eV, making it a poor absorber of visible light.

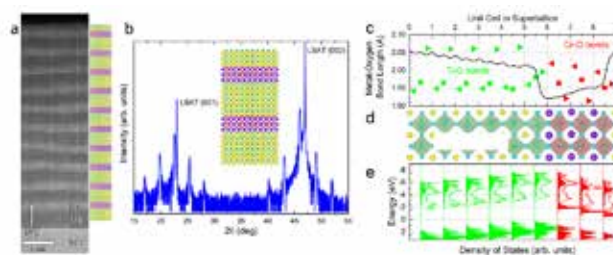
In FYs 2014 and 2015, we focused on the growth of epitaxial doped perovskite films using molecular beam epitaxy, specifically exploring the co-doped Sr_{1-x}La_xTi_{1-x}Cr_xO₃ (SLTCO) system. In this material, La³⁺ dopants stabilize defect-free Cr³⁺, reducing the optical bandgap by approximately 0.9 eV, while preserving the material's semiconducting properties. This work enables the ability to use these films for photocatalytic and photovoltaic applications, because the materials exhibit visible light absorption.

During FY 2016, we focused on a related La, Cr co-doped BaTiO₃ system to produce ferroelectric materials for a ferroelectric photovoltaic employing the bulk photovoltaic effect in the material. Photoconductivity, piezoresponse force microscopy, and electronic transport have been performed to understand the material properties. Transient reflectance measurements confirm that photoexcited carriers are present in BLTCO and work is ongoing to understand the effects of polarization on the carrier lifetime in these materials.

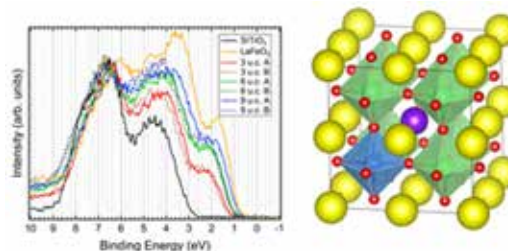
Work during FY 2016 also continued to explore SrTiO₃-LaCrO₃ (LCO) superlattice films. After achieving success earlier in the project by showing that a polarization is induced in the STO

layer due to the repeating, precisely engineered interfaces, we continued to study these materials using synchrotron x-ray photoelectron spectroscopy. These measurements allow us to examine the electronic properties of individual layers of the superlattice and have never been performed with this detail before. This work has produced unprecedented capabilities to study the electronic structure of both the LCO and STO layers of the material, allowing us to study the electronic configurations induced in the superlattice. Further measurements will help to support our current analysis, and within the next year, we expect to publish the work in a high-impact journal.

Finally, a recent study has examined the related LaFeO₃-(LFO)-SrTiO₃ interface. LFO is particularly exciting as a photocatalyst, because it has a visible light bandgap of 2.3 eV. Recent works showed that, by engineering the interface between the two materials, novel electronic behavior in the LFO film emerges to separate photoexcited electron-hole pairs. Using our unrivaled capability to measure interfacial electronic behavior via x-ray photoelectron spectroscopy, we probed interfaces engineered to produce different behavior. We found that by tailoring the interface in a certain way, we produce electronic reconstructions at the interface that are favorable for separating electron-hole pairs for ideal photocatalytic response. We expect that this work will be a major contribution to the field.



SrTiO₃-LaCrO₃ superlattice (a) scanning transmission electron microscopy; (b) x-ray diffraction; (c-e) density functional theory model.



(Left) Valence band x-ray photoelectron spectroscopy data for LaFeO₃ films grown on SrTiO₃; (right) Density functional theory model of La, Cr-doped BaTiO₃.

Optically Resonant Subwavelength Films for Tags and Seals

Kyle J. Alvine

The goal of this project was to develop a new, counterfeit-resistant, and tamper-indicating film that would be critical to identifying potential security breaches in arms control and special nuclear material tracking chain-of-custody applications.

There are multiple examples in transport and handling of nuclear materials where counterfeit-resistant tags and seals that incorporate tamper indication are critical to identifying potential security breaches. This may include diverse applications such as tags and seals with counterfeit and tamper-indicating properties for UF₆ cylinder tracking. While different applications may require unique capabilities, tags and seals require tamper indication and a unique signature to prevent and/or detect counterfeiting.

We proposed to study and demonstrate proof-of-concept tamper indication performance of newly developed subwavelength films, which are particularly well suited to applications in tags and seals. They have a unique optical reflection that is wavelength-, angle-, and polarization-dependent and can be tailored to operate from the visible to the infrared region.

In previous years, we demonstrated tamper indication and a unique signature for counterfeit detection and prevention for typical tampering mechanisms in subwavelength film.

In FY 2015, hyperspectral imaging was used to both investigate tamper detection on these samples and to aid in the design of an eventual (simple) reader for the films. Hyperspectral imaging collects the spectral response of the sample in the visible range at each pixel in the image, rather than just red, green, and blue information. Principle component analysis (PCA) is then used to look for correlation and contrast in the image from different wavelength regions. With hyperspectral imaging, we were able to locate and identify cut marks that were very difficult to find via simple visual inspection. Patching was even more apparent. Nanostructured films stood out with high contrast compared to color-matched fill material using PCA analysis. When polarization was added to

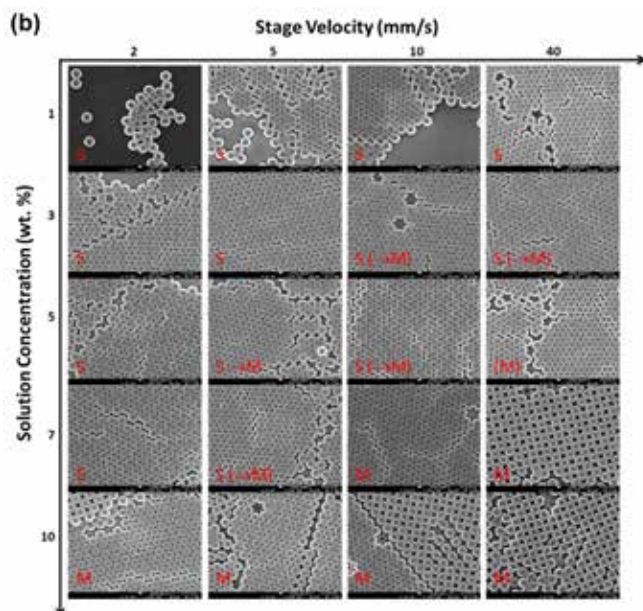
the hyperspectral setup, independent comparative rotation of the nanostructured film sections was readily apparent. These results are highly promising for tamper indication.

In FY 2016, we have been focused primarily on developing a laboratory capability for making intermediate-scale films on the order of 8 by 10 inches in size. The ability to make and work with films of this size is important in order to show both that the films are scalable and to be able to wrap or enclose complex, real-world objects for tags or tamper-indicating enclosures. This is also important for remote optical readout of the films where the stand-off distance is related to the size of the films and imaging resolution.

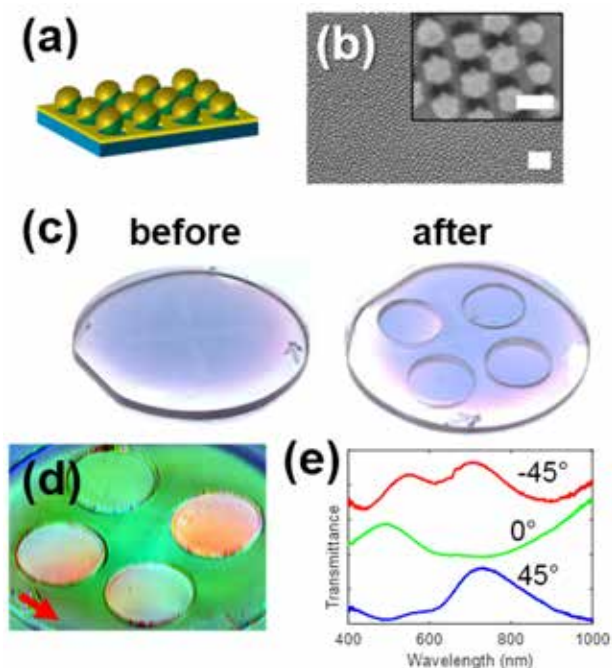
To this end, we extensively modified our existing flow coater to now incorporate both X and Y motion, a more reliable blown arc surface treatment system to replace the corona discharge system, added halogen drying lights, and options for a 9-inch-wide coating blade or three independent 2.5-inch blades for combinatorial processing. This upgraded system now allows for processing up to three nanoscale templates at one time. In addition, one can modify the surface treatment time for other combinatorial measurements.

With this machine we have demonstrated fabrication of approximately 50 films of 7 by 10 inches using multiple different size polystyrene nanoparticles. To develop a better understanding of the machine, phase space mapping was done on most of the process variables, including particle concentration, stage speed, surface treatment, and blade height. To accomplish this, we fabricated films by incrementally changing one variable while holding the others fixed to generate the phase space. This method was necessary to determine both the optimized parameters for each particle size and to develop a process for more rapidly optimizing future films.

In addition to improving the flow coater for large films, we have done tamper testing of the nanoplasmonic films, using PCA to analyze the data. The rotation of the discs within the parent film was not obvious to the eye; however, it was immediately obvious through the hyperspectral analysis.



A phase space map of nanoparticle solution concentration vs. stage velocity for the improved flow coater. The quality of the nanoparticle monolayer is highly dependent on both variables, but is roughly optimized for concentrations of 5 wt % and 5–10 mm/s.



(a) Schematic of gold nanopillar structures before lift-off with a deposition angle of 45 degrees. (b) Scanning electron micrograph of a representative sample after gold deposition. Scale bar: 500 nm. Inset: High-magnification micrograph of gold nanoshell structures showing the anisotropy due to tilted metal deposition. Scale bar: 100 nm. (c) Photograph before and after tampering with an arrow indicating the deposition angle. (d) PCA image of tampered cutouts rotated at various angles. (e) Transmission spectra of the sample before tampering recorded -45, 0, and 45 degrees with respect to the light source illustrating the anisotropic optical properties of the fabricated gold nanoshell.

Optically Stimulated Luminescence Data Storage

David W. Gotthold

We are developing a new approach for data storage using optically stimulated luminescence (OSL) that can be scaled to high data densities with excellent data lifetime and integrity.

To meet the ever-increasing data storage need for cloud computing, scientists and engineers are constantly pushing the limit of data storage density. Hard disk drive (HDD) has been the dominant computing data storage device for the past 50 years, with over 800 million units sold in 2012. Until recently, HDD density increased by 100% every year, but advancing technology has decreased this rate to 25%. Solid-state hard drive (flash) is the storage method based on ferroelectricity, where direct wiring enables immediate access to the bit. However, flash suffers memory wear and, thus, can be used only as short-term storage. Although CD, DVD, and Blu-ray are popular optical data storage methods, they have a significantly lower density (under 10% of HDD). Still another recent advanced storage method is holography, a promising technology because of its large density and fast data access rate, reading a page each time instead of a bit (like flash). However, sensitivity to vibration and extremely low speed have prevented this technology from succeeding.

We propose to develop a new data approach that is capable of making a paradigm change in storage technology. OSL has been in use for many years as a radiation dosimetry technology. OSL media has an extraordinary linear optical response (over eight orders of magnitude for LiF) that will enable ternary (higher-level) encoding of data in a single bit, thus storing more data in each defined location. The objective of this project is to prove the concept of OSL data storage by demonstrating multi-values data encoding. This information is a primary concern for potential development partners and will be key in identifying the potential market opportunities for this new technology.

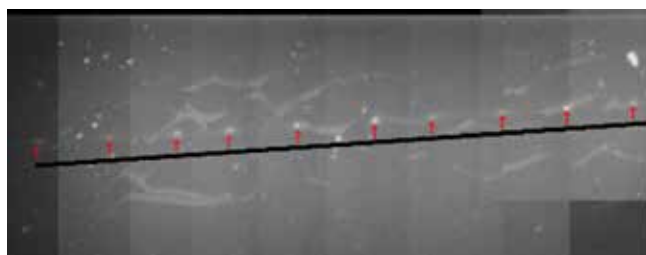
Based on work done in previous years, the lack of knowledge about the OSL process at very high power levels is identified as the critical technology gap. To address this issue, this project was started in FY 2015 to develop a very short pulse electron source capable of producing the ionizing radiation at and energy density and pulse length necessary to evaluate the dynamic formation of the F-centers responsible for the OSL signal.

To develop the ability to measure the formation of OSL centers, a high energy pulse source of ionizing radiation needed

to be developed, and it was determined that a laser pumped photocathode electron source would be the best approach to achieve the very short, high-intensity pulses needed. The completion and testing of this system has been the focus of this year's project. This system was built onto an existing JEOL 5900 scanning electron microscope to enable focusing of the electron pulse and control of the energy density. The drive laser used was a Fianium HYLASE with a frequency quadrupling crystal system in order to provide pulse of 266 nm that will be directed into the scanning electron microscope column and onto a tantalum photocathode.

Previous work focused on developing a custom photocathode with a flat emitting surface to optimize the field strength and extraction. However, additional testing showed that a standard shaped filament, made from Ta instead of W, provided sufficient extraction and improved focus due to the more optimized electric field geometry. This approach also enabled switching between photocathode and thermal emission operation, simplifying alignment and testing. While the laser drive system is capable of achieving very high repetition rates (up to 40 MHz), initial development was all done at 50–100 kHz to limit heating of the non-linear optical system and simplify the in-vacuum motion needed to scan the LiF samples by reducing the required speed. After alignment and adjustment of the filament temperature, photoemission currents up to 2.76 nA were observed, correlating to a per-pulse electron density of 3.45×10^5 , which was above the approximate saturation density of 1×10^5 estimated for a 150-nm spot based on steady state current exposures. Even at these high densities, no physical damage was observed in the LiF. By moving the sample using an in-vacuum linear electric drive motor capable of up to 0.5 m/s while operating the beam, a series of distinct, high-intensity luminescence spots were generated.

In addition to demonstrating the ability to rapidly form high-resolution data spots in OSL material, the frequency, short pulse length system complements other high speed microscopy tools, such as the dynamic transmission electron microscope, and has potential for probing a range of ultra-fast processes in critical areas such as catalysis, structure evolution in lightweight alloys, and battery electrodes.



High-intensity luminescence spots.

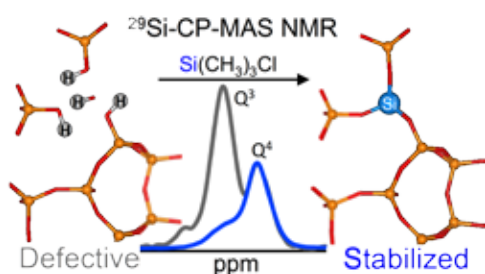
Robust Hierarchical Zeolite Frameworks

Mirosław A. Derewinski

The goal of the study is to provide molecular description on the formation and arrangement processes during synthesis of microporous crystalline silicates using PNNL's advanced measurement capabilities and use that knowledge as a basis for the synthesis of nano-sized and mesoscopically structured zeolites with tailored chemical and textural properties.

Modern industrial processes focusing on catalysis, phase separation, or ion-exchange are becoming increasingly dependent on the superior qualities of molecular sieves based on Si and Al, as well as Si, Al, and P. Their structural versatility, robustness, and chemical properties are detrimental to facilitating most processes. By tailoring these properties—the main goal of this project—upgrading processes, such as biomass conversion reactions and exhaust treatments, becomes attainable. Specifically, we focus on and have gained important insights in 1) understanding the role of structural defects and prevention of such, affecting the stability of zeolitic material in hot liquid water; 2) investigating the behavior of zeolites in the phase separation of water and organics during catalysis affected by the hydrophilicity of the system; and 3) exploring the formation mechanism of zeolite structures using Al and Na as a probe and *in situ* spectroscopic methods.

In FY 2016, a more detailed insight into the fragile nature of zeolites under hot liquid water conditions was achieved. Understanding and developing methods to increase the lifetimes of zeolite-based catalysts for the reductive transformation of biomass-derived feedstock is an integral part of developing viable methods for the production of alternative resources. Using a model system enriched with internal structural defects, the effective removal, with the help of a small reactive silane, led to materials capable of retaining crystallinity and microporosity after extended treatment in hot liquid water.



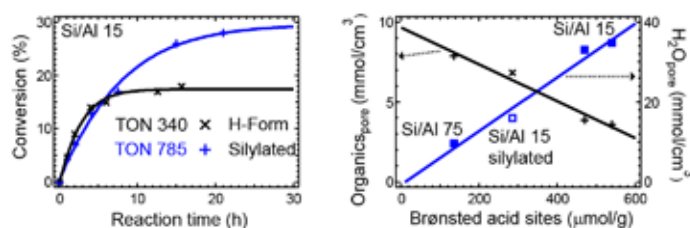
A schematic overview of healing defects, followed by ^{29}Si MAS NMR.

to depend solely on the concentration of defects, work conducted in FY 2016 suggests that the presence of organic

linity and microporosity after extended treatment in hot liquid water.

While the stability in pure hot liquid water was determined

compounds during catalytically relevant conditions leads to a different behavior. By testing commonly used Al-BEA catalysts in the Brønsted acid catalyzed dehydration of cyclohexanol, a lignocellulosic-derived model compound, their lifetime was determined. The conversion of cyclohexanol eventually stopped as the dissolution of the framework, loss of confinement, and deposition of amorphous silica blocking access to the active sites led to their deactivation. Materials of lower Al content yielded longer lifetimes, despite having a higher defect concentration. By investigating the uptake of cyclohexanol and water in the micropores of the molecular sieve, it was deduced that zeolites of high Brønsted acid site concentration adsorb significantly more water. The higher intraporous water concentration consequently led to a faster deactivation rate of the individual sites. The hydrophilicity generated by the presence of defects plays a smaller role compared to the degree of Brønsted acidity generated by isomorphously substituted Al in the framework.



The intraporous water concentration is a function of the Brønsted acidity and the correlating hydrophilicity of the system. Reduction of said properties leads to increased lifetimes for silylated materials.

By employing our previously established method of silylation, we were able to effectively increase the lifetime of Al-rich BEA zeolites. Capping of OH groups in the zeolite reduced the water uptake and prevented the condensation of water inside the pores, which would eventually culminate in the hydrolysis of Si-O-Si bonds. This result led us to investigate alternative methods of reducing the intraporous water concentration such as synthesis of extremely hydrophobic zeolites by design.

In FY 2016, we also focused our research on optimizing zeolite synthesis parameters to improve sorption capacities of CO_2 , as well as to produce stable zeolites (CHA) for use in selective catalytic reduction reactions typically found in vehicle exhaust gas treatment. Understanding how individual synthesis parameters affect the product is a crucial part of this initiative and is being aided by investigating the formation mechanism of zeolites (FAU) with the unique capabilities present at PNNL. Both *in situ* and *ex situ* solid state nuclear magnetic resonance (NMR) allowed for probing the location of Na^+ cations, charge balancing the evolving framework, during the formation of zeolites and giving insight into the mechanism.

Soil Organic Carbon/Mineral Association and Aggregation Processes

Jay W. Grate

The stabilization of organic carbon in soils is critical to understanding how terrestrial ecosystem processes relate to global atmospheric carbon levels. A new paradigm relates soil organic carbon stability to its interactions—the subject of this research—with soil minerals.

Soil organic matter represents an important pool of carbon in the global carbon cycle. The annual net change in CO₂ concentration in the atmosphere represents a small difference in yearly numbers; as a result, a modest change in the behavior of one carbon pool, leading to a change in one of those large numbers (fluxes), can have a significant effect on atmospheric CO₂ levels. Accordingly, even a moderate shift in soil organic carbon persistence could help to offset anthropogenic CO₂ in the atmosphere. Given the importance of soil carbon persistence and current knowledge gaps, the application of advanced analysis and imaging capabilities at PNNL to the interfacial chemistry of organic carbon/mineral associations in an abiotic investigation using *in vitro* model systems offers rich territory for original and impactful science relevant to DOE programmatic priorities.

This project seeks to provide a molecular-to-micron scale mechanistic understanding about how organic compounds interact with natural mineral surfaces and within aggregate structures. These issues are highly relevant to the stabilization and dynamics of soil organic carbon in terrestrial ecosystems. Contrary to the past belief that the molecular structures of carbon compounds condensed into macromolecules determined their stabilities in soil, it is now believed that the interactions of organic compounds with mineral surfaces and in aggregates of mineral particles determine the bioavailability of soil organic carbon for breakdown by biotic processes. The previous understanding was based on the extraction and organic characterization methods, while current methodology is based on actual direct observation of soil organic carbon by imaging and

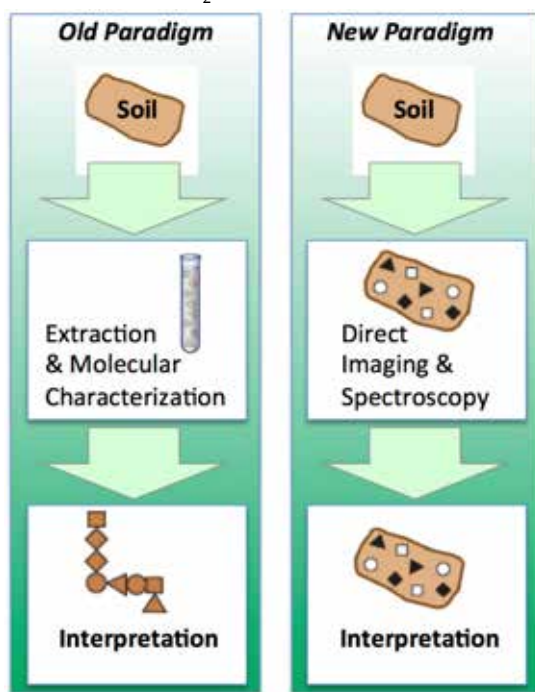
spectroscopy *in situ*. The latter methodology has shown that organic matter is present in organo-mineral aggregates. Accordingly, these interactions and their influence on aggregate formation are highly significant, yet difficult to study at molecular or nanoscopic scales directly in soil, which is a heterogeneous, opaque material.

We have developed a bottoms-up approach based on minerals and organic compounds with known functionality to establish an environmentally relevant model system for exploring organic-mineral interactions and aggregation processes. In this approach, the composition of the model system, containing a mineral, organic compound, and a solution electrolyte, is known, and measurements and observations are made. Dynamic force spectroscopy (DFS) has been used to quantify the interactions of specific organic functional groups with specific clean mineral surfaces, namely mica and goethite. Select measurements have been done that show the influences of environmental factors such as ionic strength,

cation, and pH on the strength of the organomineral interaction. This project has also used *in situ* transmission electron microscopy (TEM) and cryo-TEM to look at aggregates of mineral nanoparticles directly, where the spatial relationships of the visible mineral particles provide indications of the effects of organic adsorbates.

The DFS studies have shown that binding strength varies with the organic functional group, and that different minerals have different binding preferences. In addition, environmental factors can have a major influence. Because these environmental factors are expected to vary with time in soil materials, they could have significant effects on soil carbon persistence.

The methodology for our model system approach has been established, and measurements have quantified organomineral interactions. Using DFS, binding energies between selected organic molecules and minerals were measured, revealing trends in molecular-scale phenomena. Experiments with TEM allows one to move from organomineral interactions to assemblies of mineral particles as one scales from molecular scale to nano and micro scale.



Contrasting views of soil organic carbon.

Ultra-Low Background Polymers for Structural Applications in Radiation Detectors

Jay W. Grate

This project focuses on the materials required for ultra-low background dielectric materials (i.e., plastics) for next-generation detectors in high energy physics programs.

Ultra-low background radiation measurements are essential to several large-scale physics investigations such as those involving neutrinoless double beta decay, dark matter detection, and solar neutrino detection. Ultra-low background counting is also of value in applied radiation detection applications associated with nuclear security and treaty verification. To detect low probability events, it is essential that the background noise of the measurement systems be driven to the lowest achievable levels. Hence, the radioactive contaminant species in the materials used to make and house radiation detectors must be minimized. Radioactive impurities in materials can arise from naturally occurring isotopes, radionuclides generated by cosmic ray interactions, and anthropogenic sources. The radioisotope purity required for these detectors goes well beyond even the best industry practices for chemical purity.

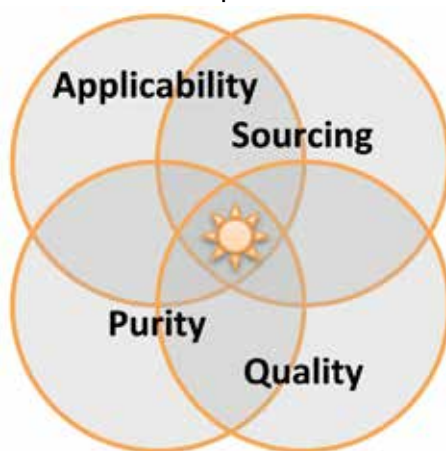
Considerable work has been done to purify germanium as the primary semiconducting detector material. Ultra-pure copper production for low background systems is well underway for structural parts requiring electrical or thermal conductivity. However, there remain needs for electrically and thermally insulating dielectric materials with extremely low background radioactivity such as polymer plastics. The materials must be especially low in uranium (U), thorium (Th), and potassium (K) and not be contaminated by radon daughter products. This project is focused on plastic polymers for use as structural materials. Specifically, thermoplastic polymers represent the class of dielectric materials capable of meeting the requirements for ultra-low backgrounds, mechanical rigidity, and suitability for forming or machining into detector parts. These materials cannot be evaluated or qualified for detector applications unless a method exists to assay the polymers for trace

contamination. The development of assay methods is coupled with sourcing, evaluating, and improving plastic polymer materials.

We made great strides in sourcing and obtaining thermoplastic materials early in the supply chain to prevent grinding, pelletizing, thermoforming, handling, and transporting activities from contaminating and confounding our ability to understand the inherent purity of the plastic materials. Both non-fluorinated and fluorinated materials with excellent structural properties have been obtained in flake, powder, pellet, or 3D printed forms.

We also made significant progress in assaying samples of these plastics for U and Th using inductively coupled plasma mass spectrometry (ICPMS). This technique is much faster and requires far less sample than traditional gamma spectroscopy. The challenge of the ICPMS approach is to get the metal atoms out of the solid plastic and into a solution for liquid nebulization. In addition, there are no reference materials as laboratory control standards, and it is hard to avoid contaminating sample solutions with traces of metals from the container materials used in sample preparation. In this regard, we developed a new dry ashing method using an ultra-high purity material to contain the polymer sample. Excellent detection limits with very low process blanks were achieved. Tracer recovery for radiometric quantification was also very high.

We have successfully sourced and analyzed fluorinated and non-fluorinated structural polymer materials with radiopurity levels in the low microBecquerel (μBq)/kg range, orders of magnitude below the 10 to 10,000 milliBq/kg of typical commercial thermoplastics. The radiopure materials we assayed have been powder or pellet raw materials. Recently, we have also examined the radiopurities of solid plastic materials and parts made from such raw materials.



To create a sustainable capability in ultra-high purity plastics for detector applications, materials must have correct structural and insulating properties and be sourced at early stages from reliable manufacturers so that subsequent handling and part formation does not cause contamination. Additionally, materials and processes must be developed to provide extremely pure parts, with quality assured throughout, requiring the development of new assays for contaminants in plastic materials at levels far below those of industrial practices for chemical purity.

Ultra-Stable, Ultra-Low PGM Loading Membrane Electrode Assembly (MEA)

Yuyan Shao

This project improved MEA performance in oxygen by 200% and in air by 25% over previous results. Efforts are ongoing to determine the performance at various loading levels and platinum weight percent in the carbon support. The results are expected to provide insights into catalyst architecture and design, in order to meet the DOE target of 0.15 mg Pt/cm².

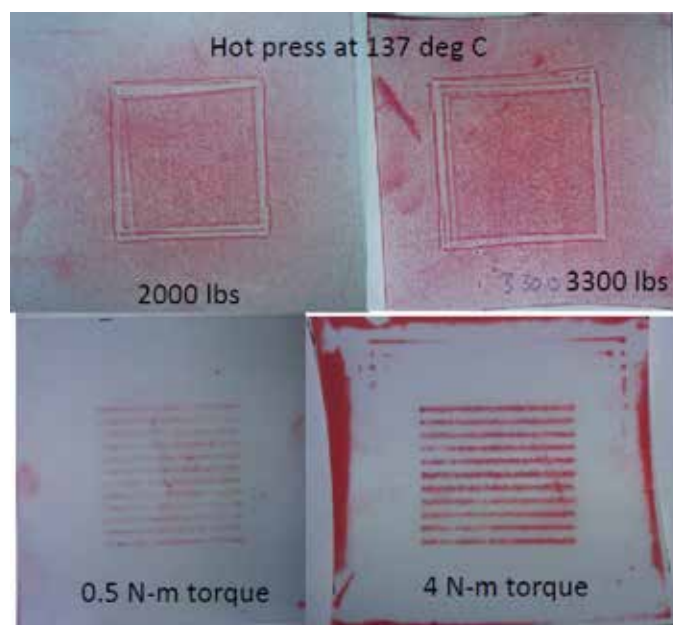
The primary objective of this project was to elevate PNNL's MEA capabilities. In the past, hot pressing conditions and torque for cell assembly were chosen based on available information in the literature. Attention was not paid to the magnitude and uniformity of the pressure distribution within the MEA. A Fujifilm prescale sensor film from Sensor Products Inc. was used to determine the surface contact pressure between the gas diffusion layer (GDL) and the membrane during hot pressing and cell assembly. During hot pressing, the temperature was maintained at 137°C, while the set point for force was varied from 1,000 to 5,000 lbs. During cell assembly, the torque was varied from 0.5 N-m to 9 N-m. The lowest force or torque that corresponded to 200 psig and a uniform pressure distribution was selected. These conditions were used for all tests, and MEA preparation was done using the spraying process on GDLs.

Test results showed that increasing the loading from 0.2 mg Pt/cm² was not desired, since the increase in performance is not in line with increase in loading, coupled with the fact that the DOE targets for the total fuel cell is 0.15 mg Pt/cm².

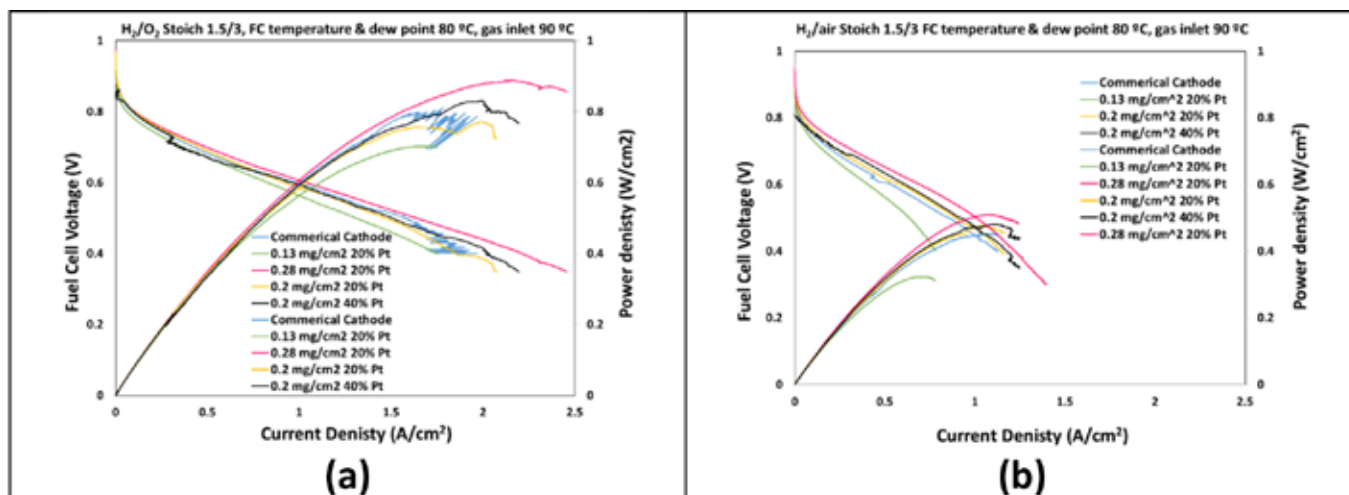
The activity indicates room for improvement by suitable catalyst architecture design. Increasing the weight percent of Pt results in improved performance, indicated by higher maximum power density and higher current density at 0.45 V. This indicated flooding was not an issue, which was verified by data showing that reducing the relative humidity (RH) did not improve the performance. This is possibly due to higher ohmic resistance at low RH. The GDL used is more effective

in reducing flooding related issues, and per vendor specification, leads to poorer performance when flooding is not an issue. Higher performance can be anticipated by improving electrode activity to oxygen reduction. The results for 10 wt% Pt in carbon were poor.

Future work will focus on optimizing the hot pressing and cell assembly conditions for each wt.% Pt, with particular focus on a loading of 0.1 mg Pt/cm² for both 20 and 40 wt% Pt. Higher Pt wt.% leads to greater flooding at high loadings, and hence, lower compressive loading during both these steps would probably facilitate better gas transport. However, since our focus will be on low Pt loadings, we intend to explore the effect of increasing hot pressing loading/time and increasing torque during cell assembly to reduce contact resistance. Lower loadings result in a thinner catalyst layer, so limitations due to gas and mass transport are expected to be negligible. At high geometric current density, the effective current density is higher for low loading. Therefore, it is critical that the activity of the catalyst be improved via ionomer:catalyst composition optimization.



Effect of hot pressing load and torque during cell assembly on pressure distribution.



(a) Performance of the MEAs of various compositions, and (b) performance of various MEAs in O₂ & air.

Mathematics and Computing Sciences



A Composable Interdependence Model for Cyber-Physical Systems

William B. Daniel

More and more, the monitoring and control of critical infrastructures is facilitated by network-based supervisory control and data acquisition systems that may fall victim to inadvertent network outages or intentional manipulation. This work will allow researchers and utilities to rapidly identify and assess systemic vulnerabilities in order to develop more robust systems and operational practices and serve critical public demands.

Past work in this field has tended to focus heavily on integrated cyber-physical system analysis. Detailed modeling has been performed of cyber networks, their software, packet-level network communications, and programmable logic controller vulnerabilities. Many commercial and research models also exist and have been used to characterize the consequences of specific contingencies. Less work has been done to effectively bridge the gap between the two areas of research, leveraging the potential for cyber-based observation and control to direct contingency analyses. The technical objective of the project is to provide researchers with novel tools to fill in gaps in infrastructure network datasets, rapidly assess the impact of individual contingency events to those networks, use notions of observability and controllability to bound the distribution of cyber threats, and identify, as a result, the most relevant contingencies and the distribution of their potential impacts.

The critical, water-distribution infrastructure sector is being used as a concrete application area within which to test theoretical results and demonstrate capability. Two different lines of work are being pursued within this context: technical and theoretical. On the technical side, an industry standard model of water distribution system performance, EPANET, has been wrapped in a Scala-based modeling framework to facilitate the analysis of contingency events. This distributed framework makes it easy to access and scale model runs across computational nodes as needed. It also makes it easy to integrate the EPANET model with other models.

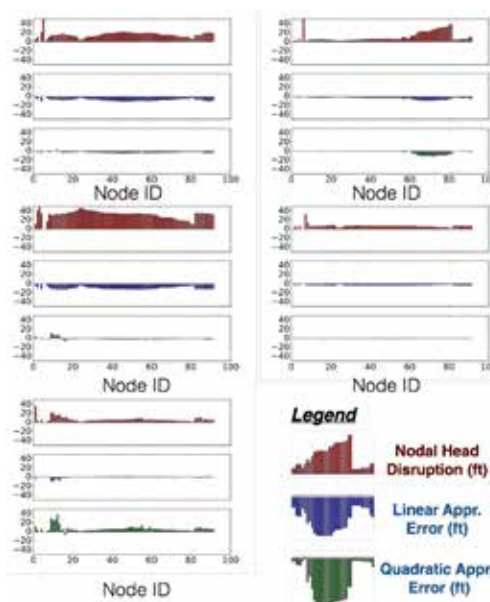
On the theoretical side, a method has been developed to rapidly estimate a water distribution system's response to alterations in component states like tank or reservoir head levels, valve settings, and/or changes in pump operation.

Historically, the analysis of a single contingency (a set of coordinated changes to component states) has required the full solution of the network head and flow equations using an iterated solver. Each such solution requires tens of matrix inversions, and for each contingency to be analyzed, the process must be repeated in full.

The approach developed within this project, instead, first performs a single, iterated solution of the system state under normal operating conditions. A second-order approximation of the head sensitivity to changes in component state is then used to assess the potential impact that contingencies may have on network behavior. Each such analysis requires only simple matrix multiplications. No additional matrix inversions are necessary. This allows the time required to analyze a particular contingency to be reduced by orders of magnitude. An initial analysis comparing the accuracy of the second-order approximation to the full solution has been performed for a small, 92-node system and submitted for publication at the 2017 IEEE American Control Conference. Additional analyses are ongoing.

Over the coming year, this approach will be evaluated in the context of larger, more complex networks and via additional metrics, such as the spatial extent of impact areas. Additionally, the work will be leveraged to develop a network

inference framework to allow researchers to fill in gaps in network topology data. This will allow researchers to better assess potential system behaviors for networks where complete information may not be available.



Comparison of first and second-order forecasts for head disruption against EPANET simulation results, given 40% tank head-level disruptions. The five contingencies correspond to the individual manipulation of the head level in each of a 92-node system's five different storage tanks.

PN1601612793

Analytics Integration and Validation Framework

Ryan R. LaMothe

The goal of this project is to enable the integration of multidisciplinary research efforts and their products into a unified framework for the discovery and validation of complex signatures.

Software tools and infrastructures for the discovery of domain-specific signatures are in use and well-known. However, none has the flexibility and the reusable methodology required to be extended into multiple domains. Typically, these tools are applied ad hoc for the task at hand, or they are integrated using workflow software, which provides a graphical interface to integrate disparate software tools. For either case, the methodology is not made explicit, in that it is tied only to a particular scientific domain or not documented systematically. This situation makes it difficult for signature development tasks to be repeated and applied in other contexts. Further, although the workflow tools help to automate and integrate distributed computing resources, they are often challenging to apply in practice and cannot individually maximize the high-performance computing resources that are required to solve many of these problems.

Our research leverages multiple developed capabilities, including analytic framework architecture, signature quality metrics, and signature processing algorithms developed

under multiple projects. To support the execution of commonly used signature discovery methods and algorithms, we created a flexible, lightweight integration platform that can incorporate existing and legacy tools into a single system known as the Laboratory Integration Framework and Toolset (LIFT), which adapts to client needs by allowing users to choose appropriate tools to address their particular signature development needs and deploy them to the environment of choice. These tools are made available via LIFT-supplied web services that are accessible from a wide range of client tools. LIFT achieves several core goals by reducing trial and error, allowing users to reuse existing tools and approaches, and providing a reproducible mechanism for constructing and evaluating signatures. This methodology encourages reusability and reinvestment and is actively being used to develop signatures in a wide variety of domains.

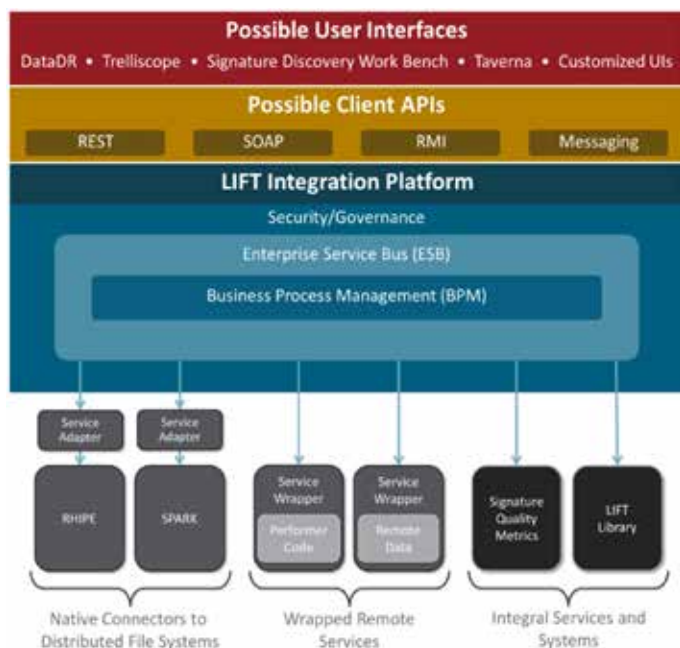
This enterprise-quality integration platform exhibits the following features:

- **Modularity** – The integration framework allows analytic services to be added and removed readily.
- **Deployability** – The system can be easily deployed and fielded through the use of flexible profiles that abstract away host-specific information.
- **Interoperability** – Web services are used to make analytic tools available to a wide range of client tools and programming languages.

In FY 2016, the analytic framework team prepared LIFT for integration with PNNL as a whole. The team began work to harden integration of existing services to become available across the Laboratory. It also supported a transition plan to move maintenance of the LIFT system to current users of the framework.

The maturity of the system is exhibited by the fact that LIFT has been adopted as the integration framework for two PNNL divisions and several external projects. Additionally, PNNL leadership has recognized LIFT's cross-cutting value and is considering supporting the framework for all applicable future research initiatives.

In FY 2016, the team will continue to support signature challenge projects through LIFT's novel AaaS platform, which will also be an integral part of PNNL's Research Cloud efforts in coming years.



LIFT architecture.

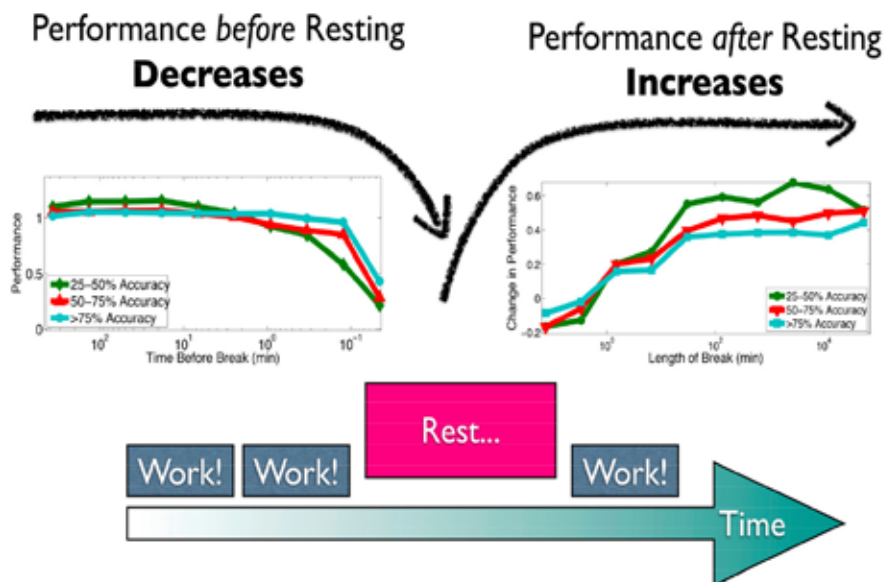
Cognitive Depletion in Streaming Environments

Nathan O. Hodas

In this project, we examine the attempts to leverage techniques for modeling cognitive resources to create adaptive systems that extend peak performance.

The problem of mental fatigue has been widely studied, but the results of past research have been highly contradictory. Although one's intuition is that mental fatigue can be offset by rest, the relationship between training, performance, and fatigue remains poorly uncharacterized. Many studies show only a weak relationship between self-assessed fatigue and actual cognitive performance; others have linked impulsive behavior (e.g., "ego depletion") to the loss of finite willpower and executive function. These latter works have been controversial, in part due to difficulty of replicating the work and also from the challenge of clearly measuring ego depletion. In short, there is no clear mechanism to model the processes proposed and no simple manner in which to design experiments or protocols to manage ego depletion and hampering progress.

We are using our domain expertise in cognitive energetic modeling, human-computer interaction, and controlled human experiments to provide unique, valuable capabilities to identify and mitigate cognitive depletion in intense streaming environments. We use these models to understand cognitive depletion and then predict the state of the user, enabling systems to weigh user actions and provide direct feedback to users, alerting them when performance decline is predicted. Using a combination of physiological models and machine learning, we have conducted user studies to measure cognitive depletion in people engaging in an extended task.



Anyone who has performed decision-making over extended periods is familiar with the feeling of cognitive depletion: increased impulsiveness and errors, slower evaluation of new information, and decreased memory retention. We identified cognitive depletion among video game enthusiasts' activity, people taking standardized tests, and document rating.

In FY 2016, we continued to explore cognitive depletion in a number of tasks, including standardized testing, video games, and document ratings. We identified that users tend to decide to take breaks when they become extremely depleted. Using these observations as a guide, we conducted a study of cognitive depletion of knowledge workers, both in their actual work environment and in controlled laboratory conditions. By comparing behavioral patterns with observed performance, we have concluded that cognitive depletion may be characterized as increased impulsiveness and errors, slower evaluation of new information, and decreased memory retention.

Using our observations and predictions from video games and standardized tests, we constructed a list of symptoms that would be indicative of cognitive depletion. These symptoms are independent of the task the user is engaging in.

We also designed a user study to evaluate cognitive depletion in expert users of nuclear magnetic resonance (NMR) spectral classification software. We conducted an ethnographic study of an expert NMR spectral classifier, observed them over the course of their work, and identified clear signs of cognitive depletion.

In addition to studying an expert at their job, we conducted a study on 40 people to see if we could quantify cognitive depletion in controlled laboratory conditions. We observed that sustained attention and effort can induce cognitive depletion after approximately an hour of work.

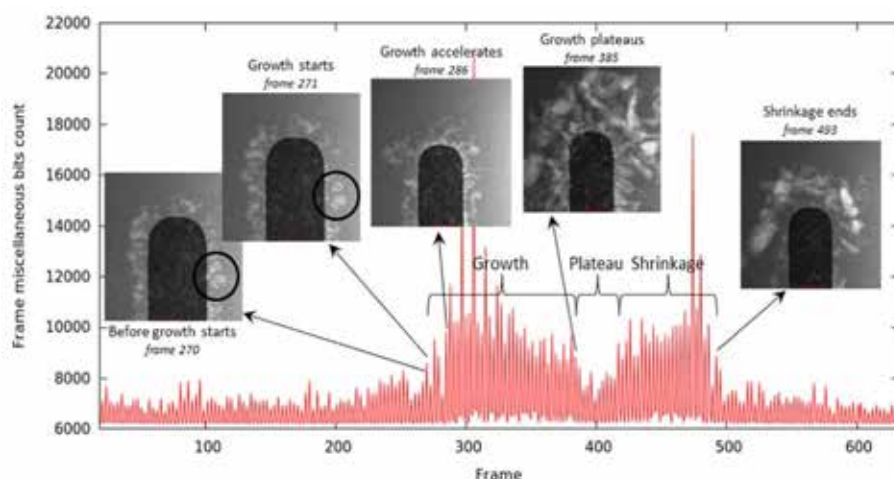
By understanding the theoretical principles of cognitive depletion and measuring the condition in controlled situations, we will be able to design more effective systems to help users avoid cognitive depletion in real time. This understanding will also enable us to provide feedback to human-in-the-loop systems regarding the fatigue state of the user.

Compression Statistics for Analysis of Streaming Data

Jeremy R. Teuton

We are leveraging metadata generated by compression algorithms to detect and potentially classify events. The value is not in the compressed product, but in the metadata generated during compression for rapid analysis of streaming or very large data sets.

Efficient compression of video uses multiple calculations comparing current to previous to facilitate compression. In this work, the value of these algorithms to speak to specific changes is to be leveraged both with native image/video data and with non-video data sources processed as video. Statistics on what percentage of an image moves, what fraction move in the same direction, contiguous shapes or isolated blocks, novel vs shifted pixels, etc., will be used in data analysis to identify and potentially classify events in streaming data. This use of compression algorithms is a novel approach to enable analysis of streaming data or to speed analysis of very large data sets in real or near real time, without having to write new customized algorithms for every new data set.



An example of how variations in video compression algorithm metadata accurately identify important events in an experiment. Still images called out by our analysis as important and categorized are connected to variations in metadata (a single piece of metadata “frame miscellaneous bits count”) and used for clarity.

This work will provide the basis for a new, flexible, modular tool for streaming data analysis support. Gaining a solid understanding of current strengths and weaknesses of this type of analysis, compared to the state-of-the-art, rule-based video and streaming data analysis, will define both its best use cases and areas for further research and improvement. In

addition, the work offers a strong basis for further development of the toolset and identification or creation of optimal algorithms and should deliver a solid research tool to determine cost benefit tradeoffs in computational power and required training.

In this project, we have validated using video compression software for data analysis/event detection and expanded the initial concept of using video compression software for analysis of data beyond the first use on electrical frequency data, demonstrating its capacity to work with multiple data types. These data sets included scene detection in normal video, event detection in nuclear magnetic resonance data, and automated analysis of electron microscopy experiments that detect the first moment of dendrite formation in a lithium ion battery (the event that eventually leads to failure of the battery).

Data quality can vary drastically by source and type; sensors are limited in quality and degrade due to time and conditions. To access our method’s sensitivity to data quality, a video file with complex scene changes was analyzed at varying degrees of degradation, from pristine to nearly unrecognizable by human eye. Despite being unwatchable with changes significant in exact value, the compression metadata did not change pattern; the method remained able to detect events despite the degradation. These results are critical as we move into use cases where, for various reasons, the data quality will be far less than ideal.

In the project’s final year, we added additional approaches to handle text event detection, as well as tracking identity. We have partnered with the Stanford National Accelerator Laboratory (SLAC) to implement our software, which automates a safety shutdown that currently requires full human attention. We have adapted our approach to handle data

collected using compressed sensing that exploits our capacity to handle very low-quality image data. We have proven the concept of exploiting complex algorithms for data analysis and event detection. We are working to continue expanding our collaboration with diverse partners like SLAC and contacts at the Air Force Research Laboratory.

Co-Simulation Platform for Rapid Prototyping of Control Algorithms

Jeffrey A. Daily

This project is developing a unique tool/infrastructure that will position PNNL as a leader in modeling complex systems that evaluate controls for the interaction of power grids, buildings, transportation, and related domains.

The ongoing modernization of power grids consists of integrating with communication networks to achieve robust, resilient control of grid operations. The typical applications of this emerging smart grid, such as demand response, dynamic reconfiguration, and distributed energy resources employ control systems that operate on mission-critical devices. Beyond the smart grid, control systems are used in additional domains, including building management and transportation systems. As such, these applications of control systems need to be tested rigorously before deployment. Developers usually do not have access to a large test bed; therefore, simulators are used for testing control system applications.

There is a dearth of simulation software available for the intricate coordination and control of power grids, building management and automation, markets, and transportation infrastructure. Further, there are few (if any) tools for developing complex control algorithms within one or across two or more specialized domain simulators. Cooperative simulation of power grid, communication network, and building simulators coupled with advanced control algorithm implementations promises to reproduce more accurately the interactions of these components. Taken together, these domain components exhibit a macroscopic behavior not predictable by examining the individual entities. This is a complex system, and there is an immediate need for the control of such a system. Co-simulation is thus the most cost-effective means of rapid prototyping and testing new control approaches.

The objective of this project is to deliver a general, scalable co-simulation platform for use in various domains. In addition to the direct support of power grid, communication network, building simulators, and advanced control algorithm implementations, our platform will support hardware-in-the-loop capabilities. We are, thus, striving to advance the state of the art in co-simulation environments, focusing on the

needs to support developing complex control algorithms across diverse simulator software packages and hardware.

The culmination of our FY 2015 efforts resulted in the release of our new co-simulation software framework: the Framework for Network Co-Simulation (FNCS) version 2.0. FNCS 2.0 features a new publish/subscribe messaging paradigm that greatly simplifies the experimental setup of a co-simulation. Data streams can be abstractly represented and fulfilled by a simple playback of data, a sophisticated simulator software package, or a hardware signal. The entire system is extensible, connecting arbitrary data sources and sinks. These data streams are fully synchronized by the FNCS 2.0 broker application and can be represented at different time scales, as needed.

A large number of simulation software packages have been modified to use FNCS 2.0. These simulators include MATLAB and, by extension, its many packages: MATPOWER, power distribution simulator GridLAB-D™, network communication simulator ns-3, and whole building energy simulator Energy-Plus. In addition, FNCS 2.0 is fully cross-platform capable, running on Windows, OSX, or Linux systems, or any combination thereof. As a result, we are now well positioned to target the integration of any simulator software on any operating system platform.

Integration with the experiment management system and other projects is well underway. This procedure highlights our success integrating FNCS 2.0 and its related simulators into our envisioned test bed. Though each project can stand on its own, the whole is greater than the sum of its parts. We successfully showcased an integrated demonstration to our internal annual review committee, receiving high marks and praise to continue with our endeavors in earnest.

In FY 2016, we focused on implementing the first hardware-in-the-loop use case, in addition to developing larger-scale use cases in preparation for FY 2017. Supporting hardware-in-the-loop was part of the FNCS 2.0 design since its inception. To that end, we began investigating the incorporation of OPAL-RT in FY 2015. Specifically, we added the Python and C programming language interfaces to aid in the integration. We have always targeted large-scale simulation; however, we have only run on the order of tens of simulators to date.

CyberFit Eddi – Living Lab Infrastructure

Chance R. Younkin

Eddi is a platform for safely removing personally identifiable information (PII) from cyber data, while preserving the structure of the data. This makes cyber security research possible without exposing PII to the researchers, enabling them to make a difference in the cyber arms race while minimizing the risk of inappropriate or inadvertent behavior.

Data sharing at the Lab level, with attention to privacy and security of the data, is an effort yet to be undertaken at any of the national laboratories. Currently, researchers set up projects independently, acquire data in their own way, and build private mini-infrastructures to meet their own requirements. This redundant work is done in isolation and is largely set apart from real-world data, leaving serious gaps between research efforts and operational solutions.

In FY 2016, the Eddi team took on additional requirements and scope by collaborating with a PNNL initiative to create the PNNL Living Laboratory. To bring the teams, the scope, and the technologies together, we gathered requirements, researched solutions, evaluated and selected technologies, and designed Living Lab (LL) 1.0.

Through these methods, the Hortonworks solution was chosen, training for the team was purchased and executed, and the system was designed on the Hortonworks HDP platform. Hortonworks was chosen for its superior approach, solid community, and because it is open source, making it a long-term solution with no required major investments.

Our team played a major role in helping to establish the Data Stewardship Board (DSB) as a PNNL institution for management and oversight of data sharing policies and guidelines. The Living Lab works closely with the DSB to implement the policies and guidelines required to make data available.

Living Lab (LL) 1.0 provides infrastructure support for ingesting, de-identifying, storing, managing, and sharing real-world data. Data sets currently available include firewall logs and host categorization data, as well as cloud telemetry data. The de-identified firewall and host categorization data will be provided to a relevant PNNL project, as will the cloud telemetry data.

This initial release of the Living Lab is a proof of concept designed to secure interest in further research development, as well as to provide additional data sets.

Demonstration of Asymmetric Resilient Technologies for Cybersecurity

Pradeep Ramuhalli

We are integrating capabilities developed through prior related projects to systematically evaluate technologies in a repeatable manner for their impact on asymmetric cyber-resilience.

Traditional cyber security has primarily focused on the strategies of detection, deterrence, denial, and delay of adversaries. This approach allows an asymmetry in favor of the adversary by requiring the defender to successfully respond to each adversary attack, while allowing the adversary to achieve their goal by learning from each failure.

Resilient cyber-systems, on the other hand, ensure mission critical functions of an organization continue to operate in the face of ongoing detrimental events. While several technologies exist that provide elements of resilience (such as moving target defense), there has been limited progress towards an integrated suite of technologies for state discovery and reasoning in cyber-systems that provide insights into decisions that can shift the asymmetric advantage to the cyber-defender.

The research conducted under this project will enable 1) a process for integration of technologies for cyber-system state discovery, reasoning, automated decision-making that takes into account potential adversary intents and associated uncertainties, and actuators that can be deployed automatically (this full process loop is referred to as “DRDA”); 2) evaluation of the integrated suite of technologies in a controlled environment for their impact on cyber-resilience relative to not having resilience technology; and 3) identification of variables affecting cyber-resilience and asymmetry.

The integration of technologies covering the different elements of the DRDA loop first required identification of key information needed by each of the technologies for each stage of this loop. This resulted in a high-level process flow diagram, linking graph models and metrics for cyber-system state discovery to methods for asset dependency and risk analysis for the purposes of reasoning based on the identified system state. The information provided by this reasoning may be utilized by game-theoretic decision-making tools that

incorporate uncertainty (about measurements, system state, and adversary intent) to determine appropriate actions. One or more actuators can then act on these decisions to influence the cyber-system state. This loop runs continuously, and the hypothesis was that such a loop, with appropriate technologies for each stage, would improve cyber-resilience.

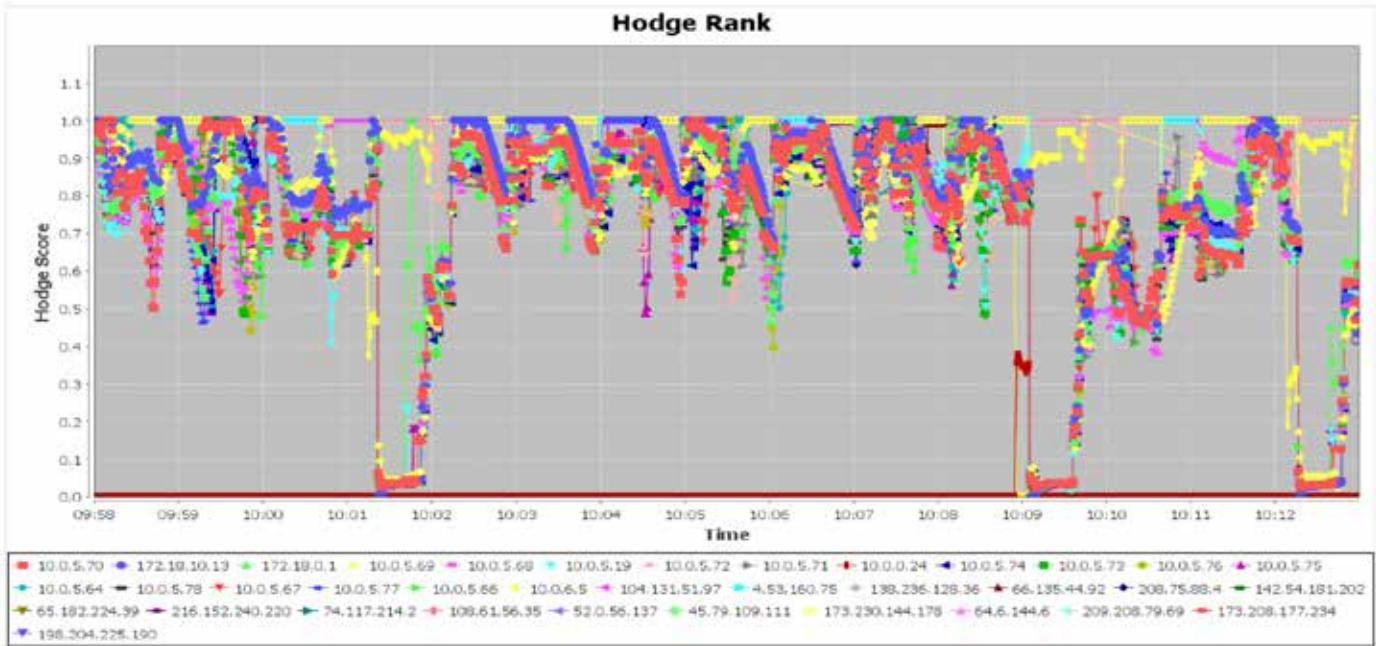
Specific technologies that were integrated during this year of the project were graph metrics based on persistent homology and Hodge rank. These quantities provide a statistical measure of changes to the graph models of the enterprise cyber-system, based on topological considerations and network traffic sources and sinks within the enterprise. Multiple types of simple actuators (for instance, actuators that cycle through different versions of the same application or operating system, and actuators that replace an application with a “gold copy”) were implemented and integrated into the platform. These actuators were specifically selected for their simplicity and their ability to interrupt the cyber kill-chain as early in the process as possible. To facilitate data exchange among these different modules, a Java-based middleware platform—developed through a different, related project—was used.

Initial experiments conducted on a small-scale enterprise (implemented using a set of virtual machines within a test-bed) provided insights into the behavior of measurements (from Netflow sensors) and the corresponding behavior of graph metrics under various types of impediments (e.g., reconnaissance, vulnerability exploits, and compromise of sensitive data). These insights were used to modify the key parameters within the graph models and to inform the decision-making process. In collaboration with the project that was developing these metrics, the metrics were also shown to be capable of identifying specific machines within the enterprise that were contributing to the anomalous behavior caused by the impediments.

A simple but realistic use case was developed, utilizing actual exploits for a specific vulnerability, to help demonstrate the integration of technologies. This initial demonstration showed that, even without identifying the specific vulnerability or exploit, decisions could be taken (and corresponding actuators deployed) that would result in moving the adversary to earlier stages of the cyber kill-chain, while potentially

denying the adversary the ability to learn from the failed attacks. The impact of this result on resilience of the system (defined in terms of the change in mission functionality relative to the perturbation to the cyber-system state, where the state perturbation is computed from the changes in the

graph metrics) and asymmetric advantage to the defender remain to be quantified. This quantification, as well as systematic experiments that incorporate additional technologies for automating reasoning and decision-making, will be performed over the next year.



Example screenshot showing graph metrics (Hodge rank) changes with repeated reconnaissance activity in a small-scale enterprise. Each curve corresponds to a different IP in the enterprise, and the curves are normalized. Time instances where the Hodge rank scores drop to 0 correspond to the reconnaissance.

Developing Signatures that Relate Fecal Microbiome Characteristics with Gastric Bypass Surgery Outcomes

Nancy G. Isern

The methods developed in this project will help us understand the biology of fecal microbial communities in relation to human hosts. These data interrogation tools will be applicable to such diverse areas as biofuel and bioproduct production.

In collaboration with Arizona State University (ASU) and the Mayo Clinic, we are exploring fecal microbiome biomarker relationships using a combination of disparate data types for anticipating patient responses to gastric bypass surgery. Our broad goal is to advance microbiome condition understanding as it relates to energy regulation and patient responses to the most commonly performed bariatric surgery in the United States, the Roux-en-Y gastric bypass. We are probing the relationship between the patient fecal microbiomes and gastric bypass health outcomes, searching for biomarkers or clusters thereof that relate to the composition of the fecal microbiome and patient health status.

Our effort differs from related research in three fundamental ways. First, we are using novel tools and statistical methods developed recently that have not been used previously in metabolomics studies. Second, we are leveraging five different data types, where previous studies have used only a subset. Finally, we possess robust datasets with at least 10 biological replicates per subject group and were able to expand our retrospective study to include new patient samples from ASU that will provide a longitudinal, or prospective, arm of the study, with samples derived from both pre- and post-surgical patients.

We believe that our project represents a unique opportunity to apply these contemporary methodologies to a rich, combined dataset consisting of multiple data types, thus facilitating predictive systems biology not only for our immediate application, but also for other systems biology research.

In total, ASU provided over 80 matched samples for data analysis, which included new samples for the prospective arm of the study. At PNNL, we finalized methods for liquid chromatography (LC)–mass spectrometry (MS) fecal bile acids

analysis and gas chromatography (GC)–MS-based fecal metabolomics, and then we collected data on matched sample aliquots for both the retrospective and prospective arms of the study. ASU provided 16S data and two classes of patient metadata for inclusion in the combined data analysis. Full-scale analysis, therefore, integrated five specific, discrete data types: nuclear magnetic resonance (NMR) 1D metabolomics (PNNL); GC–MS metabolomics (PNNL); LC–MS bile acids determinations (PNNL); parent grant 16S rDNA sequencing data (ASU); and parent grant patient metadata (ASU-Mayo Clinic). Recently, a collaboration was established with another project at PNNL to pursue identification of one of the bile acid unknowns that had a p-value of 0.08 in classification of ASU patient groups, that was also found to be significant in terms of microbiota present in consortia in that project. Our successful effort identified ursolic acid, and we are pursuing the biological significance of this bile acid within our collaborations.

Trelliscope has been used to create user interface visualizations to allow for data to be explored interactively to search for relationships and patterns between the patient fecal microbiomes and gastric bypass health outcomes. We have explored various definitions of surgical success and selected two definitions: the original ASU definition of no more than 20% regain of weight loss, and the definition that we found to be the most effective separator of surgical success, 85% excess weight loss.

We have completed univariate analysis for all retrospective samples using each single data source and recently completed full-scale statistical analysis of the combined datasets. Analysis has utilized PNNL's Tesseract Tools, including datadr to combine datasets, and Trelliscope to visualize and interrogate data. A Bayesian Integrated Feature Finder has been used to identify the analytic sets that can be used to classify each patient optimally into one of the four known patient groups. Results, including a list of significant features that classify groups, have been provided to ASU for biological interpretation. A notable finding of this project was that utilizing only metabolites jointly identified by both NMR and MS resulted in a significant improvement in classification of patient groups.

Due to a funding shortfall, PNNL was unable to analyze the prospective data that was acquired in the course of the project. In order to move the project towards completion, all data, both retrospective and prospective, has been provided to the statistics group at the Mayo Clinic in Scottsdale, who will perform the prospective analysis under the direction of ASU for a publication that is

currently being drafted by ASU. In addition to this ASU-led publication, PNNL envisions a publication elaborating on statistical methods utilized in this study and a possible joint publication with ASU on the biological significance of ursolic acid. ASU has submitted a publication on the initial aspects of the study (covered under a National Institutes of Health supplement) to the ISME Journal.

Digital Currency Graph Forensics to Detect Proliferation Finance Patterns

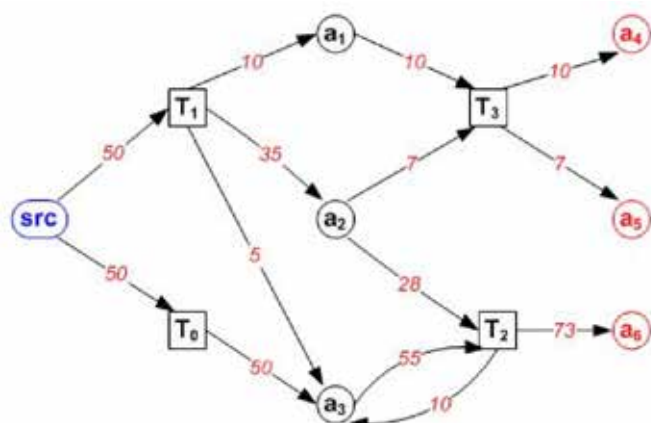
Sean J. Kreyling

We are providing methodologies for identifying patterns of illicit activity derived from large pseudonymous data networks and establishing PNNL's leadership in modeling global-scale, decentralized ledger systems.

Financial systems have matured through digitization with the rise of digital and crypto-currencies (CCs), the most prominent of which is Bitcoin. The attraction of digital currencies to launder or transfer funds for purposes like trafficking nuclear materials poses severe technical challenges to both technologists and analysts. The ability to identify patterns of behavior in publicly available information derived from large anonymous data networks requires novel technologies for graph data management, pattern mining, and modeling. Through this project, we are examining the open and publically available historical data for CCs transactions (e.g., the Bitcoin Blockchain) and attempting to identify patterns of illicit activity.

Last year the team initiated a process to identify relevant available financial datasets, including both traditional (bank or credit card networks) and CCs. We conducted research into various money laundering and money transfer methods and developed semi-formal, conceptual and/or process models for each. We also started to develop formal mathematical models of Bitcoin transactions.

In FY 2016, this led us to focus on the structural properties of the network of Bitcoin transactions. While structured mathematically as a graph, where quantities of Bitcoin stored at pseudonymic addresses are connected as inputs and outputs

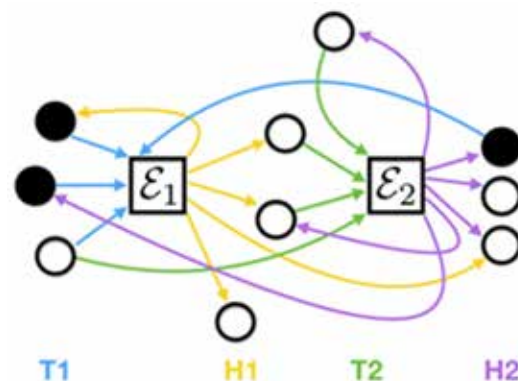


Bitcoin transaction graphs modeled as a weighted, DH (transactions as squares, addresses as circles).

of transactions, reuse of addresses means that a normal graphical structure cannot capture the level of complexity needed. To accommodate this need, we identified a new mathematical structure, being a particular flavor of directed hypergraph (DH). Whereas in graphs, transactions must have single inputs and outputs, in DHs, multiple addresses can serve as inputs and outputs, and addresses can be inputs or outputs to multiple transactions.

Of particular interest is the identification of motifs in DHs, small DH patterns with particular identifying characteristics, usually statistical. Small motifs of sizes three, four, or five are commonly used in network science, but not yet identified in the more complex DH patterns. We have identified the mathematical properties of DH motifs, and some candidate corresponding patterns of potential laundering.

In FY 2016, we also began outreach to the broader Bitcoin communities, including U.S. government sponsors, to identify critical needs. This resulted in liaison to the DOE/ National Nuclear Security Agency project, “Exploring the Potential Application of Blockchain Technologies to International Safeguards,” and a poster at the 2016 Financial Cryptography and Data Security 2016 conference, which provided access to the leading technical innovators in the field.



Simple motif of two hyperedges with Boolean labels.

As we move into FY 2017, both of these opportunities focused us on Blockchain applications outside of the purely financial. In particular, the Ethereum system provides methods for distributed, secure smart contracts involving not only financial exchange, but also arbitrary digital work. The potential for systems like Ethereum to provide disruption to a range of legal and marketing applications involving surety and distributed trust is very great, and our experience in distributed cryptographic networks is positioning us to be at the forefront of technical developments.

Discovery of Cyber/Physical Qualifiers' Relationship and Relevance to Probabilities of Detection/Non-Detection Mitigations

John R. Burnette

We are developing probability metrics for detecting and identifying disruptive, destructive blended cyber-attack tactics; techniques; and procedures for cyber and physical systems.

Critical infrastructure risk assessment approaches have evolved over the decades to assess specific organizational disciplines in an independent manner. Specifically, both physical and cyber security have robust programs to look at risk to a facility, but both focus on their discipline and not on the risk of an adaptive adversary with capabilities in both areas. Further, these security risk assessments are organized and regulated in an equally stove-piped manner, compounding the difficulty of developing an integrated approach to risk management that utilizes physical security, cybersecurity and cyber-physical security. Integrated cyber-physical attacks are seen with increased frequency, and methodologies are needed to assess risk and identify the relationships, touch points, linkages, and qualifiers necessary to mitigate blended threats against U.S. critical infrastructure.

This effort seeks to simulate the cyber-physical environment to capture corresponding data and increase the probability detection, as well as any interconnections between parameters in this environment that may arm defenders with sufficient knowledge to mitigate future attacks. These interconnections could be as simple as lapsed time or a correlation model between physical and cyber events or behavior. The data obtained from the blended cyber-physical use cases will be analyzed to identify the qualifiers for integrated threats and cross domain touch points and linkages for both cyber and physical systems.

Our work in FY 2015 identified the statistical research design for the project, developed the cyber-physical sequence of events for the PNNL Raven cyber range tools, identified the data sources (agnostic equipment and cyber-attack methods for testing the cyber/physical hypothesis), and identified the type of attacker with a “medium” level of sophistication. We looked at patterns in the Closed Circuit Television (CCTV) time

series analysis that correspond to ground truth events. Additionally, other known/ground truth events, such as proximity access with a fake proximity card, were not detected. In all cases, the early stages of this study indicate that patterns can be used as signatures to indicate malicious activity, while more extreme measures may be needed to detect other malicious events.

We also performed experiments related to unauthorized computer shutdowns. Preliminary results identify a simple pattern that can be seen in the time element, whereby an unauthorized computer shutdown can be noted quickly. The elapsed time in seconds of the activity of the administrative computer for each run clearly showed that the computer was shut down and restarted. Given that these particular computers are closely maintained, the unscheduled shutdowns should raise a flag for the system administrator. This factor, corroborated with the time of day and subsequent logon with an account, are definite indicators of abnormal activity.

In another experiment, a “rubber ducky” USB was used to simulate loading malicious commands and scripts to shut down the Industrial Control System (ICS) Human Machine Interface (HMI) by turning off all ICS components. Additionally, the rubber ducky issued a shutdown command to disable the air-gapped ICS further. The ICS HMI Event Logs did not provide information that indicated use of a USB device, such as a rubber ducky. However, a “power off” command could be seen in the ICS event logs that could alert defenders that the HMI was shut down and restarted. The abrupt stoppage of time continuity was a good indicator of abnormal activity, as an ICS is never shut down without prior knowledge, as it controls critical assets that must be operational. A defender could use software-based Intrusion Detection Software tools to monitor for unexpected shutdowns, restarts, and logons, especially during non-working hours.

Our work in FY 2016 identified cyber/physical attack scenario and tested the scenario against three levels of secure facilities—low, medium, and high—to identify overall time delay and system response. The data derived from the tests will be used in conjunction with physical data to validate a cyber/physical simulation model. This information will be used to

provide a better understanding of which type of physical, technical, or procedural security controls are most effective in deterring adversarial actions against U.S. critical infrastructure.

The cyber-physical attack methods utilized for this research project were obtained from MITRE Common Attack Pattern Enumeration and Classification. The adversary’s technical level of expertise was identified as medium for all three facilities.

A research scenario was standardized and used for all three levels of secure facilities by the adversary. The scenario accounted for a security guard stationed at the front entrance, monitoring the CCTV. The adversary would access the facility between the hours of 10:00 p.m. and 4:00 a.m. Random and scheduled foot patrols in conjunction with the security guard located at the front desk were not accounted for in the test scenario.

During FY 2016, there were 16 test-run scenarios completed for low-, medium-, and high-level secure facilities (total of 48 test runs) to establish length of time for an adversary to

complete the scenario. The total time length increased significantly from the low- to medium-level facility at 95% confidence level and decreased significantly from medium to high at 95% confidence level.

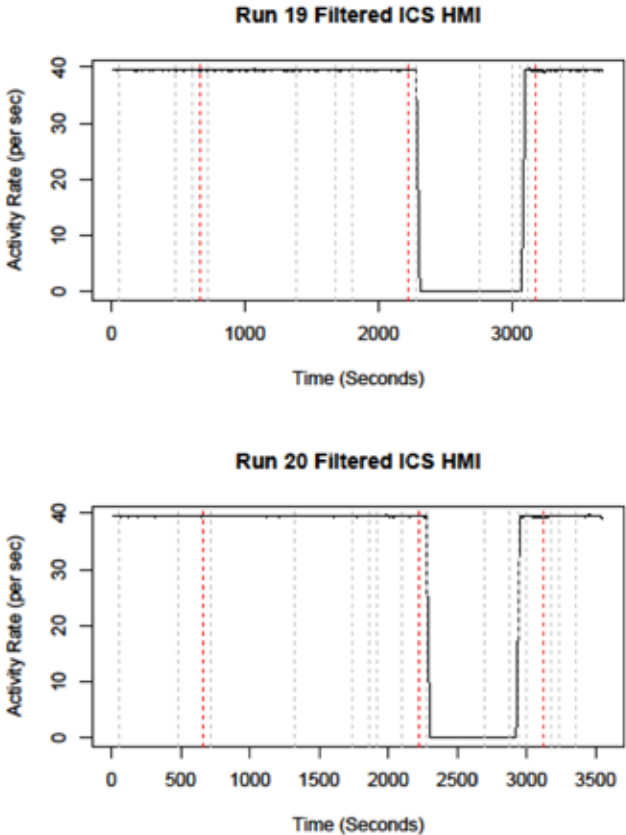
Runs 19 and 20 show the length of time to compromise the ICS HMI for the medium-level secure facility mitigation, as well as the time of day. This is an example of real-time alerts using the UNDERNET. The data is obtained from reviewing the UNDERNET and is consistent across all runs.

We determined that, while the UNDERNET provides much visibility, mitigation is only effective if alerts are built into network systems to alert defense personnel when anomalies occur. Additional training would be necessary for security personnel monitoring stations.

A probability of detection mathematical model can be developed with additional test runs for each security level. The additional test runs will assume independence between phases. An additional 220 cycles of the experiment with 30 iteration combinations of low, medium, and high adversary capability against low, medium, and high security mitigations would derive algorithms of greater than 90% confidence level, which would be used to support probability of detection.

PNNL can use the confidence limits for bin proportion to state the probability of detection. If the adversary is caught in Phase 1, the test runs will continue to Phase 2, 3, 4, etc. Analysis of the data will explain how and why the adversary was stopped from completing the remainder of the scenario, which will increase the confidence level for the mathematical model.

The research data obtained from this study provides preliminary identification for where sensors and system alerts that alert security personnel of a potential network system compromise should be placed. These alert devices include the installation of an UNDERNET to monitor activity that can be seen from “below” to identify anomalous behavior in near real-time.



Runs 19 and 20 show the length of time to compromise the ICS HMI for the medium-level secure facility mitigation, as well as the time of day. This is an example of real-time alerts using the UNDERNET. The data is obtained from reviewing the UNDERNET and is consistent across all runs.

Dorci – The Defender’s Role in Resilient Cyber Security

Roderick M. Riensche

Through Dorci, we are building a foundation of understanding the needs and roles of next-generation cyber network defenders who use resilient infrastructures for a more secure network.

Current research has explored defender roles and responsibilities; however, it has been conducted without the introduction of resilient infrastructures. A foundation of understanding roles, responsibilities, and needs of cyber network defenders can be leveraged by new and existing research in resilient infrastructures to increase utility to defenders, minimize complexity, and assist with the adoption of the new technologies. The object of this work is to advance knowledge about enabling defenders to manage resilient cyber infrastructures without increasing their cost (resources) to do so. As architectures, algorithms, and methods are developed to make networks resilient to attack, the network becomes more complex for the defender to manage. If resilient cyber security research is to tip the asymmetric advantage, it must not increase the burden on the defenders who are understanding and managing the network.

In FY 2014, we started by evaluating the importance of a “current state” study in order to provide context for future work. Preliminary questions to assist with the correlation between previous research completed were leveraged. Initially, we had planned on a scenario-based, semi-structured interview to help users to imagine their environment. Next, we explored use of a tabletop exercise within which users would be given roles and responsibilities, activities will occur, and various options for actions taken. We determined that our study should explore resilient infrastructures but needs to be believable and reasonable to defenders today. While the implementation and adoption of resilient infrastructures is not widespread (which makes technologies difficult to map), we had widespread support from other projects to help ground our research in the plausible domain.

During FY 2015, we concentrated primarily on developing a traditional technology study. Key accomplishments for FY 2015 involved design development of human subjects study and game play framework, creation of a study tracker application to track the study during participation, and additional studies, including two industry, one research, and one government study, along with several pilot studies in order to refine gameplay, as well as study effectiveness. All studies were well-received and generated results for traditional technologies with interesting findings. We also brought in a researcher from Middlesex University to assist our team with research questions regarding next year’s resiliency work. With this collaboration, we were able to establish much of the groundwork necessary to build out our planned experiment with resilient technologies.

In FY 2016, we conducted detailed surveys to gauge expert opinion regarding expected costs and effectiveness of resilient technologies. We used this information to update the game definition that was created during FY 2015 and conducted an additional study (with an academic partner). From this study, we were able to observe how participants compared new and potentially unfamiliar technologies versus traditional technologies and approaches. By completing the game definition, we have built a convenient and engaging platform that can be used to facilitate structured brainstorming exercises when considering potential impacts of future technologies.

Through the course of our interactions with internal collaborators, the PNNL advisory committee pertinent to this project, and external study participants, we identified a path to use the version of the game developed in FY 2015 (i.e., including only technologies that exist today) as a training tool. We have also engaged with groups who are including Dorci as part of a multi-day training curriculum, which will be delivered multiple times in FY 2017.

Estimation of Battery State of Health Using Utility and Literature Data

Vilayanur V. Viswanathan

This project allows the estimation of battery state of health to enable individuals to use large-scale batteries reliably.

Understanding the degradation of a battery energy storage system is key to its deployability in stationary and transportation applications. Although the transition from consumer to large-scale batteries is commencing in the automotive industry, questions about reliability and lifespan still persist. Previous work in this area includes several model types, including empirical and fundamental physics-based, electrochemistry, and equivalent circuit-based, as well as actual capacity loss measurements. In fact, the National Renewable Energy Laboratory (NREL) has developed an empirical model based on curve fitting to tease out dependence of capacity loss on time and number of cycles in the form of coefficients of time^{0.5} (a_1), time ($a_{2,t}$), and cycles N ($a_{2,N}$). However, these choices involved an empirical constant with little justification in the determination of the constant value.

In FY 2015, we developed a model to estimate the remaining energy capacity of a lithium (Li)-ion battery cell or system through an algorithm that estimates resistance increase and capacity loss for the Li-ion chemistry. While the NREL work measured remaining capacity separately, our work directly estimates the remaining capacity from the estimated internal resistance increase based on universal curves that relate capacity loss to relative internal resistance increase. Specifically, our model continuously updates the status of a Li-ion battery state of health.

In FY 2016, we tested Li-ion cells from two different vendors. We determined the degradation of these cells when subjected to Frequency Regulation (FR) grid service, and compared the degradation with baseline cells that discharged at a constant C/2 rate to the same depth of discharge. We also tested these cells by applying an electric vehicle (EV) drive cycle, along with FR and Peak Shaving (PS) grid services duty cycle while the vehicle is not being used. The degradation rate for these cells were compared with those of baseline cells, which were subjected only to the EV drive cycles.

The outcome of this work is to 1) determine the effect of grid services on battery life, and 2) determine the effect of grid services added onto EV drive cycles on battery life.

The Li-ion-phosphate (LFP) cells perform poorly when subjected to FR purely as a grid service or when added to the

Results from 140 battery cycles.

Name	RTE (%/1000 cycles)	Chg (Wh/1000 cycles)	Dis (Wh/1000 cycles)	RMS Dev between cells	RMS Dev from Trend
LFP EV FR	-12%	0.59	-1.45	0.9%	0.1%
LFP EV FR BS	-2%	-0.51	-0.56	0.4%	0.2%
LFP EV PS	-1%	0.03	-0.11	0.1%	0.1%
LFP EV PS BS	-2%	-0.07	-0.13	0.1%	0.1%
LFP FR	-9%	2.98	-2.12	0.6%	0.1%
LFP FR BS	-9%	0.07	-0.07	0.5%	0.2%
NCA EV FR	-1%	0.45	0.25	1.7%	0.2%
NCA EV FR BS	-2%	-0.81	-0.86	0.1%	0.2%
NCA EV PS	-9%	1.71	-0.11	1.0%	0.4%
NCA EV PS BS	-5%	0.53	0.33	0.6%	0.4%
NCA FR	-3%	1.43	0.24	0.2%	0.1%
NCA FR BS	-4%	0.13	0.04	0.3%	0.1%

EV drive cycle. For the first case, the round trip energy efficiency (RTE) of baseline cells and cells used in grid services decreased by 9% after 1,000 cycles. For cells used in EV drive cycle, the decrease in RTE is 12% when FR is used, while for baseline cells, the RTE decreases by only 2% per 1,000 cycles. The LFP cells do not degrade much for the PS service with EV drive cycle.

Nickel cobalt aluminum oxide (NCA) cells perform well in the FR service with and without EV, with an RTE drop of 1 to 4% over 1,000 cycles. When used in PS service, the degradation rate at 9% per 1,000 cycles is nearly double the rate for baseline cells.

The results indicate that the NCA cells do well under shallow cycles (FR and EV FR), while performing poorly at large depth of discharge of 65% (PS service). The RTE rate of decrease for NCA EV PS BS is -5% per 1,000 cycles, while for NCA EV FR BS, it is -2% per 1,000 cycles. On the other hand, the LFP cells have a -1% to -2% per 1,000 cycle rate of degradation for the EV FR BS and EV PV BS.

During the FR service, the LFP cells were subjected to a state of charge (SOC) range of 50 to 40%. The LFP crystals have a high volume change in this SOC range, resulting in poor performance.

Future work will involve investigating the effect of PS grid service. The SOC range for FR service will be shifted to 40 to 30% to avoid the volume expansion. The cells currently undergoing cycling will continue to cycle to further explore the trends.

Fundamental Mathematical Models for Human Interactions

Jennifer Webster

Human community behavior is notoriously difficult to model. There has yet to be a discovery of fundamental principles that govern how humans interact and form communities. This project is looking at mathematical models that address observational and data limitations in discrete systems.

Community detection methods are typically developed on either a theoretical mathematical graph construct with specific properties or a very specific data set. Transferring these methodologies to other real-world data sets can be problematic, as sampling schemes and data collection obscure the true underlying network and introduce a measure of uncertainty into the mathematical formalisms. The goal of this project is to bridge the gap between the theoretical development and the application of network analysis techniques. This will involve development of new community detection methods and the expansion of existing algorithms.

This year, we have focused on three thrust areas: 1) systematic comparison and development of community detection methods; 2) data fusion for the creation of a holistic, evidence-based description of human behavior; and 3) the development of methods for incorporating uncertainty quantification strategies into existing community detection methods.

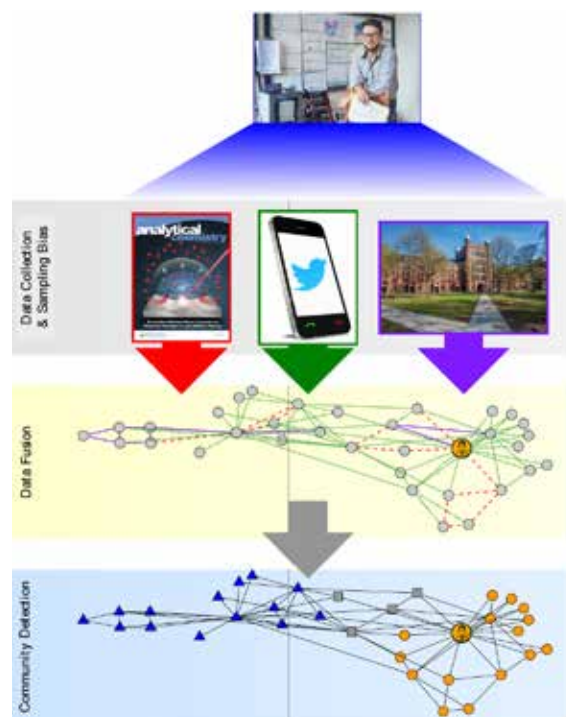
We have cataloged the most common community detection methods and translated heuristics into a single common graph theory framework. The next steps in this process include evaluating strengths and weaknesses of various algorithms and investigating their stability with respect to the various tuning parameters for each algorithm. For the comparison of methods, we have created a collection of classic academic test data sets to complement the theoretical evaluation. This theoretical framework for comparing different algorithms also allows us to determine how classic sampling strategies from the social sciences naturally impact the outcomes of the algorithms. Since many of the sampling procedures are given as a procedure rather than a probability of occurrence, we are investigating percolation theory methods of describing the behaviors of these systems.

In addition to the evaluation of the current state of the practice in community detection, we have been investigating new

community detection methods that allow analysts to use information about the relationships among the communities to improve classification of individual members. For example, it is common in politics to describe the communities as “left, right, and center,” with the understanding that the political identifiers for the left and right are more different than comparisons of either of these groups to the center. Using this knowledge of the communities, we can better identify members of the center group. Community detection methods like Min-Cut treat relationships between all communities equally and, as a result, frequently misclassify community members of the center as part of one of the other two communities.

Project team members spoke at the Society for Industrial and Applied Mathematics (SIAM) Annual Meeting and the SIAM Network Science Conference. We also organized a well-attended minisymposium at the SIAM Annual Meeting. The research team participated in several internal sponsor visits and brown bags at PNNL this year.

Next fiscal year, we will continue our research into the effects of community relations and intend to test our methods on a blind study of data collected from the University of Washington over the last several years.



Bridging the gap between the theoretical development and the application of network analysis techniques.

Geopolitical Discourse Characterization through Deep Learning in Diverse Data Modalities

Elizabeth R. Jurrus

This project will leverage and extend the latest state-of-the-art in machine learning and Deep Learning research to develop algorithms that deepen our understanding of complex narratives in social media data streams.

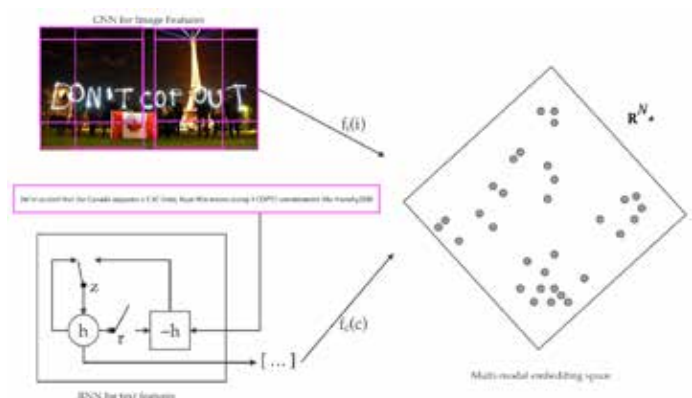
Data analytics has traditionally focused on leveraging subject matter expertise to hand-engineer features to be extracted from data to perform the analysis. This methodology has been very successful in a number of domains, but in recent years, the emergence of deep neural networks (Deep Learning) has produced phenomenal and revolutionary results in a number of tasks, including object detection, automatic image captioning, sentiment analysis, and topic analysis. While Deep Learning has stretched the bounds of what machine learning and data science can do, much remains unstudied about how these algorithms can be leveraged to solve national security, energy and environment, and other DOE-relevant problem sets. In this work, we strive to leverage Deep Learning, utilizing and extending the current state-of-the-art, to focus on addressing the complex challenge of analyzing points of view and narratives present.

The first year of this project focused on the following tasks: 1) development of a DOE-relevant use case for data analysis, 2) identification and collection of social media data, 3) a study of sentiment and emotion classification within social media as it relates to both users and groups, and 4) an evaluation and integration with automatic image captioning models.

The development of a DOE-relevant use case centered around the social media conversation about global climate change. Specifically, we targeted the global climate change conversation on Twitter during December 2015, which coincided with the Paris Agreement, an agreement established within the United Nations Framework Convention on Climate Change that sought to formalize greenhouse gas mitigations within the member states, starting in 2020. This use case was selected because it deals with the public perception of climate change and energy policy. Additionally, a number of

unique and interesting narratives could be drawn from the data supporting this use case. In the data, we found users and organizations who supported the Paris Accords, opposed it, felt it was not going far enough, or who used the media attention to further their own tangentially related cause (such as genetically modified organisms, nuclear energy and nuclear waste disposal, or even topics on the far fringe of conspiracy like chem trails and the presence of fluoride in water).

The data curated to support this use case was primarily from two social media sources: Twitter and Flickr. Flickr was selected because the data on Flickr is almost exclusively visual (some text data is available via tags, image metadata [when available], or comments). Twitter was selected as a data source, as it is a popular vehicle for social conversations. Twitter is an open platform—meaning, unless a user specifically makes their account “private,” that data is public—and one of the main mechanisms by which an individual or an organization shares their point of view with a global audience. In total, 300 thousand images were curated from Flickr and 8.5 million tweets were curated from Twitter. A significant amount of work went into cleaning and preparing the data for analysis.



Depicted is a visual representation of a multi-modal embedding system. Features are extracted from both images (purple boxes represent different image crops, which are then fed through a Convolutional Neural Network to generate a 4096-length feature vector), and the tweet text (represented by the sentence fed through a GRU RNN). From there, functions for the images $f(i)$ and captions $f_c(c)$ are learned, which minimize the error in the resulting embedded space, depicted on the right.

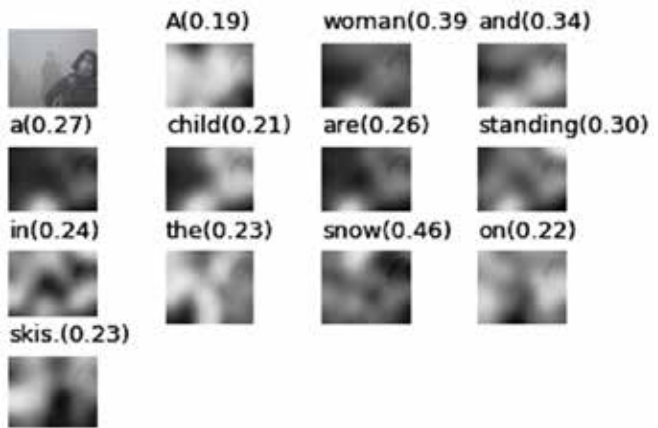
With the data ready for analysis, a study was conducted on the tweet content from the curated data set. We developed a mechanism for determining whether an account belonged to an individual or an organization. We then analyzed demographics, emotion, opinion dynamics over time (1 month), and location to draw correlations between each element, as well as behavioral differences between person and non-person account types. This work provided an in-depth insight into the behavior and opinions around a topic where multiple distinct narratives are present in the data.

Finally, work was conducted on leveraging and extending existing image captioning models. Whereas a typical image classification model will provide a ranked list of labels for an image (e.g., “dog”) describing its content, a captioning model is capable of generating a human-readable, rich description of an image (e.g., “a dog catching a ball in a park”).

While these models are an exciting advancement for both the fields of computer vision and machine learning, there still remains much to be learned about how they function and how they can be applied to aid an analyst. We evaluated a number of models, including a standard multi-modal embedding model, an order-preserving embedding model, and a visual attention-based model. We found that these models can be quite useful if the data it is applied to is well-represented in the training data sets, but that they also struggle to adapt to unique data. For example, if an image mistakenly identifies and generates the word “snow” in its caption, the presence of the previously generated word “snow” heavily biases the model to generate words later on in the caption that are often co-associated with the word “snow,” such as “skies.” Some models would also struggle to properly caption

a test image that is closely related to, but not quite the same as, data over-represented in its training data (e.g., images of people sitting at tables were routinely captioned as an individual sitting at a table “with a plate of food,” regardless of what was actually in front of them on the table).

Despite some of the limitations of the trained models, there remain opportunities for integrating additional information to make the trained models both more adaptable to seldom-seen data, as well to make the generated captions more descriptive. Next fiscal year, research will focus on building visual sentiment analysis models and new caption generator architectures to allow for better, more accurate descriptions. Additionally, work will focus on how to integrate analytics, such as text and visual sentiment, into a captioning model.



Depicted is a visualization of visual attention models for caption generation. The model generates a word with an associated confidence value, as well as a visualization of which locations within the image were being represented by the generated word.

High-Level Modeling Specification for Simulation of Control Systems

Thomas Edgar

We are accelerating the ability for control researchers to define and execute models of complex systems using simulation, emulation, and testbed tools by creating a coherent, high-level modeling specification that abstracts the differences between technologies and domains.

A key aspect of effective tool design is to understand what is required for the user to be successful. In many areas of design and engineering, such an understanding is captured in a pattern language that captures good design practices. While broadly used to teach design skills and communicate successful design, we capture pattern languages directly into tools, frameworks, and literally in high-level specification languages in computer science. When appropriately designed, these specification languages guide their users to successful solutions, as well as lead tool designers to features that reduce the cost and improve the reliability of designs, even when made by comparatively novice users.

Arion, the name assigned to this project, is a library of objects and capabilities built from the Java programming language to support specification of complex systems of interacting components. The primary use case is to enable the modeling of complex systems with control mechanisms while abstracting the different technologies and methods the simulation and experimentation tools use to execute the model. In the development of new control mechanisms for complex systems, it is common to use simulators or emulators to run experiments to evaluate the behavior of the control mechanism. While there is a range of strategies, Arion focuses on the scenarios in which the following situations occur:

- A system is described as a collection of components where each component has internal states, consumes input signals, and produces output signals.
- Once elaborated, a system may be simulated to determine how the state of the system evolves over time.
- Each component is implemented by an execution host, typically a discrete event simulator or possibly a hardware emulator or hardware embodiment.
- Assumes a coordination framework of some sort exists that mediates the global notion of time and implements inter-host communication.

We anticipate the need to generate hundreds of thousands to millions of components to form a system with multiple experi-

ments that vary the structure of the system. Today, such experiments are often run by using a programming environment, such as MATLAB® or Python, to generate input configurations for discrete simulators. Arion subsumes this step in the workflow and provide more cohesive integration, and eliminates the necessary expertise in all of the configuration and specification languages of each simulator or emulator.

An Arion script is a program that elaborates system components, their communication interconnections, the set of execution hosts, and the mapping of a component to a host. After this elaboration, the implementation of Arion generates suitable configuration information for each execution host, the coordination framework, and the computing infrastructure. We refer to this step in the workflow as compilation to distinguish from simulation. Components may be specified as external components (those coming from the existing tool libraries) or Arion components (those components defined directly in the Arion language).

In FY 2014, the Arion team took the initial steps in developing this library of objects and functionality. Our first-year goals were accomplished. The basic domain-specific language concepts were developed, and a prototype tool was created. Use cases, including a representation of the AEP gridSMART® Real-Time Pricing Demonstration project, were identified, and basic components and methodologies were extracted. GridLAB-D™ library components were added to the language to bootstrap using prior experience and extending this to a new test bed.

In FY 2015, the GridLAB-D™ library of components was made more complete. In addition, components for ns-3 were developed and added to the library to support CDMA (Ethernet like) and Wi-Fi connections. Also, the ability to compile configurations for the FNCS 2 co-simulation platform and infrastructure definitions for the Experimental Management System was created. Leveraging all of these new capabilities, a complex model replicating the AEP gridSMART project was creating and successfully tested.

In FY 2016, three things were accomplished: development and integration of common models and a capability to parse existing GridLAB-D™ models, the ability of the Arion tool to define an appropriate communications model based on a power model and control system design, and the ability to integrate external models by mapping inputs and outputs. In the final year of the project, the process has been started to open source the technical components of this project to cultivate a community and ensure the capability continues to live and grow beyond this work.

High-Throughput Genome-to-Metabolome Computational Methods for Microbiome Metabolomics and Modeling

Ryan S. Renslow

This project's goal is to develop a high-throughput metabolite-identification approach based on a quantum chemistry, super-computer-driven software engine for application in complex microbiome metabolomics.

How do we identify the preponderance (99%) of metabolites if standards do not exist or, critically, if molecular structures are not even known? Currently, metabolomic tools are limited by the ability to build libraries of compounds (e.g., chemical shifts for nuclear magnetic resonance [NMR], or collision cross sections [CCS] for ion-mobility spectrometry-mass spectrometry [IMS-MS]), which are required to identify and quantify metabolites. The premier NMR metabolomics tool, Chenomx, has a library of about 900 compounds with about 11 new compounds added per year, and one of the largest open access databases, the Biological Magnetic Resonance Data Bank, has data for about 1,200 compounds.

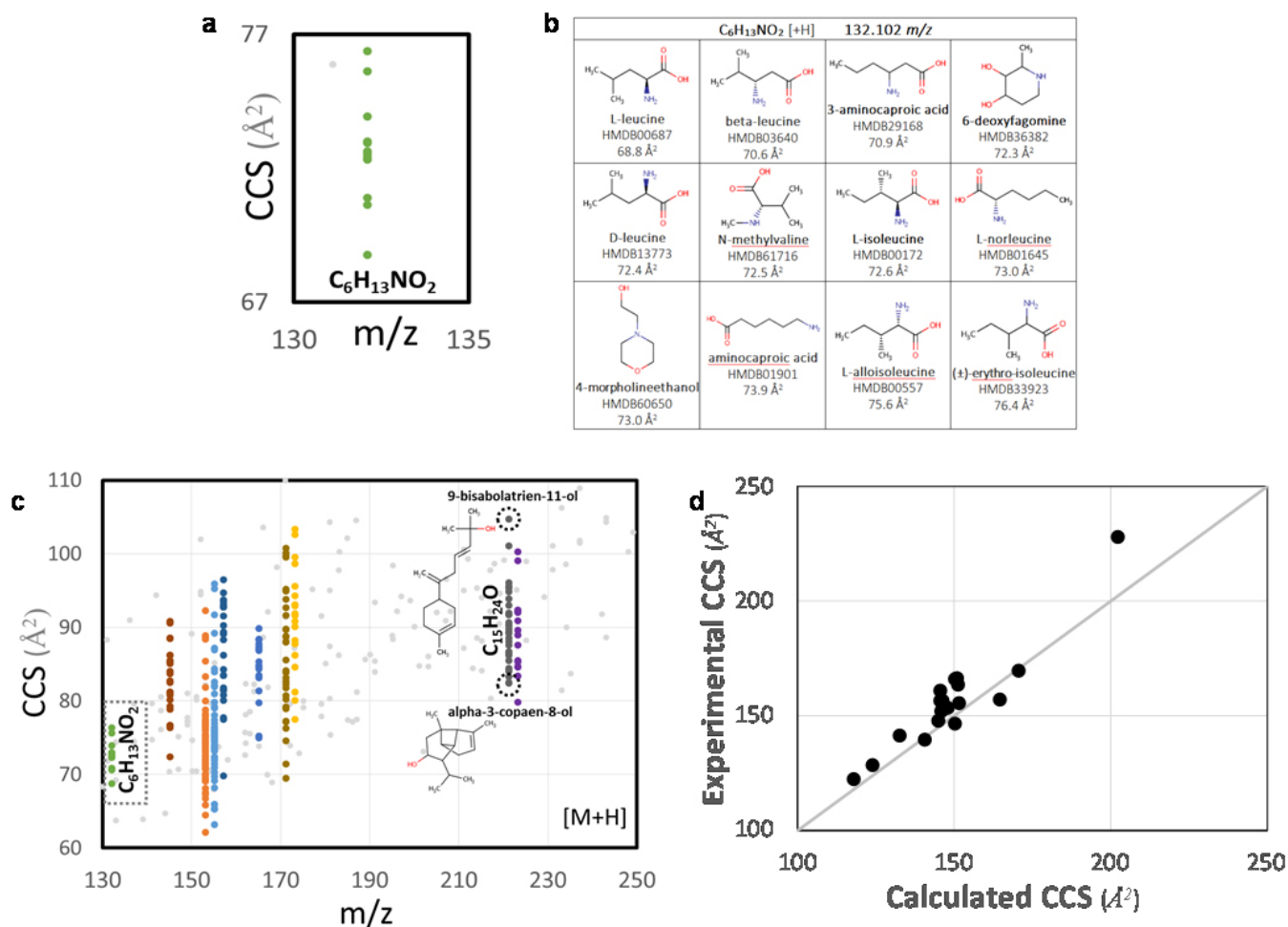
In both of these libraries, most compounds are derived from urine and human health-related samples. Currently, there is no robust environmental- and microbial-community-focused NMR metabolomics library, and well over 90% of the known metabolites in BioCyc [4, 5] are not found in an accessible library. To achieve DOE's goal of understanding, predicting, and controlling complex microbial communities, and to understand microbiomes undergoing transition, the capability to comprehensively identify and quantify metabolites is necessary.

Quantum chemical calculations have been used to predict NMR chemical shifts and IMS-MS CCS of metabolites; however, to date, these calculations have been limited to small sets of molecules. Datasets are typically arbitrarily chosen, particularly in the case of NMR, and most simulations do not emphasize compounds common in environmental sources or microbial communities. Furthermore, most simulations and current databases for metabolomics do not consider molecular conformers or custom solvent conditions. Our proposed novel computational research will expand the current sets of simulated molecules by one to two orders of magnitude and

provide a streamlined process for generating relevant metabolite libraries from microbial genomes (or community metagenomes). Currently, there is not a unified method for generating reaction/pathway lists with corresponding metabolites to build a library for identification and quantification.

This past year, we developed a supercomputer-driven pipeline to predict NMR chemical shifts and IMS-MS CCS values for any metabolite automatically, rooted in quantum chemical calculations. The software accurately calculates NMR chemical shifts of rigid body metabolites with user-selected quantum chemical methods. It also handles solvation effects around a solute molecule under any given solvent via computationally efficient implicit models, which can provide a reasonable description of the solvent behavior and enables understanding of biological, chemical, and environmental processes. The performance of the software has been tested successfully on metabolites in currently accessible databases. Another program to handle conformers automatically (via classical molecular dynamics and quantum chemical-based *ab initio* molecular dynamics) is being developed by our team, which is intended to be a part of our NMR and IMS-MS metabolite prediction pipeline.

The current lack of a unified method in the literature will be overcome in the next fiscal year via a benchmark study for quantum chemical theories. Our automated program will provide calculation of NMR chemical shifts of metabolites for a set of three to four dozen methods. Once a unified method is determined, conformers will be considered to decrease the error of our calculations compared to experimental results. Custom solvent conditions will be investigated for the water-solvated compounds common in environmental microbial communities. We hypothesize that the current implemented implicit solvation models fail to account for key chemical bonding features when water is the solvent. As a result, explicit solvent models will be placed into our automated pipeline to account for the behavior of water-solvated condensed phases with more accurate predictions and improved understanding of the physical processes. Our proposed research is expected to identify currently unknown metabolites and, overall, to build a metabolome library specifically for microbiomes and environmental samples.



Performance of the molecular modeling and quantum chemical calculation pipeline: a) a plot of calculated CCS versus m/z for molecules shown in b), illustrating the degree of orthogonality of the two metrics; b) metabolites from the Human Metabolome Database; c) a plot of CCS versus m/z for 200 metabolites, out of >11k solved structures obtained in an initial test run. Colors correspond to CCS for different metabolites with identical molecular formulae. The data for $C_6H_{13}NO_2$ from b) is outlined in the dashed box, and two anthropogenic chemicals with the formula $C_{15}H_{24}O$ are shown to have very different CCS; and d) theoretical CCS values for 11 small molecules show good agreement with experimental values.

Image Analysis Using Active Learning on Shape and Texture Features

Elizabeth R. Jurrus

This goal for this research is to produce an improved method for categorizing images from a data source given a small set of example images. Applied to nanoparticle images, our new interface offers a method for scientists to review results of scientific experiments rapidly and correlate hypotheses with outcomes.

Forensic analysis of nanoparticles is often conducted through the collection and identification of electron microscopy images to determine the origin of suspected nuclear material. Manually inspecting large image datasets takes enormous amounts of time. However, automatic classification of large image datasets is a challenging problem due to the complexity involved in choosing image features, the lack of training data available for effective machine learning methods, and the availability of user interfaces to parse through images. Therefore, a significant need exists for automated and semi-automated methods to help analysts perform accurate image classification in large image datasets. Our research, encapsulated in Instinct, is designed to quickly organize image data in a web-based canvas framework.

We demonstrate the nuclear forensic applicability of this work using particle images from the Capstone Depleted Uranium Aerosol Characterization and Risk Assessment Program. This dataset consists of a large set of backscattered electron

images from scanning electron microscopy of aerosol samples collected during perforation of vehicles with depleted uranium munitions. Our goal for examining the images of the particles is to determine the particle morphology, especially in the nano-size range. This morphology provides insight into the relationship between the chemical formation, the solubility, and the dissolution rates present during their formation.

Our Instinct framework was developed to enable efficient organization of images through the integration of a web-based interface, state-of-the-art image feature vectors, and a new clustering technique. The user interacts with the Instinct interface through a light-weight web client application, which provides a virtual canvas for organizing images and enabling different users to collaborate on the same datasets. The canvas displays images represented by a 4096-dimensional feature vector, computed from a convolutional neural network. To visualize these high-dimensional feature vectors, we attempt to map them into a 2D space while preserving their class relations.

The method we implemented to perform dimensionality reduction is an adaptive spatialization method. First, the user-selected images are clustered and “anchors” for each group are computed. The positions of the remaining images are updated in the 2D spatialization using a coordinates determined by the similarity of the images to these clusters. Similarity is measured by the angle between a feature vector and the low-dimensional subspace created from the anchor.



Example cluster of particle image. (Left) Image data with three user-created clusters of images. (Right) Layout of the image data after the distance-to-anchor diffusion map update.

The relationships between unlabeled images and small, user-selected clusters of images are visualized via diffusion map in the canvas. Our method for representing the clusters is advantageous because, in the case that the visualization does not represent all of the semantic information recognized by the domain expert, the visualization can be globally updated by moving a small number of samples closer to or further from the clusters.

The work presented here represents an exploration into an adaptive data visualization technique through the representation of high-dimensional spaces. Our results appear to reflect semantic relationships in a way that is hopefully valuable to human operators. We envision this tool as a unique processing capability for triaging large quantities of images or simply qualifying groups of similar particles.

Impediments

David O. Manz

Cyber security testing and evaluation is currently a manual and expensive process. This effort will develop capabilities to automate and improve the reproducibility of cyber security testing.

This project will characterize and reproduce threats and vulnerabilities on the basis of a network with real-world traffic to increase accuracy and resiliency. This effort will be used in testbeds and other realistic sandbox environments that are safely contained from operations.

Traditional network analysis projects or penetration tests have been limited to non-realistic networks composed of static servers, workstations, firewalls, etc. The testbed used for this project comprised powerful Red/Blue Team tools and network traffic generating workstations to rationalize a real network. Eventually, this project will result in a design framework with resiliency that detects and resolves unknown attack patterns and unpredictable threats.

This effort moves beyond the static networks of point solution testbeds and adds the dynamic and co-evolutionary nature of sophisticated adversaries engagements. There are no one-offs in the real world, and in a small way, this project introduces that complexity to the laboratory environment.

The current year was spent in preparation stage by researching state-of-the-art Red/Blue Team practices, real network traffic generating agents, and testbed configuration. Not only using the given tool itself, we also modified and implemented some applications (e.g., Metasploit attack automation tool).

For Red Team simulation, we installed exploitation tools running on the Xenial server and optimized the cloud network. Security Onion, Etherape, TNV, Netgrok, Kali, Core Security, Mutillidae, Virtual Hacking Lab, WebGoat, and Metasploitable are running on the testbed. Furthermore, attack automation application written in Python code will be used to increase complexity and efficiently send malicious packets to the testbed. Intrusion detection systems such as Snort and Bro are installed and successfully detect malicious packets.

The preparation/configuration stage is almost finished, and we are going to configure all applications and tools that we can use and implement under legitimate license into the testbed and simulate Red/Blue Team scenarios with real traffic in the next fiscal year. Based on the results and the strengths and weaknesses from each experiment, a new design of framework with enhanced security and resiliency will be designed.

This will culminate in an extensible framework for adversarial hazards action to be used in scientifically reproducible and validated, useful generation of datasets and experiments.

Integrated Adaptive Resilient Asymmetric Data Security

Jian Yin

This project developed an approach to cyber security that significantly shifts advantage toward cyber defenders and makes it possible to protect against zero-day security exploits.

Data confidentiality becomes increasingly important as more valuable and sensitive data are used by many essential online web services. It is desirable to build the capability to achieve resilient data confidentiality and integrity, in addition to availability in distributed environments. This capability can allow us to counter a wide range of security threats, including zero-day attacks and insider threats. One of the distinguishing requirements is resilience. That is, we can prevent leaking of sensitive data or subverting integrity even when hackers compromise parts of the systems.

The traditional approaches for cyber security are mostly reactive and labor intensive. Cyber defenders must intensively monitor and analyze the system using various tools to detect and counter attacks. Once an attacker breaks into a system, at least some aspects of availability, confidentiality, and integrity are compromised.

In our approach, we can continue to provide data availability and integrity for mission-critical applications and protect confidentiality of highly sensitive data even when attackers manage to break into or hijack a part of system. Even though there are some very recent commercial systems that are designed to achieve resilient availability in the face of machine crashes or network partitions in distributed environments, including cloud computing environments, none of those systems address resilience against malicious attacks. If this project is successful, the technologies and mechanisms developed in this project can be highly useful to DOE and other U.S. government agencies who have highly sensitive data and mission-critical applications to protect. Our approach significantly shifts advantage toward cyber defenders and makes it possible to protect against zero-day security exploits.

This year, we completed implementation of our proposal algorithm and systems. Additionally, we explored and evaluated various techniques to improve performance, which

included offering a range of tradeoffs between security guarantees, performance, and using distributed schemes. We demonstrate these techniques of improving performance with experimental evaluation.

We have completed the development of algorithms with associated secure proofs of these algorithms, which allows us to establish a blueprint for implementation. We have shown that it is theoretically impossible to use only homomorphic encryption to provide a sufficient set of operations for data in common real-world applications. To solve this problem, we extended and combined homographic encryption, Byzantine fault tolerance, and dynamic adaption. We have developed a set of algorithms and refined it for efficiency. Our algorithms are based on learning with error in polynomial rings. Trap-door functions are used to allow zero knowledge verification, which reduces both space and time complexity. We have implemented the comparison circuits and chose one that offers lowest time complexity.

We also observed an interesting tradeoff between time complexity and space complexity: with a little bit of pre-computing and increased space complexity, we can reduce time complexity. When we detect increasing threat levels, we would like to increase the threshold number of machines that an attacker must break into to compromise data confidentiality. We devised a scheme that, as long as the number of machines that an attacker has broken into is less than the current threshold, the system can adapt to increase the threshold and data confidentiality is preserved.

We have produced a set of experimental measurements that quantify the overhead introduced by our proposed solution and determined the set of acceptable overhead thresholds, which establish the targeted level of performance to make our scheme practical. We investigated a few techniques in improving efficiency of resilient privacy assured computation. Privacy assured computation relies on ciphertext packing in reducing overhead. With ciphertext packing, permutations are often needed to rotate the positions of plaintexts for them to interact with each other. Permutation of plaintext positions is mapped to permutation of coefficients of polynomials that use ciphertext packing to encode the plaintexts.

Moving data around not only introduces overhead, but can also cause unaligned memory access and pollutes caches. We developed the mechanisms of virtual permutation to address this issue. Virtual permutation includes two mechanisms. One of the mechanisms is indirection, which allows us to translate memory access to a different location. Another mechanism change references algorithms themselves. With virtual permutation, we can reduce data movement, which can reduce memory access overhead (the major overhead in resilient privacy assured computation). We also examined how to exploit application-specific data semantics to improve efficiency. We designed a protocol to allow dynamically changing the threshold in face of increasing security threats

and developed a scheme that allows us to mix ciphertexts with plaintexts. This scheme reduces the amount of memory or disk space that needs to store the data and computation overhead. Moreover, we also reduce the increases of noise level. By reducing noise, we can reduce the numbers of bootstrapping and further improve efficiency.

We have published a paper in a workshop sponsored by DOE Office of Science Advanced Scientific Computing Research, Advanced Scientific Computing Research (ASCR), which allow us to provide input for future calls for proposal by ASCR.

Future work could include identifying some potential applications and deploying our algorithms to those applications.

Modeling Continuous Human Information Processing

Leslie M. Blaha

We seek models and related metrics for dynamic decision-making to support the development and evaluation of adaptive interfaces for interactive streaming analytics.

Analyst behavior in a streaming visualization environment demands online, continuous decision-making. Modeling human decision-making, in general, requires formal mechanisms to characterize information accumulation and choice biases.

Models of decision-making, particularly from the human information processing perspective, are capable of teasing apart the mechanisms involved in the decision-making process based on non-invasive behavioral data, namely response choice and response time. Existing models capture discrete decision events in static environments.

To capture continuous decision-making, we seek to extend information processing models of decision-making to capture both the discrete decision events and the cognitive dynamics for waiting and monitoring in between explicit decision events. This requires theoretical extension of response time modeling approaches, incorporating exploration of the correlations between sequential decisions. We additionally want to test novel measures of performance based on both total response time and motor dynamic of executing the response. The full dynamic profiles provide insights about the decision-making as the process unfolds. We will develop appropriate metrics to enable use of these dynamics to inform and evaluate the design of analytics interfaces.

The first step of the research was to evaluate models of decision preparation dynamics as candidates for modeling the cognitive behaviors between specific decision-making events. We identified a successful candidate model of decision anticipation, which predicts the likelihood that an event requiring a decision and response will occur at different points in the foreperiod (the time prior to the onset of the stimulus or event cue). The model captures probabilities of cuing events occurring as a function of the time since a response or an alerting event indicating the start of a decision-making trial.

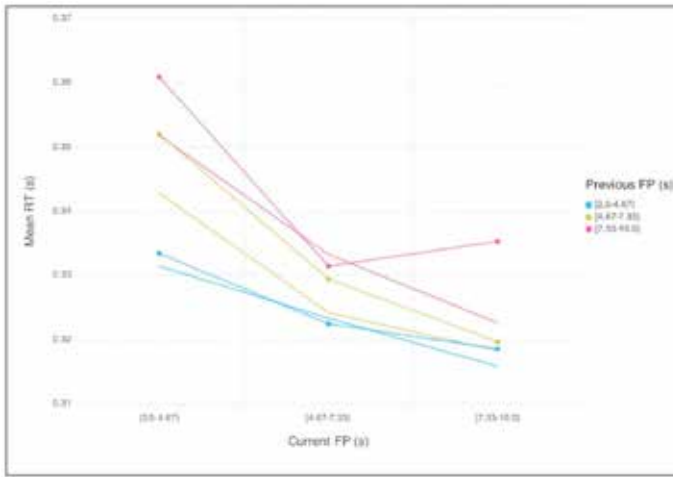
Model simulations show that the model predicts a series of important response time effects, including the foreperiod effect, in which the duration of waiting time affects the speed of responding. Preliminary model fits have been conducted against the average data values from a continuous detection task requiring sustained attention, the psychomotor vigilance task.

We have made additional, parallel strides in the use of Gaussian process models for functional analysis of hand and eye movement behaviors. A new functional version of Fitts' Law is the result of modeling hand movement data, which we have demonstrated on touchscreen interaction data for 2D movements. Related models of eye movement dynamics are in development.

To support the study of dynamic time series of data simulated or captured by these models, we collaborated with the User-Centered Hypothesis Design program to develop a storyline visualization technique for eye movement data. This technique leveraged dynamic time warping for aligning data from multiple human observers viewing movie clips.

Over the coming year, we will integrate our initial decision preparation model with sequential sampling models of the explicit decision process. The goal is to create a continuous cognitive model that captures all cognitive information processing from the start of a continuous task until the end. We will test this model on a continuous even detection task. If successful, we will extend it to dynamic multitasking, requiring human observers to respond to multiple, threaded simple decision tasks in a dynamic environment. We will further extend the storyline visualization to incorporate the time series statistics and metrics derived from the Gaussian process models in order to capture eye and/or hand movements in the dynamic continuous cognitive tasks.

The results of this work will provide a new set of models, metrics, and visualizations that can be used to characterize user performance in interactive streaming analytics tasks. This will advance the science of usability evaluation, while also enabling the study of the cognitive mechanisms of interactive streaming analytics decision-making.



Empirical and predicted mean response time data based on parameters from preliminary model fitting for continuous detection data. Data show the sequential effects for short, medium, and long inter-trial intervals. Lines with points give the empirical data; plain lines give the best fitting model predictions for each foreperiod (indicated here as FP) length.

Module Integration Interface for Resilient Cyber Systems (MiiRCS)

Jeffrey L. Jensen

This project focuses on integrating research (and possibly other third party) applications to operate in various stages of the cyber decision cycle, as well as the development of actuators, or pieces of software that are specifically executed to perform certain actions deemed advantageous for the defender.

MiiRCS is a specialized software integration architecture that provides secure data transport, sharing, and routing between cyber security applications that act in a larger integrated solution to achieve greater resilience in the enterprise while facing attack.

The project focuses on two areas supporting the demonstration and measurement of a resilient enterprise. The first emphasis is on integrating research (and possibly other third party) applications to operate in various stages of the cyber decision cycle. These applications perform numerous functions that can support resiliency and mission preservation while under attack. There are several common enterprise integration patterns from which MiiRCS has adopted an asynchronous messaging architecture using Java Enterprise Edition and the Java Message Service application programming interface (API). This process allows potentially any of the research applications to integrate in a loosely coupled, distributed fashion to overcome issues surrounding remote communications such as unreliability, latency, and availability.

The second area is focused on the development of actuators, or pieces of software that are specifically executed to perform certain actions deemed advantageous for the defender. Most of these actuators are designed to work in a virtual environment, taking advantage of APIs to control virtual components of the enterprise, and some can operate in a traditional network, as well.

During FY 2016, a version of MiiRCS was created using WildFly 10 to integrate a netflow collector, two topological graph models, and four actuators. MiiRCS was deployed in an enterprise environment, modeling a fictitious company named

ViSR, using OpenStack cloud computing technology. MiiRCS supported several experimental campaigns designed to produce data sets and exercise multiple topological data models. The automation and collection capabilities provided by MiiRCS allowed for fast, repeatable, and controlled experimental executions to be performed and analyzed on a regular basis. This deployment was used at the annual review to demonstrate a full cyber kill chain where an adversary performed reconnaissance, exploitation of vulnerability, root ownership of a ViSR server, and exfiltration of sensitive data. Four actuators were available during the live demonstration and were invoked at various stages of the kill chain, forcing deviations in the adversary's strategy and ultimately protecting the enterprise.

Models based on two different areas of graph theory were integrated into MiiRCS to act in the orient phase of the decision cycle. One of the research models is based on Persistent Homology, a method for computing topological features of a space at different spatial resolutions. To define that space, vectorizations of various netflow attributes were constructed to form n-dimensional point clouds over time. The other research model is based on Hodge theory. It ranks the importance of data sources and sinks and looks for ranking instabilities over time. Both operate on netflow-based data that were previously collected and stored during the observe phase of the decision cycle. The collection of netflow data was accomplished using a combination of tcpdump scripts, CouchDB databases, and a Node.js service.

Eight actuators were developed during the year, and the most appropriate (four) were deployed for the scenario demonstrated at the annual review. These included software to reboot selected machines, adjust firewalls at the host and network level, modify user access levels, create machine snapshots, quarantine machines from the network, recycle and replicate machine snapshots, and operating system (OS) rotation. A human in the decision cycle was used to interpret the orient models' output and invoke actuators.

In FY 2017, MiiRCS will support four quarterly demonstrations similar to the single demonstration given at the end of FY 2016. With each experiment, new technologies will be evaluated using the scientific method. MiiRCS software

architecture will continue to mature as new components are integrated. The human in the decision cycle will be supplemented with a new software component based on game theoretic research. In addition, the existing research models for topological graph analysis will be adapted for streaming processing of netflow records as opposed to sequential batch processing. Other sources of observable data will be considered, such as OS process and system log records. These models will continue to be developed and evaluated against other threat types.

Post processing of model outputs will be improved for better understanding by the human and data sharing with other system software components. Realizing the possibilities of integrating other technologies are endless, MiiRCS will focus on a generalized approach for software component integration and data exchange to support resilient cyber systems. An API and set of guidelines will be created to establish a “best practices” approach, which researchers and other software developers can consider when designing new algorithms that operate in the decision cycle for resilient systems.

Multiscale Modeling and Uncertainty Quantification for Complex Non-Linear Systems

Alexandre M. Tartakovsky

Our research is creating novel dimension reduction methods for large-size, non-linear systems of deterministic and stochastic equations, potentially leading to an explosion of results in mathematics and various engineering and science applications.

We have developed multiscale models for complex non-linear systems. Our general idea is that one needs to exploit, as much as possible, the inherent structure of a problem before engaging in computations. Features such as time and/or scale separation, variable and component dependencies, and randomness should be used as guides to reduce the actual computational work. At the same time, the need and usefulness of the elegant formulations that encompass many different cases should not be ignored: they allow us to view the big picture, make connections between seemingly unrelated subjects, and serve as starting points for controlled approximations.

Stochastic basis adaptation for uncertainty quantification.

In this work, we have addressed the curse of dimensionality by using the basis adaptation representation of random space for subdomains of interest. We demonstrated that the adapted space can be significantly smaller (in terms of the number of random variables) and the governing equations can be solved much faster than the original problem. The resulting solution is highly accurate within the subdomain of interest and is less accurate outside of the subdomain. If solution is needed in the entire domain, then it can be discretized in several subdomains, and our approach can be used to obtain solutions in each subdomain. Finally, the solution in the entire domain is obtained as a superposition of the subdomain solutions. In this problem, the operation count to obtain the exact solution is 8761 but is only 1541 in our approach, while the maximum error does not exceed 5%.

Smoothed particle hydrodynamics (SPH) model for high-energy-density lithium (Li) batteries. Dendrite formation on the electrode surface of high-energy-density Li batteries causes safety problems and limits their applications. We developed an anisotropic diffusion reaction model to study the anisotropic mixing effect on dendrite growth in Li batteries using the SPH method to model dendrite growth in an aniso-

tropic electrolyte solution. Several parametric studies of dendrite growth in an anisotropic electrolyte were performed, and the results demonstrated the effects of anisotropic transport on dendrite growth and morphology, showing the possible advantages of anisotropic electrolytes for dendrite suppression.

Incompressible smoothed particle hydrodynamics (ISPH) for large-scale multiphase flow simulations. In this work, we implemented a multiphase model into a consistent second-order ISPH multiphase code built on top of the massively parallel particle library LAMMPS and the linear algebra library Trilinos. This code allows largest-to-date, incompressible SPH multiphase flow simulations. We are using the SPH method to study stability of the triple-fluid interfaces.

Non-local model for multiphase flow. The main advantages of the non-local model for multiphase flow that we developed is that there is no need to compute interface curvature (to impose the Young-Laplace boundary condition at the fluid-fluid interface) and contact angle to impose a boundary condition at the fluid-fluid-solid interfaces. To test this model, we have implemented it in a finite-volume MATLAB® code. Currently, we are implementing it in the massively parallel open-source code OpenFOAM.

Multiscale hierarchical stochastic model for advection-diffusion-reaction systems. We developed a novel hierarchical model for solute transport in bounded layered porous media with random permeability. The model generalizes the Taylor-Aris dispersion theory to stochastic transport in random layered porous media with a known velocity covariance function. In the hierarchical model, we represent (random) concentration in terms of its cross-sectional average and a variation function. We derived a one-dimensional stochastic advection-dispersion-type equation for the average concentration and a stochastic Poisson equation for the variation function, as well as expressions for the effective velocity and dispersion coefficient. We observed that velocity fluctuations enhance dispersion in a non-monotonic fashion: the dispersion initially increases with correlation length λ , reaches a maximum, and decreases to zero at infinity. Maximum enhancement can be obtained at the correlation length about 0.25 the size of the porous media perpendicular to flow.

Projection formalism analysis for polymer looping. Polymer looping concerns the study of the approach of the ends of a polymer so that its two ends come within a certain distance (called the capture radius). The problem of predicting the mean time for the looping of polymers is important for theoretical but also practical reasons. For example, different functions of a DNA chain (which can be considered a polymer) are determined by the looping of the ends (or even internal parts) of the chain. Even though the first theoretical models for polymer dynamics, namely the Rouse and Zimm models, appeared more than 50 years ago, the development of techniques to accurately predict the looping time is still an open problem. There have been proposed several asymptotic theories that operate on different regimes (e.g., length of the polymer, capture radius) and under various assumptions concerning the timescales during which the phenomenon takes place. Together with a PhD student from the Applied Mathematics Department at the University of Washington, we are in the process of establishing the first complete derivation of the different asymptotic theories based on a projection formalism. In particular, we aim to show that the multitude of asymptotic theories in the literature are actually different limiting cases of one theory. To do that, we use the Mori-Zwanzig (MZ) projection formalism, which has been previously used to construct reduced models for systems of differential equations.

Analysis of deep neural network training and deep reinforcement learning. We have started exploring the mathematical reasons behind the success in training deep nets. In particular, we have provided evidence that the main reason deep nets can be trained efficiently resides in the fact that the large number of hidden layers allows the network to learn slowly the features of the data. This promotes the correlation between the different layers of the network, which in turn allows the training algorithm to discover a low-dimensional feature manifold and stay on it. The result of such a process is the significant shrinking of the (usually astronomically large) number of possible network weight configurations to something manageable, with moderately large sample sets. As part of our analysis, we have also discovered novel architectures for reducing further the computational time needed for training.

Improved single noisy image super-resolution. In collaboration with non-PNNL scientists, we have developed a unified framework for performing image super-resolution and denoising based on wavelet domain dictionaries. Within this framework, problems with missing high-frequency subbands are handled simultaneously with denoising. We have proposed two trained models: one based on a wavelet analysis dictionary and one on a wavelet learning dictionary. Experimental results on a variety of noisy images demonstrate the effective-

ness of the proposed algorithms. Our methods outperform the state-of-the-art super-resolution methods both in quantitative and visual results, especially in high-density, noisy images.

Mori-Zwanzig reduced models for uncertainty quantification. The MZ formalism has become, in recent years, one of the most popular methods for constructing reduced models for systems of differential equations. It has been applied to various types of systems, from molecular dynamics to fluid dynamics and tracking of singularities of partial differential equations. In recent work, we have applied the formalism to the problem of constructing reduced models for uncertainty quantification (UQ). In particular, in the construction of reduced models for systems of differential equations resulting from polynomial chaos expansions of solutions of differential equations depending on uncertain parameters and/or initial conditions. The most difficult part of constructing a reduced model is to obtain an accurate representation of the memory term. In the course of applying the MZ formalism to UQ, we have devised a new reformulation of the memory term, which involves a hierarchy of differential equations that can be solved in tandem with the reduced model to result in a more efficient, yet accurate, representation of the memory. This reformulation allows the clear distinction between cases of short and long memory and treats them differently, so that one can utilize simplifying features that can appear in these different cases of memory length.

Improved conditional path sampling algorithms. The problem of conditional path sampling is central in different contexts, ranging from filtering to the sampling of paths between metastable states of molecular systems. The inherent difficulty with conditional path sampling is that, in many applications, such conditional paths have a low probability of occurring. Thus, whether one opts for straightforward Markov Chain Monte Carlo or for evolving a system of (stochastic) differential equations, the computational time needed to sample an adequate number of such paths in order to obtain converged statistics can be extremely long. In previous work, we have devised a homotopy method for the sampling of such paths, which starts from a modified system for which it is easier to sample such paths, and then slowly morphing the path so it conforms with the original system dynamics. This algorithm has been applied to the difficult problem of multi-target tracking, as well as to path sampling between potential wells. Together with a different PhD student from the University of Washington, we have initiated a more detailed study of the underpinnings of the algorithm and how it can be improved. In addition, we have been collaborating with the Chemical Engineering Department at the University of Washington to apply the algorithm to sampling of metastable states of different protein systems.

Multiscale Modeling Using Particle-Based Methods

Wenxiao Pan

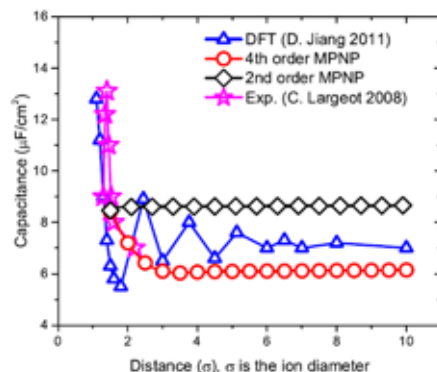
This project focused on the development of numerical schemes for multiscale modeling of systems characterized by multiple time and length scales. Specifically, we studied ion transport and its coupling with hydrodynamics, which has a broad impact on applications associated with various electrochemical systems such as fuel cell, flow battery, capacitor, and others.

To the best of our knowledge, there is still an absence of any analysis conducted using a consistent multiscale modeling approach, particularly consisting of modified mean-field continuum models and atomistic material chemical and structural properties. Moreover, the prior studies were restricted to small-scale computing, and they increased scientific knowledge and understanding for these application systems in some degree, but are still very limited in their predictive capabilities. The developed computation methods in this project can be extended to investigate multi-physical and multiphase transport processes, and the highly scalable implementation allows for large-scale high-performance computing of real systems and, hence, serve to guide design and decision-making for the development of next-generation technologies.

In FY 2016, we systematically studied the numerical scheme for solving fully coupled Navier-Stokes (NS) and Poisson-Nernst-Planck (PNP) equations. In particular, we discussed at mesoscale the effects of ion size and ion correlation that are described by modified PNP (MPNP) equation with finite correlation length.

A multi-time stepping approach was explored for the time integration of the coupled equations with different time scales.

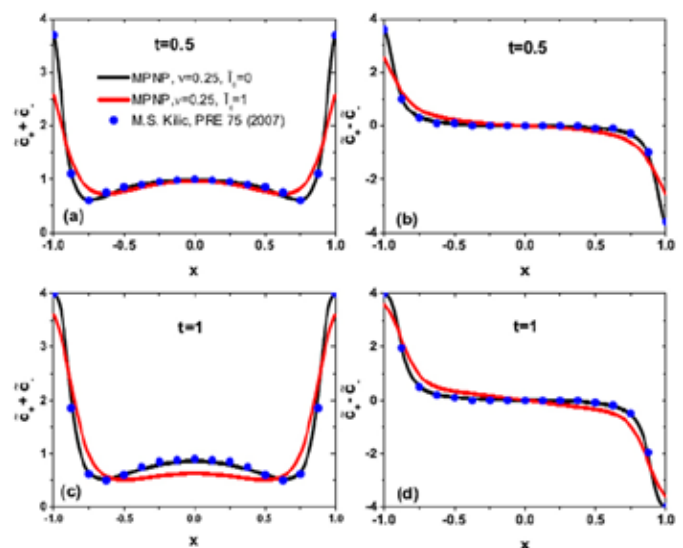
Steady state ion distribution. We investigated the electrostatic double



EDL capacitance in real porous electrodes of capacitors.

layer (EDL) capacitance in real porous electrodes of capacitors by numerically solving the modified Poisson Boltzmann equation with ion size and correlation effects. We demonstrated the ion correlation effect is important to capture the EDL structure in nanometer length-scale pores. The simulation results were compared with both density function theory predictions and experimental data.

Unsteady ion transport. We examined how unsteady ion transport is affected by ion size and correlation. The finding is important to augment the literature studies about ion transport predicted by original PNP equation without taking into account ion size and correlation effects at mesoscale. The numerical results were validated by literature data.



Unsteady ion transport with ion size and correlation effects at different times.

Ion transport coupled with hydrodynamics. The fully coupled MPNP and NS equations are numerically solved to capture charge inversion and flow reversal in a nano-channel electro-osmotic flow, which was first examined by molecular dynamics (MD) simulations. We compared our numerical prediction with the MD simulation results and also investigated the impact of correlation length on the electro-osmotic flow. In addition, we examined ion size and correlation effects on the electroconvective instability and chaotic hydrodynamics.

NOUS: Incremental Maintenance of Knowledge Graphs

Sutanay Choudhury

This project seeks to develop a large-scale knowledge graph system that will continuously learn from data added to it, which will reduce the expense and time domain experts spend constructing and maintaining these graphs.

The term “knowledge base” has been around since the early days of artificial intelligence and expert systems research. One may think of knowledge graphs as knowledge bases storing facts about inter-related entities, where entities may refer to people, organization, location, or even words, scientific topics, etc. Knowledge graph construction and maintenance is an expensive process involving manual curation by domain experts. Such approaches become infeasible as data sources grow in volume and variety and motivate the need to explore automated approaches that scale with these data sources.

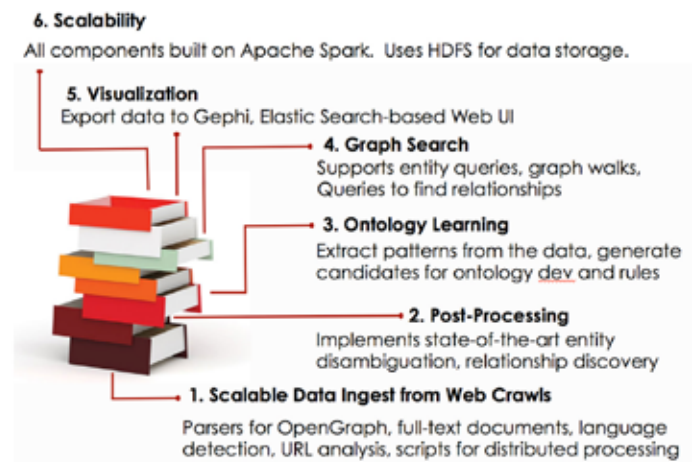
Data-driven applications critically depend on human experts who learn about a given domain through their interaction with data over time: the expert gains experience and familiarity with the domain. The surge in data volume, throughput and diversity pose newer challenges for training experts. Codification of domain requisite knowledge and its persistence for classification and reasoning is critical.

We are focusing on three science questions: 1) How do we learn the ever-evolving set of patterns or rules for a domain from a data stream? 2) How do we determine the veracity of facts that arrive in a stream? 3) How do we infer new facts by observing the stream of information?

Work on this project so far includes development of a scalable prototype that incrementally constructs a knowledge graph from large text-streaming data such as the *New York Times* and *Wall Street Journal*.

In a pilot exercise earlier this year, we demonstrated how our system could monitor large-scale data from the internet and find emerging trends. We processed millions of web pages collected from drone-related sources and showed how our algorithms can find new trends, such as autonomous drones, and narrow down the information to manufacturers of the product and its components.

We also submitted a software invention disclosure for the system prototype and released our work as open source to the community. We continually discover new users beyond PNNL that find this work extremely relevant and useful.



Example of how the knowledge graph process works.

Proteomic Signatures of Wild versus Lab-Adapted Pathogens

Eric D. Merkley

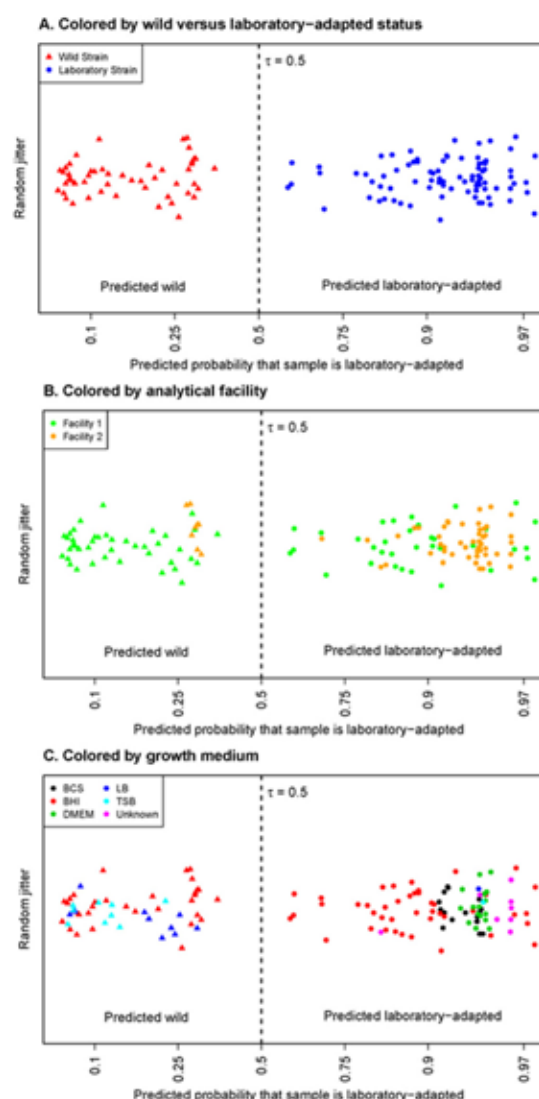
We have developed an approach for mining archived mass spectrometric proteomics data and applied it to discover signatures that distinguish wild from laboratory strains of *Yersinia pestis*.

Category A Select Agent bacterial pathogens are all endemic in certain environments, and distinguishing naturally occurring organisms from laboratory strains remains a challenge for the biodefense community. We recently isolated the Select Agent *Yersinia pestis* from fleas collected in two distinct regions of the United States and serially passaged multiple subcultures of each isolate in a standard rich medium. We discovered that there were consistent changes in protein expression among the passaged descendant strains that, surprisingly, were *not* obviously associated with underlying genomic changes. These protein abundance changes were evident, even though both the wild isolates and their descendants had been cultured in the same laboratory growth medium prior to analysis, which suggested that proteomic data might provide a general signature of laboratory adaptation. Our project was a test of that hypothesis.

For this effort, we took advantage of the archived data at the proteomics facility located at the Environmental and Molecular Sciences Laboratory (EMSL) at PNNL, which has, over a period of years, performed mass-spectrometry-based proteomic analysis for numerous studies, addressing diverse biological questions and targeting hundreds of different organisms. We extracted all *Y. pestis* proteomics data collected on non-fractionated samples from the EMSL archive, combined it with our data from the previous study, as well as from our own unrelated proteomic studies of *Y. pestis*, for a total of over 381 datasets collected on 137 independent cultures. We developed an approach for using quantitative as well as presence/absence data and tested whether proteomics data could differentiate wild isolates from the laboratory strains by developing logistic regression classifiers (LRCs) with the data.

We showed that, despite different genetic strains, growth media, growth temperatures, growth stages, sample preparation procedures, and analytical instruments, protein abundance data could reliably differentiate wild isolates from

laboratory-adapted strains. In addition to pointing towards a direct signature of laboratory adaptation in *Y. pestis*, our work also suggests that proteomic data (both protein identification and abundance data) from unrelated experiments and facilities can be combined and mined to garner useful information. The ability to mine mixed, archived datasets for multiple signatures would be a boon to biomarker discovery efforts in many contexts.



Output of the LRC to distinguish wild from laboratory-adapted strains using relative protein abundance data. Each symbol represents the prediction of the LRC for an independent culture. Triangles represent cultures of wild strains. Circles represent laboratory-adapted strains. The horizontal axis value is the predicted probability that a culture is laboratory-adapted and is non-linear; points are separated vertically in a random fashion to improve the visualization.

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Rendezvous: Optimization and Stochastic Algorithms for Asymmetric Resilient Infrastructure

Samrat Chatterjee

By developing a framework for uncertainty quantification of attacker payoff utilities in this project, we enable the application of game-theoretic methods for improving the resilience of cyber systems.

Cyber security is a multifaceted, challenging research problem. Defenders typically function within resource constraints, while attackers operate at relatively low costs. Design and development of resilient cyber systems that support mission goals under attack must also account for dynamics between attackers and defenders. Game theory provides a mathematical framework to model and analyze dynamic interactions between multiple strategic decision-making agents (e.g., attackers and defenders). Game theory has, therefore, been applied to solve several cyber security problems, such as intrusion detection and network interdiction, within non-cooperative settings. However, the practical application and scalability of game-theoretic methods is significantly limited by uncertainties in attacker payoffs and accurate identification of system states.

Game theory is the mathematical study of the interaction between two or more entities acting to achieve their individual goals. Game-theoretic methods have been successfully applied to solve several challenging problems related to cyber security. Various taxonomies for classifying game-based modeling approaches exist. These game formulations contain assumptions about rounds of game plays, past player actions, types of players, number of cyber system states, number of player actions in a given system state, and payoff (reward or penalty) functions associated with player actions. However, a significant limitation of current approaches is the lack of rigorous treatment of uncertainties in payoffs and system states. A thorough investigation of uncertainty quantification methods applied to cyber security games will, therefore, benefit the cyber security research community and help advance the state-of-the-art in this domain.

In a realistic setting, a defender cannot assume that all necessary information—both about the attackers and their own system—will be available. Since a cyber attacker's payoff generation mechanism is largely unknown, appropriate representation and uncertainty propagation is a critical task. One must also account for the lack or absence of perfect cyber system state information; such uncertainties may arise due to inherent randomness or incomplete knowledge of the behavior of or events affecting the system. For example, partial observability may make a cyber system's state uncertain over time. Moreover, multiple types of attackers could potentially target a system at a given point in time.

Advances in state-space modeling of cyber systems and reinforcement learning approaches for Markov decision processes have inspired the development of partially observable stochastic games (POSGs) and their potential applications for cybersecurity. Solving such problems involves iteratively finding policies (that map system states to actions) that achieve high rewards, on average, over the long run. However, POSGs are very general formulations and become intractable.

We previously developed a probabilistic framework for quantifying attacker payoff uncertainty within a stochastic game setup that accounts for dependencies among a cyber system's state, attacker type, player actions, and state transitions (received a best paper award in cyber security at IEEE-Homeland Security Technologies [HST] 2015 conference).

This approach adopts conditional probabilistic reasoning to characterize dependencies among these modeling elements. The application of probabilistic theories (such as total probability theorem) and functions (such as marginal and conditional) may then lead to simulation of attacker payoff probability distributions under various system states and operational actions. The framework is flexible and accounts for multiple types of uncertainties—such as *aleatory* (statistical variability) and *epistemic* (insufficient information)—in attacker payoffs within an integrated probabilistic framework. Statistical probability distributions typically address aleatory uncertainty, while mathematical intervals address epistemic uncertainty. Depending on these representations,

uncertainty propagation methods may include Monte Carlo sampling analysis, interval analysis, and/or probability bounds analysis. Application of uncertainty propagation techniques generate probability distributions, intervals, or intervals of distributions associated with attacker payoffs that serve as critical inputs within stochastic cybersecurity games. These probabilities may be informed and updated based on empirical event and system data, simulation experiments, and/or informed judgments of subject matter experts.

The game-theoretic and uncertainty quantification methods outlined above model the dynamics between cyber attackers and defenders, and have real-world potential to address proactive resource allocation challenges within resilient cyber systems. However, challenges to their implementation exist, including real-time, data-driven system state determination, “realistic” payoff uncertainty representations, and scalability of uncertainty propagation and stochastic game algorithms.

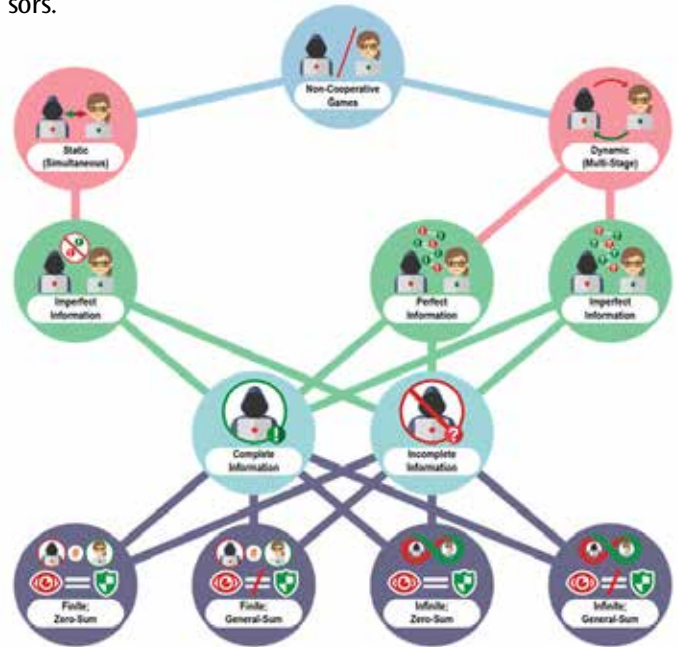
In FY 2016, we focused on four research areas that led to the development of rigorous mathematical formulations of the optimal defender strategy problem and potential solution approaches. The first focus area involved computational experiments (using a notional cyber system), with the representation and propagation of attacker payoff uncertainties (using two-phase Monte Carlo sampling and probability bounds analysis) as statistical probability distributions, mathematical intervals, and distributional bounds. This resulted in an article in the National Cybersecurity Institute journal and a conference paper at the 2016 IEEE-HST conference. Our journal article was also nominated for the 2016 Best Scientific Cybersecurity paper completion organized by the National Security Agency.

The second focus area was on the development and application of approximation algorithms for real and synthetic attack graphs to characterize cyber system vulnerability and criticality. This resulted in our second conference paper at the 2016 IEEE-HST conference.

The third focus area involved the development of a stochastic network interdiction problem with probabilistic attack graphs as a two-stage stochastic mixed integer linear program. The goal of this formulation was to generate optimal interdiction plans that minimize the expected maximum risk from a cyber attack. The sample average approximation scheme along, with Benders decomposition technique, was implemented to solve the mixed integer program. This resulted in our third conference paper at the 2016 IEEE-HST conference. Finally, we developed a formal approach for resilient reachability based on end system route agility in the context of

network security. This involved formalization of efficient and resilient End to End (E2E) reachability problem as a constraint satisfaction problem, that identifies potential end-hosts to reach a destination, while satisfying resilience and quality of service constraints. This resulted in a fourth conference paper for the Association for Computing Machinery 2016 Workshop on Moving Target Defense. We also published a second (invited) article in the *Society for Industrial and Applied Mathematics News* journal focusing on game theory and uncertainty quantification for cyber defense applications.

In FY 2017, we plan to continue our work in three areas. Our first focus will be on the design and development of an agent-centric decision-making approach to formulate the cyber defense problem as a partially observable Markov decision process (POMDP). The output of the POMDP solver will be recommendations for optimal cyber defense responses. Secondly, we will integrate data in POMDP format from other services under the ARC initiative. Finally, we will integrate a prototype implementation with ARC Technologies for resilience. We will also continue collaborations with our academic partners and develop research proposals for external sponsors.



Types of non-cooperative game models for cybersecurity.



Probabilistic attacker payoff framework.

Robust Statistical Data Exploration and Analysis for Microbiome Metabolomics

Lisa M. Bramer

As capabilities to detect and quantify abundances of small molecules grow, the need for integrative and robust statistical methods for exploratory data analysis, data normalization and quality control, and biomarker discovery are needed for impactful and interpretable biological research. This project aims to improve analysis methodologies, develop a robust and reproducible biomarker discovery process, and improve the quality and reduce the time to reach meaningful results integrating multiple heterogeneous data types.

The microbiome response to perturbation is first observed as changes in the metabolome, followed by changes in the metatranscriptome and metaproteome, and ultimately, to the metagenome in the case of a chronic perturbation. However, the analysis of metabolic data from complex microbiome communities poses several challenges: multiple analytic platforms are available, each with unique capabilities and differences in run-time variability and, therefore, varying data analysis requirements.

The ability to perform exploratory data analysis (EDA) is the critical first step in analyzing such data. EDA is of paramount importance for the purposes of assessing data quality, determining relationships among explanatory variables, assessing relationships between explanatory and outcome variables, and making a preliminary selection of appropriate models. Metabolomics data from a microbiome has unique challenges for the EDA process. The biochemical relationships between variables (metabolites) is often complex and in the presence of other confounding variables, such as censored or missing data; traditional statistical metrics (e.g., correlation) are insufficient; and data quality procedures become extremely difficult, sometimes resulting in large amounts of data going unused or being underused.

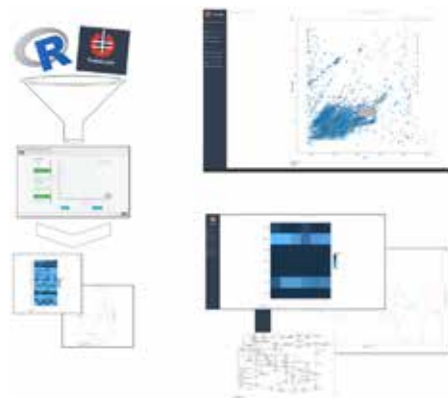
This project aims to modify existing methodologies and develop new ones for EDA, data quality control (QC), and normalization to identify important biological relationships based on multiple sources of omics data. These methods and algorithms developed will be deployed in an interactive framework, allowing researchers to actively investigate data in a manner flexible to multiple data types. Finally, this project aims to be scalable to large amounts of data that are

produced in biological experiments. It will do so by using a divide and recombine paradigm that allows for high-performance statistical computing and integration into high-powered visualization tools that can be combined with statistical metrics.

In FY 2016, we developed a streamlined framework for data management that is generalizable across data types. This structure was developed in the form of an R package, providing the framework for reproducible research and automated reporting and tracking of data filtering, normalization, etc. A data QC package for metabolomics, proteomics, and lipidomics was released, and a corresponding publication was submitted and is under review. Additionally, a data QC package for analytical platforms generating count data (e.g., metagenomics, 16S amplicon, etc.) is under development. Several new methods are under development for sample outlier detection and normalization, as well.

We have collaborated with several microbiome research projects at PNNL to test the developed packages and methods data generated from their experiments. These collaborations led to several invited talks and publications. Finally, we developed functionality for analyzing and visually exploring data generated by PNNL's Fourier transform ion cyclotron resonance instrument, which is utilized across many biological experiments. These functions were implemented in an interactive environment, allowing the researcher to effectively explore and interact with large amounts of data.

In FY 2017, we will finish the development of two R packages currently under development for the visualization of multiple data types and robust statistical tests and models. These packages and new methods included in them are expected to lead to several publications. Additionally, we will focus on models for integrating heterogeneous data types generated by biological experiments, while still providing statistical results that are interpretable by biologists. Finally, we will work to implement developed methods into our established interactive environment to facilitate collaboration with domain experts.



Interactive data visualization and EDA framework developed for integration of heterogeneous data sources.

Scalable Feature Extraction and Sampling for Streaming Data Analysis

Abhinav Vishnu

The objective of this project is to design a set of large-scale, parallel sub-sampling algorithms for execution on cloud computing systems and supercomputers.

Today, we are producing exorbitant volume of data using mobile devices, network sensors, scientific simulations, and other instruments. Machine Learning and Data Mining (MLDM) algorithms, which analyze these data and produce useful information, such as classification models, are critical in science domains (such as distinguishing between signal and background for identifying the Higgs Boson particle) and cyber security, regardless of whether a network system is under attack. With increasing volume and velocity, several attempts to design parallel MLDM algorithms have been proposed.

However, not every data point is equally important. Usually, only a subset of data points define the model, yet most approaches use the entire dataset for model generation using MLDM techniques. In addition, these approaches are geared toward batch processing of data, even though we are ushering in an era in which data velocity is increasingly the primary issue. Upon in-depth analysis, we observed that there is a gap in parallel algorithms that incrementally sub-sample a high velocity stream and learn the model iteratively over the lifetime of the stream.

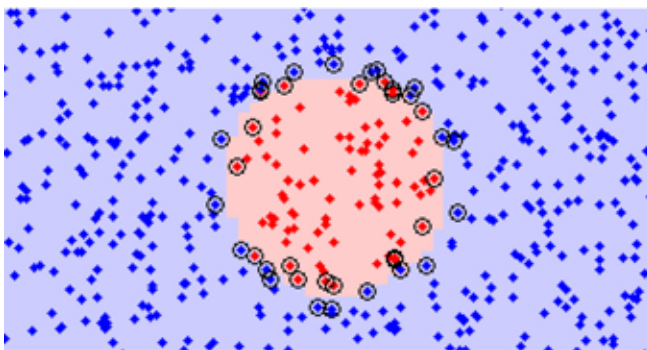
Our project will provide a mechanism to address data velocity by learning model only on a subset of data, while still gathering nearly accurate information. Our research in feature extraction and sampling by using parallel computing systems will create a fundamentally new approach for analyzing very large datasets with potentially minimal accuracy loss. In addition, our project will develop techniques to automatically select a subset of dimensions, which are useful for learning the model. The project outcome will provide better models

for science domains, human behavior, and cyber security. A specific expected outcome is an open source software that would be used by scientists and other researchers for faster and better scientific discovery.

We expect to build faster MLDM algorithms by using smart mechanisms for sample elimination (sub-sampling) and executing these algorithms using computational features of architectures available today. As an example, modern systems consist of graphics processing units and Intel Many Integrated Cores, which are rarely used in conjunction with the system

core (such as Intel Haswell) and high-performance network. We intend to build MLDM algorithms, such as Support Vector Machines (SVMs), clustering algorithms, and Deep Learning algorithms, which are still in their infancy in terms of large scale. We expect that the impact of the existing project will transcend national laboratories and academia.

In FY 2016, we have conducted research on designing scalable Deep Learning algorithms, which can execute effectively on supercomputers, cloud computing systems, and are built on widely used systems including Berkeley Caffe and Google TensorFlow. Our research included methods to scale these implementations using Message Passing Interface (MPI), which makes it suitable for supercomputers. We have also conducted research on sample elimination with Deep Learning algorithms, techniques to adaptively prune neural network while training the network, and novel algorithms for fault tolerant, frequent pattern mining and SVMs for handling the increasing fault rate in supercomputers. We have also conducted research in applying these techniques to several domains, which had not considered Machine Learning. Specifically, we conducted joint research with SLAC National Accelerator Laboratory for applying Deep Learning on their use cases; applied particle detection techniques for Higgs Boson particle detection; better localization for underwater acoustic telemetry, and generated high-fidelity models for fault modeling of extreme-scale applications on supercomputers.



Our developed parallel SVM algorithm, processing samples and reducing the working set by observing a pattern.

In FY 2015, we made significant strides in designing large-scale MLDM algorithms, which iteratively reduce the overall computation while maintaining the accuracy of the proposed solution. As an example, we developed a novel parallel SVM algorithm that eliminates the unnecessary samples from the computation. While several sampling algorithms reduce the cardinality randomly, the proposed algorithm processes the samples and iteratively reduces the working set by observing the following pattern, readily observed in many real-world datasets. As observed in the figure, only a few samples actually result in defining the boundary between blue and pink points. The proposed algorithm and its implementation using MPI with evaluation using up to 4096 cores shows its efficiency. Specifically, we evaluated the proposed algorithm using Higgs Boson dataset, and we observed that, in comparison to the algorithm that does not conduct any elimination, our proposed algorithm can reduce the overall execution time by 2 times. We conducted similar evaluation with other real-world datasets in many domains, and the results were similar.

We also designed and developed a novel parallel algorithm for frequent pattern mining. We observed that the algorithms proposed in the literature conclude that load balancing and communication are the primary problems with the frequent pattern mining algorithms. Specifically, we considered the FP-Growth algorithm, now considered the default frequent pattern mining algorithm due to its low time and space complexity. We developed a work-stealing algorithm using MPI one-sided model and created a new method for merging distributed trees such that the overall communication time reduces by up to 38 times. We will be presenting this work at an IEEE conference.

For FY 2017 and beyond, we plan to continue to pursue this research by collaborating strongly with academia and industry. In addition, we are working strongly with application domains and national laboratories, such as SLAC National Accelerator Laboratory, who would leverage the research and software produced for deployment on production systems.

Scalable Hierarchical Validation & Calibration for Robust Distributed Control of Large-Scale Complex Systems Under Uncertainty

David W. Engel

This project will develop a general framework and toolkit that provides a systematic treatment of the uncertainties involved in the control of complex systems.

Many state-of-the-art control techniques rely on accurate predictions of the physical processes involved in complex systems. Such a prediction can be achieved with computer simulations of a detailed mathematical model. However, when some aspects of the model (e.g., property parameters, initial and boundary conditions, etc.) are not known exactly, the prediction is subject to uncertainty. Making control decisions without considering the uncertainty in the model prediction could cause unexpected control results. In contrast, making robust control decisions and designing systems requires a quantification of the uncertainties in the model.

Approaches for validation, calibration, and uncertainty quantification (UQ) that are data-driven may be capable of providing estimates of uncertainty that are time-varying as the quantities being measured vary with time. Such a capability provides the option of adjusting acceptance criteria and, potentially, setting points in a time-varying fashion to meet the needs of the next generation of complex systems.

The majority of control systems do not adequately quantify the uncertainties that occur within complex systems, such as the power grid. The identification and quantification of different sources of uncertainty is needed, since it leads to an estimate of confidence about the results of a control strategy. To help with the design of robust control strategies, our goal has been to incorporate UQ techniques to develop a general toolkit in MATLAB® for uncertainty quantification, optimization, and model calibration and validation; discover the major sources of uncertainty and quantify how much error/uncertainty we currently have in our control feedback; and determine efficient addition of sensors to reduce uncertainties and achieve more accurate control.

Despite the relatively simple mathematical formulations involved in a UQ analysis, the UQ problems are generally

difficult to solve. One cause of this difficulty is that the simulation model and the measurement model in many real applications are nonlinear and often lack analytical representations. Consequently, the solutions to the UQ problems also need to be sought with some numerical approximations. Another difficulty is seen in high-dimensional UQ problems (i.e., the problems involving a large number of uncertain parameters). This is because the computational cost required by many UQ approaches grows exponentially as the dimensionality of the problem increases and, hence, could become unaffordable.

To accomplish our goals, we are developing a general framework and toolkit that provides a systematic treatment of the uncertainties involved in the control of complex systems. The four major UQ problems in this framework are the identification, propagation, and reduction of uncertainties, as well as making control decisions under quantified uncertainties. A specific group of UQ methods (i.e., the ensemble-based methods) were adopted and implemented to solve these problems.

As a result of our first year's and this year's development, several methods have been incorporated into the UQ toolkit. These methods were then applied to two different applications (demonstration) problems.

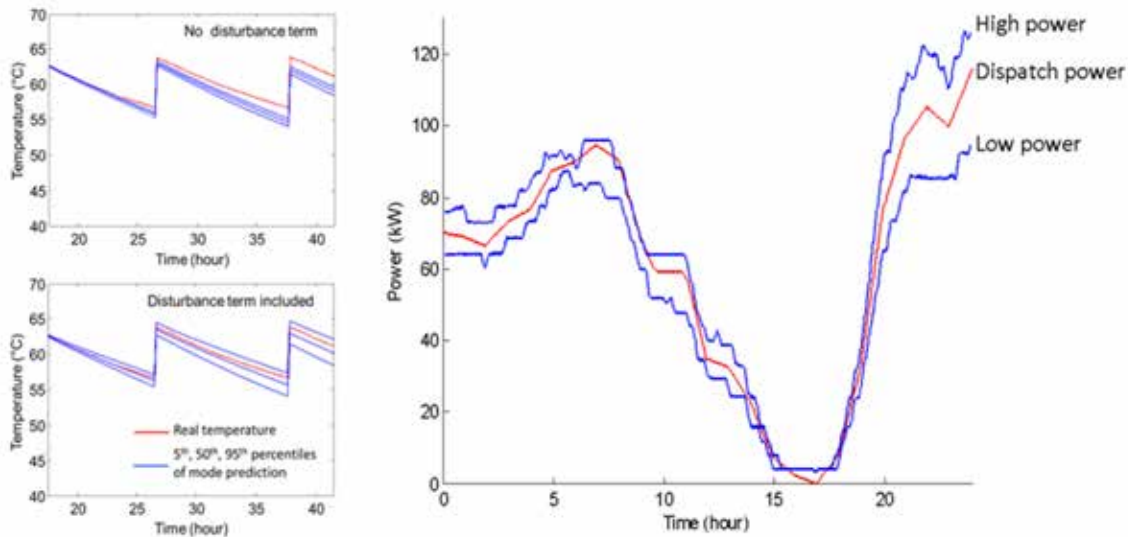
The UQ capability was illustrated with a case study, in which a group of thermostatically controlled loads are controlled according to the simulation results of a second-order equivalent thermal parameter model, while some of the model parameters are subject to uncertainty. In this case study, we found that uncertainties increase as the simulation time (i.e., the length of a market cycle) increases, but cannot be eliminated by choosing an extremely short market cycle. The initial mass temperature was the dominant contributor to the uncertainty. Thermal properties of the house do not significantly affect the demand curve. Using the measurements of air temperature and the ensemble Kalman filter method, we saw a major reduction in the demand curve uncertainty after only a few data assimilation steps. We also discovered that, with quantified uncertainty, the coordinator can achieve the control objective fairly accurately.

In the second case study, UQ was applied to the optimal scheduling of a population of water heaters, where the temperature changes and energy consumptions needed to be predicted with simulations. The control algorithm can manage load to shift away from peak periods, while simultaneously allowing increasing load during off periods (e.g., help deal with morning photovoltaic ramps). Actual water heater testbed data was used in a Bayesian calibration. Three uncertainty parameters were considered for each water heater.

Some major insights and findings in this case study were that uncertainty results from various reasons; in this case, model mismatch between real and simulated water heaters. Uncertainty also impacts physical operations, so system operators may need to hold additional reserves. And finally, uncertainty can impact economic benefits, business decisions, and financial settlements; a utility/aggregator may need to acquire fewer or more resources based on confidence bounds. Posterior parameters resistance and disturbance were negatively

correlated, as would be expected, and including the disturbance term in the modeling produces a more realistic representation of the uncertainties and, thus, the true state of the system.

In FY 2017, we are planning to tie the UQ toolkit with more control models/capabilities, specifically control systems being developed, as well as visualization tools and the testbed workflow. We will incorporate data from the testbed and demonstration cases to implement advanced UQ methods, including model reduction (truncated Karhunen Loeve expansion that results in a reduced set of uncertain parameters) and using more efficient stochastic optimization methods, such as generalized polynomial chaos expansions to quantify uncertainty propagation from the input parameters to the object function and constraints. We will also use stochastic collocation based on sparse grid, nonlinear regression, or compressive sensing.



Results from the water heater demonstration case illustrating the effects of including the disturbance term in predicting temperatures (left plots), and then the propagation of those uncertainties to estimate the confidence interval for the power level (right plot).

Scaling Multisubject Cortical Parcellation

Mahantesh Halappanavar

The goal of this project is to explore novel representations of the surface of the human brain based on the unique structural neuronal connections and functional activities of individual human brains. In doing so, we aim to create a representation of the brain surface at a resolution yet unseen in neuroimaging research in order to better understand how variability across people with regards to factors like environment and genetics affect the cortex via development, aging, cognition, and disease progression.

Using the Human Connectome Project data, we initiated this project with two goals in mind: 1) to build a new population-based atlas of the human brain surface, and 2) develop new parallelizable algorithms to generate and process this atlas and apply the atlas to new patients. Neuroimaging data is often very high-dimensional and parallelizability of algorithms is key to being able to not only produce initial results, but also replicate them.

Previous research has attempted to build new mapped representations of the human brain surface based on connectivity information using various clustering algorithms to these connectivity profiles. A variety of interesting research has resulted from these studies, but the majority of these studies stop short of building a population-based representation of the human brain, use algorithms that suffer from computational inefficiencies or heuristic bias, and do not address how to apply these new mapped surfaces to new patients.

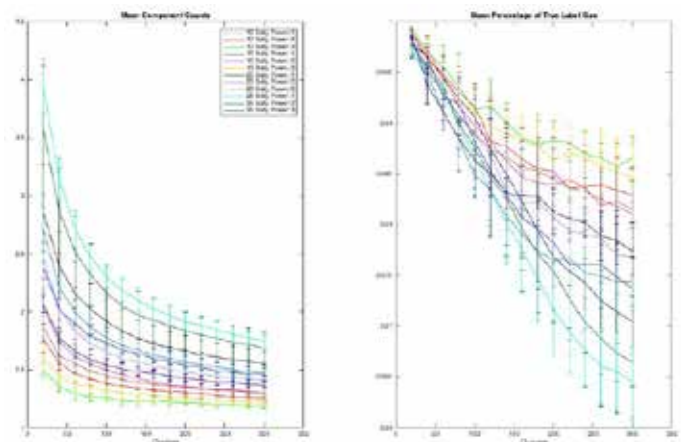
In FY 2016, we developed a workflow to generate novel, high-resolution representation of the human brain surface capable of producing cortical labels that can be shared across individual patients. The workflow is also capable of jointly parcellating (clustering) multiple brains (subjects) in concert.

For each subject, K , in the Human Connectome Project dataset, we have $N = 32,492$ data points. From this data, we generate an $N \times N$ similarity matrix derived from the connectivity profiles of each point. We then apply a surface-matching algorithm to identify points on the brain surface

that are similar between two subjects' brains. This algorithm decomposes each brain surface into its Laplacian graph and then performs Coherent Point Drift point set registration between the eigenfeatures of each surface. Each of these matching matrices is also of size $N \times N$. We incorporate the similarity matrices and inter-subject surface matches into one large block matrix of size $(K \times N \text{ by } K \times N)$. The diagonal blocks represent each individual subject's similarity matrix, and the off-diagonal blocks represent the surface matches between subjects.

We then apply spectral clustering to this large matrix, thereby guaranteeing that multiple subjects are parcellated (clustered) simultaneously and that labels are shared between subjects. The off-diagonal matching blocks in the input matrix guarantee that labels share rough spatial correspondence across subjects.

In FY 2016, we developed prototype implementations for computation of similarity matrices for each subject, as well as computation of eigenvalues and eigenvectors of the full block matrix using a trace minimization algorithm. Further, we conducted several experiments to examine the quality of the produced labels from our workflow.



Preliminary results representing the mean number of compartments per cluster label, which varies with the power to which the similarity matrix is raised. Higher powers produce lesser compartments, but compartments increase in size.

Signature Development for Identifying Human Trafficking in the Online Landscape

Sean J. Kreyling

We explored various data features relevant to developing a signature for identifying the presence of Domestic Minor Sex Trafficking (DMST) victims advertised in the online marketplace for prostitution; a population that is both hidden and non-self-identifying. Given that human trafficking and smuggling share characteristics with other types of transnational threats, it is hypothesized that the knowledge gained during the signature development process could be used to inform signature discovery for other types of illicit trafficking and/or national security priorities, as well.

The DMST market is currently poorly characterized, in part because it is embedded within, and obscured by, the larger commercial sex industry. Through this project, we used a PNNL initiative analytical framework to identify and select the best features from multiple measurement sources to examine the various attributes of a signature. Expert knowledge was also leveraged to assist in identifying other attributes, features, and/or relationships of the advertisements which are not intuitive to the non-expert observer.

Approaches and tools from similar projects helped us identify ways to integrate expert-driven and data-driven approaches. Our goal was to increase understanding of methods to develop signature(s) for DMST victims by integrating expert-driven and data-driven approaches to victim discovery, leveraging tools and methodologies from other PNNL projects (e.g., Trelliscope, Fishing for Features, Signature Quality Metrics, etc.), in order to facilitate the future identification of DMST victims within this online landscape.

The data we examined were publically available, open source, online advertisements. The data often contains a combination of text, symbols, and images. There is significant meta-data and potential features available, including, but not limited to date and time of ad placement, website of ad placement, frequency of ad placement by same phone number (on single or multiple sites), image contents (e.g., known hotel décor patterns, presence or absence of a face, tattoos or lack thereof, etc.), Natural Language Processing analysis of text to determine if gender of author is different than the person advertised, use of certain key words or descriptions, hash value of image, telephone numbers and area codes, and more.



A map produced from a sample of online advertisements for adult commercial sex work.

In FY 2016, the team began developing fundamental engineering capabilities for analyzing the data. We started by adding records to our existing database and obtaining the appropriate source code for the tools. We parsed the records and began the initial data exploration. Additionally, we identified existing academic research on online advertisements and human trafficking and elicited information from law enforcement about the significance of attributes in the advertisements themselves and obtained ground truth data.

The team initiated a process to identify relevant available attributes of a signature. We created a comprehensive, structured database of online advertisements that had both data-driven and expert-driven features, including features specifically identified by U.S. law enforcement, DARPA's Memex project, and derived features from our own analysis on text-based, gender classification and image analysis. We employed Trelliscope to evaluate features and cross-correlations in the features. This allowed us to develop hypotheses relevant to producing metrics of DMST behavior and a category set of structured variables. We also evaluated additional tools, Fishing for Features and Signature Quality Metrics, for applicability.

Lastly, the team explored other approaches, including Deep Learning. For this approach, we are using a deep bimodal auto encoder structure to encode both the features of each advertisement, as well as their images into a combined set of features. This combined encoded representation of our features is then clustered using an unsupervised clustering technique called affinity propagation to converge on an optimal number of clusters for our data, allowing us to see which advertisements have a similar signature to our ground truth data.

Software for Analysis of Metabolite Data from Ion Mobility/Mass Spectrometry Systems

Sam H. Payne

Metabolite concentrations and fluxes are one of the most data poor areas of systems modeling for biology, driven primarily by the absence of a reliable and high-throughput measurement platform and data analysis methods. Our goal is to dramatically improve computational methods for metabolite data processing.

To model and understand metabolic exchanges and fluxes between organisms and their environment, we need to study the ecosystem in great spatial and temporal detail. The level of characterization we can achieve is primarily limited by the sample throughput. Metabolomics measurements are frequently carried out with gas chromatograph-mass spectrometry (MS) and require a significant manual curation during data analysis. This severely limits our ability to perform enough experiments to sufficiently describe the biological system under inquiry.

PNNL is working on a technology platform to dramatically increase the throughput of measurements, by up to 8,000 samples per day. The new platform involves the integration of a high-throughput sample injection system with an ion mobility spectrometer/time-of-flight (TOF) mass spectrometer (IMS-TOF) instrument. The current computational capability for the data produced by the platform does not take full advantage of the spectra from multiplexed fragmentation. This information provides the necessary specificity to uniquely identify metabolites in complex environmental samples such as soils and biofilms.

In FY 2016, we identified two primary roadblocks to high-throughput metabolomics analysis on the IMS-TOF platform. The first issue was the identification of features from raw MS

data. The extraction of features is necessary for all downstream data processing and must be both sensitive and specific. In our manual analysis of thousands of features extracted from datasets (using multiple algorithms) we identified several characteristic problems. We observed that the score of the feature (again using multiple algorithms) did not necessarily correlate well with the classification (good/bad feature) that manual data quality inspectors would assign. We saw that feature abundance had a significant, sometimes overpowering, effect on the final quality score (as opposed to the objective quality of feature extraction).

The most significant issue that we observed happened when adjacent features in the 4D space were not baseline resolved. In this instance, multiple features were often inappropriately grouped, and other correctly segmented features were scored poorly. These and other issues were binned, and multiple test areas were established. We created a large corpus of test data for each area. In analyzing these test data, we created new metrics to attempt to better describe the quality of a feature.

The second roadblock that we tackled in FY 2016 was related to the database comparison of identified features. During the course of the project, we had a workshop with the instrument vendor and the alpha-testing laboratories to discuss data pipelines for metabolite identification. In this workshop, we discussed both the computational complexity/time spent in identification, as well as methods for false-discovery rate estimation.

Finally, we worked to smooth the downstream bioinformatics modeling and analysis of metabolic data by working with the Systems Biology group at PNNL to map the metabolite database into proper biological context of networks and pathways.

Sparsity-Based Data-Driven Learning Method for Complex Systems

Xiu Yang

Our research aims to develop new, data-driven machine learning (ML) methods for scientific computing. It will provide alternatives to the first principle rule approaches to studying complex systems, and it will lead to an explosion of new discoveries in science and engineering applications.

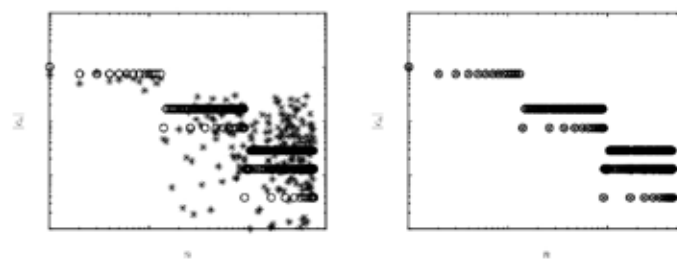
We are developing new, accurate, and efficient predictive capabilities for complex systems using data-driven approaches. The project's goals include: 1) the integration and development of the emergent and novel state-of-the-art data methods of compressive sensing (CS), ML, sensitivity analysis, and uncertainty quantification, with more traditional methods for dimensionality-reduction and data-assimilation; and 2) the application of this approach to the discovery of fundamental relationships, model equations, and improvement of existing models for complex systems. This proposed research is a significant improvement over the existing efforts in this area, because they mainly rely on the proper orthogonal decomposition (or singular value decomposition) analysis of data, which does not capture the dynamical information. Also, the existing techniques using “sparsity” structure of the complex systems do not explore the low-dimensional property of the system sufficiently; hence they cannot exploit information from limited data efficiently. Moreover, most techniques developed in this area only concentrate on the simple prototype systems and the efficacy for complex systems is not verified.

We developed a new framework of ML based on sparse measurement. This framework enhances the sparsity of the orthogonal polynomial based sparse regression model for complex systems. Given the limited data from measurement, sensor, experiment, etc., we used an iterative, rotation-based method to transform the original regression model into a new one, which bears a more sparse structure. The improvement of the sparsity facilitates the CS method to recover the full-state space accurately with only a few data. This is a breakthrough within the sparse recovery techniques of the past two years, as it intrinsically explores the low-dimensional structure of the systems and yields more accurate results than the existing CS method. Moreover, it also helps to identify optimal sensor locations, in order to maximize the information extraction. This technique can be applied to the physical

space to recover dynamical systems (e.g., in climate, subsurface studies). It can also be applied to parameter space to construct a response surface of a system, thus improving the efficiency of experiment design, parameter inference, etc. We have verified the efficiency of the new methods with prototype systems. Moreover, we are using the dynamic mode decomposition (DMD) methods developed by our collaborator at the University of Washington to investigate the dynamical properties of a stochastic subsurface flow. This system is complicated in that it has a much smaller ratio of snapshot over the degree of freedom in physical space, which is common in the systems we specified studying in the proposal (e.g., climate, subsurface, and chemical imaging). This experience is very important, since it provides us insight to further improve the existing DMD framework.

In the first year, we focused on reconstructing the dynamical system, and in the second year, we will focus on building learning dictionaries based on the data itself. This will enable us to achieve three critical tasks in studying realistic problems: 1) classification and/or categorical decision-making about the data, 2) reconstruction of the full-state space, and 3) in the case of dynamical data, an efficient prediction of the future state. The method facilitates a family of local, reduced-order models for the various physical regimes measured, thus allowing for an accurate reconstruction of the full state of the system, along with a low-dimensional and accurate prediction of its future state, even under noisy measurements.

Our plan for the second year is to integrate the DMD method with the sparsity enhancing technique and dictionary learning method to provide a new, data-driven model for recovering the full dynamical state space and optimally informing categorical decisions, as well as characterizing and modeling complex, nonlinear dynamical systems over a range of dynamical behaviors.



The coefficients of the sparse regression model by the existing CS method (left); the coefficients by our new method (right). Circles are the exact solution and stars are the results by different methods. The comparison demonstrates that the new method is much more accurate.

Stream Adaptive Foraging for Evidence (SAFE): Human-Computer Co-Assisted Signature Discovery and Evidence Generation for Streaming Data with Deep Learning

Sean M. Robinson

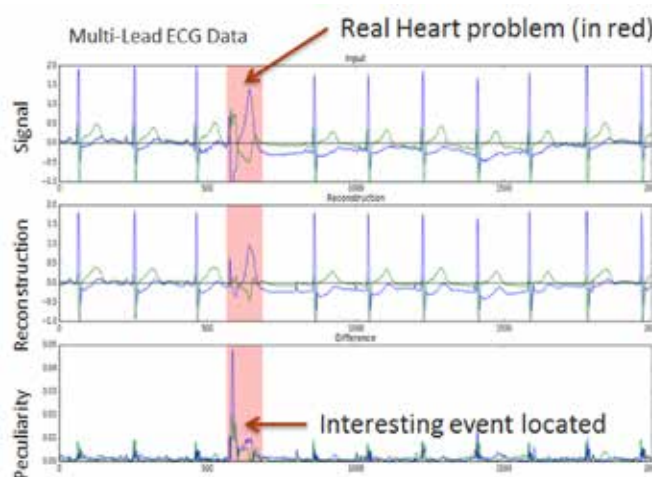
Some of the most pressing machine learning applications, such as cyber security and object recognition, lack enough ground-truth training data to build a classifier. We solve this problem by using unsupervised deep learning techniques to determine when data were anomalous or deviated from the norm.

Traditional methods require either specific context-dependent expertise to construct models or prior examples of events and large amounts of well-labeled data to perform machine learning. Some cutting-edge deep learning approaches have been used to characterize complex events, but these have not been applied to the streaming environment and require a great deal of data. We demonstrated the use of an autoencoder and relevant featurization techniques to both learn a feature space and then identify anomalous portions without the aid of labeled data or domain knowledge. In this way, we demonstrated the detection of anomalous features without domain knowledge or tagged examples, and delivered these features to users.

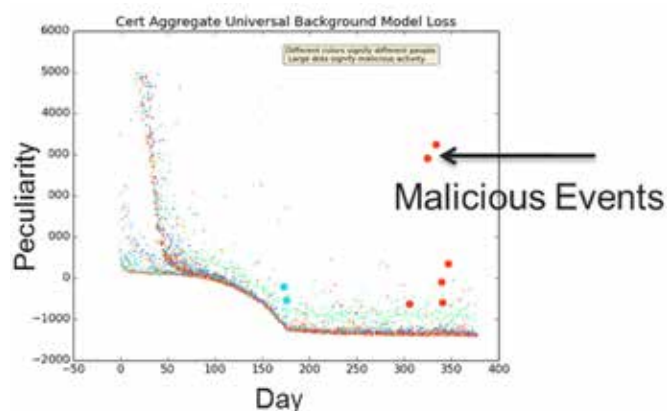
The SAFE project utilized autoencoders and other deep learning techniques to develop architectures for finding the most relevant data from a stream. Autoencoder stages were devised to dynamically train on numeric or event-based data streams and generate best reconstruction estimates for these same streams. The difference between the input and reconstructed data at every point was compared and used to produce a “peculiarity” metric, which in turn was used to determine where rare or unexpected behavior happened in the data set. This method was used to find real and synthesized heart conditions with no foreknowledge of any kind, and these results were published and presented. We also utilized featurization techniques to demonstrate detection and delivery of interesting events in cyber data and showed the capability of these methods to both enhance the likelihood of detecting these events in real time and also deliver to a user likely reasons that behavior was anomalous.

In the next fiscal year, we will apply the extremely positive results of the methods developed in FY 2016 to apply to sev-

eral additional data domains such as cyber, power grid analysis, medical imaging, and radiation detection. We will also produce a system in which prior methods select data elements from a stream that a user should find most interesting. The elements represent a selection of features and time windows, and will be presented to users in a way the user can determine utility. This will require presenting visualization, summary statistics, or other summary representation of the data. These techniques will be applied to realistic data sets, allowing for presentation of useful data subsets in a way relevant to these problem spaces, and user responses will be employed to retrain and enhance method results.



SAFE technique operating to find anomalies in EKG data.



Analysis of CERT cyber-security data with post-hoc labeling of malicious events.

Streaming Data Characterization

Mark T. Greaves

We are creating an integrated resource of high-speed, streaming data characterization and analysis algorithms upon which future projects can build.

Initial processing of streaming data sources is an area in which there is substantial previous work. We planned a set of related projects that will survey the scientific literature on initial stream processing algorithms for co-design domains, implementing the promising projects and tasks, as well as evaluating their performance. We will use these studies to qualify potential university partners for their suitability within longer-term cooperative research and development relationships. There is currently no repository that gathers together the state-of-the-art in powerful online analytic algorithms that can process data on the fly. With this in mind, our repository will be not only critical for success, but also a lasting resource for PNNL.

We directed four exploratory projects within different organizations, as described below.

Foundational streaming data preparation methods. This work was performed by Rensselaer Polytechnic Institute (RPI) and carried many initial research tasks, including a categorization, survey, and component listing for popular open-source and commercial off-the-shelf (COTS) streaming frameworks; an analysis of the specific uses of the selected frameworks that identified the overall strengths and any breakthroughs in these frameworks; and a new formal ontological classification of the chosen frameworks based on the technical approach.

Visualization of hypotheses about streaming data. This work was performed by the University of Utah's Scientific Computing and Imaging (SCI) Institute and addressed statistical hypothesis testing as a visual tool for quantifying and understanding complex phenomena. An approach was developed that relied on simultaneously leveraging the analytic capabilities of both humans and computers, using visualization as a knowledge-sharing interface between the two. For example, computers are good at extracting linear models, dealing in high-dimensional spaces, and processing large quantities of data. On the other hand, humans tend to be much better at more abstract tasks, such as identifying non-linear models and classifying data. In this work, the human helped to build the computer model, while the computer simultaneously helped the user to build the mental model of the data that resulted in a prototype.

Data summarization for streaming scientific video. At the University of Washington, this project supported initial architectural work to address the significantly growing scale of microscopy video data. The scale of imagery coming from new systems, such as PNNL's dynamic transmission electron microscope, requires developing new streaming algorithms even for common tasks such as retrieval, interactive browsing, or compressing video content. This project developed architectures and basic summarization algorithms to identify pertinent scientific content from sequential imagery on the fly and produce a condensed version of video as an output. The techniques also meet single-pass and high-throughput requirements.

Event summarization in microscopy imagery. At the SCI Institute, we collaborated with biologists that had streaming image analysis needs. For two selected bioimaging problems, we determined the best set of nonstreaming algorithms and proposed modifications to methods to adapt them for streaming image analysis in experimental settings. Expected impacts include techniques for automated analysis of image data from microscopes to drive experimental techniques, as well as the development of new techniques for quickly post-processing acquired image data.

In FY 2015, we expanded and formalized our collaboration with university partner RPI. Together, we focused on studying specific symbolic techniques to consume and reason over a volatile data stream. The project leveraged selected COTS and open-source components of the Semantic Web software stack and explored the interaction of these different software frameworks on choices for streaming data models. Along with RPI interns onsite at PNNL, we commenced work on developing a library of algorithms to optimally manage a cache for symbolic data. This new family of algorithms allows us to account for the interaction between the particular symbolic knowledge representation formalism (e.g., description logics of various expressive powers), the different reasoning engines provided by the Semantic Web software stack, and the parameters of the streaming analysis task.

In FY 2016, we supported an RPI doctoral student to further develop the cache management algorithms created in FY 2015 and published three papers weaving their operating principles into a new account of semantic importance. We provided a formal ontological account of importance ranking and cache replacement based on priority vectors. We used this account to build streaming cache management algorithms for offside detection and real-time insider threat detection in cyber networks. In each case, we were able to experimentally show that algorithms based on semantic importance exhibited higher performance under resource limitations than traditional approaches to cache management.

Temporal Modeling in Streaming Analytics

Lisa M. Bramer

As data recording capabilities continue to grow, research on classification and predictive modeling on data in near real-time plays an important role in modern data analysis. This project aims to improve current streaming analysis methods, making model output more useful and timely to decision-makers, by leveraging the temporal nature of this data.

Machine learning algorithms can easily be implemented on streaming data. In traditional machine learning settings, both supervised and unsupervised learning algorithms operate under the assumption that the data is stationary (i.e., the mean and covariance structure are the same for all parts of the dataset) and observations are independent of one another. Analytic methods for streaming data must differ from standard machine learning practices, because in the streaming context, there is a continuous flow of data rather than a static set of data, the data distribution is expected to evolve over time, and observations are seen over time in sequential order and likely are not independent.

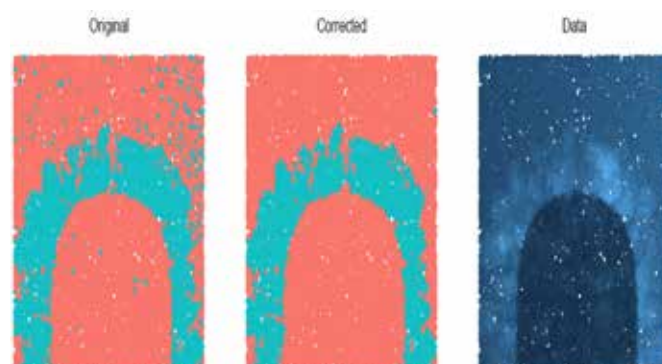
This project aims to broaden the understanding of the role that temporal dependence has on the performance of state-of-the-art algorithms for streaming data. This is to be accomplished by leveraging the temporal nature of streaming data sources to improve prediction and classification analytics capabilities by incorporating temporal information into algorithms and models for streaming data. These new methods will be computationally scalable to operate within a streaming environment. Additionally, we will strive to provide insight into quantifying the benefit of incorporating temporal information into predictive models over a variety of conditions. Finally, we aim to develop methods for integrating multiple data sources that may be observed at different and/or irregular time scales, which generate reliable predictions at user-specified time steps.

In FY 2016, we conceptualized several models and algorithms that incorporated temporal dependence and applied these algorithms to four unique domains—cyber security, weather related to tropical cyclones, mass spectrometry, and electron microscopy—with data collected in a streaming context. We developed models for forecasting episodes of rapid intensifi-

cation events for tropical cyclones based on weather sensor and satellite data streams. These methods demonstrated the ability to more accurately predict events of interest compared to current state-of-the-art multivariate regression models. We also conceptualized and implemented automated methods of establishing typical/baseline behavior for multivariate data streams. We defined metrics, incorporating temporal dependence, to measure the similarity of real-time observations compared to the aforementioned baseline.

These methods were applied to cyber security data for detecting when a user's behavior has significantly changed. Additionally, these methods were applied to mass spectrometry data for the purpose of quality control (QC) and were shown to significantly improve the detection of low-quality sample runs. The final product for mass spectrometry QC has been implemented and connected to the mass spectrometry instrument and is being utilized in real-time, with instrument operators acting as humans-in-the-loop. Finally, we implemented and augmented the aforementioned multivariate models on electron microscopy (EM) videos for the purpose of detecting changes in lithium growth/dissipation. We developed further models that characterize the growth/dissipation of lithium by quantifying the amount of material and rate of growth. These models were demonstrated to be robust when less than 5% of the sampled images were made available (and still give reasonable results at 1%), a major requirement to make analytical methods useful to the scientists.

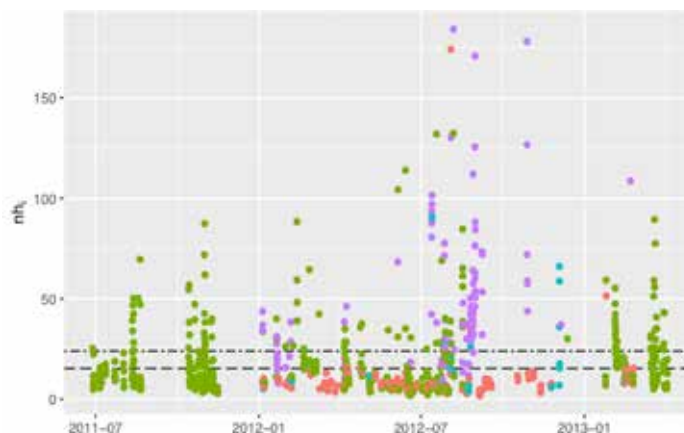
In FY 2017, we will focus on the continued partnership and collaboration with the EM and mass spectrometry scientists.



Classification lithium growth vs background of EM video at one time point, using Gaussian mixture model algorithm. Classification results based on 25% sample of data with lithium growth in blue (left), classification corrected by spatial correction method (center), and full image with all values plotted.

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We will continue the development of additional algorithms and work to improve existing algorithms. Additionally, we will augment and develop methods for dealing with data that have multiple streams that may or may not be coming in simultaneously, at regular or irregular time scales, while still leveraging the temporal structure in the data. Finally, we will pursue other relevant application areas and corresponding partnerships with domain experts.



Scaled influence values computed using the model-based cutoff, with a human-in-the-loop for the mass spectrometer, Orbitrap instrument.

Testbed Federation Tools for Control of Complex Systems Research

David O. Manz

We are enabling high-fidelity experiments on complex systems by designing and implementing tools for federating testbed resources from research collaborators for modeling full complexity across large computational platforms and with high quantities of physical equipment.

The highest form of research evidence is gained through experimentation with real systems. However, when attempting to research systems like the buildings and their interaction with the power grid, it is impossible for one research organization to fund and support enough equipment to model the scale and complexities. The most reasonable solution is to collaborate among the research community to achieve the desired scale and complexity to fully explore new control architectures, paradigms, and algorithms for operational use. This project's focus is on developing the tools necessary to enable federation among individual testbeds that are geographically dispersed.

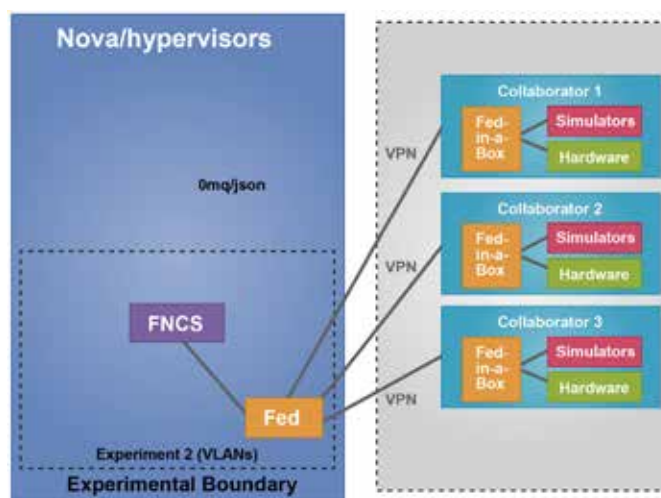
A goal of the Control of Complex Systems initiative is to form a coalition at the facilities of multiple participating organizations (anticipated to be three to six in a pilot). The tools needed for enabling this collaboration include creation of secure tunnels across the internet, access controls, experiment model sharing, and information and control transmission. Specific needs for tools will be defined in a parallel effort of all participating organizations.

This project will support identification of the needs for tools, design a system necessary to fulfill requirements, and implement and test the tools. To foster collaboration and speed progress, the tools developed will be packaged such that they are easy to distribute to partners for operation within their testbed environments. This packaging can include the creation of an operating system image, a virtual machine, or even a prebuilt and preconfigured piece of hardware. As every organization has their own policies and acceptable risk levels, the tools are going to be developed with as much flexibility as possible. This flexibility can extend through every layer of federation, including multiple types of tunnels with varied security protocols, multiple field protocols, and multiple methods of integrating equipment.

As the goal of the testbed is to study new control methods, the federation tools also need to enable the distribution, management, and operation of control logic. In support of

this goal, the VOLTTRON™ platform developed at PNNL is being investigated as one of the tools to enable the distribution of control agents into collaborator testbed environments.

In FY 2016, the initial design of the federation tools has been created. This design includes the packaging of the tools within a virtual machine that can be shared with collaborators. Within the virtual machine, multiple virtual private network tools will be present to enable secure tunnels across the internet and support crossing enterprise boundaries. The highest performant, but least applicable to most environments, is the OpenVPN tool, which establishes an IPSEC layer 3 secure tunnel. The least performant, but most easy to get through border security policies, is the OpenSSH tool, which establishes a layer 4 secure tunnel. A layer 2 tunnel will be established on top of either tunnel to enable the devices on each side to communicate as if they are on the same local area network. The VOLTTRON™ platform provides a translation interface that allows communication with devices on collaborator sites, with operational protocols like Modbus to a routable protocol like the VOLTTRON™ Interconnect Protocol. Initial implementations of this design were created to start discussions with collaboration partners.



Over the next 2 years, this initial design and implementation will evolve into tools that support the broad requirements laid out by the coalition participants and are robust and easy to use. Further tools will be integrated and software will be developed to integrate them into a cohesive package that can be easily configured and executed automatically. The methods of packaging might also change based on what best suits their operation and management within collaborator systems. Within this project, these tools will be tested, and federated experiments will be supported.

Topological Analysis of Graphs in Cyber Security (TAGs-CS)

Emilie A. Purvine

We are discovering shapes, structures, and overall cyber system behavior using topological data analysis of graphs derived from data, providing analysts with information to determine the state of their cyber system.

Topological methods have been deployed in real-world problem domains with great success (e.g., signal processing, medical imaging, and drug discovery). These methods were created to identify the largest, most robust shapes and structures in data sets and to discover average system behaviors. In cyber systems, the network is dynamic as elements (e.g., computers, printers, and mobile devices) sign on and off, as communications begin and end, and when interactions with other networks initialize and complete. In this project, we analyze these dynamic systems using topological methods and create new tools for system administrators to judge resiliency and ensure that missions can still be completed.

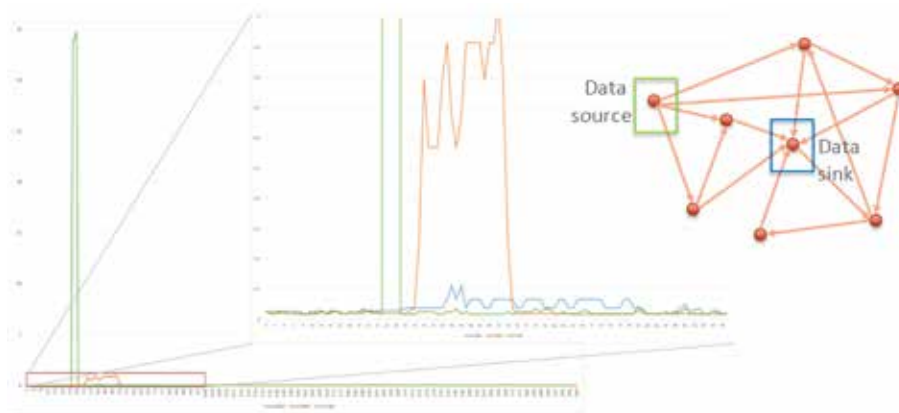
We hypothesize that this analysis will provide a way to differentiate between resilient and non-resilient system states, possibly even identifying different causes of non-resilient states. Within the broad theory of topological data analysis, we are using two major techniques, both coupled with novel statistical graph analysis. The first technique, persistent homology (PH) is used to discover the most prevalent higher order structures within a topological space. We use a time interval of NetFlow data to generate a point in a high-dimensional space by calculating relevant features such as number of IP addresses seen, number of flows, and total number of bytes transmitted. Over time, these points accumulate into a point cloud that we consider as a topological space. The PH of a space can serve as a fingerprint of the structures contained within. We compare the fingerprint of a query space—corresponding to the current time period—to that of a known baseline space using established distance metrics on these fingerprints. In FY 2016, we showed by example that this methodology can be used to identify instabilities or unusual events in test NetFlow data sets (e.g., a port scan or a data exfiltration).

Our second technique tracks the behavior of each individual IP in a network

according to its level of data source or data sink. We consider cyber traffic as a dynamic “flow” on a graph where information (bytes) moves between nodes over time. This flow is dynamic, since communications only occur for short time periods, and some network components can be turned on and off regularly. By applying discrete Hodge theory, an area of differential topology, to the dynamic flow, we derive an ordered list of all IP addresses (i.e., a ranking), starting with those that are most like data sources (where information mostly flows from) down to those which are most like data sinks (where information mostly flows to). The ranking is computed for each time period so that when a particular IP address has a major behavioral change, we flag it for further investigation. When our PH metrics indicated some level of instability in the system, we were then able to discover the perpetrators of port scans and data exfiltration in test data sets during FY 2016.

Also in FY 2016, we explored the use of information measures on neighbor distributions in NetFlow graphs. An IP address with all communications going to the same destination IP has a different behavioral profile than one who spreads out communication across multiple destination IPs. The former may be a server that is only used by a single user, whereas the latter could be an email or web server. We used metrics developed under a prior LDRD project to create behavior profiles for the network, which evolve as IPs change their communication. We observed that deviations in this behavior profile can indicate the occurrence of an anomalous event.

In FY 2017, we will conclude this project by generalizing our methods beyond NetFlow, testing on more robust and realistic data sets, and combining results from all methods into a more full picture of the system.



Anomaly spikes from PH method, corresponding to three port scans at different levels of evasion.

Toward Enabling Complex Sensemaking from Streaming Data

Lyndsey Franklin

The goal of this work is to increase the cognitive complexity of analytical tasks that can be performed by analysts in environments that use streaming data.

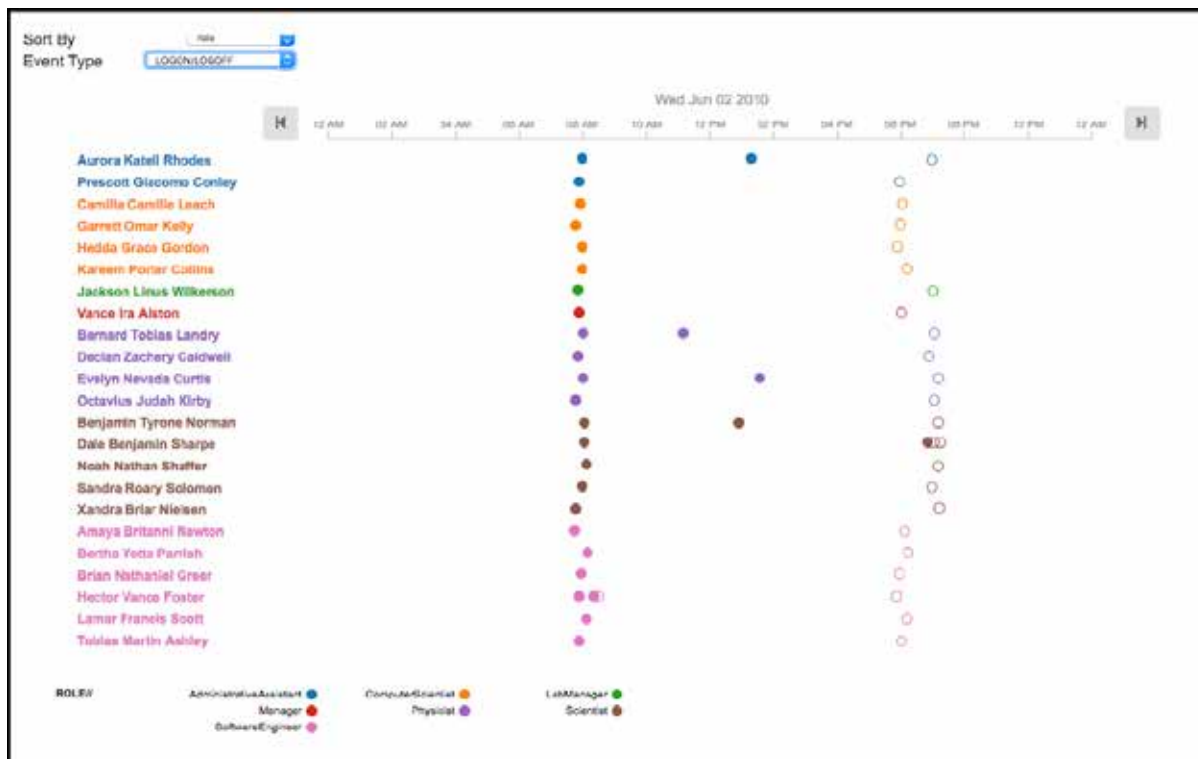
Cell population measurements provide averaged values often dominated by the most robust or abundant processes, where molecular details and variability between cells are buried in the averaged measurements. Analysis techniques such as single cell transcriptomic analysis have the potential to unmask cellular heterogeneities and, for example, identify a cancer cell among healthy cells in a tissue or a cell with a critical function in complex microbial communities. Specifically, single cell RNA-Seq is a young field, with the first publication appearing in 2009 and remains largely missing statistical tools for detecting unique signatures of individual cells that may be buried in the natural noise created by the normal variability between cells.

We are establishing new tools to identify signatures of cell abnormality or responses to perturbation that may be buried in the natural noise created by the normal variability between cells. These tools could be also applied to decipher the role of individual microbial cells in complex microbial communities to understand their function in C cycling, biomass degradation, plant resistance, or the release of volatile organic compounds. Specifically, we have focused on single cell RNA-Seq datasets for cells perturbed by engineered nanoparticles (NPs). The exploding growth in the use of nanomaterials in a wide range of applications is expected to increase both the intended and unintended human exposure to NPs, but a great deal of confusion exists about the properties that make a NP toxic or biocompatible. Distinct physical and chemical properties of the NP engage and activate distinct proteins and cellular pathways that in turn govern the fate of the NP and its impact on the cell and, ultimately, on human health.

The relationships between NP properties and key cellular processes and response are far from being understood. This knowledge gap is largely due to experimental challenges that

NPs present, including the difficulty to achieve uniform NP distribution over the exposed cells. The distribution typically spans 2 to 3 orders of magnitude, indicating that some cells are “overloaded” with hundreds or thousands of NPs, while other cells are loaded with only few or no NPs. Responses such as regulation of gene expression or pathways have always been measured in the population as a whole. This approach identifies averaged, often most common or generic processes, while leaving other critical processes undetected due to the dilution of signals across many cells, each carries a different number of NPs. As a result, it is unclear whether population studies identify only generic, emergency state responses in a subset of “overloaded” cells, and whether responses unique to the properties of the NPs at lower load levels, where many cells might carry not even one NP, are diluted and missed altogether. To answer these questions, we exposed alveolar epithelial cells, which present a vulnerable target to airborne NP exposure and sorted individual cells by their NP load for single-cell RNA-Seq analysis. Using these datasets, we have been developing analytical tools to enable the detection of unique response signatures in these datasets.

Quantum dots (QDs) have been generating interest in a growing range of applications from solar and interfacial charge transfer, including photovoltaics and solid-state LED lighting to biological research, drug delivery, and medical diagnostics. However, controversy still exists about the type of coating and functional groups that could make these promising NPs biocompatible. By developing new approaches to analyze single cell RNA-Seq data of cells carrying lower or higher loads of animated or carboxylated QDs, we have begun to uncover unique response patterns to distinct QD types. Using classification trees recursively, we identified the top 100 genes that contributed the most to the separation between the treatment groups. We found that these top 100 genes could more effectively segregate cells by their treatment groups than all 9233 genes found in all the single cell RNA-Seq datasets. This separation is demonstrated by the principal component analysis (PCA) shown in the image.



This figure shows two screen captures from the rate-of-stream study. Each row represents a “user” of the simulated network. Color indicates either each user’s role (top) or department (bottom). The individual dots represent log on/log off times (top) or internet activity (bottom). All participants in all conditions had access to these color and data options. The different conditions differed on how quickly information updated and whether or not the participant had direct control.

Transpire: Transparent Model-Driven Discovery of Streaming Patterns

Aritra Dasgupta

In streaming data environments, “the only thing that is constant is change.” To the human observer or analyst, changes are often too fast to notice, too many to remember, and too complex to understand or make predictions. To help analysts discover and reason about changes, we will be developing a transparency-based, explanatory visual analytics framework for integrating streaming data visualization techniques with automated methods that increase confidence and utility of human judgment for streaming data analysis.

Most existing visual analytics techniques are based on assumptions about how human-stream feedback should work and are not supported by concrete empirical evidence. In our work we fill these gaps through three main contributions.

First, we test how varying levels of model transparency can best leverage the high bandwidth of human perception system for communicating key insights. By conducting qualitative studies through surveys and interviews of analysts from diverse domains, we will develop a characterization of model transparency and its impact on analysts’ perception of streaming patterns.

Second, we use this transparency characterization for developing visual analytics methods and techniques for context-aware human reasoning about streaming data. We study how differences in model transparency affect efficiency, effectiveness, and the trustworthiness of human insights.

Finally, we aim to demonstrate the efficacy of our methods through case studies and quantitative user studies that demonstrate how these visual analytics methods can significantly improve human interpretability of machine-detected streaming patterns.

To date, we have established a streaming data visualization design framework for analyzing trade-offs related to change perception (paper under review at Computer Graphics Forum). We also conducted a user study for demonstrating the explicit relationship between transparency of analytical methods and human trust (paper accepted at IEEE VAST 2016).

Additionally, we implemented two explanatory visual analytic techniques: 1) *SeekAView* for high-dimensional data analysis (paper accepted at IEEE LDAV), and 2) *ClassSignatures* for explaining outcomes of binary classifiers (paper accepted at IEEE VDS Symposium).

And finally, we implemented a cyber visual analytic system, called *ActivitySignatures*, for supporting the insider threat detection research program.

In FY 2017, we will focus on applying the design framework for implementing and evaluating streaming visualization techniques and studying how velocity and volatility of streaming data affects change perception. We will leverage outcomes from FY 2016 to develop explanatory visual analytic techniques for streaming data. Finally, we will continue to investigate the issue of domain experts’ trust in automated methods and visual analytics by conducting qualitative and quantitative evaluations with cyber defenders for improving the tools.

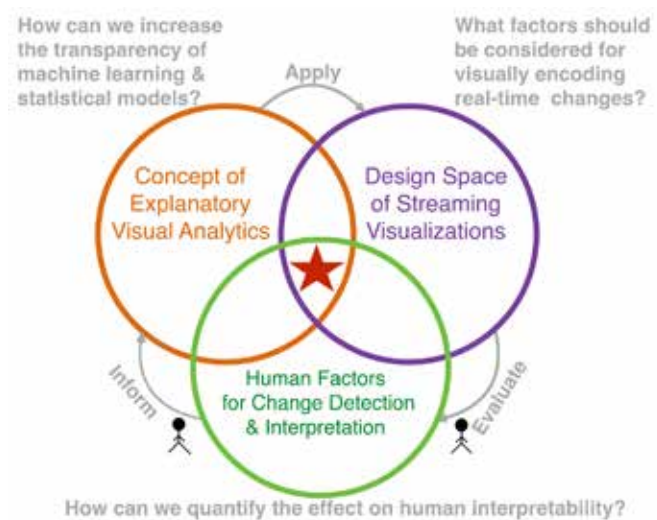


Illustration of the three-fold objectives of Transpire.

Unmask Signatures of Cell Perturbation Hidden in the Normal Variability Between Cells

Galya Orr

We will build on approaches developed by previous projects to establish new tools that will identify signatures of cell abnormality or responses to perturbation that may be buried in the natural noise created by normal variability between cells. These tools could be also applied to decipher the role of individual microbial cells in complex microbial communities to better understand their function in carbon cycling, in biomass degradation, in plant resistance, or in the release of volatile organic compounds.

Single-cell RNA-Seq is a relatively young field, with the first publication appearing in 2009. The field is still largely missing statistical tools for detecting unique signatures of individual cells that may be buried in the normal variability between cells.

To develop methodologies that could accomplish this task, we have been focusing on single-cell RNA-Seq datasets for cells perturbed by engineered nanoparticles (NPs). The exploding growth in the use of nanomaterials in a wide range of applications is expected to increase both the intended and unintended human exposure to NPs, but a great deal of confusion still exists about the properties that make a NP toxic or biocompatible.

This knowledge gap is largely due to experimental challenges that NPs present, including the difficulty to achieve uniform NP distribution over the exposed cells. The distribution typically spans two to three orders of magnitudes, indicating that some cells are “overloaded” with hundreds or even thousands of NPs, while other cells are loaded with only few or no NPs. Yet responses, such as regulation of gene expression or pathways, have been always measured in the population as a whole. Cell population measurements provide averaged values that are often dominated by the most robust or abundant processes, where molecular details and variability between cells are buried in the averaged measurements. Single-cell analysis techniques, such as single-cell transcriptomic analysis, have the potential to unmask cellular heterogeneities and identify, for example, a cancer cell among healthy cells in a tissue or a cell with a critical function in complex microbial communities.

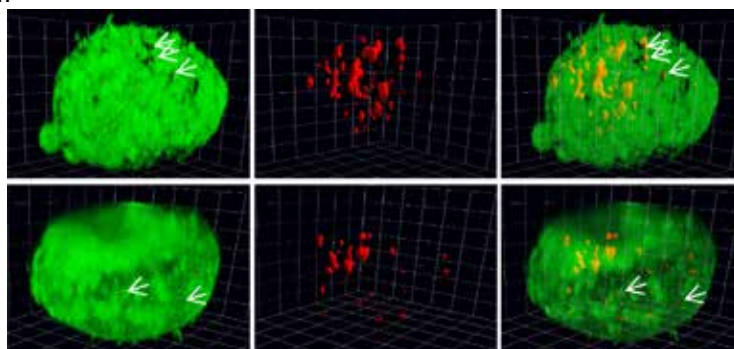
To answer these questions, we exposed alveolar epithelial cells, which present a vulnerable target to airborne NP exposure, and sorted out individual cells by their NP load for single-cell RNA-Seq analysis.

Quantum dots (QDs) have been generating interest in a growing range of applications, from solar and interfacial charge transfer, including photovoltaics and solid-state LED lighting, to biological research, drug delivery, and medical diagnostics. However, controversy still exists about the type of coating and functional groups that could make these promising NPs biocompatible.

By developing new approaches to analyze single-cell RNA-Seq data of cells exposed to aminated QDs, which are more toxic, and carboxylated QDs, which are less toxic, we uncovered unique response patterns to each type. We found that cells carrying lower QD loads responded with multiple strategies, mostly with upregulated processes, which were nonetheless coherent and unique to each QD type.

In contrast, cells carrying higher loads responded more uniformly, with mostly downregulated processes that were shared across QD types. Strategies unique to aminated QDs showed strong upregulation of stress responses, while strategies unique to carboxylated QDs showed upregulation of DNA repair and RNA activities.

Together, our studies suggest scenarios where higher QD loads lock cells into uniform responses, mostly shutdown of cellular processes, whereas lower loads allow for unique responses to each QD type that are more diversified, proactive defenses or repairs of the NP insults.



Confocal fluorescence images in 3D of cells carrying higher (A) and lower (B) QD loads. Membrane-specific fluorescent probe was used to outline the membrane (green). By removing or dimming the green color in images 2 and 3, respectively, the QDs (red or yellow) inside the cells can be seen, with few QDs found at the cell surface (examples indicated by the arrows).

User-centric Hypothesis Definition

Dustin L. Arendt

Our research aims to reveal effective techniques for visual communication of machine learning output to non-expert users in a streaming environment.

The rate of adoption of machine learning and analytics is rapidly outpacing the comparatively small group of expert users who understand the technology. While there are many visualization tools and techniques to support data scientists, software engineers, and statisticians who produce sophisticated machine learning models, there is relatively little support for the typical “naïve user” who consumes these models. These tools, which generally aid with high-dimensional data exploration, feature selection, and parameter fitting, are well-suited toward post hoc analyses in a forensic setting with an expert user. However, they are not appropriate in a deployed space where decisions are required in real-time from analysts who, we assume, wish to leverage sophisticated analytics without comprehending the underlying algorithms.

Our research goal is to gain insight on how to support analysts through visual communication when they want to leverage machine learning analytics to understand a complex changing system. As visualization researchers, we achieve this goal by designing visualizations for a use case and evaluating the effectiveness of our techniques in a controlled human subjects experiment.

Our hypothesis is that the level of abstraction of the visual communication has a strong influence on the user’s ability to accomplish tasks related to streaming analytics. Specifically, we believe in the fairly counter-intuitive assertion that “less is more;” showing less detailed information in a more abstract manner can actually improve the user’s speed, accuracy, and confidence.

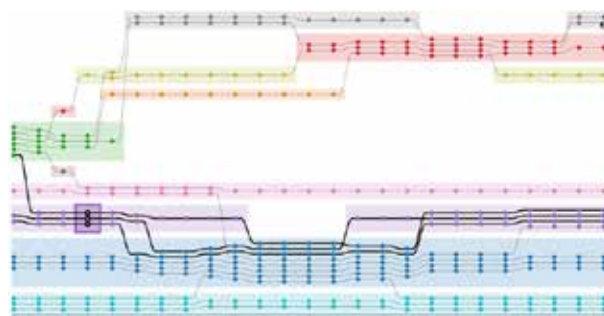
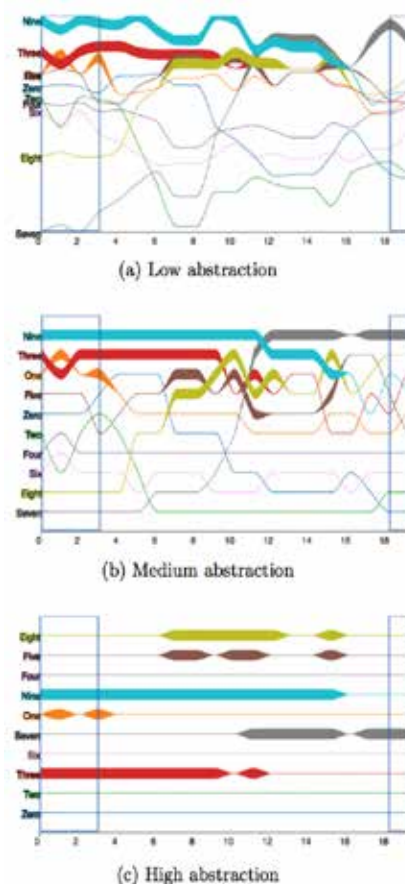
We tested this hypothesis using visualizations in a user study. We measured the performance of experts and non-expert users at detecting change in predictions made by machine learning classifiers on streaming data. What we found confirmed some of our hypotheses. We found that users detected change more slowly with the most abstract visualization, but conversely, they were more accurate at pinpointing when the change occurred.

We developed new visualizations and algorithms for showing how machine predictions about a collection of entities are changing over time. Our research focused on storyline visualization techniques, which show how entities are related over time. These are well-suited for streaming data because of

their intuitiveness and their capacity to show change with historical context.

We used our storyline visualization techniques as part of a user interface for semi-supervised learning. This helps a user to train the machine to assign labels to objects according to the user’s own mental model. As the user provides feedback to the machine, the model changes, and the storylines illustrate this change. This allows the user to more quickly and easily spot errors in the model so they can be corrected through further training. Our research has paved the way for future work on streaming time series analysis and pattern mining.

This figure illustrates how we vary the level of abstraction while keeping other aspects of the visualization, constant. In each visualization, streaming output of a binary classification model is represented by a line, which is thick when the model’s confidence in a positive classification is high. In (a), the least abstract visualization, the model’s confidence is directly encoded on the vertical axis. In (b), the relative ranking of each model is encoded on the vertical axis. In (c), the most abstract visualization, no data are encoded on the vertical axis, and models are ordered alphabetically.



This figure is a storyline visualization that shows how entities are related over time. In this case, it is showing when individuals are in the same place at the same time.

Visual Analytics Platform for Large-scale Hierarchical Control System Data

George Chin Jr.

To enable control system designers to develop and evaluate modern-day control systems more effectively, advanced visual analytics capabilities may be developed and deployed to allow designers to better explore, interpret, and analyze the behavior of complex systems and the data and results they generate.

Infrastructure systems today are generating ever-increasing amounts of data that are highly complex, relate to large numbers of sensing and/or control end-points, and involve multiple scales of operation. Innovative and advanced visualization tools and platforms are needed to enable control system designers to

- Visually test, validate, and refine control systems.
- Effectively explore big data to identify critical events, relationships, and patterns.
- Access, evaluate, and compare experimental results at multiple levels of detail and aggregation.
- Understand how controls manage, impact, and interact with devices, agents, and systems.
- Provide control-system-specific and general modeling visual analytics capabilities currently unavailable to control system designers.

We are focused on researching and developing novel visualization techniques and tools for navigating and exploring large-scale, massive-entities control system data. Tools will feature intuitive visual representations, zoomable interfaces, multi-scale layout and navigation, and scalable performance. In addition, the project is developing a visual analytics platform to manage and integrate visualization tools with complex models and data that will combine and examine results across multiple experiments. This platform will provide connections to data sources, coordinated views of data and models, and capabilities to search, filter, and integrate complex control system data.

In FY 2016, we continued to work closely with control theory projects to gather their visualization requirements. Derived visualization requirements centered on comparing the results

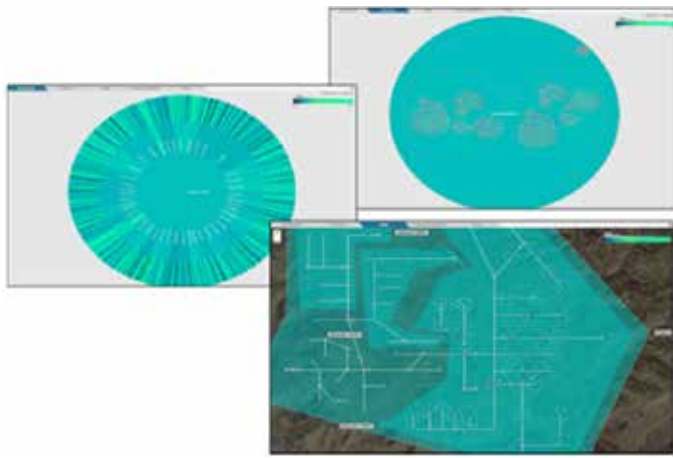
of the new control systems against theoretical or competing systems and evaluating the effects on the entities and end-points they control.

To address these visualization requirements, we developed a visual analytics prototype that allows system designers to examine control system behavior and compare experimental results using time plots. The user may select a particular time point or interval to access and display higher resolution views of controlled entities and/or end-points, including their states. Three different visualization techniques (geospatial view, sunburst, and packed circles) were developed to show end-point results from different logical perspectives (geospatially, hierarchically, and within clusters). Through each of the visualizations, the user may animate the time-series data over a selected time interval to rewatch specific behaviors emerge in time. Users may also instrument the visualizations to produce visual alerts when simulated power grid measurements move beyond minimum or maximum thresholds.

The project is also developing a new Tree View visualization to display control system communications and expose control system structure and network communication characteristics and impacts. The Tree View visualization reveals communications as dynamic edges occurring in a hierarchical network structure, which may be expanded or collapsed to better explore specific areas or regions of the communications network.

The project is continually evaluating the usefulness and usability of visualization tools and visual analytics platform against experimental results generated from other projects. Here, control system designers perform the same analysis process to examine and validate their systems, but use the visual analytics platform to carry out the analysis. We then evaluate whether the analysis was more effective using the visual analytics platform compared to the original analysis approach.

In FY 2017, we will continue to collaborate with control system designers to further refine the visualization tools and visual analytics platform. Additional development of the Tree View visualization also will continue. Furthermore, the project will integrate the visual analytics platform with the experiment management system to draw control system experimental data from its database, which will enable visual data mining and analysis of experimental results.

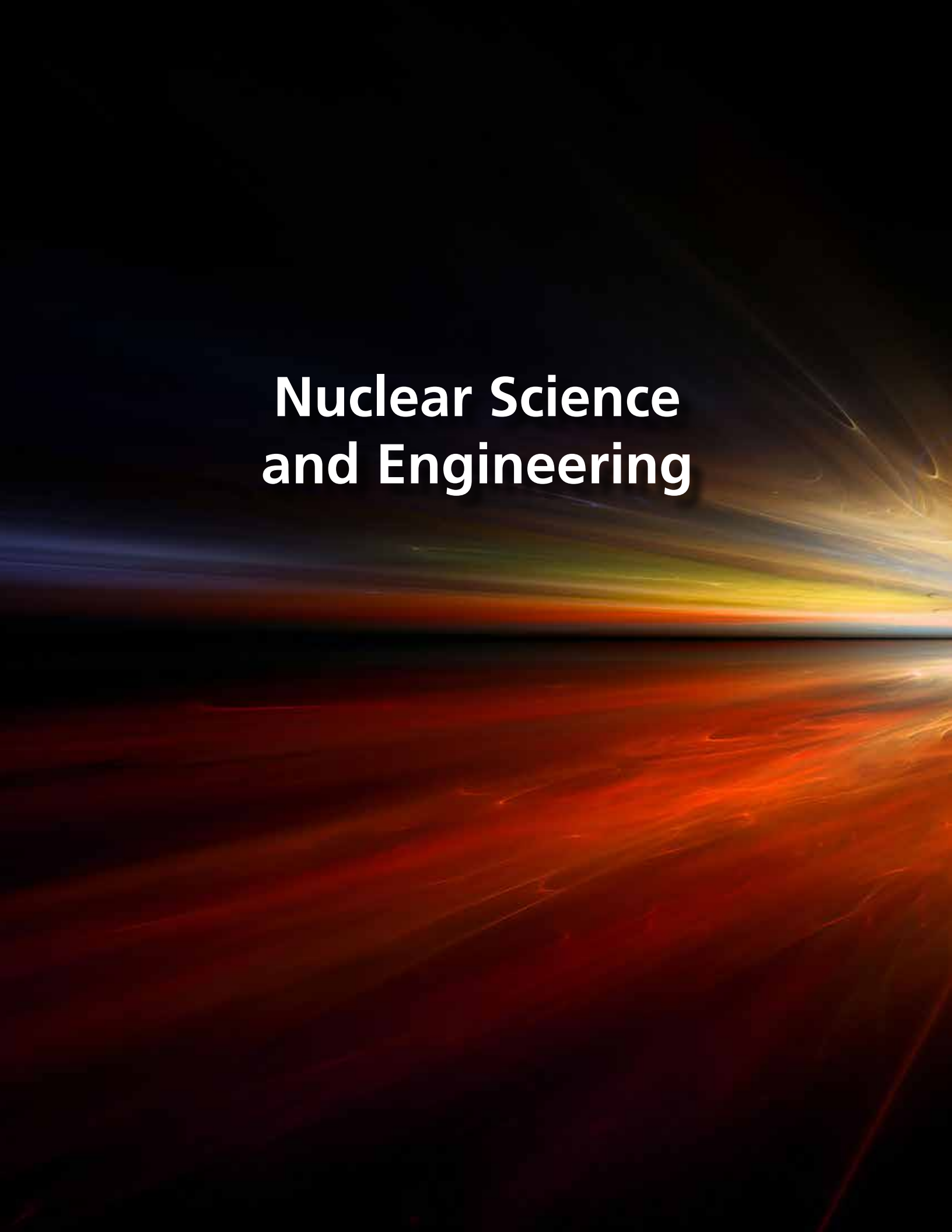


Map view, Sunburst, and Packed Circles visualizations provide alternative visual summaries of experimental results.



Time-series plots link to experimental data, which may be replayed to examine temporal data.

Nuclear Science and Engineering

The background of the slide is a dark, abstract image. It features a horizontal band of light, possibly representing a sunset or sunrise, with streaks of orange, yellow, and blue light. The light streaks are more prominent on the right side of the image, creating a sense of motion and energy. The overall color palette is dominated by dark blues and blacks, with the bright light streaks providing a strong contrast.

An *In situ* Investigation of Gamma-AlOOH Dissolution under High pH Conditions

Edgar C. Buck

Hanford boehmite is an order of magnitude more resistant to dissolution than normal forms. We are using a combination of advanced atomic resolution and in situ electron microscopy techniques, combined with theoretical calculations, to determine why this is the case and how we might be able to speed up this process.

The dissolution of boehmite is a technical issue for the Al industry because of its recalcitrant nature. However, a similar problem exists with boehmite in the nuclear waste sludge at the Hanford Site in eastern Washington State. We are exploring potential intrinsic and extrinsic effects that may limit boehmite reactivity; one clue comes from microstructural analyses that indicate an association of Cr with Al in the Hanford nuclear waste, as well as the presence of Mn and Fe. Hence, in this first year, we have investigated the potential role of Cr, Mn, and Fe on the reactivity of boehmite in caustic solution. The testing platform that we are developing will have wide application in other areas of nuclear processing science.

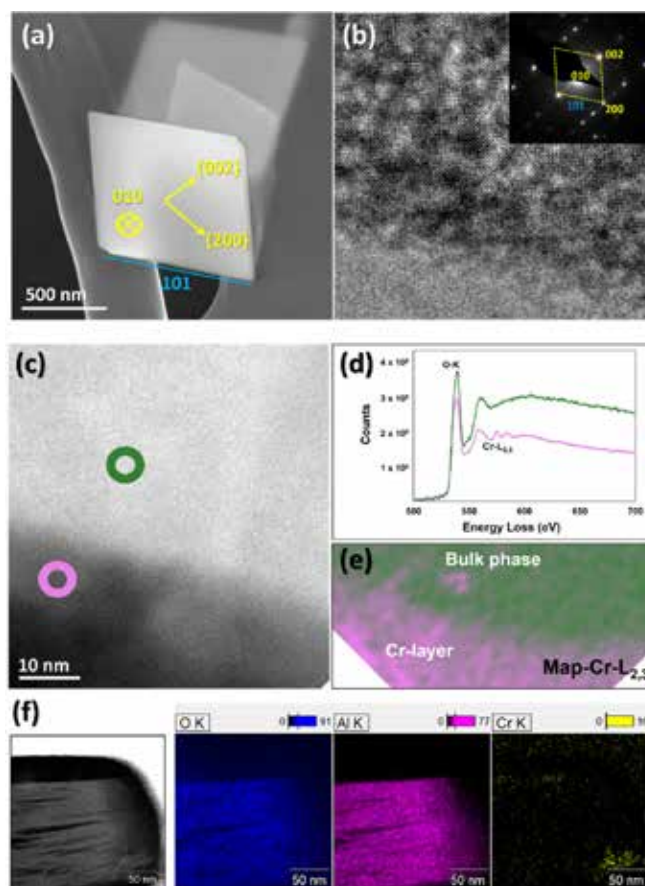
An important finding in the case of Cr was that, irrespective of the synthesis pathway, amount of Cr(III), or the resultant morphology, there was no evidence for Cr incorporation in the bulk structure, in agreement with quantum mechanics calculations. In fact, microscopic and spectroscopic analyses (X-ray photoelectron spectroscopy, transmission electron microscopy [TEM], scanning TEM-energy-dispersive spectroscopy [-EDS], -electron energy loss spectroscopy [EELS]) showed that Cr was enriched at the (101) edges of the boehmite. However, Cr had no measurable effect on the morphology during the synthesis step. In contrast, comparison of the morphologies of the synthetic Cr-doped and pure boehmite samples after exposure to caustic solutions provided evidence that Cr inhibited the corrosion. TEM showed that Cr was not homogeneously distributed at the surface. Consequently, Cr may have partially passivated the surface by blocking discrete energetic sites on the lateral surfaces of boehmite.

There was clear evidence of changes in the corrosion behavior of boehmite in the presence of Fe and Mn. Microscopic

analyses showed that the Fe and Mn reduced the extent of corrosion. In the case of Mn, this was the most significant. Cryo-TEM investigations of the boehmite system have examined the particle-particle association. Cryo-TEM is an excellent method for probing dynamic processes in solution.

Our preliminary observation that Cr enrichment at rhombic boehmite surfaces (thus far there is no evidence for bulk incorporation of Cr) affects the reactivity of boehmite during caustic leaching has implications for the behavior of boehmite (and possibly other Al phases) in the Hanford tank waste.

In the next year, we will examine the role of uranium interactions with boehmite and continue to develop *in situ* liquid cell platforms for testing out dissolution processes that may have wide applicability for Hanford-related problems.



Microanalyses of boehmite 1 and Cr-doped boehmite 1 Cr: Representative (a) scanning electron microscope, (b) TEM with selected area (electron) diffraction inset, (c) high-angle annular dark-field imaging, (d) EELS spectra of (c), (e) EELS mapping of (c), (f) STEM energy-dispersive X-ray spectroscopy mapping of focused ion beam cross section.

PN1509012765

Chemical Consequence Assessment Methodology and Code Assessment

Pete Lowry

This project seeks to develop improved methodologies for evaluating complex chemical releases that support documented safety analysis. Improved methodologies for evaluating chemical releases will produce more accurate results with a conservative bias.

The DOE standard for documented safety analysis (DOE-STD-3009-2014) requires inclusion of chemical hazards. The current guidance provided in the standard is not immediately suitable for evaluating complex chemical releases over significant distances, as may be required for determining the need for safety-related controls. Having improved methodologies and guidelines capable of evaluating a broad spectrum of chemical releases will provide results that are more accurate, while being able to demonstrate a conservative bias. Benchmarking will be required to validate improvements in the methodologies for evaluating chemical releases.

The DOE-STD-3009-2014 guidance recognizes that a number of variables can influence chemical dispersion. Therefore, it may not be appropriate to use the radiological dispersion analysis or DOE “Toolbox code” for all chemical dispersion analysis

In FY 2016, we selected ammonia as the example chemical, since it is a commonly used chemical and has the potential for significant health effects. Ammonia at standard temperature and pressure is gas, but it is often stored as a liquid under pressure/temperature. The effects the initial conditions of ammonia have on the release characteristics were researched. Secondly, the effects of the chemical form and the nature of the release on the subsequent transport of the chemical were evaluated.

Key initial conditions of ammonia after a release include the density of the gas and physical form. Ammonia, when released, can be considered either a dense gas or neutrally buoyant gas, depending on factors such as storage conditions and ambient temperature. After a sudden release, the initial state of the gas in transport can be assumed as a mix of vapor and droplets that do not fall to the ground.

For ammonia, the initial conditions that were evaluated further were liquefied under pressure and liquefied by refrigeration. Pressurized liquid ammonia was evaluated as a gas release. The flashing portion of a refrigerated liquefied ammonia release was also evaluated as a gas. The release due to evaporation from a pool of liquefied ammonia greater than 1 cm in depth from the liquid fraction of the release was also included.

For the evaluation of the release, the reaction between ammonia and water vapor was conservatively ignored.

The scenario evaluated is a rapid release from a storage vessel, thus the impact of a jet-plume was included prior to modeling with a standard Gaussian distribution profile.

Comparison of the above modeling considerations against prior work performed utilizing the existing DOE guidance showed a greater than 50X reduction in concentrations at evaluation points often associated with the distance to public receptors (e.g., greater than 1.5 miles) at DOE sites.

The improved methodologies for evaluating chemical dispersion will aid in determining appropriate analysis to use. This will also aid in the proper selection of safety-related controls. Using the appropriate dispersion analysis for chemicals can also avoid over specifying costly safety-related controls.

Correlation of Colloidal Interactions and Macroscopic Rheology in Concentrated Electrolyte Solutions

Jaehun Chun

This project will develop a “science-based” fundamental understanding of macroscopic rheology of slurry under conditions relevant to harsh storage and treatment of nuclear waste treatment, especially by connecting phenomena at different length scales (i.e., particle interactions, aggregate structures, and bulk rheology). The project will build a scientific framework to accomplish safe and efficient nuclear waste treatment processing, in addition to proper design and operation of waste disposition processes (e.g., pretreatment facility in the Hanford Waste Treatment and Immobilization Plant).

Hanford tank wastes are radiological mixtures of insoluble and soluble solids in highly concentrated salt solutions, leading to complex non-Newtonian waste rheology that must be considered during design and optimization of Hanford wastes disposition processes. The rheology of colloidal dispersion in high salt concentrations is expected to be governed by long-range ordering of individual particles or particle aggregates. This microstructure results from particle interactions that are themselves moderated by interfacial phenomena at molecular and colloid scales. Particle interactions in simple colloidal systems (e.g., single component mono-disperse spherical or other simple particle shape colloids suspended in dilute single component electrolytes) have been studied and well understood; it is modeled using the Deryaguin-Landau-Verwey-Overbeek (DLVO) theory. Resultant rheology of such suspensions has been reasonably understood via the DLVO theory.

The DLVO theory cannot be simply applicable to Hanford waste slurry. Its high salt content induces the loss of electrostatic repulsion. Owing to existence of various minerals in the dispersed phase, an aggregation can result from an electrostatic “attraction” between mineral phases with opposite surface charges. Furthermore, steric hindrance and solvent structuring can fundamentally influence particle interactions, which allow non-DLVO effects.

This project seeks to quantify these unique particle interactions (i.e., those which are not explained by the DLVO theory),

to understand how they arise in terms of the chemical physics at solid-liquid interface, and to determine how these interactions influence long-range ordering of colloidal waste particles and bulk rheology in tank-waste-relevant systems. Furthermore, a simulation framework to predict bulk suspension rheology would be built, based on such correlations.

To limit the complexity of test systems, a single solid mineral phase, boehmite, is selected for our study because 1) it exists prevalently in tank waste, and 2) it is one of the important components with respect to waste rheological properties. Colloidal interactions between boehmite particles will be measured directly under tank-waste-relevant chemical conditions using colloid probe atomic force microscopy (CPM) and will be modeled using theories/simulations that implement the relevant water and dissolved ion structuring at the particle-liquid interface.

The aggregate structure and its evolution will be investigated by two different experimental tools: *in situ* scanning electron microscope (SEM) and nuclear magnetic resonance (NMR) spectroscopy implementing System for Analysis at the Liquid Vacuum Interface (SALVI) to couple particle interactions with the microstructure of aggregates. Furthermore, the rheology of the model suspensions will be characterized using a standard bench-top rheometer (and possibly a micro-rheometer based on SALVI). As a complementary tool, colloidal scale simulations will be performed to link particle forces, aggregate structures, and bulk rheology.

Using high-resolution transmission electron microscopy (HR-TEM), we characterized the size, shape, and surface of boehmite particles and further identified crystallographic faces along with selected area diffraction. Using the 3D unit cell/2D projection analyses, we further obtained the density of surface charge groups (-OH) for different crystallographic faces (e.g., [010] vs. [101]). These details will be critical inputs to simulations of bulk rheology.

Combining such details, along with zeta potential measurements, we found that the electrostatic repulsion between boehmites is dependent on crystallographic faces; the repulsion between (101) faces is higher than that between (010) faces. Furthermore, based on the Lifshitz theory and available dielectric properties of boehmite, we calculated a correct Hamaker constant (2.1×10^{-20} J) which is about one third of the reported value. Both findings will play significant roles for rheology. We will continue to study unique particle

interactions involving water structuring at the interfaces, the effect of high salts on van der Waals forces, and hydration forces. This will provide critical components to simulations of bulk rheology.

The second thing we worked on was imaging of a single boehmite particle (less than 100–200 nm), based on the *in situ* liquid SEM equipped with SALVI. We optimized parameters and procedures using boehmite particles in liquid SEM. Important points for the optimization include 1) the usage of carbon coated devices to reduce charging during image acquisition and 2) the usage of *in situ* SEM/focused ion beam (FIB) along the microfluidic channel to provide more observation windows in a given experiment.

The optimization enabled us to acquire both secondary electron and backscattered electron detection images of boehmite particles in the high-vacuum mode, compared to other wet SEM techniques. Our results illustrate the possibility to observe particle morphology and aggregates (possibly with NMR), which would be within the scope of FY 2017.

In order to obtain a better understanding of boehmite-boehmite interactions at various conditions, such as pH, salt concentrations, ionic species, and crystal orientations, we used atomic-force-microscopy- (AFM-)based Dynamic Force Spectroscopy (DFS); DFS is necessary to study the interactions between mineral crystals, in comparison to a conventional CPM intended for spherical polymer latex particles.

We have developed a novel method for attaching thin platelets onto AFM tips for colloid probe microscopy. A boehmite crystal (thin platelet) was deposited on a Si substrate. A layer of C/Pt protective layer was deposited on top of the boehmite crystal. The AFM tip attached with boehmite crystal as a force tip was obtained via FIB cutting.

We performed force measurements on a gibbsite substrate crystal in aqueous solutions, owing to unavailability of large boehmite single crystals (greater than 5–10 μm). The value of the retract velocity was changed after every approach/retract cycle in order to acquire the distribution of the rupture forces at different retract velocities. Using these loading rates, the rupture force has a linear relationship with the retracting velocity. Therefore, we were able to obtain the equilibrium rupture force from extrapolation. We found that the rupture force has a strong relationship with pH; in water, the rupture force is highest and decreases with either increasing or decreasing pH. Based on the novel fabrication method and results, we will study forces involved in the approaching step as a function of physicochemical parameters in FY 2017. Concurrently, we will also look for a possible way to use a boehmite substrate with existing small boehmite single crystals (approximately 5 μm).

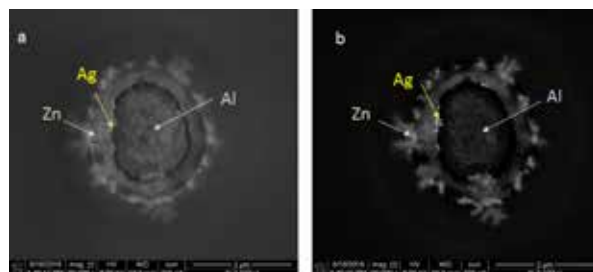
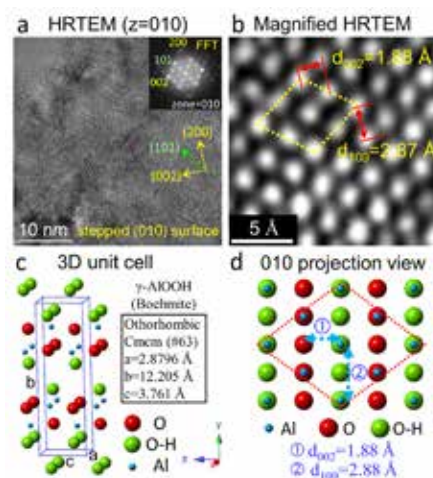
In FY 2017, we will also use several approaches to grow large boehmite single crystals, because they are not commercially available.

Lubricated Flow-Discrete Element Method (LF-DEM) has been identified as a relevant simulation framework. LF-DEM is desirable to overcome the limitations as to the size variation and surface geometry (SD) and the critical contact interactions (DPD).

We performed the simulations for bi-disperse Brownian colloidal dispersions (1.4:1 for the size ratio), varying particle concentrations and colloidal/friction forces. A cohesive force (i.e., attractive colloidal forces) between particles is known to increase suspension viscosity and even cause yield stress behavior. We found that this depends on the strength of cohesion and the volume fraction of suspension.

We observed that the effect of increasing magnitude of cohesion on the rheology of Brownian suspensions at $\phi = 0.5$, a condition chosen to allow combined examination of yield stress and shear thickening behavior. The yielding behavior has been probed down to $\phi = 0.15$. The Brownian shear thinning at low cohesion strengths is replaced with yielding shear-thinning behavior with negative one slope. The cohesive bonds and associated aggregate contact network which resists deformation are torn apart at higher shear rates. We found that, at high shear rates, the amount of cohesion in the system is immaterial to the rheology as hydrodynamic forces dominate. We will continue to study rheological behavior at a higher bi-dispersity in FY 2017, along with implementation of additional finding of particle interactions.

HR-TEM and subsequent unit cell analysis: (a) HR-TEM for (010) surface of boehmite, (b) corresponding magnified HR-TEM for (010), (c) 3D unit cell, and (d) (010) projection view from the analysis.



In situ liquid SEM with SALVI: (a) secondary electron image and (b) backscattered electron image of a mixture of ZnO, Ag, and AlOOH particles.

Detection of Production at the Source

Ron P. Omberg

Research reactors are often the first technology that a nation intent on producing unauthorized plutonium will procure. Larger research reactors can produce a significant quantity a year, if properly configured. This research will determine if such a reconfiguration will produce revealing transient operating signatures.

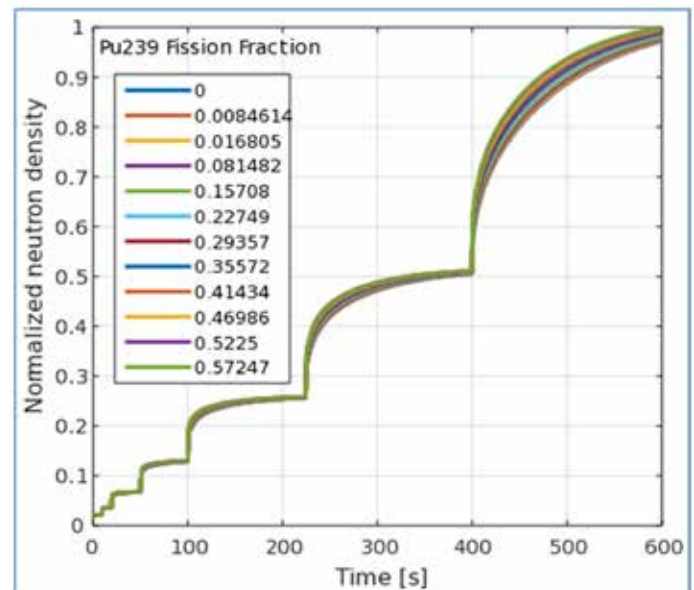
Previous efforts to detect unauthorized use of a research reactor were based on a thermal or calorimetric balance, which is performed on the primary heat transfer system. Such balances are known to be inaccurate and only determine the thermal power (i.e., the power at which the reactor is operating), rather than detecting unauthorized use. This project will examine the neutronic behavior of the reactor, which has the possibility of detecting unauthorized production by different means. The technical achievement associated with this project is to provide a signature technique that will allow declarations made by nation states to be validated with objective evidence, thereby revealing any unwarranted use of the reactor.

Research in 2015 explored the possibility that unauthorized use of a research reactor would produce a transient behavior with a revealing signature. It was found that such use would produce a different signature when the reactor is ascending to power after being shut down for refueling, and the magnitude of the signature depended upon the extent to which unwarranted use was occurring. It was also found that the magnitude of the signature could very well be small, and accurate detection techniques may be needed if transient behavior is to be the only indication.

While performing this research, it was found that there may be other possible indicators of unauthorized use that are related but not identical. These were investigated in 2016. For

example, one thing found was that reactivity changes during an operating cycle will not only affect transient behavior, but will also affect both the operating cycle length, as well as the position of the control rods during that operating cycle. The possibility that multiple signatures exist is important, because multiple signatures will always provide more reliable, objective evidence than evidence based on a single signature. And it was found that these two signatures are more revealing and are neither easily obscured, nor do they require accurate or sophisticated measurement techniques.

In summary, the reactivity changes associated with the transient behavior of a reactor during power ascent also affect the length of the reactor's operating cycle, as well as the control rod positions during the operating cycle. While not conclusive simply by themselves, when compared to the declared operation of the reactor, they can be better, more reliable indicators of use.



Transient behavior of a reactor during power ascent with differing levels of unwarranted use.

Development of a PNNL Underground Nuclear Explosion Simulation Tool

Justin D. Lowrey

We have developed a PNNL simulation capability with the aim of understanding some of the more challenging aspects of post nuclear explosion environments and the evolution of underground nuclear signatures so that we can improve detection technology design and implementation. Additionally, this tool will complement field and laboratory experimentation designed to test similar phenomena.

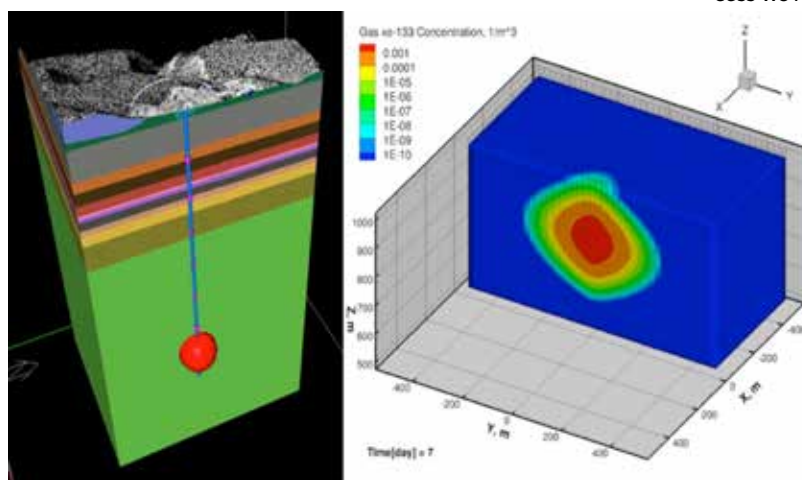
Monitoring the world for evidence of underground nuclear explosions represents a significant technological challenge and constitutes a major priority for the United States as well as the international community. For over two decades, numerical underground gas transport models have been used to increase understanding of how post explosion gases migrate through the ground and leak at the surface. The goal of developing such post explosion models is to explore how best to collect and target environmental samples that might indicate whether a nuclear explosion has occurred. This includes investigation of when, where, and how to look for explosion evidence and how to distinguish it from other civilian sources. A greater effort has been made in recent years to examine underground gas transport from a more fundamen-

tal level, as past experience demonstrates gaps in scientific understanding.

We have developed a simulation tool suite—based on the PNNL Subsurface Transport Over Multiple Phases (STOMP) environmental fate and transport software—that is capable of simulating post underground nuclear explosion scenarios by combining detailed nuclear, chemical, and environmental problem components. This tool not only allows for realistic modeling of the physical geology of such scenarios, but also facilitates simulation of the complex geochemical and hydrological interactions that post nuclear explosion radionuclides can experience underground. The simulation suite has been built to study how these fundamental processes, coupled with radioactive decay, can impact large-scale mass transport and potentially alter expected nuclear signatures.

Initial work in FY 2015 began with a broad assessment of STOMP capabilities and deficiencies for application to exploring underground nuclear explosions. This included a detailed verification of STOMP's ability to handle radioactive decay chain processes, updating STOMP equations of state to cover a wider range of temperature and pressure conditions, and running of a set of 3D non-isothermal test problems. Efforts in FY 2015 culminated in building a generic, large-scale post explosion test case, consisting of a problem domain with variable geologic media, as well as initial fission product deposition into the subsurface by high temperature and pressure.

Into FY 2016, this test case was used to define a general process flow for how to build generic scenarios in the simulation tool. The resulting framework was then utilized to conduct a large sensitivity study covering, among other parameters, detonation depth, rock porosities, permeabilities, temperature, and pressure. Results from this study helped illuminate those parameters that had the greatest impact on nuclear explosion xenon gas reaching the surface where it might be detected. With close to 1,000 simulated scenarios, various uncertainties can be quantified based on the output of the model and, ultimately, used to evaluate the predictive capability of the new underground nuclear explosion simulation suite. Lastly, how the new tool suite could be integrated into other dynamic tools and frameworks to create end-to-end models was examined.



Geologic environmental detail that can be used by the post nuclear explosion simulation tool suite being developed by this project. Left: an EarthVision geologic framework model incorporating detailed information about the problem domain; right: a 3D visualization of the ^{133}Xe gas concentration simulated from an explosive dispersal of ^{133}I in the ground with the concentration front breaching the surface.

D-T Neutron Generator Based Standard to Replace ^{252}Cf

James R. Hilliard

Through the use of a 14 MeV neutron generator and various moderating materials, we will develop one or more neutron reference field alternatives to the use of ^{252}Cf in the process of calibrating and/or testing personnel dosimetry and other neutron detection devices employed for radiation safety assessments at nuclear power plants, DOE, sites and other industrial occupational neutron radiation environments.

Californium-252 is relied upon, nationally and internationally, to produce fission neutron reference radiation fields for calibrating neutron radiation measurement devices, because of its high specific activity. For emulating the occupational neutron radiation environments of various nuclear facilities, especially nuclear power plants (NPPs), ^{252}Cf may be further placed within a sphere of heavy water (D_2O) moderator. High activity sources (e.g., greater than 10^8 n/s), needed for establishing high dose equivalent rates, must be replaced about every 5 to 7 years, due to a relatively short half-life. Such encapsulated sources have become very expensive to procure, and a former DOE loan/lease program has been discontinued. Neutron generator (NG) produced neutron radiation fields have been applied for isolated cases of neutron survey instrument evaluation, mostly outside the United States. Some laboratories have developed NPP-surrogate “workplace” neutron reference fields using NGs. This project explores the challenges and feasibility of applying the deuterium-tritium (D-T) fusion reaction, yielding approximately 14 MeV neutrons, to evaluate and/or calibrate health physics survey instruments and personal dosimeters. In addition to facilitating a 14 MeV neutron field, researchers seek to reshape spectra to serve as surrogates for the ^{252}Cf fission spectrum, U.S. workplace spectrum from D_2O -moderated ^{252}Cf or even more appropriate workplace surrogate spectra.

In the initial year of this research, it was found that the current U.S. “workplace” neutron spectrum, yielded by D_2O -moderated ^{252}Cf , is not unilaterally representative of all workplace spectra, and its overreliance could lead to substantial inaccuracies of estimated personnel dose equivalent for neutron-exposed workers. A review of previously measured NPP workplace spectra was used to propose an alternative surro-

gate NPP workplace spectrum. Monte Carlo modeling was conducted, using simple spherical geometry, to establish a library of beam shaping light and heavy element candidates of multiple thicknesses. The influence of these materials was gauged relative to various desirable and undesirable characteristics (e.g., fluence preservation, energy reduction, prompt gamma production, etc.). Then, various combinations of two to five different materials were evaluated for shaping 14 MeV neutrons to approximate the target NPP spectrum. A rating system was developed to aid in the objective selection of various resulting shaped spectra. A common facet of the various beam shapers considered was their significant mass, which could be up to 2,000 kg. In anticipation of developing a practical beam shaper to attain the surrogate NPP spectrum using the PNNL-owned Thermo Scientific D-711 NG, the NG target/head was moved to a position for which floor loading specification would accommodate such a massive object. A conceptual beam shaping assembly, fitting the neutron tube of the D-711 NG, was designed to achieve the surrogate NPP workplace spectrum.

The practical aspects of employing an NG for the high-accuracy requirements of calibration and testing were explored. Calibration and monitoring of a NG-produced 14 MeV or shaped alternative neutron spectra are key challenges. Through a survey of measuring possibilities, a favorable method emerged utilizing a fission chamber transfer-standard to determine the fluence rate of the NG-produced spectra relatable to the fluence rate of a ^{252}Cf source calibrated at National Institute of Standards and Technology (NIST). This method relies on highly accurate modeling of both fields. In order to develop the necessary models for the chosen calibration methodology, a thorough evaluation was made of the D-T neutron production methodology employed in the D-711 NG. Two neutron detection methods, including a fission chamber and tissue-equivalent proportional counter, were explored for monitoring the NG-produced neutron beam, both of which were found to be well-suited.

In addition to the mechanisms for accurately identifying the neutron field characteristics, challenges with use of the D-T system were addressed. As a result of observed instabilities in the PNNL system during ramp-up and extended operation at full power, it was determined that operating the D-711 NG at the minimal power level would optimize the accuracy of the

produced/delivered neutron dose equivalent. Additionally, this would also potentially extend the lifetime of the neutron tube and reduce the facility neutron background and potential neutron-activation of nearby components.

Through collaboration with NIST, the means to generate a fission-like neutron spectrum were explored (i.e., to replace bare ^{252}Cf). Although such a reference field is also not representative of workplace spectra, it is viewed by NIST and their counterparts at other National Metrology Institutes, as being highly important for interlaboratory comparison and proficiency testing. Two conceptual shaping approaches were developed. One method relies on the use of a fission reaction in a depleted uranium converter within the beam shaping structure, while the other relies on (n,2n) reactions and inelastic scattering in heavy metals to attenuate the nominal 14 MeV primary neutron component to shape the field in to the desired spectrum.

Spectra for the best and fourth place shaping configurations in comparison to the surrogate nuclear power plant spectrum. The D_2O -moderated ^{252}Cf spectrum is shown for comparison. Ratings beneath each shaping configuration (including D_2O -moderating sphere) represent *spectrum shape matching* (S_I); lower values represent better matches, *neutron fluence preservation* (Φ), the *fraction of 14 MeV neutrons* remaining in each shaped spectrum (f_{14}), *mass* and *volume* of each shaper assembly relative to the mass and volume of the D_2O moderator used with ^{252}Cf (M and V , respectively), and the *prompt gamma-to-neutron ratio* (γ/n).

Gamma-Gamma Coincidence Analysis Algorithms

Glen A. Warren

This project will advance our capabilities in gamma-ray signature analysis, enabling more efficient and accessible evaluation, in turn providing greater sensitivity than traditional single gamma-ray spectroscopy.

Gamma-gamma coincident signatures improve detection sensitivity of select radioactive isotopes in a variety of applications. It is particularly powerful for conditions that render single gamma-ray spectroscopy methods less effective such as high backgrounds, high count rates, and low-resolution detectors. Coincident gamma-ray signatures have the potential to be exploited for a variety of nuclear science applications, including low-background measurements, post-detonation forensics, and environmental monitoring.

Despite these proven advantages, the nuclear science community has been slow to utilize gamma-gamma coincident signatures in applied nuclear research. The main barrier to a large-scale use of this technique is the complexity of the analysis: no publicly available tools are available to streamline the analysis, and there are few publications on the underlying methodology. Our project aims to reduce that barrier greatly by developing well-vetted algorithms for gamma-gamma coincident signature analysis. We have developed algorithms for this type of analysis, tested them on simulated and real data, and are in the process of publishing our results.

There were three main accomplishments for the project in FY 2015. First, we completed a review of prior work in the

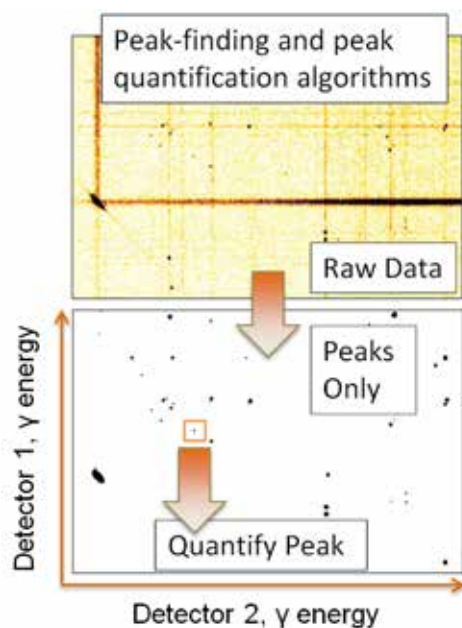
field, including publications, existing analysis tools at PNNL, and engaging subject matter experts. This review confirmed that there is no existing solution to this problem, and that there is significant interest in the community in developing a solution. In addition, we commissioned two gamma-gamma coincident detection systems that increase PNNL's capability to perform gamma-gamma coincident measurements. These

two systems enabled us in the project to test algorithms against well-known calibration sources and leverage a variety of experimental sources from other ongoing PNNL work. Finally, the project began work on algorithm development, identifying several possible algorithms. The ground work for a testing framework to evaluate algorithms has been developed. Simulated and measured data on fission products have been used to support initial algorithm development.

For FY 2016, we focused on developing and testing analysis algorithms. Five novel algorithms for signature quantification were identified for further examination. These algorithms were refined and applied to a set of test data. The results of the five algorithms were compared to commonly used manual gamma-gamma coincidence analysis techniques. Significant improvements were demonstrated in some of the novel algorithms.

In addition, two algorithms were developed for peak-finding, which refers to the ability to locate a signature and distinguish correctly between real signatures, uninteresting spectral features, and random noise.

A peer-reviewed journal article on our findings is in draft form and nearing completion. It is expected to be published in FY 2017.



This pair of spectra show one of the algorithms devised in this project in action. The top image shows raw gamma coincidence data from a pair of detectors; the round dots are the signatures of interest that need to be separated from other features. The bottom image shows the isolated peak data after one of our algorithms is applied.

Hot Particle Analysis Aided by a State of the Art Focused Ion Beam

Dallas D. Reilly

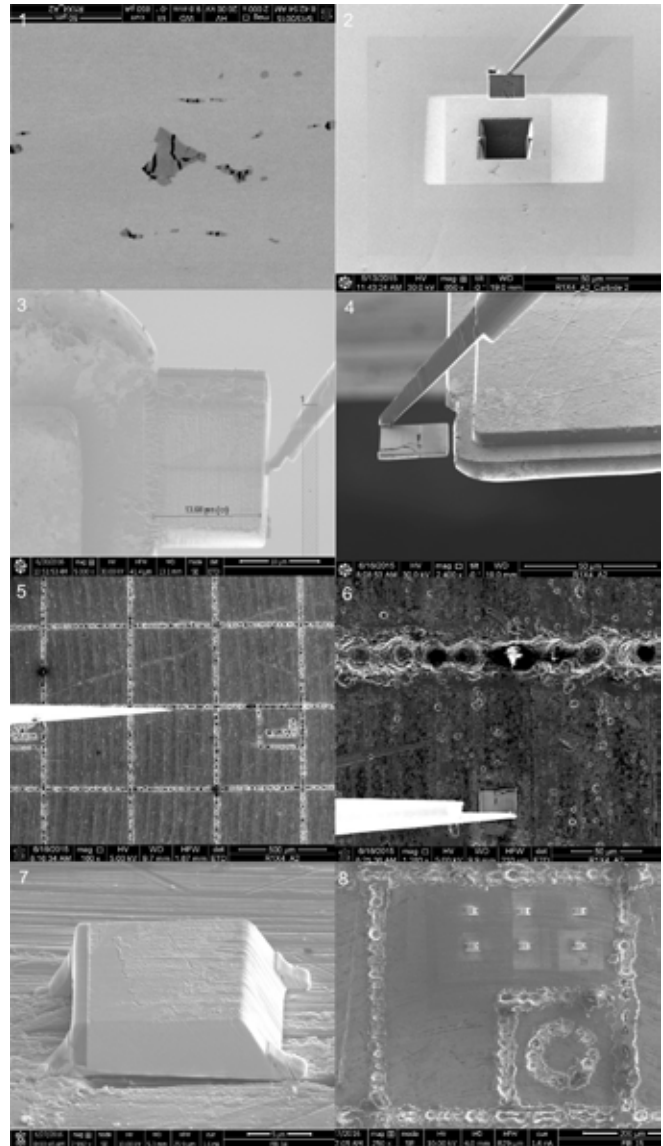
This project seeks to use focused ion beam (FIB) technology to subsample highly radioactive materials for analysis, advancing the capabilities for radioactive materials study around PNNL and the DOE complex.

The main intent of this project is to use a combination FIB/scanning electron microscope (SEM) to analyze and subsample highly radioactive materials for movement into laboratories and instruments at PNNL that do not currently or regularly accept highly radioactive samples. The Radiochemical Processing Laboratory (RPL) is a facility at PNNL that has the capability to perform chemistry and analysis on many radioactive materials up to very large quantities. However, it lacks analytical capabilities in which other parts of the laboratory excel.

A state-of-the-art FIB/SEM has been acquired at RPL for subsampling and analysis of radioactive materials. These instruments have been used previously in the semiconductor industry for microscopy sample preparation and analysis, and show a lot of promise when applied as radioactive material subsampling tools.

Thus far, this project has focused on various subsets of samples, including low-enriched uranium metal fuels, used nuclear fuel remnants, and plutonium hot particles to prove the worth of this technology for national security, basic actinide sciences, and areas relating to the nuclear fuel cycle. These materials have been “free-released,” a process that uses Nuclear Regulatory Commission allowances for low-level surface contamination to determine if there is any risk associated with a given material. The paperwork for these releases is in place, and those samples have been moved to both ultra-trace and completely non-rad facilities for advanced analysis, including nano-secondary ion mass spectrometry (nanoSIMS), large geometry SIMS, atom probe tomography, other mass spectrometry methods, and transmission electron microscopy. These analyses have resulted in new science, publication potential, and a high likelihood of follow on funding.

In FY 2017, this project will continue and focus on various plutonium samples for national security, energy, and environmental management mission areas.



Workflow for using the FIB/SEM to locate areas of interest, extract, move to a new substrate, and weld down for analysis on other instruments.

Interfacial Diffusion and Crud Formation at the Liquid:Liquid Interface of Solvent Extraction Processes

Amanda J. Casella

Tributyl phosphate is an important extractant in liquid:liquid reprocessing; it undergoes radiolytic degradation, yielding species that participate in formation of unwanted solids at the liquid:liquid interface (e.g., cruds). These cruds sequester metal cations and contribute to materials unaccounted for. The aim of this work is to determine the mechanisms of transfer and solid phase nucleation at the liquid:liquid interface.

The transport of butyl phosphate extractants and actinides across the liquid:liquid interface determines the efficiency and effectiveness of solvent extraction processes. In addition, these species can organize at the phase boundary, sometimes leading to the formation of unwanted interfacial solids. In this project, the rates and mechanisms of solute interfacial transfer are being investigated.

Tributyl phosphate, an extractant in liquid:liquid reprocessing, undergoes radiolytic degradation, yielding dibutyl and monobutyl phosphate. The solubilities and rates of transfer for these species vary as a function of diluent and process scale. This investigation will also look at the scale dependence for interfacial processing and the resultant signatures by comparative studies on macro- and microfluidic scales.

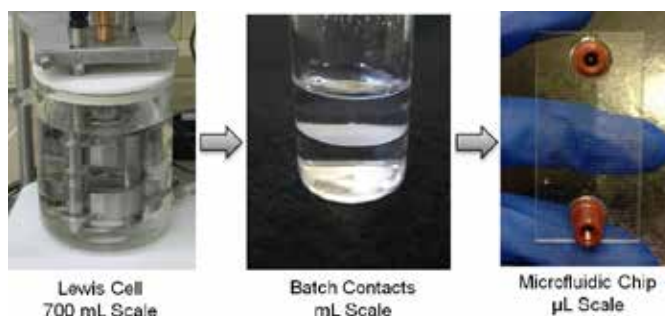
This project is in its first year and had a mid-year start. Extensive document reviews have been performed of historical Hanford records mapping crud occurrence to process and stream conditions for both classic plutonium uranium extraction (PUREX) systems and those used specifically for Pu purification. This mapping provides the framework for Year 2 experimentation, investigating the rates and mechanisms controlling the transport of butyl phosphate species and the formation of interfacial solids at the liquid:liquid interface.

Year two will focus on macro-scale testing, using real-time monitoring with Raman spectroscopy. Specifically, this work

will target examination of the fundamental kinetic drivers governing chemical signature evolution for solvent extraction. The progress into the third year will examine how these rates and signatures vary as a function of scale by performing the extractions in microfluidic cells.

In addition to experimental work, computational approaches will also be used to gain further molecular insights about liquid:liquid interfacial processes. Examination of the relationship between adopted configurations of the solute, orientation of the solvent, and the ability of the solute to enhance microsolvation, may provide clues regarding molecular organization of precursors to solid phase nucleation and formation of interfacial solids. Computational results will be integrated with experimental results to link spectroscopic signals from Raman and other experimental approaches in order to gain molecular insight.

The specific aims of this work seek to provide a scientific basis for anticipation of and exploitation of chemical signatures in liquid:liquid reprocessing both under traditional PUREX conditions (e.g., n-dodecane as the diluent) and those targeted for Pu purification (i.e., CCl_4 -based systems). In addition, by detailing the scale dependence for chemical processing and the resultant signatures in liquid:liquid reprocessing, new signatures can be obtained to reveal operational scale and rates of processing.



Monitoring Diffusion of Actinide Daughters and Granddaughters in Metals for Chronometer Applications

Dallas D. Reilly

This project seeks to understand trace elemental fractionation at the solid-liquid interface during casting or remelting of uranium (U) metal, a process important to the nuclear forensics community for the application of radiochronometers of process age and the development of new, process-derived signatures. The basic science generated here will also benefit efforts currently underway by DOE to develop lower-proliferation-risk, low-enriched uranium metal-alloy fuel alternatives to high-enriched uranium oxide fuel forms currently in use by many research reactors.

Past studies have observed trace elements like thorium (Th), the daughter product of U decay, fractionate quantitatively during metal casting. However, the mechanisms controlling this phenomenon are not understood. The goal of this project is to elucidate these mechanisms by combining high-temperature controlled experiments, molecular-scale observations, and theoretical modeling. If successful, determining the fractionation mechanisms of Th during U metal casting will not only confirm the use of U/Th parent/daughter radiochronometers as an endogenic nuclear forensic signature of U casting process age, but also lay the groundwork for interpreting the next generation of chemical process signatures within actinide metals.

In order to ease the burden of analysis, initial experiments have been commissioned using U metals doped with high concentrations of Th. The primary mission for these initial experiments was to identify the most likely chemical and physical phenomena controlling Th fractionation. A prototype induction furnace system capable of melting U metal samples has been constructed and tested, although further refinement of this system is planned. This system will be used to force fractionation of Th across the solid/liquid boundary under highly controlled conditions. A newly constructed metal production capability at PNNL has also been utilized to produce U metals with measurable amounts of Th in order to track the total inventory of the element within various regions of the

cast. Regions of interest within these samples were extracted for mass spectrometry measurements of Th and U isotopes. These analyses are ongoing. Modeling efforts have been undertaken to both interpret experimental observations and test our understanding of the mechanisms controlling Th fractionation during U metal casting.

Our plans for next fiscal year and beyond include 1) further refining the induction furnace system, 2) initiating an extensive analysis campaign of well-pedigreed high-enriched uranium metal being shipped to PNNL from Y-12, and 3) expanding our focus to other parent/daughter relationships, including $^{231}\text{Pa}/^{235}\text{U}$ —a potential signifier of historic chemical processing age of the material prior to its conversion to an oxide and casting as a metal.

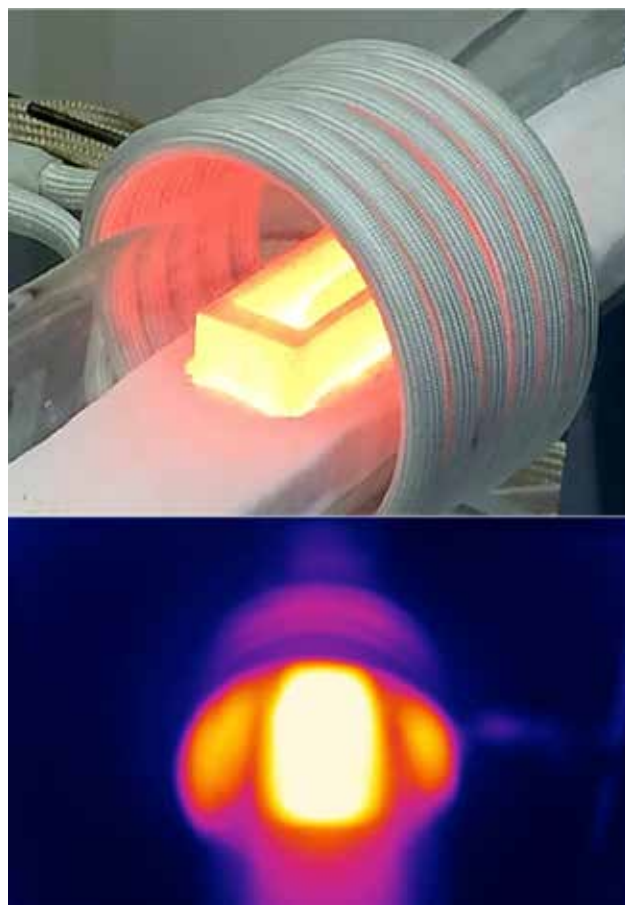


Photo of the induction furnace melting a sample of uranium metal (top) and the image that the induction furnace's infrared camera produces (bottom).

Optimization of PUREX for Centrifugal Contactors

Gregg J. Lumetta

This project seeks to determine optimal operating conditions for running a typical PUREX flowsheet in 2-cm centrifugal contactors and to determine the effects on plutonium recovery and purity if the contactors are not operated in the optimal manner.

Solvent extraction is a well-established method for processing of nuclear materials and the PUREX process is the world-wide industrial standard for recovering uranium and plutonium from irradiated nuclear fuel. The PUREX process uses tri-butyl phosphate (TBP) dissolved in an aliphatic diluent to co-extract uranium(VI) and plutonium(IV) from strong nitric acid solution. Under these conditions, most of the fission products remain in the aqueous phase and are, thus, separated from the uranium and plutonium as the latter two elements are transferred to the organic phase. The plutonium is separated from the uranium by reducing the plutonium to the +3 oxidation state. Plutonium(III) is poorly extracted by TBP, so it partitions to the aqueous phase, while the uranium(VI) remains in the organic phase.

Most fuel reprocessing plants today rely on using pulsed columns in the first cycle of PUREX. Use of centrifugal contactors in this application has been limited. Centrifugal contactors offer significant potential advantages over pulsed columns for use in fuel reprocessing. Specifically, 1) they are easier to model, in that each individual contactor represents a single separation stage, and 2) the short residence time in the contactor minimizes the radiological dose received by the solvent, thereby reducing the degradation of the solvent. The latter consideration is important with respect to modern operation of commercial nuclear power reactors, which are tending towards increasingly higher burnups.

Centrifugal contactors consist of a cylindrical well, typically milled into a stainless steel block. A hollow rotor is inserted into this well. When operating, the rotor is spun at a high rate and the organic and aqueous phases are fed into the annulus between the spinning rotor and the sides of the well. In this region, the spinning rotor results in efficient mixing of the two phases, leading to phase transfer of the components as

desired (e.g., extraction or stripping). The mixture flows over a series of veins into the inner part of the rotor, where they are separated by the centrifugal force of the spinning rotor. The separated phases then flow out the top end of the contactor and over a set of weirs, into separate streams. To execute a process flowsheet, several contactors are connected in series, and the organic and aqueous phases are flowed in a counter-current direction. The key parameters that influence the efficiency of the separation include the flow rates (both the organic and aqueous solutions), the rotor speed, and temperature. The size of the contactor (i.e., the diameter of the well and rotor) is also important, but this is not a parameter that can be varied on a given contactor.

A set of existing 2-cm centrifugal contactors is being used in this work. As designed, the rotor speed on these contactors is fixed at 3,600 RPM. Because we sought to probe the effect of rotor speed on the PUREX extraction efficiency, the first task undertaken was to modify the contactors so that the rotor speed could be varied. This was achieved by routing the power to the contactor motors through a frequency modulator. The contactors use small, single-phase synchronous motors designed for 3,600 RPM. A single-phase, 120-volt frequency controller was procured to control the speed of the motors. With this controller, the contactor motors can be operated at speeds from approximately 2,100 to 3,600 RPM.

An experiment was performed to determine the effect of rotor speed on the extraction of U(VI) and Th(IV) from HNO_3 solution by 30 vol% TBP dissolved in *n*-dodecane. Raman spectroscopy was used to monitor the U(VI) extraction in real time. Selected grab samples were also taken and analyzed by gamma spectroscopy and inductively coupled plasma optical emission spectroscopy. The Raman system was calibrated by flowing the HNO_3 solution through the contactor system and increasing the U(VI) concentration in a stepwise manner. The intensity of the Raman band at 871 cm^{-1} , attributed to the UO_2^{2+} ion, was measured and plotted against the U(VI) concentration to generate the calibration curve.

A single 2-cm centrifugal contactor unit was operated with an aqueous 2.9 mol/L HNO_3 solution containing 86 g U/L and 0.7 g Th/L and an organic phase consisting of 30 vol% TBP in *n*-dodecane. The experiment was commenced at full rotor

speed, with incremental decreases in the speed until phase separation ceased to occur. No statistical change in the U(VI) or Th(IV) distribution ratios was observed as the rotor speed was lowered to the point of phase separation failure. This suggests that the PUREX process is robust, and foreseeable variations in the rotor speed would have no significant impact on process performance.



Frequency controller to modulate the rotor speed on centrifugal contactors.

Particle-Filter Surface Interactions and Dynamics in the Presence of Cross-Flow

Richard C. Daniel

By direct observation of the accumulation of mineral oxides on sintered steel filtration media, this project will provide unique characterization of the fouling processes that hinder efficient and sustainable filtration of industrial slurries. These measurements, which hereto have been limited to biological systems on morphologically simple filter media, will guide development of predictive models that, given knowledge of chemical properties of the slurry being treated, will allow prediction and optimization of industrial slurry filtration processes, like those used in the disposition of nuclear waste treatments.

Filtration of mineral oxide slurries, including nuclear waste slurries stored at the Hanford Site in Washington State, has been studied extensively. However, most studies in the peer-reviewed literature focus on conditions that are not relevant to industrial processing, including filtration at low solids concentration (0.01-wt% or lower), with solids of small particle size (1 μm or less), under laminar flow conditions, and using dead-end configurations. In contrast, industrial processes operate at relatively high slurry concentration (1-wt% or greater) and involve complex dispersions with both colloidal and granular particles (submicron to 100 μm or greater) processed under turbulent flow in crossflow filter configurations.

Few predictive models relevant to industrial processing exist, and for Hanford applications, use of existing crossflow filter models does not yield an accurate prediction of the rate and extent of filter fouling observed. Filtration of Hanford wastes is unique in that it requires use of a sintered stainless steel filter media that can withstand long-term (40 years or more) contact with the highly toxic, corrosive, and radioactive wastes. Sintered stainless steel media are rarely used in scientific filter studies and little if any data on how particles interact and accumulate on these filters are available in the general literature. Furthermore, several Hanford-specific filter studies suggest that crossflow filtration of mineral oxide slurries has not been able to obtain the performance expected from this configuration (i.e., a rapid and relatively large

steady-state filter permeate production rate). As the permeate production rates can often limit throughput in operations that rely on crossflow filtration of solid-liquid separations, the need to understand, and with that understanding, control how mineral solids accumulate on sintered filters in the presence of crossflow is crucial to understanding how to improve and optimize industrial filtration operations.

Studies relevant to mineral oxide filtration have, to date, relied solely on post-measurement interpretation of declines in permeate production to understand how solids accumulate on the filter surface. Recent studies of filtration in biological systems have used direct visual imaging of the filter process to understand how cells and bacteria accumulate and interact with filter media. This project aims to apply recently developed visualization techniques to image how solid waste simulant particles accumulate on sintered stainless steel filter elements in the presence of crossflow. This imaging, coupled with simulation of particle-filter interactions under flow conditions, will help improve understanding of the unique filtration behavior observed for mineral oxide slurries undergoing crossflow filtration, lead to the development of improved predictive models, and help engineers improve optimization of industrial crossflow filtration operations.

Design of the Crossflow Visualization Cell (CFVC) began in February 2016 and included both 1) the design of a flow visualization cell that was representative of the filter elements used in typical Hanford waste processing and that would allow observation of the filter element and pressures representative of typical commercial filtration (20 to 60 psi) and 2) the selection of associated systems like the suspension reservoir, pump, and flow metering devices. Commercial options for the CFVC were explored and found to be inadequate, both in terms of being able to accept stainless steel filter coupons and in their ability to visualize crossflow filtration. As such, a custom CFVC was designed and built at PNNL. Following completion of the CFVC design, evaluation of the pumping power and configuration needed to reach the target flow velocities (up to about 15 ft. s⁻¹) and pressure (up to 40 psi) and selection of the make and model of a pump capable of meeting these system demands was completed. By May of 2016, fabrication of the CFVC and delivery of all CFVC components (with exception of the pump) was completed. By late May of 2016, assembly of the CFVC and subsystems had been completed,

with a lower power gear drive pump (capable of 0.5 ft. s^{-1}) substituted for the more powerful pump (whose delivery was delayed until August 2016). In addition, a second CFVC was designed to support an external collaboration with Howard University.

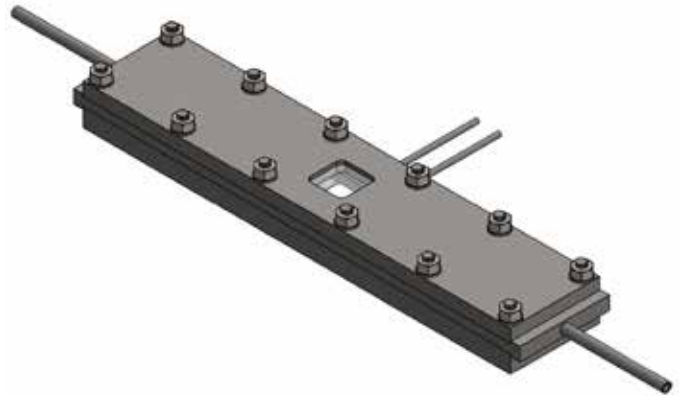
As part of the effort, PNNL formed a collaborative partnership with Dr. Kimberly Jones (Professor and Chair of the Department of Civil and Environmental Engineering, Howard University). Since the commencement of efforts, PNNL staff members have worked directly with Dr. Jones to design the CFVC and experiments to provide insight on the dynamics of particle-filter interactions and how those interactions impact the magnitude and rate of decline in filter performance. As part of the collaboration, three undergraduate students from Howard University, Sekayi Curtis, Diswash Bajracharya, and Preety Shakya, engaged in a Minority Serving Institutions Partnership Program internship at PNNL from May until August 2016. The students helped perform preliminary shakedown and scoping testing on the CFVC and continued collaborative research with PNNL after their return to Howard University for the 2016 school year.

PNNL staff members, with the help of the three summer intern students from Howard University, performed shakedown and preliminary testing of the CFVC system. Shakedown tests included microscopic characterization of a Mott Grade 0.2 sintered stainless steel filter and characterization of the clean-water flux of these filters. Two test dispersions composed of nominally $5 \mu\text{m}$ blue fluorescing polyethylene (PE) beads and $40 \mu\text{m}$ red fluorescing PE beads, respectively, were prepared and characterized. Preliminary filter testing of a 1-ppmv red bead dispersion demonstrated that accumulation of fluorescing beads on the filter surface could be reliably measured and quantified as a function of time and correlated to filter flux decline as a function of time. Methods for cleaning latent bead contamination from the CFVC system were developed and refined to support future testing.

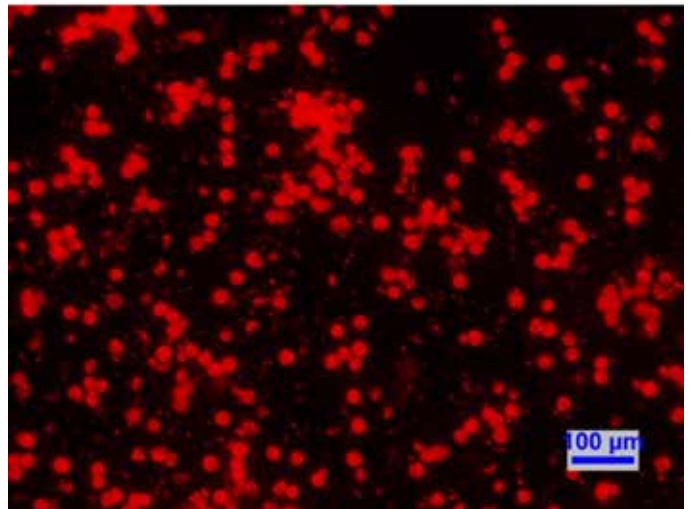
Continued work on the filtration project in late FY 2016 and FY 2017 will involve the following actions:

- Finalize and improve CFVC design by installing a more powerful pump and adding a permeate flow meter.
- Perform well-controlled, single component dispersion filtration tests with 1 ppmv dispersions of $5 \mu\text{m}$ and $40 \mu\text{m}$ fluorescing PE and glass beads.
- Perform well-control filtration tests of bi-disperse mixtures of $5 \mu\text{m}$ and $40 \mu\text{m}$ beads.

- Explore computational fluid dynamics modeling of particle-filter interactions.
- Develop publication based on findings of single component testing for publication in early FY 2017.
- Develop publication based on findings of bi-disperse mixture testing.



Schematic of filtration visualization flow cell.



Direct visual observations of red fluorescing solids on a sintered stainless steel filter element.

Provenance and Pathways Investigation of Uranium Oxide Particles Using Oxygen Isotope

Dallas D. Reilly

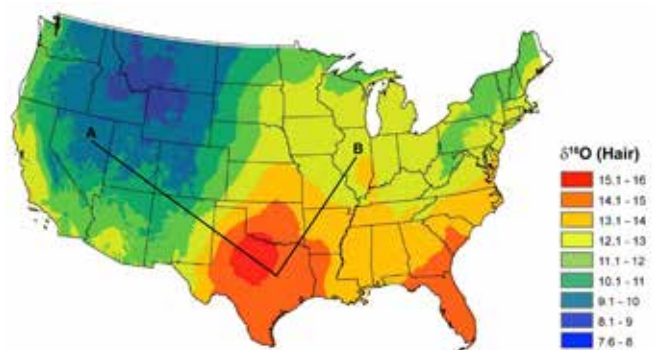
This project is developing new ways in which nuclear forensics scientists can look at material to gather the most information possible for a given investigation. When successful, this research will allow end users to understand questions about nuclear material such as: Where was this material produced? Where was it stored? When was it chemically altered?

This project was initiated because there has been a significant amount of groundwork laid by the geochemistry and climatology communities on oxygen isotope (oxygen-18 compared to oxygen-16) fractionation to study the history of our planet, and there is a potential for using similar studies on materials of interest to the national security community. For example, there is a very good understanding of the oxygen isotope ratio in water vapor across the planet; if a material like a uranium oxide is diverted from a known location and is interdicted at another known location, the water vapor absorbed by that material may indicate which route it took between those points. That information may help nuclear forensics scientists answer important questions relating to their investigation. By the end of this project, we hope to be able to apply oxygen isotope measurements to compounds like uranium oxides to provide qualitative information on its origin.

The main focus in the first year of this project was to develop capability to measure oxygen isotope signatures on the bulk

(about 5 mg via fluorination separations) and particle (via large geometry secondary ion mass spectrometry [SIMS]) scales. The bulk analysis will be used for larger samples when available, as well as to standardize samples for particle analysis, since matrix and standard calibrations are vital for proper performance. The bulk separation and analysis system is ready for testing, and measurements have already been made on uranium oxides via SIMS.

In FY 2017, the bulk separation and analysis systems will be tested on various uranium oxides, experiments will be conducted to monitor the uptake of water vapor for uranium oxides, and test cases will be run for various uranium oxide fuel pellets produced at different locations.



Map of oxygen isotope ratios of human hair in the United States; uranium oxides likely absorb local water vapor in a way similar to how the human body does. As an example, if a material was lost at point A, traveled through a region with a high concentration of oxygen-18, and was recovered at point B, investigators may be able to determine a likely travel route.

Signatures of PuF₄ and Pu Metal Processing

W. Karl Pitts

The goal of this project is to reconstitute subsets of Hanford's Plutonium Finishing Plant (PFP) processes used to produce plutonium (Pu) metal and recover Pu from various waste forms.

The production of plutonium metal from plutonium tetrafluoride (PuF₄), its shaping and forming, and the recycling of scrap materials are all of interest to the nonproliferation community. These production steps are beyond those required to form plutonium oxide for mixed-oxide (MOX) reactor fuel and are indicative of weaponization activities. However, these same processes might be used legitimately to produce metallic nuclear fuel for fast reactors. Specifically, PNNL is converting PuF₄ from the last remaining Hanford stock to Pu metal in order to understand the process that was used to produce Pu metal and weapons components at U.S. production sites during the Cold War. This reduction process was developed at Los Alamos in 1944 during the Manhattan Project, and it became the baseline process for most of the plutonium metal produced by the United States during the Cold War.

During FY 2015, legacy material was analyzed using a variety of analytical techniques to characterize the plutonium isotopic composition with mass spectroscopy and the chemical form and properties with a variety of spectroscopic analyses. These analyses confirm that the material was chemically separated from other radioactive materials in late 1966 with subsequent conversion to PuF₄. Numerous characterizations showed that, while the PuF₄ has undergone metamictization during its 50 years of storage as a reference neutron source, it retained its particulate morphology and structure known from historic studies performed at the PFP. In FY 2016, further measurements conclusively showed that simple techniques, such as heating, could reverse some of the observed changes.

The reduction process takes place by reacting the PuF₄ feedstock with calcium metal at high temperatures in a reaction similar to a thermite process. Calcium metal, iodine, and PuF₄ are placed in a ceramic crucible and heated in an induction furnace. The iodine and calcium react at several hundred degrees to quickly bring the PuF₄ and calcium to ignition. After cooling, there is a dense button of plutonium metal and a slag of calcium fluoride. The PNNL system uses a custom-designed induction furnace that is compatible for installation into a glovebox certified for plutonium operations. In addition

to the induction coil, the system includes a vacuum and inerting chamber to maintain atmosphere inside and around the reaction vessel, a load cell designed for sealing the reaction vessel and serving as pressure relief, and temperature control using an infrared pyrometer with a thermocouple readout for over-temperature control.

During FY 2016, the reduction apparatus was installed in a glovebox for plutonium operations. Before the installation, a series of tests were successfully performed using a CeF₄ surrogate and a high-quality cerium button was formed. Very similar process parameters were used for the first 10-gram PuF₄ reduction. The reduction also gave a dense, well-formed metallic button with good separation from the slag. The plutonium button is being polished and prepared for further analysis in FY 2017. The expected outcomes during FY 2017 include at least one more reduction to produce gallium alloyed plutonium and an extensive series of tests on the chemical and physical properties of the metal.

Results of a 10-gram "cold test" reduction with a CeF₄ surrogate. Note the dense button and good separation between the metal and the slag.



The results of a 10-gram reduction with Hanford-produced PuF₄ material. The metal is the button on the right. Note the presence of small, well-formed plutonium beads on the slag.



Solving the Pu-238 Problem

Jeffrey A. Katalenich

We are working to improve fabrication techniques for radioisotope heat sources to enable future space exploration missions that rely on plutonium-238 for heat and power.

Current techniques for producing plutonium-238 (Pu-238) oxide heat source pellets are based on powder processing that generates dispersible plutonium powders. Processing Pu-238 oxide powders in gloveboxes and hot cells results in plutonium dust dispersal and increased potential for a breach in the engineered containment. To solve this problem, PNNL is investigating sol-gel techniques as a way to keep plutonium in solution until macroscopic oxide microspheres are formed, eliminating the powder generation and handling steps. Our research is focused on demonstrating the production of Pu-238 microspheres for processing into heat sources. Production of Pu-238 oxide microspheres by internal gelation has not been previously attempted, so project tasks include determining the optimal chemical parameters, defining desirable gelation conditions, overcoming radiolysis effects, and modifying sol-gel equipment for glove box operation.

To demonstrate the production of Pu-238 sol-gel microspheres and subsequent pellet pressing, work is divided into three main tasks pursued in parallel. First, we are bringing a new glove box in the Radiochemical Processing Laboratory operational to process gram-quantities of plutonium. Second, we are developing the optimal process chemistry for neptunium and plutonium using gram-quantities in a glove box. Third, we are operating a non-radiological sol-gel apparatus in the Applied Process Engineering Laboratory (APEL) as a testbed for new sol-gel equipment and procedures. Cerium oxide microspheres produced in APEL are also being used by collaborators to test pellet pressing methods.

Sol-gel chemistry and solution preparation methods are established in the literature for cerium and uranium, but a technique for producing plutonium-only feed solutions has not been published. To develop a method for preparing plutonium oxide microspheres, we began developing procedures using a neptunium-237 (Np-237) surrogate material. This work has led to a procedure for preparing a feed solution



Plutonium oxide gel in a test tube produced while optimizing sol-gel chemistry.

with the relatively low acidity and high metal ion concentration that is required for effective internal gelation to occur. Related procedures were prepared and tested for preparing plutonium feed solutions. Optimization of feed solutions parameters are ongoing with neptunium and plutonium and are beginning

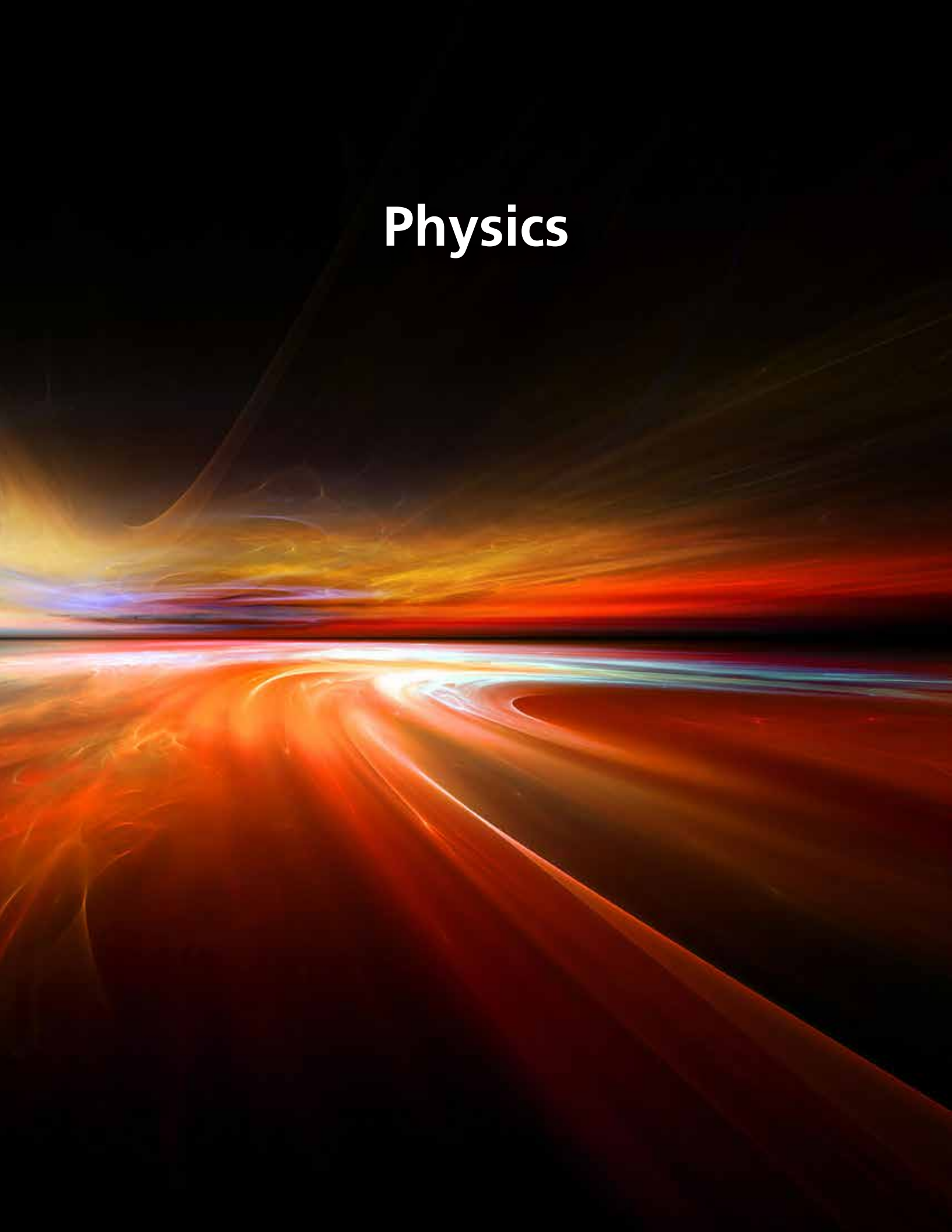
with Pu-238. Strong gels of Np-237, Pu-239, and Pu-238 have been demonstrated.

Non-radiological equipment in APEL was improved and modified to ease operations of a duplicate unit in a radiological glove box. Sol-gel equipment for radiological work was assembled and installed in a glove box and procedures were developed for its use. Additionally, a liquid nitrogen transfer system was designed and installed for introduction of liquid nitrogen into a glove box for a surface area analyzer instrument, which will be used to analyze neptunium and plutonium microspheres. Procedures for liquid nitrogen transfer were also developed.

Non-radiological equipment and cerium oxide microspheres were also used to test non-flammable chemicals for washing sol-gel microspheres. While the hazards of flammable chemicals are simply managed in non-radiological spaces, use of flammable liquids in glove boxes poses risks that are difficult to mitigate. Non-flammable chemical testing has led to several promising alternatives that are under continuing investigation.

In FY 2017, we plan to begin radiological sol-gel microsphere production in a glove box to demonstrate Np-237, Pu-239, and Pu-238 oxide microsphere production. Work will include process optimization and characterization of microspheres by thermal gravimetric analysis, microscopy, and pycnometry. If possible, non-flammable wash techniques will be employed for glove box processing. Cerium oxide pellet pressing by external collaborators will also continue.

Physics



Accelerator Neutrino Physics in Liquid Argon Time Projection Chambers

Eric D. Church

The U.S. High Energy Physics plan, following recent P5 recommendations, has committed to a long-term, 1 billion USD investment in Liquid Argon Time Projection Chambers (LArTPCs) and related facilities. These unique detectors offer unprecedented event resolution and full calorimetric reconstruction. The central justification for this investment is for large international collaborations to measure properties of neutrinos interacting in detectors, such as MicroBooNE and Short-Baseline Near Detector (SBND) in the Booster Neutrino Beamline (BNB) at Fermilab (FNAL), as well as those of neutrinos in the 34-ktonne DUNE detector in South Dakota.

The full program of LArTPC detectors on the BNB, consisting of MicroBooNE, SBND, and T600 (Icarus), will be able to determine whether MiniBooNE and Liquid Scintillator Neutrino Detector results are real, or it will implicate some non-oscillation background. Either result is profoundly important. Everything learned in these detectors also will accrue to DUNE.

We want to understand the nuances of the detectors that allow us to make very accurate cross-section and oscillation measurements, and eventually be able to demonstrate development of data acquisition systems that will allow for paradigm-changing proton decay measurements at DUNE.

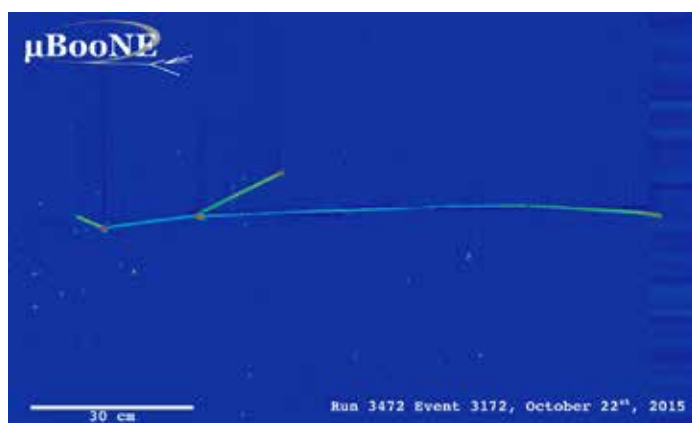
For SBND, PNNL demonstrated in 2016 a workable trigger and timing distribution system in the laboratory. This demonstration of the use of commercial-off-the-shelf (COTS) products, with firmware and software implementation by the primary investigator (PI) in the laboratory, will lead to adoption of the so-called White Rabbit technology in U.S. neutrino experiments to precisely measure time at all detector sub-components.

MicroBooNE physics results were presented at the big summer conferences (e.g., Neutrino 2016), in which the PI made

a large contribution, both to the data acquisition (DAQ) system that took the data and to its 97%+ running uptime, as well as to the papers and posters that were presented. These posters showed expected and achieved physics measurements in MicroBooNE. Further, for MicroBooNE, we built and showed the running time advantage, at minimal cost to memory, of multi-threaded code running in the large software framework called LArSoft. All the LArTPCs use LArSoft for their simulation/reconstruction/analysis work. We might expect this to have large benefits to the community and bring further demand for our software skillset.

Further, PNNL established itself as the leader of a COTS DAQ system, for which there is strong reason to believe DUNE will adopt for its approximately 2024 DAQ. A DUNE demonstration experiment will also use this DAQ in 2018 at the European Organization for Nuclear Research (known as CERN). This leadership is demonstrated in protoDUNE meetings and calls in which we push the agenda, showing work accomplished and work that must be accomplished in coming months, and collaborating with CERN and other institutions.

We expect to use the MicroBooNE data to do analysis and publish a paper in FY 2017 on proton decay backgrounds in LArTPCs. We will also demonstrate a functioning COTS DAQ that can read out the protoDUNE experiment by end of 2018.



A neutrino data event observed in the MicroBooNE detector. This collection plane view shows the remarkable event fidelity and calorimetric information available in LArTPCs.

Advanced Detection Techniques for a Linear Collider Detector

Jan F. Strube

This project will develop technology to improve the reconstruction performance of next-generation detectors at high-energy particle colliders. The physics aim of this project is to increase the precision of Higgs boson measurements and the detection capabilities for signatures of dark matter.

Detector concepts for the next generation of linear accelerators have been designed for precision measurements of Higgs boson properties and to be sensitive to a wide range of new phenomena. In particular, they measure the energy of quark jets with a resolution of 3.5 to 5% over relevant energies, compared with 10% to 30% at the Large Hadron Collider (LHC) Compact Muon Solenoid (CMS) experiment. To this end, the Calorimeter for Linear Collider Experiment (CALICE) collaboration has developed sampling detectors with extremely fine granularity (a technology that was also chosen for the upgrade of CMS). The information that these detectors provide per particle is at least an order of magnitude higher than what the current generation of detectors provide. This additional level of detail calls for new algorithms to fully exploit the potential of these detectors.

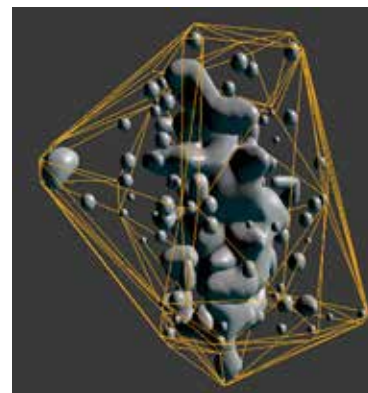
PNNL is developing algorithms that bring techniques from image reconstruction and recognition into the domain of event reconstruction for particle physics experiments. These algorithms will be developed with current and future generations of computing hardware in mind, to facilitate efficient running at the time when the experiment becomes operational. The developments of these algorithms will be guided by physics simulation studies and evaluated on beam test data from the CALICE collaboration.

Since the start of the project in April 2016, the preparation phase of the project has successfully completed. PNNL is member of the CALICE collaboration, which affords the group access to the data samples of the test campaigns and allows the validation of reconstruction algorithms on measured data.

We have started to design an analysis of invisible Higgs decays that will eventually be used to benchmark improvements to the reconstruction. This analysis is extremely challenging at the LHC and its upgrades, and it benefits from the precision available at a next-generation lepton collider. A paper on the achievable precision has been published in collaboration with Japanese colleagues.

We have created a first prototype implementation of algorithms that take techniques from computer graphics and apply them to the reconstruction of events in a collider detector. Preliminary attempts at using these techniques to classify the particles look promising and encourage us to develop the methods further. These preliminary results have been presented at a workshop of the CALICE collaboration.

In FY 2017, we will continue developing the reconstruction techniques to further extend the physics reach. In particular, we plan to extend the tracking into the calorimeter, which allows the reconstruction of long-lived particles that would otherwise be missed.



Visualization of the 3D measurements of a 50 GeV pion in a CALICE detector.

Atomic Tritium for Project 8

Brent A. VanDevender

Determination of the absolute neutrino mass is one of the priorities of modern nuclear and particle physics. Although extremely light, neutrinos are also extremely abundant, possibly outweighing all of the normal matter in the universe and affecting the evolution of large-scale structures such as galaxy clusters. This project will support the development of an atomic tritium source for Project 8, the next-generation experiment to measure the mass of the neutrino.

The 2015 Nobel Prize in Physics was awarded for the discovery of neutrino oscillations, which show that neutrinos have non-zero mass, in contradiction to the Standard Model of Particle Physics. However, that discovery does not determine the absolute mass, apart from setting a lower limit of about $0.01 \text{ eV}/c^2$. Past experiments to directly determine the absolute mass have not yet had sufficient sensitivity, resulting in an upper limit of $2 \text{ eV}/c^2$, with a new experiment coming online that will reduce that to $0.2 \text{ eV}/c^2$ by about 2022 if neutrinos are still lighter than that. Even between those bounds, the implications of neutrino mass for the evolution of the universe and the development of a quantum field theory to replace the Standard Model of Particle Physics are profound. Project 8 anticipates sensitivity to neutrinos as light as $0.04 \text{ eV}/c^2$.

The atomic tritium source to be developed by this research will improve the sensitivity of neutrino mass measurements by removing an irreducible systematic experimental uncertainty that appears in any experiment done with a molecular tritium source. Like the experiments that set the current upper limits, Project 8 will use the tritium endpoint method, which determines neutrino mass by its effect on the energy spectrum of electrons emitted in the beta decay of tritium. In its natural state, tritium gas exists as a two-atom molecule. When one of those atoms beta decays, the daughter molecule

rotates and vibrates, resulting in a smearing of the electron energy spectrum. That smearing limits sensitivity to the mass of the neutrino also emitted in the decay. That limit can only be removed by using a source consisting of individual tritium atoms. Confining those atoms in the sensitive region of an apparatus and preventing them from recombining to molecules is a major challenge to be addressed by this research.

The first step is to develop a conceptual design of the source and an understanding of how its parameters affect the neutrino mass sensitivity of the Project 8 experiment. The pressure of tritium in the experiment will be very low, so that recombination of atoms to form molecules will happen only on the walls of the containment vessel. We plan, therefore, not to have a physical vessel at all, but rather a magnetic field configuration that will confine magnetic tritium atoms while nonmagnetic tritium molecules quickly escape. Such a source by itself would self-heat because of its radioactivity and evaporate away, so we will add a small amount of nonmagnetic helium which will pass freely through the magnetic trap to exchange heat with the cryogenic physical vessel.

This year, we developed a plan for a simulation-driven design effort. That plan will address the trapping and cooling issues above and allow us to specify quantitative design parameters, such as the required purity of tritium atoms relative to molecules and the strength of the magnetic trap. We have chosen an existing simulation framework that will naturally handle the magnetic trapping of atoms and the paths of decay electrons in the magnetic field. Next year we will extend that framework to include the thermodynamics of the source to account for the helium heat exchange and thermal evaporation of tritium atoms from the trap.

The design parameters will be driven by their effect on the neutrino mass sensitivity of the experiment. We must, therefore, also develop the uncertainty budget of the experiment next year, so that in the third year of research, we can complete a quantitative conceptual design with the ability to map the design parameters to their contributions to the total uncertainty.

Axion Dark Matter Experiment (ADMX)

Noah S. Oblath

Axions are one of two theoretically well-motivated candidates for the dark matter that is known to make up about one quarter of the total mass of the universe. The ADMX experiment is searching for cold dark matter in the local galactic dark-matter halo. The existence of the axion would not just solve the mystery of dark matter, it would also explain an apparent “fine tuning” question in particle physics, known as the Strong CP Problem. At the heart of ADMX is a tunable microwave resonant cavity that increases the ability to detect an axion of a particular mass by a factor of approximately 10,000.

A variety of techniques have been used to try to detect axions, but only ADMX would be able to detect an axion with the appropriate properties to make up a significant portion of the cold dark matter in the universe. Those axion properties include its mass and the strength with which it interacts with normal matter. ADMX searches for different axion masses by changing the resonant frequency of the cavity. The ADMX experiment started in 1995, and has been relentlessly working to improve its ability to detect axions. The current experimental plan will cover the theoretically best-motivated range of axion properties, and PNNL has capabilities that will enable ADMX to be successful.

In 2015, the ADMX collaboration went through a process of expanding the collaboration membership to bring in new expertise that would be needed to enable the experiment to be sensitive to higher axion masses. They sought the involvement of PNNL, in particular, to take advantage of our expertise in microwave engineering, as well as to grow the existing relationships between PNNL and the University of Washington (where the ADMX experiment is based). We will use our expertise in microwave engineering to provide the receiver electronics that process the signals coming out of the experiment and to do the modeling of the microwave resonant cavity that helps us understand the cavity resonant modes that are critical to the experiment's success.

This year, the main contributions to ADMX from the PNNL group included work on cavity modeling, a new radio-frequency (RF) receiver, and the data-acquisition and experiment controls systems. The ADMX experiment started a new data-taking phase in August, which will last for approximately one year. To fully cover a particular range of potential axion masses, the experiment operators need to understand how to tune the cavity resonance frequency. The PNNL group used advanced RF simulation tools to model the cavity and its tuning mechanisms and produced a reliable mode map that is being used to guide the cavity tuning during experiment operations.

For each year-long phase of ADMX, a new receiver is needed that is matched to the frequency range being covered during that time. As the experiment moves to higher frequencies, the design and construction of the receiver becomes more challenging. This year our RF engineers and technicians designed, built, tested, and delivered a new RF receiver. This work was completed ahead of schedule and under budget. The receiver was delivered to ADMX in August.

The next phase of the ADMX experiment will present a variety of challenges in the coming year. We are working to increase our level of involvement in the ADMX collaboration by applying our expertise to these challenges.

A new design for the microwave cavity is required to be sensitive to higher axion masses. We have already participated in the design process for the new cavities. We will continue to work with collaborators at Lawrence Livermore National Laboratory in building and commissioning the cavities and putting them into use in the experiment. Multiple cavities will be used simultaneously, which requires adding a system to lock the resonant modes of the cavities, such that they are sensitive to the same axion mass range. PNNL has expertise in cavity locking and will be contributing to the development of this critical system.

In FY 2017, we will continue to improve the data-acquisition and experiment controls systems, and we will develop new analyses to increase the scientific output of the ADMX experiment.

CLEAN Detection of Dark Matter and Low Energy Neutrinos

Christopher Jackson

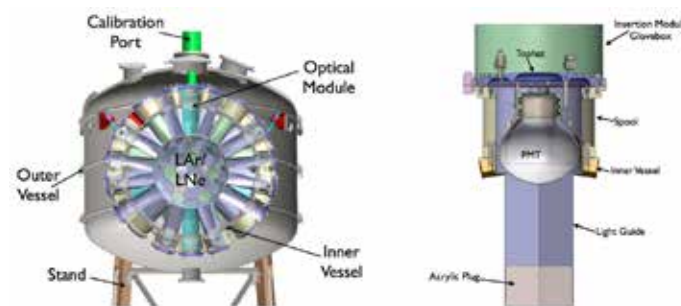
Monolithic detectors using noble liquid targets hold promise to significantly extend the sensitivity and potential discovery of particle dark matter and new physics in the neutrino sector well beyond the state-of-the-art anticipated for the Generation-1 (G1) and G2 experiments. The efficacy of the so-called “single-phase” approach for the detection of dark matter and neutrinos will be examined with the MiniCLEAN and DEAP-3600 experiments operating deep underground at SNOLAB.

The evidence for dark matter in the universe is familiar and well established. This is one of the very few places where there is direct experimental evidence for physics beyond the standard model. Primordial nucleosynthesis argues that the dark matter is non-baryonic. Observations of structure formation indicate only a small contribution from neutrinos. Dark matter is, therefore, exotic, and Weakly Interacting Massive Particles (WIMPs) are a compelling candidate for dark matter that provide a cold, dark relic of the Big Bang and could be directly detected as they scatter from massive, ultra-pure detector targets operating deep beneath the earth's surface.

The challenge to realizing sensitive dark matter detectors is in separating backgrounds from the nuclear-recoil events characteristic of the WIMP signature. Future progress hinges on improving sensitivity by several orders of magnitude beyond existing or planned experiments before the background floor imposed by the coherent scattering of neutrinos from the atmosphere and from the sun will limit sensitivity to WIMPs. Detectors based on the noble liquids are particularly well suited to achieving the target masses necessary to pursue this challenge.

CLEAN (Cryogenic Low-Energy Astrophysics with Noble) liquids exploit a *single-phase* detector where only the scintillation light is measured. Simplicity of design affords the construction of a very large detector at low cost, while pushing the major sources of background outside of the region of interest for a sensitive WIMP search. Such large, monolithic detectors benefit not only from signal rate, but are also significantly easier to model, allowing precise predictions of response to signal and background.

This project recognizes the utility of liquid argon as a sensitive dark matter target, the basic principle of which has been demonstrated in small prototype detectors. MiniCLEAN will serve as technical demonstration of the principles of a single-phase detector using 500 kg of liquid argon (LAr) as the target material and including 3D event reconstruction. The MiniCLEAN detector is deployed in the Cube Hall, 6,800 feet underground at SNOLAB.



(Left) Model of the MiniCLEAN central detector with its 4π target viewed by 92 optical cassettes. (Right) The optical cassettes are 30 cm long and consist of a 10-cm thick acrylic plug, the front surface of which is coated with a wavelength-shifting fluor (tetraphenyl butadiene [TPB]), and 30-cm light guide leading to the photomultiplier tubes. The inner target, defined by the TPB surface, contains 500 kg of LAr within a nominal radius of 44 cm.

The first cool down of MiniCLEAN to 87 K showed a difficulty in cooling the detector, with the rate falling from 5 K/day at the start to 0.8 K/day at 130 K. A significant upset in schedule and progress occurred with the leak that opened up during structural improvements required by SNOLAB. The addition of an LAr condenser has increased the cooling rate, and the cool down of MiniCLEAN is in progress.

PNNL has established the data acquisition and analysis plan for the MiniCLEAN science campaign with LAr and designed the test bench to assay the ^{39}Ar content of sample for potential use as a calibration source. Commissioning of the MiniCLEAN detector at SNOLAB with an LAr target using argon extracted from the atmosphere is underway.

The next fiscal year will see the execution of a science run and calibration program using an atmospheric LAr target and the analysis of these data samples. The plan to deliver and inject ^{39}Ar -spike source into the MiniCLEAN detector will be established and executed. The data acquisition will be upgraded to accommodate the enhanced data rates of the ^{39}Ar -spike data run.

Dark Matter Physics

Raymond A. Bunker, III

The dark matter that makes up 85% of matter in the universe is unknown. This project focuses on analysis of current data and future experiments to address this scientific priority in the field of cosmology and particle physics.

The nature of the matter in our universe is currently unknown. Cosmological observations—on scales of galaxies to galaxy clusters to the thermal photon relic of the Big Bang—suggest that the standard model of particle physics accounts for a mere 15% of the matter in the universe.

Baryonic matter has been largely ruled out as a candidate for dark matter, which leaves new stable fundamental particles as the favored possibility. Weakly Interacting Massive Particles (WIMPs) are a broad class of fundamental particles similar to the neutrino that are proposed as a solution to the dark matter issue. Fundamentally, if there is a new symmetry of nature, then there could be massive, stable particles filling the universe that have limited interactions with normal matter.

Through this project, we established critical roles in the community effort to make progress in dark matter. PNNL is now a member of several large collaborations dedicated to searching for WIMP dark matter: MiniCLEAN, DarkSide, SuperCDMS, and PICO. SuperCDMS employs unique germanium detectors to search for dark matter interactions, using cryogenic bolometry to identify background events. PNNL scientists have recently published SuperCDMS data using a novel, high-voltage technique to search for low-mass (less than 5 GeV/c^2) WIMPs. DarkSide and MiniCLEAN approach dark matter detection using liquid argon as the target material. The scintillation light of liquid argon fundamentally contains information on the nature of particle interactions. MiniCLEAN is a small but critical demonstration for the envisioned third-generation experiment, a high-priority, long-term strategy of the high energy physics community.

PNNL's expertise in ultra-low-background materials helps to reduce background that could overwhelm the extremely

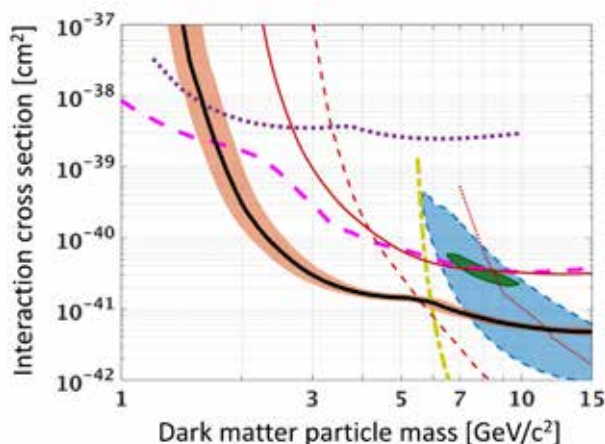
rare dark matter events, which is the common challenge for all these experiments. PNNL's expertise in copper electroforming, uranium and thorium assay, and detector assembly are helping these experiments make order of magnitude improvements in sensitivity.

In addition to the low energy signals, a dark matter signal is expected to vary annually due to the earth's orbit varying with respect to the Milky Way's center. PNNL developed a maximum likelihood analysis technique to search simultaneously for the low energy and time varying signature of dark matter interactions in deep underground experiments. The software analysis method development is sufficiently generic to be applied to future dark matter experiments.

In addition to participation in the planning, execution, and analysis of large, complex experiments to detect dark matter, PNNL is involved in detailed calculations to determine the ultimate sensitivity of specific approaches to WIMP detection. Scientists are using modern computation techniques to estimate the backgrounds produced in unavoidable high energy, cosmic ray interactions. Additionally, scientists are calculating the signals that are expected from the lowest energy interactions in semi-conductor detectors, a crucial detail that has no data and no theoretical predictions. With these two calculations,

both the signal and the backgrounds will have better predictions, leading to a better understanding of the ultimate sensitivity of future experiments.

In FY 2016, we focused on the first-generation PICO and SuperCDMS dark matter experiments and cross-cutting transformational technology. New analyses with these generation-one dark matter projects were published. The sensitivity to dark matter in the region studied has improved by an order of magnitude due to this research. We additionally demonstrated measurement of the radioisotope ^{40}K in materials relevant for future dark matter experiments and with new world-leading sensitivity. Finally, we demonstrated an ultra-sensitive radon emanation system for screening materials relevant to future dark matter experiments. The system takes advantage of custom PNNL gaseous detectors that have an electroformed copper construction to achieve ultra-low backgrounds.



From a 2016 Physical Review Letters article, illustrating the scientific impact of recent SuperCDMS data. The black line shows the data sensitivity in the low-mass region using the high-voltage operational mode proposed and led by scientists at PNNL.

Discovering CENNS in MiniCLEAN at Fermilab

Christopher Jackson

The detection and study of coherent elastic neutrino nucleus scattering (CENNS) will open a broad window to previously unexplored regimes in neutrino physics. The research supported by this project will define the capabilities for mini cryogenic low-energy astrophysics with noble (MiniCLEAN) and future detectors scaled in target mass using the CLEAN liquids technology.

CENNS is a fundamental process in the Standard Model of elementary particle physics, yet it has eluded detection since it was first predicted over four decades ago. Innovations in dark matter detector technology, in concert with the Booster Neutrino Beam (BNB) at Fermilab, now make the unseen CENNS testable. Key to measuring CENNS is a detector with a sufficiently low-energy threshold to reveal a clean nuclear-recoil signal that is free of background. The basic approach we have taken for the direct detection of dark matter has culminated in the design and construction of the MiniCLEAN detector. The detector requirements for a CENNS measurement are strikingly similar to that for dark matter detection, but with a key difference: dark matter detectors need to be operated deep underground and free of cosmic-ray-induced background, while a CENNS detector would be placed near the surface in a neutrino beam with its associated beam-related backgrounds.



Installation of the MiniCLEAN detector deep underground at SNOLAB. Presently under commissioning, this prototype detector of dark matter by design exhibits all of the salient specifications for a CENNS detector. Performance of MiniCLEAN, in concert with simulations, will provide the necessary testbed of data to evaluate capabilities for future CENNS experiments.

The technical approach and methodology of this project focus on the primary detector performance parameters for a single-phase liquid argon detector, as it concerns the challenges for measuring CENNS in a neutrino beam at Fermilab's BNB. The technical outcomes are congruent with the need to establish a detector with a sufficiently low-energy threshold to afford meaningful sensitivity of the CENNS signal. The ability to reach this energy threshold hinges on a complete understanding of the light yield and radioactive backgrounds at the analysis threshold, and the related experimental uncertainties that translate to uncertainties in the measured cross-section and differential energy spectrum. The results from the research supported by this project will define the capabilities for MiniCLEAN and future detectors scaled in target mass using the CLEAN liquids technology.

In FY 2015, we evaluated nuclear-recoil scintillation efficiency, in light of available data, and revised the model accordingly. We calculated Coulomb corrections to ^{39}Ar beta spectrum, modeled uncertainties, and evaluated uncertainty in energy scale impact on precision in CENNS cross-section and differential energy spectrum measurements. Additionally, we defined requirements for water shielding and active muon veto and evaluated the costs and project plan for moving MiniCLEAN from SNOLAB to Fermilab.

In FY 2016, we started benchmarking the light yield model and the radioactive background model against data from MiniCLEAN.

Image Fusion – Secondary Ion Mass Spectrometry (SIMS) and Microscopy

David G. Willingham

Image fusion of SIMS and Energy-dispersive X-ray Spectroscopy (EDS) is a powerful correlative imaging capability that enables the analysis of isotopic and elemental microstructures at spatial resolutions approaching those typical of electron microscopy techniques. This research improves the ability to identify U-bearing particles for non-ideal sampling conditions for nuclear safeguards, analysis of Al-rich domains within lithium aluminate (LiAlO_2) ceramics used as tritium-producing burnable absorber rod materials, and Mo-rich domains within uranium-molybdenum (U-Mo) alloys developed for light water power reactors.

Image fusion is the process of combining input from two or more images into a single image. The resulting image is, by definition, more informative than any of the stand-alone images individually. Previously, this type of correlative imaging has been primarily applied in the areas of satellite and medical imaging. Image fusion as it applies to microanalysis of nuclear materials was initiated to capitalize on the power of combining two or more analytical technologies to enable the observation of spatially unique domains with high precision isotopic specificity at superior spatial resolution. The expectation is that the successful development of an image fusion capability involving SIMS and EDS will lead to a novel approach to the analysis of isotopic and elemental spatial distributions aiding analytical measurements ranging from the identification of actinide-containing particles to the irradiation behavior of micro-structured nuclear fuels.

Nuclear Safeguards. Image fusion of SIMS images and X-ray elemental maps from EDS was performed to facilitate the isolation and re-analysis of isotopically unique U-bearing particles, where the highest precision SIMS measurements are required. Image registration, image fusion, and particle micromanipulation were performed on a subset of SIMS images obtained from a large area pre-screen of a particle distribution from a sample containing a mixture of several certified reference materials (CRM) U129A, U015, U150, U500, and U850, as well as standard reference material 8704 (Buffalo River Sediment) to simulate particles collected on swipes during routine inspections of declared U-enrichment facilities by the International Atomic Energy Agency.

In total, 14 particles, ranging in size from 5 to 15 μm , were isolated and re-analyzed by SIMS in multi-collector mode, identifying 9 particles of CRM U129A, 1 of U150, 1 of U500, and 3 of U850. These identifications were made based on the measured isotopic composition, which was accurate to a few percent of the certified value and which proved to be consistent with the respective National Institute of Standards and Technology certified atom percent values for ^{234}U , ^{235}U , ^{236}U , and ^{238}U for the corresponding CRMs. This work represents the first use of image fusion for nuclear safeguards application, resulting in improved accuracy and precision of isotope ratio measurements for U-bearing particles. Implementation of image fusion is essential for the identification of particles of interest that fall below the spatial resolution of the SIMS images.

Nuclear Fuels. SIMS analysis of a U-Mo foil sample was performed with the width of three fields of view (FOVs) (about 150 μm each) used to cover the width of the foil. Five individual masses (^{12}C , ^{90}Zr , ^{98}Mo , ^{235}U , and ^{238}U) were measured for each FOV.

This SIMS image analysis is indicative of several key characteristics of the U-Mo foil system. First, Mo-rich grains were observed in the SIMS images. These grains are highly elongated along the length of the foil (this result confirms previous electron microscopy measurements). Additionally, there is significant penetration of Mo grains into the Zr cladding. In general, Mo appears to be quite mobile within the U matrix. Second, carbon inclusions, likely a result of uranium carbides, were found in the SIMS images. These inclusions can be seen to track with the previously observed Mo-rich elongated grains. Third, Zr is located in the cladding region only and is not observed within the U-Mo foil itself. Last, the U within the foil is much more homogenized than the Mo and is less mobile. There are slight ion signal increases in the U signal within the Mo-rich areas of the foil, but this variation is likely due to enhanced ionization because of the presence of the Mo rather than an actual concentration variation (this finding would need further investigation). Additionally, the boundary between the U and Zr layers is more defined than the Mo and Zr layers. Finally, the $^{235}\text{U}/^{238}\text{U}$ is consistent with the known value at about 20% enrichment of ^{235}U . More interestingly, the U isotopic distribution is homogenous in all regions of the U-Mo foil. Therefore, it is likely that non-uniform swelling of these foils under irradiation is due primarily to elemental heterogeneities rather than isotopic variations.

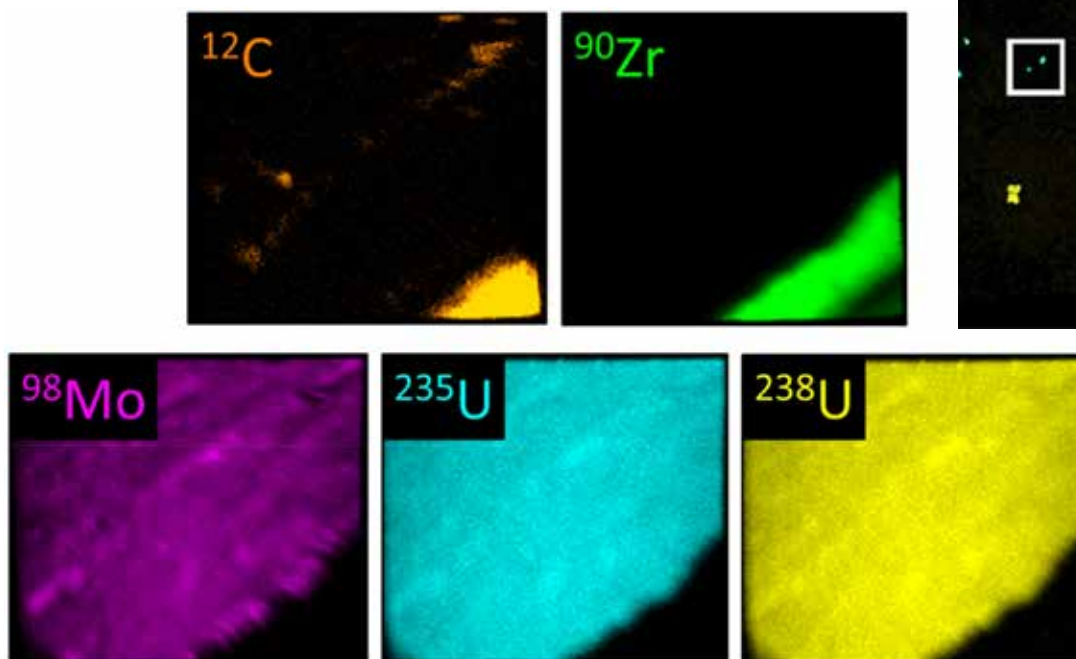
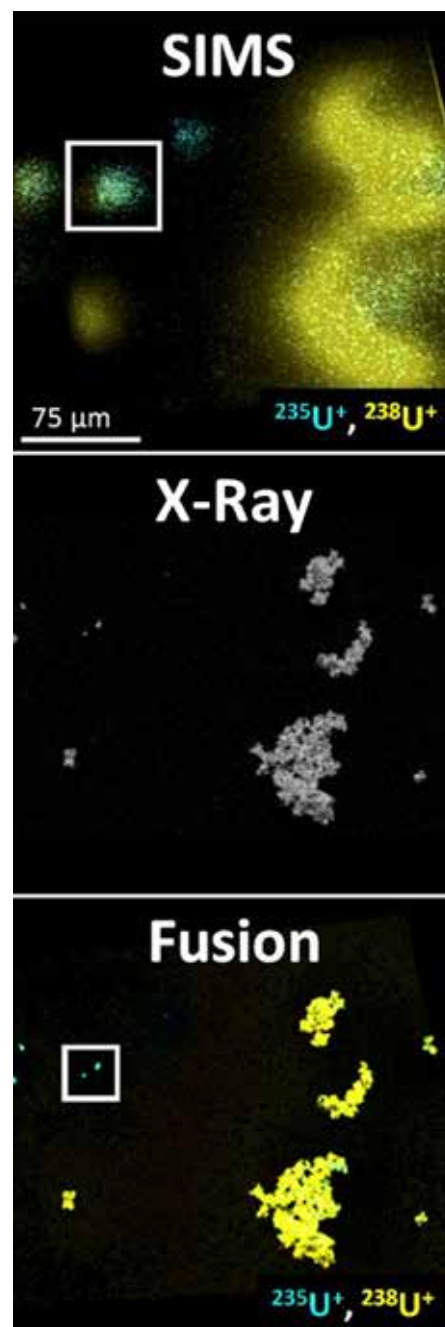
Future plans are to develop a method for analyzing particles containing both U and Pu with image fusion using pyrolyzed

resin beads as particle surrogates. The concentration and isotopes of U and Pu on these beads can be finely tuned and quantified through solution chemistry prior to SIMS and EDS analysis. Building on the U-only image fusion methodology, the U/Pu beads can be isolated and re-analyzed following SIMS pre-screening; however, unlike the U-only particles, the U/Pu beads would be re-analyzed by thermal ionization mass spectrometry. The U/Pu image fusion aims to resolve two main challenges for SIMS measurements of U/Pu containing particles: 1) the unresolvable $^{238}\text{U}^{1}\text{H}$ isobaric interference on ^{239}Pu and 2) the unequal detection sensitivity for U and Pu inherent in all SIMS instruments that limits quantitative analysis.

In addition, because SIMS is not a completely non-destructive imaging technique, the depth of the sample (z-axis) may be probed along with the x- and y-axis. Therefore, given a sufficient number of analysis cycles, a 3D representation of the analyzed sample can be rendered using a method known as isosurfacing. The resulting 3D rendering can be rotated, sliced through, and manipulated in a number of useful ways. Thus, a new image fusion capability could be developed by expanding upon the 2D image fusion methodology using correlative analysis of micro-computed tomography ($\mu\text{-CT}$) and SIMS image depth profiling (3D SIMS). Because $\mu\text{-CT}$ is a non-destructive analysis technique, it pairs seamlessly with 3D SIMS. Correlative analysis of $\mu\text{-CT}$ and 3D SIMS is likely to yield similar benefits as the previous image fusion

methodology, but in three dimensions instead of two. The 3D imaging SIMS capability could provide additional information for accurate particle identification, as well as 3D information about the microstructure of LiAlO_2 and U-Mo nuclear fuel systems.

Results of fusing a region of interest:
1) registered overlay SIMS image of $^{235}\text{U}^{+}$ (cyan) and $^{238}\text{U}^{+}$ (yellow); 2) uranium EDS image; 3) pan-sharpened image of 1) and 2). All images are the same size.



SIMS measurement of a U-Mo foil indicating five individual masses (^{12}C - orange, ^{90}Zr - green, ^{98}Mo - magenta, ^{235}U - cyan, and ^{238}U - yellow). The individual images represent the summation of the total SIMS analysis of this FOV (about 150 μm), containing 1,000 individual images at each mass. This particular FOV is of the edge of the U-Mo foil and contains signals from the foil itself, the Zr cladding, and the edge of the carbon containing epoxy.

Laser-Ablation Based Multimodal Tool for Nuclear Forensics

Sivanandan S. Harilal

The aim of this project is to understand the physics and chemistry of laser ablation (LA) as it pertains to optical emission and particle formation, with a goal of optimizing the performance of combined laser-induced breakdown spectroscopy (LIBS) and LA-inductively coupled mass spectrometry (LA-ICP-MS) sensors.

LIBS and LA-ICP-MS analytical techniques are routinely used for various applications; however, there is a lack of complete understanding of fundamental physics problems, and such a study is ultimately important for improving the analytical capabilities of both techniques and moving towards sensor fusion. This project addresses some of the long-lasting fundamental scientific questions important for LIBS and LA-ICP-MS sensors independently and will provide a basis for developing an improved multimodal sensor with unique capabilities found in both LIBS and LA-ICP-MS.

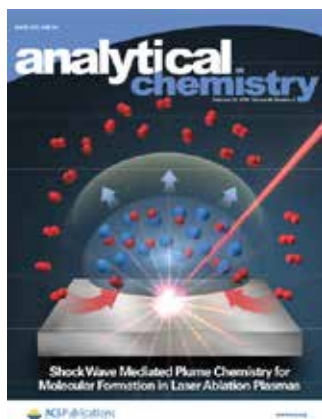
During FY 2016, we investigated the role of the plume dynamics and plasma chemistry on molecular formation in an Al laser plasma expanding into various ambient pressures. Our results clearly showed that the plume hydrodynamics play a great role in redefining plasma thermodynamics and molecular formation. The shock waves at the plume front act as a barrier, keeping ambient oxygen away from the plume during early times of plasma expansion. The molecular formation is found to be prevalent after the shock wave collapse. A thermodynamic analysis of the standard Gibbs energies and the equilibrium constants (K_{eq}) values for the reaction pathways for the combustion of Al support the coexistence of Al and AlO over the estimated molecular temperature ranges found in this study.

Also in FY 2016, we investigated mechanisms for U optical signal variation

under various environmental conditions. Potential explored mechanisms for signal quenching related to ambient conditions include plasma chemistry (e.g., uranium oxide formation), ambient gas confinement effects, and other collisional interactions between plasma constituents and the ambient gas. Absorption studies showed that the persistence of the U ground state population is significantly reduced in the presence of air ambient compared to nitrogen. Emission spectroscopic results yield congested spectra. The plume hydrodynamic features and plume fundamental properties showed similar results in both air and nitrogen ambient. Investigation of U oxide formation in the LA plume from U metal and sample containing U as a trace suggests that higher U concentration in the plume is necessary for consistent detection of UO molecular spectra.

During FY 2016, we also evaluated and compared molecular formation in plasma systems generated by nanosecond (ns), and femtosecond (fs) LA. Our results show that, at a fixed laser energy, the persistence of molecules is found to be highest for ns LA though the molecular emission peaks at later times. For fs LA, the molecular species are formed at early times; however, the persistence of its emission is significantly limited. The delayed molecular emission from ns LA is related to hotter initial conditions and, hence, the presence of a stronger shock wave that controls the combustion process. The observation of molecular species at early times for fs LA compared to ns LA is due to a range of processes, which includes lower plasma temperatures, reduced thermal effects during ablation, increased ablation efficiency, and the presence of an atomic plume. Spectral fit analysis provided molecular temperatures, which showed temperatures of ns LA are significantly higher compared to fs LA.

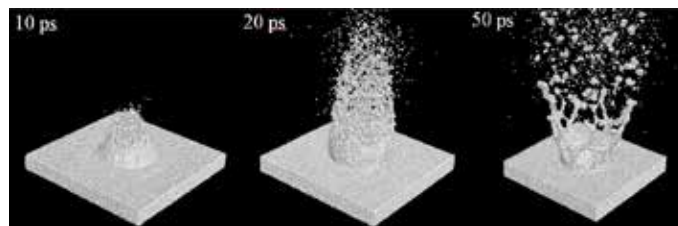
In collaboration with Purdue University, we made a comprehensive comparison of local plasma conditions between ns and fs LA sources in a combined LIBS and LA-ICP-MS system. The ability to actively monitor fundamental parameters of the plasma provided interesting insights into the effect of various parameters on plasma conditions and its correlations to LIBS and LA-ICP-MS signals. The optical emission spectra and ICP-MS signal were recorded simultaneously for both ns and fs LA, and figures of merit of the system were analyzed. Characterization of the plasma was conducted by evaluating excitation temperature and electron density of the plume under various irradiation conditions using optical emission spectroscopy.



Pictorial view of shock wave control over molecular formation in LA plumes; feature story in Analytical Chemistry.

copy, and correlations to ns- and fs-LIBS and LA-ICP-MS signal were made. This study provided useful conditions for a multi-modal system, as well as giving insight into how laser ablation plume parameters are related to LA-ICP-MS and LIBS results for both ns and fs LA.

In FY 2016, we also combined Momentum Scaling Model and molecular dynamics methods for studying the non-equilibrium process of ultrafast laser-material interactions at an atomic level, the nature and behavior of materials in a highly non-equilibrium state, microscopic mechanisms of melting, disintegration of material, and formation of nanoclusters. The size distribution of nanoclusters, their velocities, and mass fractions were investigated for various laser fluence levels. Our results show that laser fluence greatly affects the nanoclusters' size distribution, magnitude, and direction of their velocities and polar angle.



3D snapshots of Al plume at different time moments.

Low-Mass Dark Matter Backgrounds R&D

Raymond A. Bunker, III

Dark matter makes up 85% of matter in the universe, but its composition is unknown. This project focuses on understanding the backgrounds expected to limit the sensitivity of direct searches for low-mass dark matter and on the development of background reduction strategies, with the ultimate goal of specifying the design of a future experiment that will explore dark matter interaction rates down to the solar neutrino floor.

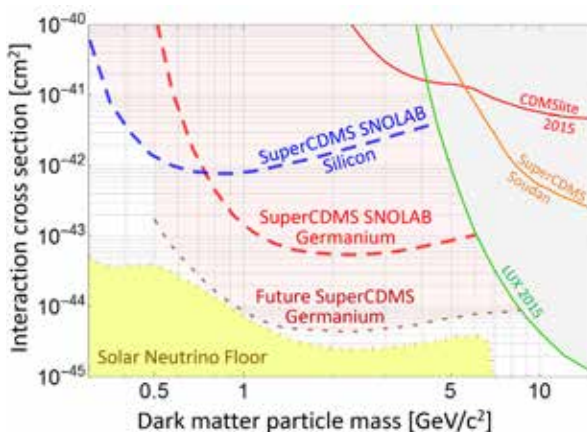
The nature of dark matter is one of the most fundamental questions of modern physics. There is a wealth of evidence supporting its existence—from the orbital velocities of stars and galaxies, to temperature fluctuations in relic photons from the Big Bang—but its composition is unknown. Experimental data disfavor normal (baryonic) matter as a solution to this mystery. Weakly Interacting Massive Particles (WIMPs) are a well-motivated class of fundamental particles similar to the neutrino that could make up a majority of the dark matter and could be detected directly by an Earth-based detector. WIMP searches are at a mature stage, capable of measuring rates as low as a single interaction per year in a 10-kilogram detector. The background requirements are becoming extraordinary, and progress in the field is directly tied to the backgrounds achieved.

In recent years, the hunt for WIMPs has pushed into the “low-mass” regime, in which solid-state detectors are used to search for the scattering of dark matter particles with masses less than $10 \text{ GeV}/c^2$ ($10\times$ the mass of the proton). The premier next-generation low-mass experiment is the Super Cryogenic Dark Matter Search (SuperCDMS SNOLAB), which will use germanium and silicon detectors installed within radiopure shielding and located about 2 km underground at SNOLAB in Sudbury, Ontario.

An important milestone in the search for low-mass dark matter is the detection of coherent nuclear scattering of solar neutrinos. This benchmark is often referred to as the solar

“neutrino floor.” Although the next-generation SuperCDMS experiment will explore a broad range of previously untested low-mass parameter space, it will generally fall short of the solar neutrino floor by 1 to 2 orders of magnitude. Trace levels of radioactivity in the detector’s materials of construction are expected to limit the experimental sensitivity.

The goals of this project are two-fold: understand and characterize the backgrounds expected to limit the sensitivity of low-mass dark matter searches and develop strategies for the reduction of these backgrounds in order to design a future experiment capable of reaching the solar neutrino floor. Backgrounds from sources intrinsic to the detector substrates are expected to dominate, particularly those created through exposure to cosmic rays (e.g., tritium and ^{32}Si) and to environmental radon (e.g., ^{210}Pb). Research and development will focus on methods for measuring and mitigating these sources of background, utilizing PNNL’s expertise in ultra-low-background materials, low-background detectors, and ultra-sensitive radiopurity assay.



Dark matter sensitivity (vertical scale) vs. dark matter particle mass. Solid curves show first-generation experiments. The next-generation SuperCDMS expected sensitivities (thick dashed) will explore low-mass parameter space but fall short of reaching the solar neutrino floor (yellow shaded). This project will develop background reduction strategies to enable a future experiment to reach this important milestone and potentially detect dark matter.

Progress in FY 2016 was made in several areas. Particle physics simulations were conducted to develop an understanding of the rates at which radioisotopes are produced in materials due to exposure to cosmic rays at sea level. There is significant uncertainty associated with the tritium production rates in germanium and silicon, motivating development of a method to measure tritium contamination in these materials in FY 2017. Preliminary plans were made to explore the feasibility of removing tritium from detectors by “baking” them at high temperature. These plans will be finalized and executed in FY 2017. An initial survey of the commercial processing stream for detector-grade silicon was conducted to obtain

samples for ^{32}Si measurements, and the possibility of obtaining isotopically pure ^{28}Si was explored. The purpose of the former is to establish a firm ^{32}Si background expectation for silicon-based dark matter detectors, while the latter is a possible mitigation strategy. A publication featuring the results of these investigations is planned for FY 2017. Finally, progress was made on the development of advanced background modeling tools, which will be used to construct a background budget at the solar neutrino floor. Further progress will be made in FY 2017, and radon-related measurements and mitigation strategies will also be pursued.

Mitigating Challenges Toward an Enduring Supply of Low-Radioactivity Argon for Ongoing PNNL National Security and Basic Science Programs

Henning O. Back

Argon produced from underground sources has been found to be nearly free of the radioactive isotope ^{39}Ar . We are exploring the contaminations in the CO_2 source that hampered the original underground argon production facility, as well as the residual ^{39}Ar and ^{85}Kr source. Additionally, we will be exploring contamination mitigation techniques.

While argon is easily purified to remove natural radioactivity, argon that is derived from the atmosphere contains a small fraction of the radioactive isotope ^{39}Ar . Due to the difficulty of separating argon isotopes, the ^{39}Ar is the limiting background in all very low-level radiation detectors that use argon, particularly argon-based dark matter detectors. The future successes of these programs depend on the establishment of an enduring source of low-radioactivity argon that is free, or nearly free, of ^{39}Ar .

Argon from active CO_2 wells in southwestern Colorado have been found to contain very low levels of ^{39}Ar , with upper limits of ^{39}Ar shown to be 0.07% atmospheric concentrations. For more than 5 years, a dark matter experiment (Darkside-50), extracted and purified this argon for the target material in the Darkside-50 detector.

The production of the argon began in southwestern Colorado with a vacuum-pressure swing absorption (VPSA) unit, where CO_2 , methane, and other unwanted gases are absorbed onto zeolites. The VPSA plant produced an argon-helium-nitrogen mixture, which was then sent to Fermi National Accelerator Laboratory (Fermilab) in Batavia, Illinois, where the final purifications were performed through cryogenic methods. At Fermilab, the argon and nitrogen were liquefied at high pressure to remove the helium, and then the gas was passed through cold traps to freeze out unwanted contaminants, some of them unknown. Oxygen was removed with activated copper getters, and finally the argon and nitrogen were separated in a cryogenic distillation column. The final product is

pure argon with a nitrogen contamination level at the 10-100 ppm level.

With PNNL support, the DarkSide-50 collaboration was successful in producing more than 150 kg of low-radioactivity underground argon (UAr), however the largest challenges to overcome were known and unknown contaminations in the CO_2 . The VPSA plant required continuous monitoring and upkeep to maintain UAr production; however, it also suffered from long-term degradation. At Fermilab there are clearly contaminants that made it through the VPSA plant and would then concentrate and freeze in the cryogenic systems.

These challenges were overcome with short-term stopgap measures, but the next generation of UAr production facility needs to find permanent solutions to these challenges. The goal of this project is to investigate the contamination issues encountered in the production of the DarkSide-50 target, ensuring the success of the next generation of UAr production facility.

With samples of the adsorbents from the VPSA plant in Colorado, we extracted and identified the contaminants that permanently adsorbed onto them and degraded the adsorbent's performance toward CO_2 , CH_4 , and other gases. Additionally, samples were taken from the cryogenic plants at Fermilab as the plant was warmed up for dismantling. The contaminants found in the cryogenic plants were nearly identical to the ones found in the adsorbents.

Examples of high mass compound contaminants found in VPSA and Fermilab purification plants.

C_3H_8	C_6H_{12}	C_8H_{16}	$\text{C}_5\text{H}_{10}\text{O}$
C_5H_{10}	C_6H_{14}	C_8H_{18}	$\text{C}_6\text{H}_{12}\text{O}$
C_5H_{12}	C_7H_{14}	C_9H_{20}	$\text{C}_5\text{H}_8\text{O}_2$
C_6H_6	C_7H_{16}	$\text{C}_6\text{H}_{13}\text{I}$	$\text{C}_6\text{H}_{10}\text{O}_2$

The work in this project has led to collaborating on an international effort to upgrade underground argon extraction from the southwestern Colorado site with a plant named Urania. Within this collaboration, PNNL is meant to lead the operation of underground argon extraction, once the plant has been installed in Colorado. Princeton University has

obtained funding from an Italian funding agency to build the plant, and they have submitted a proposal to the same Italian funding agency for installation in Italy and for plant operations. The success of this proposal should be known by FY 2017 Q3.

Through the collaboration on the Urania project, the concentration of the contaminations identified by PNNL were measured by the University of Houston. These results are now being used as an important input to the design of the Urania plant underway in Italy.

An additional part of the project was to explore the production of ^{39}Ar through cosmic ray interactions on the UAr. What we found is that, although simulations software exists, the production of isotopes through cosmic ray interactions and the production of ^{39}Ar is not well understood at sea level.

A highlight of the UAr production is that it was presented on the floor of congress as it relates to helium production in the United States by Rep. Jerry McNerney on June 16, 2016.

Modeling Underwater Sound in Coastal Environments to Accelerate Development of Renewable Ocean Energy

Wen Long

This project developed a cutting-edge modeling tool called FVSOUND for assessing the impacts of noise generated by the construction and maintenance of renewable ocean energy developments on marine mammals and fish in the coastal oceans.

The increasing demand for renewable energy from off-shore sources has raised concerns that underwater noise from construction and operation of off-shore devices may interfere with marine animal communication and navigation, and may cause injury. Based on a finite volume method, an underwater sound model called FVSOUND was developed to simulate sound propagation from marine-hydrokinetic energy devices and off-shore wind energy installations. The FVSOUND model accounts for four major mechanisms of ocean sound propagation to determine: the sound level distance from the source; the change of sound direction due to factors that affect the non-uniform distribution of ocean sound speed; reflection of sound from natural boundaries (surface, seabed, coastal lines); and transmission and attenuation of sound into the seabed. Prior models have not considered all mechanisms, so they do not fit the requirements for sound assessment for coastal oceans. The current model is built on a highly scalable, high-performance parallel computing library with a fast numerical solution method, providing sound levels rapidly for any location within 50 miles of the noise source.

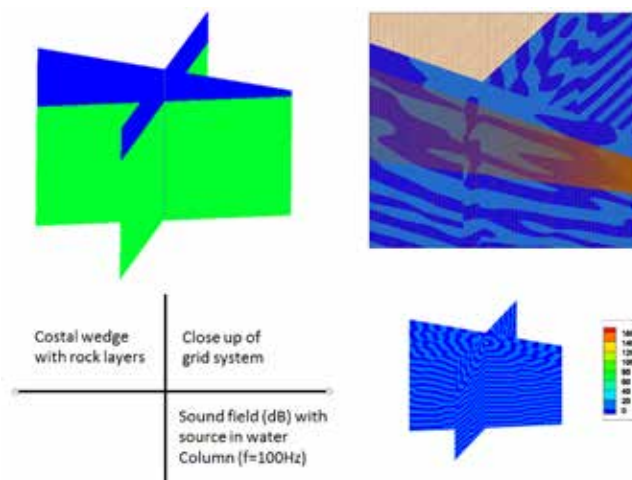
Sound transmission is closely related to ocean water density, which is, in turn dependent on water temperature, salinity, and pressure (depth). Sound transmission in coastal waters is more temporally and spatially variable than in the open ocean due to seasonal influences of river plumes, the effect

of meteorological wind stress and heat fluxes, and reflections of sound from complex coastal geometries. An advanced model can be used to account for this variability to predict sound fields accurately. However, coupled hydrodynamic and acoustic modeling has not been developed because the intensive computational resources needed for coupled modeling have not been readily available until recently.

Through this project, we have worked to improve the ability to predict sound propagation in coastal regions by providing two major innovations in the field of acoustic modeling. First, we formulated the acoustics problem mathematically for meeting the challenges of coupling a time domain hydrodynamic model with a frequency domain acoustic model. Second, we coupled the well-known 3D ocean hydrodynamic circulation Finite Volume Community Ocean Model (FVCOM)—which resolves the variations of water density and sea surface elevation—with an acoustic model that solves underwater sound transmission loss, reflection from lateral, bottom, and surface boundaries, as well as refraction due to the varying density field.

Sound in a 3D coastal domain is challenging to solve because of complex geometry and the short sound wavelengths, and requires many grid points to resolve the sound field. This situation increases the number of unknowns in the resulting discretized equation system for moderate (e.g., 100 Hz) to high frequency sound (e.g., 1,000 Hz).

In FY 2016, we succeeded in developing the FVSOUND software for simulating sound through the following achievements: 1) developing a Discretizing Helmholtz sound equation with terrain, using a coordinate system in a finite element grid system; 2) solving the resulting equation using the Complex Shifted Laplacian Preconditioner method with parallelization; 3) implementing



3D benchmark testing with an idealized coastal wedge (top left). The grid is shown in top right. A sound source of 100 Hz is placed in middle of water column, and the sound pressure field is shown in bottom right.

a radiation boundary condition; 4) adding rock layers, as sound can propagate into sediments; and 5) coupling the acoustic model with the FVCOM model.

Steps towards validation of FVSOUND were made with the collection of a sizable field dataset from Sequim Bay, Washington, and setting up the model to run there.

The output of this project includes a peer-reviewed paper published for the prototype in Marine Technology Society Journal, drafting a new manuscript of the full model, and licensing of FVSOUND v.1 underway. Finally, interest has been expressed in FVSOUND by university faculty and other researchers associated with ocean energy development.

Search for Lepton Number Violation

John L. Orrell

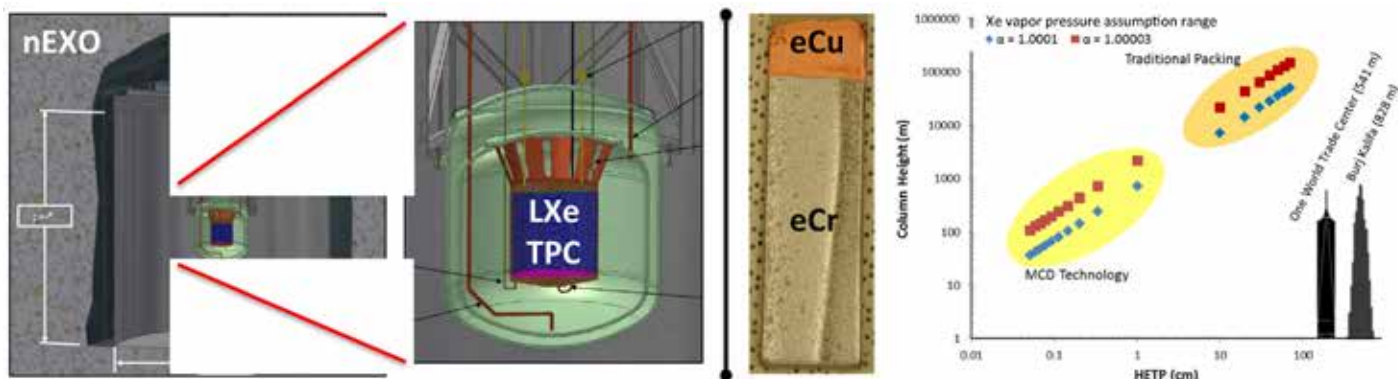
This project will assist development of a ton-scale neutrinoless double beta decay experiment employing ^{136}Xe operated as a liquid xenon time projection chamber by working with the nEXO Collaboration. The focus of this project is on background estimation, material radio-impurity assay, low background materials development, and isotopic enrichment of xenon, which are areas of PNNL expertise.

PNNL is investing in the development of the next generation of neutrinoless double beta decay research, an experimental program recommended by the scientific community in the 2015 Nuclear Physics Long Range Plan to search for explicit lepton number violation and the nature of the neutrino. Lepton number—the quantum number identifying electrons, muons, taus, and their respective neutrinos—is conserved in all observed Standard Model particle interactions. However, due to their electrically neutral nature, neutrinos and their antimatter counterpart anti-neutrinos may be unique among the Standard Model particles, in that they are actually the same identical particle, proving lepton number cannot be a conserved quantity. Neutrinoless double beta decay ($0\nu\beta\beta$)—the double beta decay of an unstable nucleus without the emission of the requisite two anti-neutrinos—is potentially an experimental means to identify the matter-antimatter uniqueness of neutrinos. The discovery of lepton number violation through $0\nu\beta\beta$ decay would open the door to a possible explanation of the matter-antimatter asymmetry of the universe: the fact that only matter predominates and antimatter has only a fleeting existence in our world.

The nEXO Collaboration is an international organization of research institutions developing a ton-scale, next-generation $0\nu\beta\beta$ decay experiment employing a target mass enriched in ^{136}Xe and operated as a liquid xenon time projection chamber. PNNL expertise in background estimation, material radio-impurity assay, low background materials development, and isotopic enrichment are significant contributions to the research and development program to demonstrate the feasibility of the nEXO experimental design.

In FY 2016, the PNNL team has measured radioactive impurities in materials under consideration for use in the nEXO experiment. Commercial copper was measured using inductively coupled plasma mass spectroscopy to determine the trace impurities of uranium and thorium. These trace radioactive impurities can create a background to the observation of $0\nu\beta\beta$ decay. Measurement of these isotopes for the purposes of mitigating their presence in the experiment is crucial to demonstrating the design feasibility. The copper was measured to have levels of U and Th at 0.2 and 0.1 parts per trillion (ppt). The team also measured trace radioactive impurities in the Si of Si photomultiplier photon detectors at levels below 1 ppt. This material assay analysis is used by the nEXO Collaboration to guide design and select material alternatives. The PNNL team is developing an electro-deposition method for producing a chrome-copper alloy that may provide both a higher purity construction material, as well as satisfying the structural design requirements of the time projection chamber vessel.

The PNNL team has also worked to understand the potential for a new means to enrich xenon using micro-channel distillation (MCD) to produce the several tons of ^{136}Xe needed for a next-generation experiment with sensitivity reach recommended in the 2015 Nuclear Physics Long Range Plan.

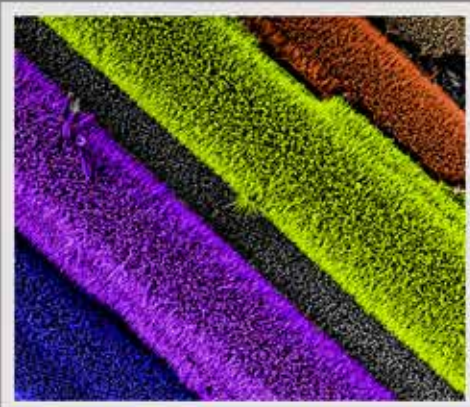


The two panels on the left show the nEXO neutrinoless double beta decay experimental design concept. The two panels on the right show (from left to right) electrodeposited chrome (eCr) on top of electrodeposited copper (eCu) and the first analysis of linear column heights estimated for a vapor pressure isotopic enrichment distillation to produce ton-scale quantities of ^{136}Xe using either traditional distillation technology or a MCD technique developed at PNNL.

ON THE COVER



At Pacific Northwest National Laboratory, researchers are enhancing the fundamental understanding of nucleation sites and growth characteristics of zinc oxide plates (shown here) and how these plates nucleate and grow as secondary structures on zinc oxide surfaces. This is a vital step in making zinc oxide a more effective material for use in the development of high-energy storage systems, such as lithium-air and zinc-air batteries. The research is being conducted in facilities at PNNL and EMSL, a DOE-SC national scientific user facility located at PNNL. This research is being funded by DOE's BES.



PNNL researchers are designing novel metal alloy anodes that improve the effectiveness of sodium-ion batteries. Within these anodes, nanofibers created through an electrospin technique serve as key components. The tiny fibers, shown in this colorized image, were captured with a helium ion microscope. The research is based on a design material synthesis approach that achieves targeted nanoarchitectures by controlling chemical reactivity at interfaces. The goal is to create a low-cost storage technology that makes it possible to more effectively capture energy and manage the nation's power grid. This research was funded by DOE's BES.



PNNL researchers are framing a magnified view of a microbe on Arabidopsis plant roots to provide a "window" into the rhizosphere – the world of soil, roots, and microorganisms. The image was captured with a Helios Nanolab dual-beam focused ion beam/scanning electron microscope at EMSL. Funded by DOE's BER, this research includes scientists from EMSL, PNNL, DOE's Joint Genome Institute, Brookhaven National Laboratory, and the Universities of Minnesota and Missouri.

2016 Annual Report

Laboratory Directed Research & Development

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