Pacific Northwest NATIONAL LABORATORY

PNNL-37398

Enhancements to Lab-on-a-Fish Technology

Final Report for I3T Project 81245

March 2025

Daniel Deng
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U.S. DEPARTMENT OF ENERGY

Prepared for the U.S. Department of Energy
under Contract DE-AC05-76RL01830

PACIFIC NORTHWEST NATIONAL LABORATORY
operated by
BATTELLE
for the
UNITED STATES DEPARTMENT OF ENERGY
under Contract DE-AC05-76RL01830

Printed in the United States of America

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Summary

PNNL has engaged in discussions with several organizations regarding potential future collaborations. PNNL has developed the Lab-on-a-Fish prototypes and produced video instructions for an organization conducting research on shark eggs. Several organizations have expressed interest in using the Lab-on-a-Fish for their studies. We've also updated two versions of the Lab-on-a-Fish to expand its applications to a wider range of animal studies and accelerate its commercialization. The new design of the Lab-on-a-Fish, which incorporates PCB-based electrodes, has demonstrated the ability to capture clear ECG signals without the need for additional needle-shaped electrodes. This innovation eliminates the need for implanting electrodes beneath the fish's skin at specific locations, which is required with the current version of Lab-on-a-Fish. This change significantly simplifies the manufacturing and implantation process. For the Lab-on-a-Fish design that uses an optical pulse oximeter, the measurement results were primarily influenced by respiratory activity rather than heart rate. This occurred because the oximeter was placed beneath the operculum. Further research and development are needed to explore more suitable locations and methods for accurate pulse oximeter measurements.

Acknowledgments

This research was supported by the Strategic Investment Laboratory-Directed Research and Development (LDRD) Program at Pacific Northwest National Laboratory (PNNL). PNNL is a multi-program national laboratory operated for the U.S. Department of Energy (DOE) by Battelle Memorial Institute under Contract No. DE-AC05-76RL01830.

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Figures

1.0 Introduction

PNNL successfully developed a working prototype of a Lab-on-a-Fish sensor and implanted the device in Rainbow Trout through a small-scale study[1]. Compared to current biotelemetry tags that are constrained to limited types of measurements, our device integrates temperature, pressure, magnetic field, acceleration, gyration, ECG, and EMG sensors simultaneously for fish behavior, physiology, and environmental sensing on a platform with low weight (2.4 g) and small dimensions (⌀: 7 mm, L: 37 mm). The device can wirelessly transmit data in real-time as well as store it in a Flash memory. These features open new possibilities for studying behavior and physiology in smaller fish with extended periods and challenging locations.

The Lab-on-a-Fish's ability to store and transmit historical sensor data on both environmental parameters and the tagged animals' bioactivity will provide valuable information for long-term animal behavior studies, leading to critical insights into the correlation between physiological behavior and physical stressors. Therefore, it will assist in strategy and decision-making for hydropower and marine energy operations and help solve the challenges of hydropower and marine energy production related to the potential for animal injury and mortality from hydropower and marine energy deployment.

The objective of the project is to provide Lab-on-a-Fish devices for third-party testing and evaluation by industry and research organizations. Additionally, it aims to optimize the Lab-on-a-Fish by incorporating non-invasive sensing methods to measure heart rate and oxygen saturation sample, and manufacture samples for third-party testing and evaluation. This feedback and development will allow us to refine the Lab-on-a-Fish, greatly expand its applications of using Lab-on-a-Fish to study a variety of animals, such as sharks and tuna, and accelerate commercialization.

2.0 Collaboration and marketing

PNNL has discussed potential collaborative efforts with Stanford University aimed at testing the application of Lab-on-a-Fish on tuna.

We manufactured the Lab-on-a-Fish prototypes and documented step-by-step software installation guidelines and comprehensive user instructions in video format. The prototype and instructional materials were sent to Wageningen University in the Netherlands for research on studying shark eggs. Moreover, we participated in multiple virtual meetings to discuss the functionalities and potential applications of these Lab-on-a-Fish prototypes.

We have received interest from the Georgia Institute of Technology (Georgia Tech), the University of Chicago, and Oregon State University (OSU) to try the Lab-on-a-Fish. The delivery timeline is currently being discussed.

3.0 Develop non-invasive sensing methods

3.1 Two non-invasive Lab-on-a-Fish designs

We developed two designs for the Lab-on-a-Fish: one incorporates a PCB-based electrode, while the other utilizes an optical pulse oximeter sensor, both of which are shown in Figure 1. The PCB-based electrode structure is an extension of the existing Lab-on-a-Fish circuit board, with electrodes connected via flexible interconnections to the main circuit board. With all sensor components and electrodes integrated into a single PCB, this design anticipates cost savings through simplified manufacturing and implantation, eliminating the need for additional wire connections and needle-shaped electrodes. The optical pulse oximeter design comprises two components: the optical pulse oximeter itself and a microcontroller (MCU) prototype board to capture and store the oximeter readings. The upper oximeter sensor can be disassembled to allow the connection of additional wires, enabling live fish testing to be conducted.

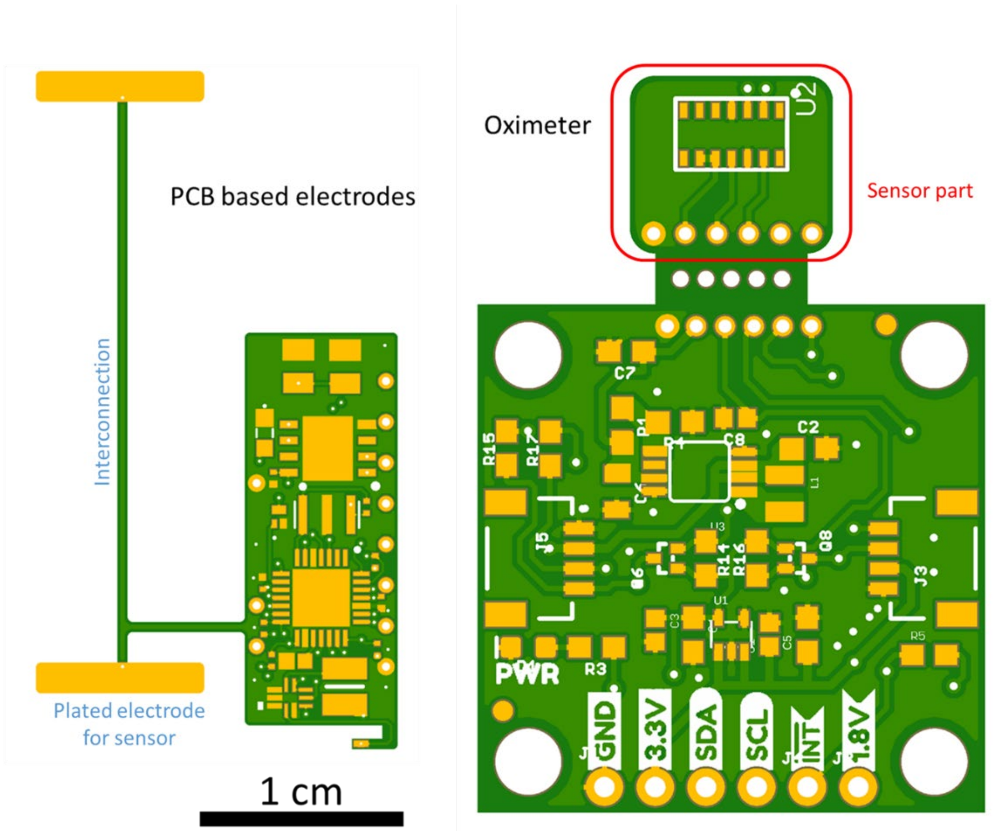

Figure 1. Images of Lab-on-a-Fish design for PCB based electrode and pulse oximeter

To facilitate the attachment of the optical pulse oximeter to the fish more conveniently, we designed a clamping-type sensor implantation technique for oximeter evaluation, as shown in Figure 2. This technique ensures secure integration between the inner oximeter sensor and the outer circuit board. The oximeter is the only part that closely attaches to the skin surface, adjacent to the blood vessels. The other components, including power, communication, MCU, and supplementary sensors, are consolidated within an external board linked to the sensor.

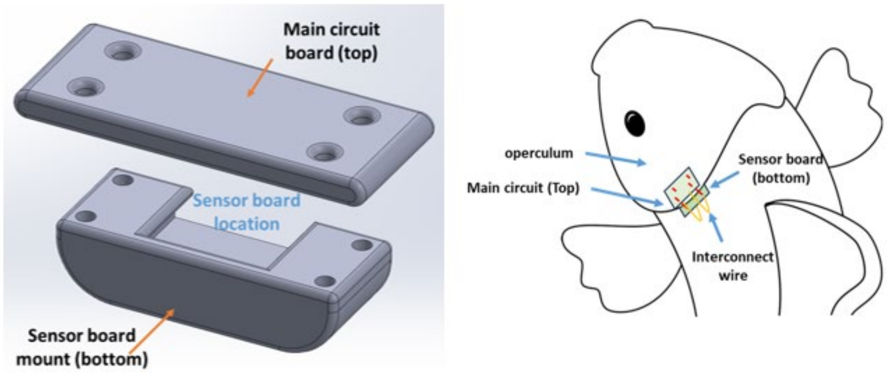

Figure 2. Explanation images for clamping design for the optical pulse oximeter

3.2 Two non-invasive design verification

To validate the sensor's performance, we created a sensor device equipped with an external sensing circuit for data recording. The PCB-based electrode was connected to the analog-front-end (AFE) IC for ECG measurement. The optical pulse oximeter sensor transmits data through serial communication to a main board.

To verify the performance of the optical oximeter sensor design, we conducted pulse rate measurements on the finger (Figure 3). The pulse rate data is shown in Figure 4a. The corresponding heart rate can be further derived by applying real-time data processing algorithm, presented in Figure 5b. Additionally, we can also incorporate moving window for averaging heart rate to obtain stable and consistent heart rate data.

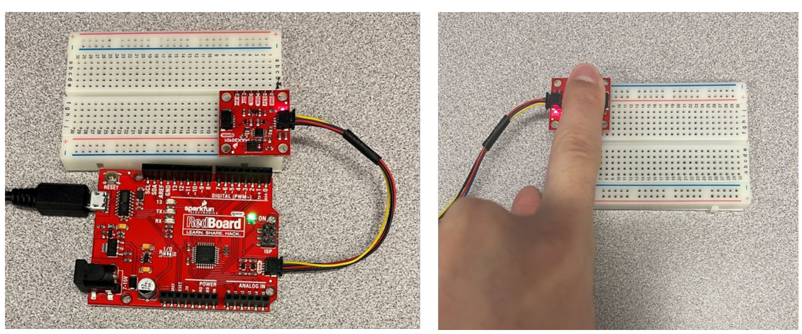

Figure 3. Photograph for on-body test setup for optical pulse oximeter

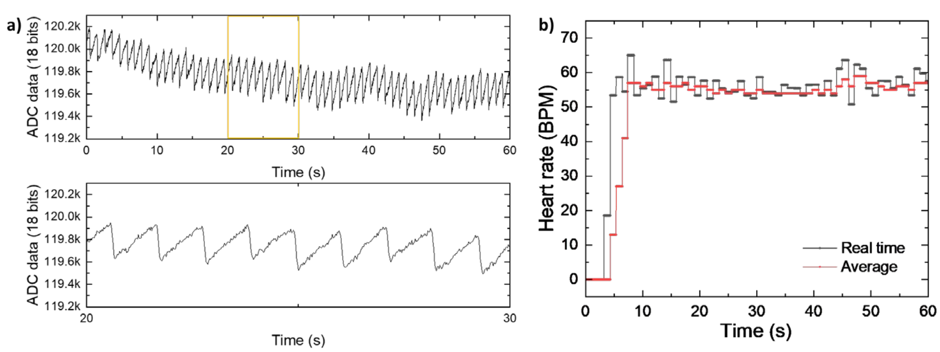

Figure 4. a) Obtained oximeter data from human finger and b) processed data for heart rate

3.3 Live fish test

After validating these two non-invasive methods on the human body, we proceeded to evaluate their performance on Spring Chinook fish at the Aquatic Research Laboratory (ARL). Figure 5 shows a PCB-based electrode Lab-on-a-Fish prototype with extended wires for recording ECG data, which will not be included in the final Lab-on-a-Fish product. A Spring Chinook, measuring 272 mm in length and weighing 252 g, was used for testing.

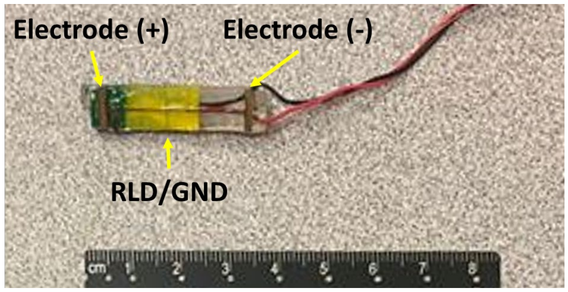

Figure 5. Photographs of PCB-based electrode sensor dummy

Initially, we administered an anesthetic to the fish, provided water through its mouth, and then inserted the PCB-based electrode Lab-on-a-Fish in close proximity to the fish's heart, as shown in Figure 6. After insertion, the incision was sutured using a stitching needle. ECG measurements were conducted continuously while the fish was in three states: under anesthesia, conscious, and during euthanasia. As shown in Figure 7, the resting heart rate under anesthesia exhibited a lower tendency, at approximately 30 bpm. Following awakening, the heart rate increased to around 72 bpm. During the euthanasia process, there was a significant drop in the heart rate to approximately 23 bpm.

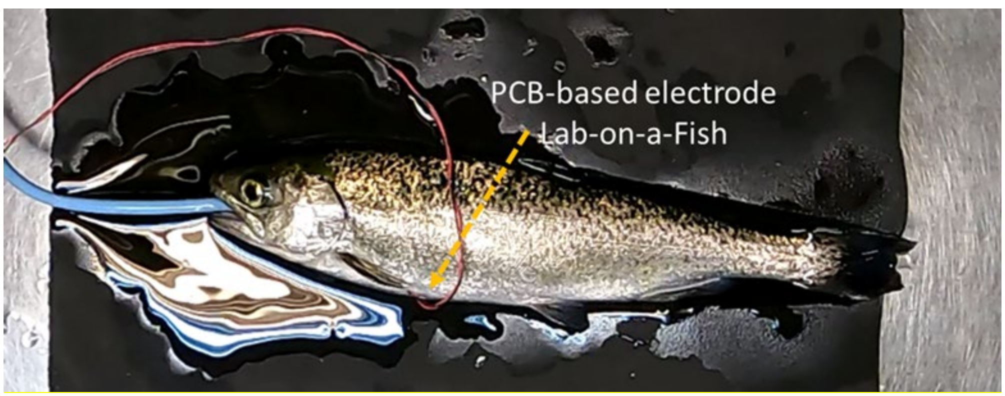

Figure 6. A photograph of set up for fish test with PCB-based electrode Lab-on-a-Fish

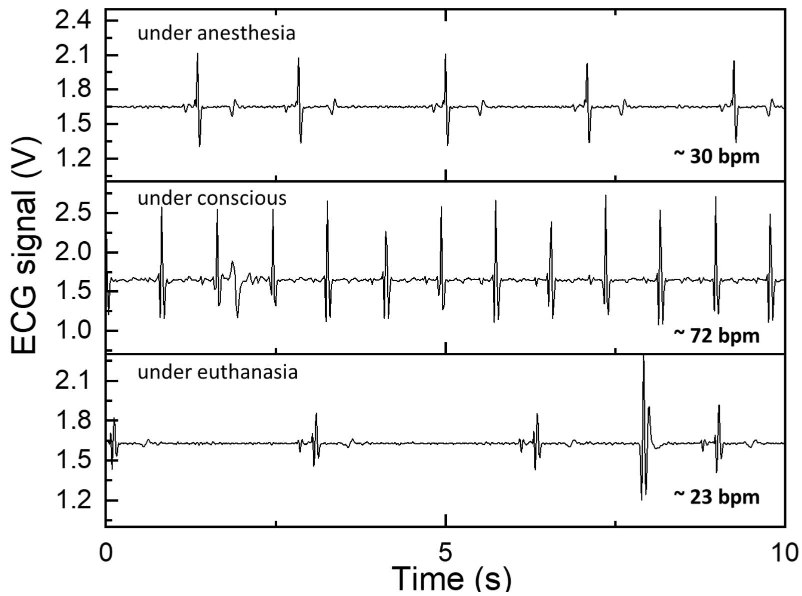

Figure 7. ECG signal of chinook from PCB-based electrode Lab-on-a-Fish under anesthesia, conscious and euthanasia

We also conducted another test using an optical pulse oximeter (Figure 8) to study the correlation between heart rate and the data detected by the oximeter. The oximeter was positioned facing the inner side of the operculum and securely attached to the main board, which was positioned on the outer side of the operculum through sewing. The PCB-based electrode Lab-on-a-Fish was inserted near the heart under the dermis (Figure 9). The oximeter readings and ECG measurements were recorded simultaneously.

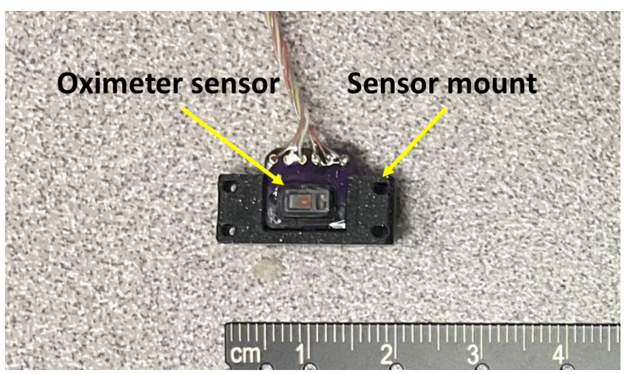

Figure 8. Photograph of optical pulse oximeter with sensor mount for clamping

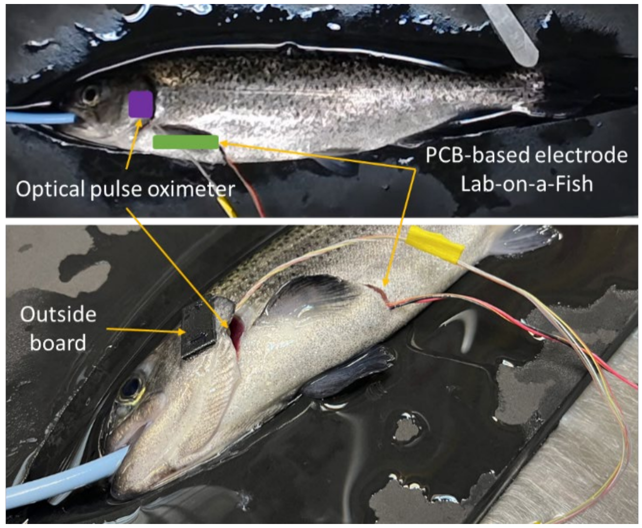

Figure 9. A picture depicting the insertion of an oximeter and a PCB-based electrode sensor into a fish

The measured data are illustrated in Figure 10. Our goal was to derive the heart rate using the oximeter readings on fish. However, the experimental results indicate that the oximeter data are impacted by the operculum's movement caused by respiration. After analyzing the oximeter data, it became evident that the fish displays a fairly consistent cycle, regardless of whether it is at rest or engaged in activity. This observed pattern differs from the ECG signal obtained from the PCB-based electrode Lab-on-a-Fish. The recorded ECG signal shows a lower bpm when the fish is under anesthesia and a higher bpm when the fish is awake. Therefore, to ascertain the suitability of the oximeter on fish, it is important to identify alternative locations on the fish where the heart rate can be monitored. We plan to conduct additional experiments to explore implantation locations for the oximeter sensor.

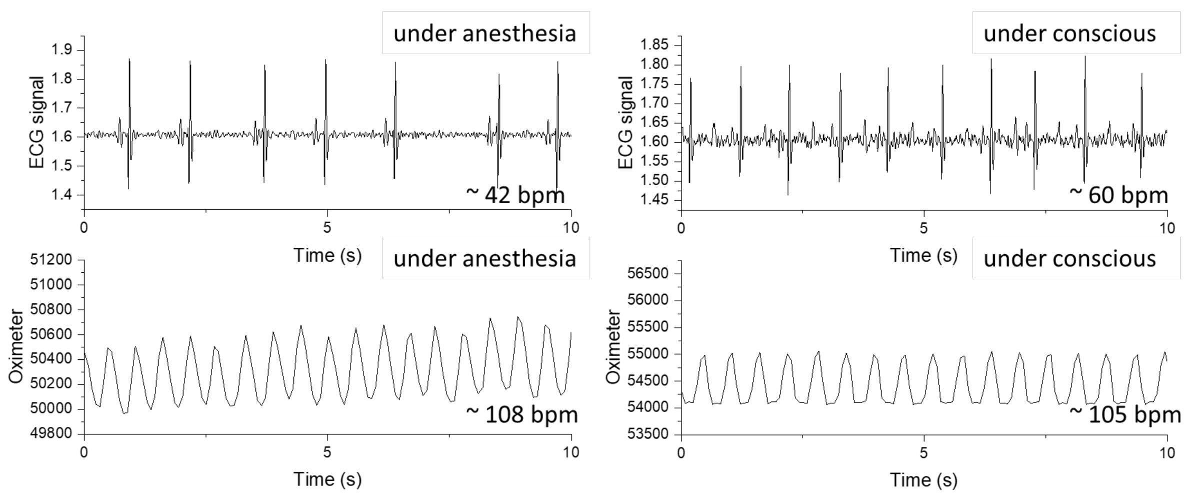

Figure 10. The ECG data from PCB-based electrode Lab-on-a-Fish (top), and oximeter data (bottom) during under anesthesia and conscious condition

4.0 References

1. Yang Y, Lu J, Pflugrath BD, et al. Lab-on-a-Fish: Wireless, Miniaturized, Fully Integrated, Implantable Biotelemetric Tag for Real-Time In Vivo Monitoring of Aquatic Animals. IEEE Internet Things J. 2022;9(13):10751-10762.

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