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	Characterizing Bioavailable Soil Nutrients Across Coastal Systems
	Western Lake Erie and Chesapeake Bay
	August 2022
	Jared Musci Kaizad Patel
	U.S. DEPARTMENT OF ENERGY Prepared for the U.S. Department of Energy under Contract DE-AC05-76RL01830

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Characterizing Bioavailable Soil Nutrients Across Coastal Systems: Western Lake Erie and Chesapeake Bay

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Abstract

Terrestrial Aquatic Interfaces (TAIs), defined as where land meets water, exhibit important hydrological, biological, and biogeochemical dynamics and play a critical role in the Earth's carbon cycle. Current Earth System Models improperly represent this role due to the complexity of these systems and the disproportionally large impact TAIs have relative to their area. To improve our predictive capacity of coastal system dynamics, a better understanding of carbon and nutrient cycling across TAI's is needed. Coastal Observations, Mechanisms, and Predictions Across Systems and Scales (COMPASS) aims to achieve this through field observations and laboratory incubations throughout the Western Lake Erie and Chesapeake Bay regions. Here, we report on preliminary results from a soil characterization experiment across the WLE and CB regions. This experiment was conducted as part of the 2-year field, measurements, and experiments (FME) pilot study portion of COMPASS to provide initial baseline data for the sites and to characterize the spatial variability of our analytes. Soil samples were collected from three locations along the TAI transect (upland forest, transitional forest, and wetland) and analyzed for bioavailable nutrients using three types of extractions: (a) water extractions, (b) 2M KCl extractions, and (c) 1M NH4Cl extractions to analyze (a) water soluble organic carbon (WSOC), (b) ammonium and nitrate, and (c) base cations, including Ca, Mg, Na, K, and Fe. These analyses, in conjunction with other bulk physicochemical analyses, will help establish baseline understanding of these sites to drive future research and design focused experiments that will ultimately improve our predictive understanding of these freshwater and marine coastal TAIs and their response to short and long-term changes.

I. INTRODUCTION

Carbon and nutrient cycling are a fundamental part of the Earth system and play a significant role in ecosystem function and climate change. Although this cycling occurs at every stretch of the Earth's surface and atmosphere, when viewed 2-dimensionally, this system can be conceptualized as cycling that occurs on land, on water, and where the two meet. Current Earth System Models do a good job at representing the land and water areas of the Earth but have trouble adequately representing the cycling that occurs where they intersect. These intersections where land and water meet are called Terrestrial Aquatic Interfaces (TAIs) and they play a huge role in the carbon and nutrient cycling of the Earth. Although they only account for a small percentage of the Earth's total surface area, they impact the global climate at a ratio that greatly exceeds the area they occupy [¹]. There is therefore a TAI knowledge gap in which these coastal systems are not represented well enough in Earth System Models, resulting in insufficient model dynamics. TAIs are extremely dynamic and constantly changing due to tides and periods of drought or inundation. This dynamic nature results in them exhibiting unique and significant hydrological, biological, and biogeochemical dynamics. To improve scientific understanding, modeling, and predictive capacity of these dynamics, Coastal Observations, Mechanisms, and Predictions Across Systems and Scales (COMPASS) was formed [2]. The project is beginning as a 2-year pilot study in the Western Lake Erie (WLE) and Chesapeake Bay (CB) regions meaning freshwater and saltwater coastal systems. It is a large project with many moving parts, but this paper focuses on the bioavailable nutrients soil characterization experiments that fall within the Field, Measurement, and Instrumentation (FME) component of COMPASS.

II. BACKGROUND

A. Soil Characterization

Soil characterization is simply analyzing soil to gain a baseline understanding of the properties and characteristics of that soil. Soil characterization is not an experiment with a tested hypothesis but is an overall analysis of the current state of the soil to better understand the study sites. There are many ways to characterize soil including soil biology, chemistry, physics, and hydrology. A combination of these characterizations was used, but this study specifically focuses on the bioavailable nutrients of the soil.

i. Bulk Analyses

Bulk Analyses refer to analyses that look at the characteristics of the soil sample as a whole. The bulk analyses examined in this characterization study were Total Carbon, Total Nitrogen, Total Sulfur, pH, Conductivity, Water Retention, and Soil Texture. These are talked about more in depth in a partner paper.

ii. Bioavailable/Extractable Nutrients

Bioavailable nutrients refers to nutrients that are readily available to plants and microbes. Operationally, this is defined as the nutrients present on the soil surface that are easily extractable when the samples are shaken with a liquid medium like water or salt solution. This is important because it distinguishes between what nutrients plants and microbes can access and what is locked up in the soil. The three types of extractable nutrients examined in this characterization study were (a) Water Soluble Organic Carbon (WSOC), (b) Base and Acid Cations (Ca, Mg, Na, K, Al, and Fe), and (c) Extractable Inorganic Nitrogen (Ammonium and Nitrate).

B. SHIMADZU TOC-L Analyzer

Dissolved organic carbon (DOC), interpreted as WSOC in our case, was measured by catalytic combustion using a Shimadzu TOC-L analyzer. It is a quantitative analysis because it tells us how much DOC is present in our samples. The samples are heated and combusted at 680 °C in purified air to induce oxidation. The carbon dioxide generated by oxidation is cooled, dehumidified, and detected as TC using an infrared gas analyzer. Due to the large percentage of Inorganic Carbon (IC) often present in environmental samples, Non Purgeable Organic (NPOC) is used to avoid significant errors in Total Organic Carbon (TOC) analysis. This is done by acidifying the sample and sparging it to convert any inorganic carbon into carbon dioxide. Once that carbon dioxide is removed, what remains is NPOC which is measured as TC using Nondispersive Infrared (NDIR) detection, as discussed above (Figure 1) [³].



Schematic Diagram of NPOC Measurement

Figure 1: How DOC is Measured in the Form of NPOC (Source: $[^3]$)

C. Fourier Transform Ion Cyclotron Resonance Mass Spectrometry (FTICR-MS)

FTICR-MS is used to characterize the molecular composition of the DOC. It is a qualitative analysis because it tells us what kind of DOC is present in the sample but does not provide certainty on the magnitude of specific molecular contents. It is a non-targeted approach because it examines what is seen across the 200-900 m/z (mass to charge ratio) range as demonstrated in Figure 2. FTICR works by ionizing the molecules, trapping them in a Penning trap using a magnetic field and electrical trapping plates, and exciting the ions using a radio frequency (RF) signal so they oscillate in phase at their respective cyclotron frequencies (a function of their m/z) [^{4,5}]. The ions induce a charge, and the recorded signal of those charges can be transformed into a mass spec using a FT [⁵]. This allows for extremely high resolution so we can distinguish between ions with very similar masses in highly complex systems [⁶]. One of the ways we can visualize these complex systems is via Van Krevelen Plots (Figure 3).



Figure 2: FTICR Results Example (Source: [⁷])



Figure 3: Two Van Krevelen Plot Examples for Various Soil Samples (Source: [⁸])

D. Inductively Coupled Plasma Mass Spectrometry (ICP-MS)

ICP-MS is used to determine the amounts of specific elements and molecules in a sample (Ca, Mg, Na, K, Al, and Fe in our case). It is a targeted approach compared to FTICR's non-targeted approach because it focuses on specific peaks across the mass spec range as demonstrated in Figure 4. Additionally, ICP is an elemental analysis technique, as it measures elements rather

than molecules or compounds like FTICR-MS does. ICP works by atomizing the sample and creating ions using a plasma, which are then detected using mass spectrometry by filtering the analyte ions by mass [⁹].



Figure 4: ICP-MS Results Example (Source [¹⁰])

E. Colorimetric Analysis

Colorimetric analysis uses absorbance to determine the amount of nutrients present in a solution. This analysis is founded on the Beer-Lambert law which relates absorbance (color intensity) to concentration. The analysis process is typically automated using a flow injection system such as the Lachat QuikChem 8500 Flow Injection Analyzer shown in Figure 5. Solution is run through a tested and calculated chemical process to isolate the nutrients of interest (Ammonium and Nitrate in our case). Once the nutrients are isolated within the solution, absorbance is measured via incident and transmitted light as shown in Figure 6. This absorbance is then related to a nutrient concentration using a Beer's Law Plot (Figure 6).



Figure 5: Lachat QuikChem 8500 Flow Injection Analyzer (Source: [¹¹])



III. METHODS

A. The Study Domain

There are three sites in the WLE region: Old Woman Creek (OWC), Portage River (PR), and Crane Creek (CC); and two sites in the CB region: Money Stump Marsh (MSM), and Goodwin Islands (GWI). Each site consists of a TAI. TAI transects consist of three sections: an upland forest, a transitional forest, and a wetland as seen in Figure 7. Multiple samples were taken from each section along these TAI transects at each of the five sites within the study domain to determine spatial variability. Additionally, soil from different horizons was collected at each sample point when possible. This setup resulted in 206 samples in total that each went through 7 specific analyses, not including 3 bulk analyses for each transect location as well.



Figure 7: Sections of a Terrestrial Aquatic Interface (Source: [²])



Figure 8: Soil Profile Graphic and Example (Source: [^{13,14}*])*

B. Soil Processing

Soils were sieved through 4 mm mesh and homogenized to create uniform samples. They were subsampled and weighed into centrifuge tubes for subsequent analysis. All samples were refrigerated at 4 °C until ready for analysis.

C. Soil Extractions

Once processed and needed for analysis, the samples were extracted. Soil extractions are when a liquid medium removes the most readily available nutrients from the soil surface, representing what is most available to plants and microbes. To perform the extraction the desired amount of extraction solvent (1:5 soil to liquid in our case) was added to the pre-weighed soil to begin the extraction. The soil solution was then shaken for 1 hour at 200 rpm to enhance the extraction process. The test tubes were then placed in a centrifuge for 15 minutes at 7000 rcf and 21 °C to separate the solid soil from the extracted solution. The solution was then filtered using a vacuum system or a syringe depending on the nutrient being analyzed. The result is a concentrated filtered extract ready for nutrient analysis.

i. Water Soluble Organic Carbon (WSOC)

For the WSOC extraction process, 8 grams of field moist soil were shaken with 40 mL of Milli-Q water for one hour. Milli-Q water is ultra-filtered water with extremely low carbon contents (4-18 ppb TOC). The samples were then centrifuged and filtered into 40 mL glass vials using 0.45 μ m PES (polyethersulfone) syringe filters. The filtered extracts were then refrigerated at 4

°C until ready for TOC analysis (using the Shimadzu instrument) and Solid Phase Extraction (SPE).

To prepare for Shimadzu, three test samples are run to ensure the limits of the DOC calibration curve are sufficient. Once the calibration limits are confirmed or adjusted, 9 mL of extract are transferred into glass culture tubes and run on the Shimadzu TOC-L system. As mentioned in the background, DOC is measured, via catalytic combustion, as NPOC, wherein dilute hydrochloric acid is used to react with any inorganic C in the samples, and leave behind organic C.

After the samples are run on the TOC analyzer, SPE can be performed. The DOC amounts inform the desired concentrations for SPE. The objective of SPE is to remove interfering compounds (e.g. salts and iron) and concentrate the sample in preparation for FTICR. First, the extraction solutions must be diluted to obtain the desired concentration of DOC in the SPE sample. The diluted samples are then acidified to around pH 2 using 85% Phosphoric Acid. Once acidified, SPE can be performed. SPE involves activating a porous membrane using methanol, capturing the liquid sample in the membrane, rinsing HCl through the membrane to remove any salts and minerals, then eluting the sample from the membrane into glass Microsolv vials using methanol. Now that SPE has occurred, the soils are ready for FTICR.

ii. Base and Acid Cations (including Ca, Mg, Na, K, Al, and Fe)

For the Base and Acid Cations extraction process, 5 grams of field moist soil were shaken with 25 mL of 1M NH4Cl. NH4Cl was used because NH4 has a positive charge, and therefore facilitates cation exchange, attaching to the soil particles and removing the cations into solution. Once shaken and centrifuged, the solution was vacuum-filtered into 50 mL Olympus plastic test tubes using Whatman 42 filters. The filtered extract was then refrigerated at 4 °C until ready for ICP analysis.

iii. Ammonium and Nitrate

For the Ammonium and Nitrate extraction process, 5 grams of field moist soil were shaken with 25 mL of 2M KCl. KCl was used because unlike NH4, KCl does not contain any forms of Nitrogen and therefore will not interfere with the Nitrogen based extraction. Once shaken and centrifuged, the solution was vacuum filtered into 50 mL Olympus plastic test tubes using Whatman 42 filters. The filtered extract was then refrigerated at 4 °C until ready for Colorimetric analysis.

IV. RESULTS/DISCUSSION

Due to the preliminary nature of this soil characterization data, much of the work done has not reached the test and analysis phase. For this reason, this paper specifically focuses on Dissolved Organic Carbon (DOC) and Nitrate across the TAI transect and soil horizons. DOC is covered for both CB and WLE whereas Nitrate is only covered for CB. Additionally, DOC (bioavailable) is compared to TC (bulk analysis) to illustrate the difference between bioavailable and total nutrients. The Nitrate results are less comprehensive but focus on variability across study sites.



Figure 9: Bulk vs Bioavailable Nutrients

This first graph illustrates, at a broad scale, the large difference between total carbon and DOC. This emphasizes the significance of distinguishing between what is present in soil and what is actually available to plants and microbes.



Figure 10: Bulk vs Bioavailable Nutrients - Refined

This next graph fine tunes the scale of our analyses and highlights the general correlation between TC and DOC while also displaying a more legible difference in scale compared to the previous graph.



Figure 11: DOC vs Transect

When initially looking at DOC trends along the TAI transect for both sites, we see that CB has higher amounts of DOC than WLE, and that a downward sloping trend seems to exist in coordination with the TAI transect as referenced in Figure 7.



Figure 12: DOC vs Transect - Chesapeake Bay

When we take a closer look at CB alone and examine the soil horizons, we see that the O horizon samples are much higher than the A and B horizon samples as expected, and that all horizons decrease in DOC as they travel down the transect gradient.



Figure 13: DOC vs Transect – Western Lake Erie

When we take a closer look at WLE alone and examine the soil horizons (no O horizons present in WLE), we see that the results are not as distinct with no obvious trends along the transect or the soil horizon. This indicates the importance of characterizing these sites before future experimentation, as disparity in trends can be present. Alternatively, when we compare back to CB (Figure 12), we see that CB DOC is slightly higher by horizon and transect than WLE.



Figure 14: Nitrate vs Transect

Shifting over to Nitrate in WLE, we see that the results are relatively similar with some variation between range and site. No clear trends are seen here, but it is important to note that these are all A horizon soils. So, when we look at the soils specific to each site, we see that PR and CC have much higher Nitrate values in the wetland than does OWC. This signifies the importance of

characterization because it shows that different sites can have different amounts of NO3 even with the same experimental setup.

Overall, although the data/results are limited and the analysis is brief, these graphs highlight an important need to always consider all the variables. With datasets like these, where several different variables are influencing your soil samples (transect, horizon, site, region), it is easy to illustrate an incomplete story with your data - as seen in the transition between Figure 11 and Figure 12. Therefore, caution should be used when doing bulk comparisons across regions and variables.

V. CONCLUSION

Characterization of the soil in this study domain is extremely important because these analyses will help establish baseline understanding of these sites. This understanding will lay the groundwork for expectations and drive future research. This research will be driven by focused experiments that test specific hypotheses to answer more applied and tailored questions about these dynamic systems. Knowledge gained from this characterization experiment and the experiments that it will create will improve fundamental representation in Earth System Models and ultimately improve our predictive understanding of these freshwater and saltwater coastal TAIs and their response to short and long-term changes. Short term being flooding, drought, and agricultural runoff and long term being anthropogenic climate change, and sea level rise. As climate change continues to amplify and changing environmental conditions continue to occur, a need for understanding these dynamics and their role in the carbon and nutrient cycle is paramount. Better understanding, predicting, and modeling TAIs is a step to doing just that.

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