Clam Gape Sensing Equipment For Water Monitoring

Entire Organism Broadband Toxicity Sensors in the Form of Freshwater Clams are Instrumented to Monitor Gape Behavior

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The research and development L effort discussed in this article has been carried out as a Phase 2 small business innovative research (SBIR) contract under the auspices of the U.S. Army Center for Environmental Health Research. The Phase 2 effort includes developing a second generation biobay (biosensor) monitoring device for recording the gape of living bivalves mounted in an autonomous vehicle and characterizing the response of bivalves to a range of environmental variables and aqueous toxicants. The biological response-measured as change in shell gape and measurements of water quality with standard sensors-has been characterized in an effort to draw conclusions about the suitability of common freshwater clams (Corbicula) as mobile broadband toxicity sensors. The objectives of the Phase 2 SBIR were to characterize Corbicula as a broadband biosensor, design the biosensor suite for mobile platforms, implement a sensor fusion algorithm, and test and demonstrate the complete system. This article primarily addresses the development of the clam gape monitoring hardware.

Clam Fixture Development

Clams typically respond to irritating stimuli by closing (they pull their two

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Second revision clam fixture showing the clams arranged below the sensing circuitry, with the magnet plungers in contact with the top of each clam shell.

shell halves together and close). Otherwise, they are typically feeding and respiring with their shells open. In order to quantify the clam response to stimuli, equipment had to be developed. Past work in this area used inductive proximity sensors to sense when the clam was open or shut. For this project, several sensing technologies were developed, with Hall Effect sensors emerging as the sensor of choice. Hall Effect sensors are small, inexpensive and easy to waterproof. Linear Hall Effect sensors vary their voltage output with respect to magnetic field intensity. This signal change can be interpreted with a microprocessor. A magnet is glued to the top half of the clam's shell, which moves with respect to the Hall Effect sensor. As the magnet/clam shell moves, the magnetic field varies causing the Hall Effect sensor voltage to vary. The analog-to-digital converter and microprocessor convert this voltage into an integer value that corresponds to the extent of the clam gape. This data is logged onto a laptop or other data logging device.

The clam fixture went through two design revisions over the course of the Phase 2 SBIR, and then a third revision for use by the U.S. Environmental Protection Agency (EPA).

First Clam Fixture Revision

The first clam fixture design consisted of a central pressure vessel flanked by two rows of clams—one row on each side with eight clams per row.

This design required the user to glue a small magnet to one half of the clam, and then glue the other half of the clam to a stationary support. The magnet side of the clam moved with respect to the central pressure housing. Just inside the pressure housing, Hall Effect sensors sensed the variation in magnetic field as the clams moved. (Top) Third revision clam fixture designed for the EPA, with clams installed. This design can accommodate a wide variety of clam species, and can be deployed in any orientation. The design can also survive in situ for extended durations.

(Bottom) A tow fish equipped with clam fixture and water quality sensors.

This design was effective, but the gluing and orientation of the clams and magnets was cumbersome and time consuming. This design was retired shortly after the start of the Phase 2 SBIR.

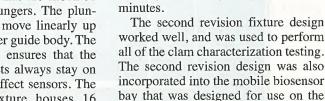
Second Clam Fixture Revision

The second clam fixture design is a multi-layer design comprised of an electronics housing on top, a plunger housing in the middle and clam supports on the bottom. The second revision clam fixture was designed to make use of the Hall Effect sensor circuitry and analog-to-digital converter circuitry developed for the first revision. The problems experienced with the first revision were strictly mechanical issues: the sensing circuitry worked well and was reused in all second revision units.

Instead of gluing magnets to one half of the clam, the second revision units have magnets that are mounted in stainless steel plungers. The plungers are allowed to move linearly up and down in a plunger guide body. The plunger guide body ensures that the plungers and magnets always stay on axis with the Hall Effect sensors. The second revision fixture houses 16 clams in two rows of eight.

The clams are glued to moveable platforms that can be advanced toward and away from the magnet plungers to allow for variability in clam size and shape.

Each clam fixture contains a power and data cable that runs from the clam fixture all the way up to the surface. The clam fixtures are powered with 12 volts, and data is returned as an RS-232 American Standard Code for Information Interchange data string. Each data string contains start characters, a sequence number and the 16 clam gape values. The clam fixtures can be programmed to return all 16



Third Clam Fixture Revision

cle (AUV) and towed platform.

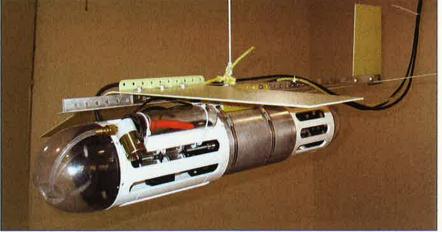
clam channels at a rate of 33 hertz, all

the way down to just once every 30

Ranger autonomous underwater vehi-

The third clam fixture revision is a design effort that was conducted separately from the Phase 2 SBIR. The U.S. EPA has been working on a multi-faceted source water monitoring initiative. As part of this, the organization is investigating different monitoring techniques. The entire organism clam biosensor is one of those techniques. The EPA evaluated the first revision clam fixture and provided valuable feedback on its usability and potential upgrades. After the evaluation





period, a design and production effort was undertaken to provide the EPA with a clam fixture that would meet their operational criteria.

The clam fixture had to be redesigned from 12 inches long to 24 inches long. This permitted the use of a wide variety of clam species, from the small freshwater *Corbicula* to three-inch-wide marine mussels. The EPA also required the capability to deploy the clam fixtures in any orientation.

The second revision clam fixture design requires gravity to load the plungers into contact with the clam shell. The plungers in the EPA design must keep contact with the clam shells regardless of clam fixture orientation. At the same time, the contact force could not be so high as to limit the ability of the clam to open. A magnetic repulsion design was implemented to ensure the plungers always made gentle contact with the clam shell, regardless of clam fixture orientation. So far, the EPA has been pleased with the clam fixture units. The units have been deployed in a number of locations throughout the United States. Clam gape data is being recorded along with standard water quality data, such as conductivity, temperature, depth, dissolved oxygen, pH, turbidity and chlorophyll. The data will be compared to see if the clams behave consistently in the presence of certain conditions that can also be sensed and verified by the water quality sensors.

Characterization and Field Testing

The clam characterization tests were carried out in a way that would simulate clam exposure on a mobile platform in a real environment. The clam fixtures were arranged in a recirculating flume. This simulated the movement of the clams through a body of water. The flume was fed by one reservoir full of standard condition clean water. This reservoir was circulated through the flume so the clams could acclimate to clean water. After the acclimation period, a valve was switched to shut off the clean water and introduce test condition water in a seamless transition. The test condition water was prepared in a separate reservoir.

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This change simulated the clams being exposed to changing water conditions as the clams moved through a body of water on a mobile platform. In order to provide a statistically significant sample size, each sample parameter was tested with 48 clams in the test flume, and each test was performed twice to check for variability in the results.

The clams have quantifiable gape response to a variety of stimulants. It is reasonable to attribute the gape response to the introduction of a test condition in laboratory flume conditions. During field testing, the clams were towed through the environment on a tow fish instrumented with water quality monitoring sensors. During field testing, it is difficult to attribute gape changes to any particular change in environmental conditions. In the real environment, several parameters can change at once, making it difficult to determine whether the clams are responding to the presence of a possible chemical irritant, or to the irritation associated with being pulled through the water. Further field testing and correlation with water quality sensor data is necessary in order to draw reliable conclusions about clam behavior on a mobile platform.

Conclusions

Biosensors have the potential to revolutionize water monitoring. Toxicity is a nebulous parameter that has to do with the physiochemical reaction of an organism, not just the readings of a few water quality sensors. This is why it is crucial that organisms be used to monitor water that humans will come in contact with. Entire organisms identify toxic events that water quality sen-

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"The clam characterization tests were carried out in a way that would simulate clam exposure on a mobile platform in a real environment."

sors miss. Entire organism biosensors have been used to warn humans of dangerous conditions for a long time in a number of industries, with effective and reliable results. The biosensor bay effort is a natural extension of this, and it is being done in a way that quantifies entire organism response, while checking it against water quality parameter measurements.

It is now possible to quantify clam gape response as a result of this development effort. It is possible to deploy clams in a variety of environments and monitor their gape behavior. The clam fixtures are enabling technology for organizations interested in entire organism biosensing. This technology is certainly finding application in source water monitoring, as demonstrated by the EPA. The clam fixture devices are small and rugged, making them suitable for integration on AUVs and other mobile underwater platforms. The deployment versatility lends the clam fixtures to a variety of U.S. Department of Homeland Security applications.

With further development, biosensors may be useful for forward deployed troops, or field test kits for industry.

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