Designing a pH Sensor Based on Spectrophotometry to Understand Chemical Changes in the Puget Sound

Introduction to Ocean Technology: OCEAN 216

LAG: Lauren Bayne, Xavier Giomi, Edward Cheng, Sophie Goddard, Hailey Holder, and Josie Adams

December 1st, 2021

Ocean Technology Center in Oceanography Teaching Building

1431 NE Boat St Seattle, WA 98195

Executive Summary

Changes in pH within the Puget Sound has effects on calcifying organisms and the food chain. Salt water that comes from the ocean and brackish bay water fed partially by runoff has different pHs. In the Puget sound estuary this is a large problem for organisms and has economic and societal impacts, so we want to test this in our area. Our goal is to measure pH in brackish water with different levels of salinity to test how it changes across a salinity gradient. The most effective way to test pH is to have a sensor in the Puget Sound waters directly. We want to see if this sensor will be able to detect pH changes and how accurately it will be able to show differences in pH. We will attempt to utilize a light sensor, non-toxic dye sensitive to pH, and water pumps to look at changes in the color of treated waters gathered.

While creating this sensor the team learned many new skills such as soldering, drilling, calibrating, fritzing, and 3D printing. We also learned about LED lights and light sensors, because the red and blue LED lights output a variable luminosity to the light sensor that we couldn’t fix. To solve the problem, white LEDs were used and blue and red filters were placed over them. We also learned how to use water pumps with switches. Together these created a functioning circuit that a microcontroller triggered through code.

Throughout the process we were productive and created a functional sensor. The device can be placed into the water to get pH measurements. The team calibrated the device and placed the floating sensor into the water. The pumps brought the water, and then the calibrated dye, into the cuvette. Next, the LEDs go off and the light sensor takes luminosity data from each. This data is then calibrated with previous known pH solutions. The pH retrieved from the field test was 6.7 and a pH strip showed a pH of 6 at this location. The in situ pH sensor worked and was relatively close to the pH of the water.

Introduction

The Puget Sound estuary holds a very large ecosystem that has diverse organisms and is important to humans for food, land and economics. However the impacts of increasing anthropogenic carbon dioxide are harmful to this ecosystem. It is affected by changes in pH due to its varying chemical water properties. The Puget Sound is initially fed by the ocean and then as the distance increases from the ocean there is an addition of runoff and freshwater from rivers. Some regions can have high salinity oceanic waters and others can have low salinity with runoff contaminants included (Feely et al. 2010). The team's motivation was to help fill gaps in the estuarine pH data and improve monitoring. This is important because pH can affect water quality and the marine organisms that live in these conditions. Due to the addition of contaminants carried down in rivers and runoff the Puget Sound Estuary chemical water properties change with location. The pH of salt water, that is directly connected to the ocean, is less acidic than brackish bay water that is fed by runoff.

There are natural processes that increase acidity in the Puget Sound due to its estuarine processes. The deep basins for instance are greatly affected by remineralization and stimulated respiration processes (Feely et al. 2010). Understanding these natural processes and their changes are important to be able to assess anthropogenic changes. The estimation for pH decreases caused by anthropogenic carbon dioxide increases is 24-49% of all acidification (Feely et al. 2010). This is a significant amount and has caused changes to the ecosystem. It is important to be able to continue to record these changes for the future.

An approach to measuring pH is spectrophotometry. This is the measure of light absorbance through a substance and its intensity. This approach uses indicator dyes that are added to seawater that act as absorption spectra for visible light (Carter et al. 2013). The dye in the water changes color when the water's pH changes. In the Carter et al. study an equation was used to calculate pH and the precision reached was high due to the dye used. This study utilized non natural dye and created a sensor for pH that can accurately detect small changes in pH with a standard deviation of 0.00042 pH units (Carter et al.).

The paper “Seawater pH measurements in the field: A DIY photometer with 0.01 unit pH accuracy” by Yang et al. is about creating a low cost portable photometer that gives pH measurements. This design was also a photometer that used LED lights. Their measurements were based on absorbances and this calculation was helpful to look over. The calibration was ratios of absorbance from the photometer to a spectrophotometer. The main point of their system was that it is cheaper and still accurate. Compared to other instruments such as electrodes which can range from $50-1500 and spectrophotometers that range from $8000-20000 (Yang et al. 2014). Their sensor also didn’t need to be calibrated. This information was really beneficial when deciding what path to go down.

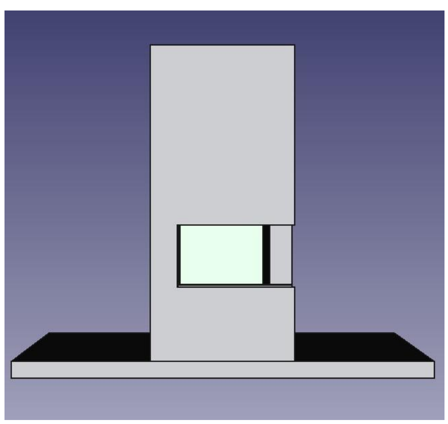
Another paper “Connecting chemistry concepts with environmental context using student-built pH sensors” by Sasha et al. demonstrated how a class built a pH sensor. This paper discussed using a light sensor and LEDs that were different colors, one red and one blue. As well they used cabbage juice for their dye because it is non toxic and a good standard for pH indication. Their 3D printed model was useful to look at for constructing something similar with a closed top for light control and holes for a light sensor and LEDs (Sasha et al. 2020). Our beginning steps were to recreate their sensor to understand each moving part. After doing so it was more straightforward how we wanted to go about creating our sensor. Keeping some of the same basics and changing the pieces that we needed to so that we could deploy this in the water.

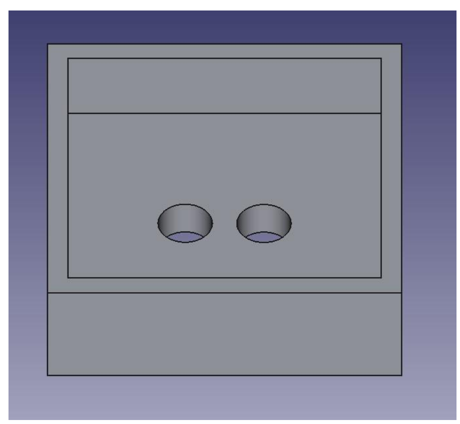
Using these approaches we have come up with the idea to utilize the LED lights and a light sensor as well as two water pumps. As the Sasha et al. paper described we created cabbage juice and are going to calibrate it to convey pH from luminosity. One pump brings in one second of outside water that we are measuring, and the other pump brings in half a second of cabbage juice dye. The pumps work using time in the code and we used these times to get a one third dye to two third sea water ratio. The LED lights each go off at separate times through their respective red and blue filters. The light sensor then measures luminous intensity and we will utilize our calibration done beforehand to relate this to get a pH measurement. This sensor, unlike the others cited, can be used in situ and is completely natural. The sensor was designed to be moveable and take samples straight from the water beneath it without harming the environment.

The team aimed to create a sensor that provides pH data. We created a working circuit that incorporates all of these sensors and gathers data that we can to get pH measurements. If the sensor can tell the differences of waters with pH differences then it is a success. If the pH is more acidic in the bay compared to closer to the ocean then our null hypothesis is proved wrong.

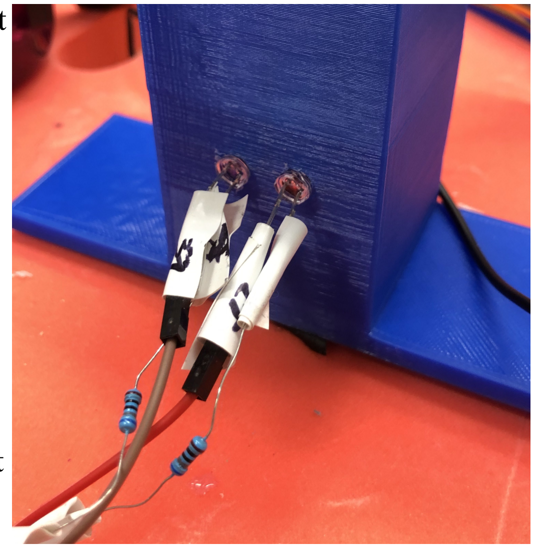
Instrument design

Our original thoughts about the design of the project were based on Dr Seroy's project where they fill a cuvette with a non toxic dye and some water of a certain pH. They then put the cuvette in a housing much like

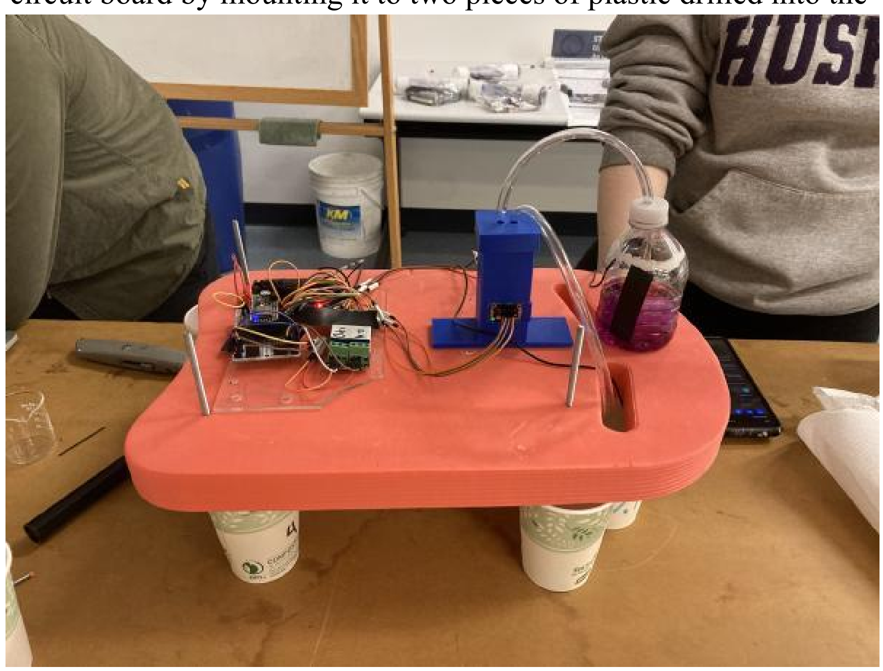




our own with two LEDs and a light sensor. For our project we had to make some simple adjustments to the Size of our cuvette holder and created a lid that limited outside light and allowed us to pump water and dye into the cuvette. Post printing of the CAD design we had to drill holes in the back of the cuvette housing for the lights.



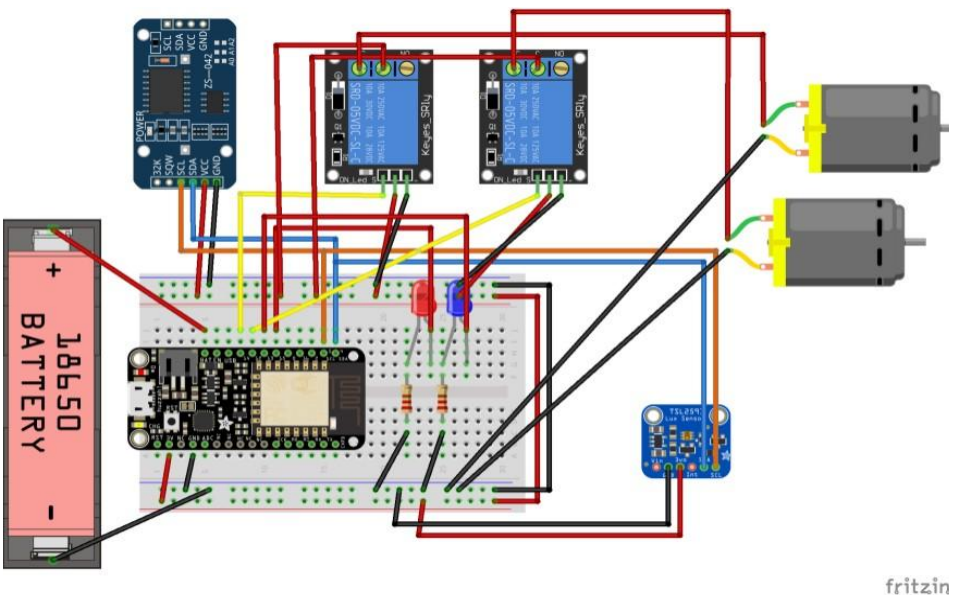
Once we had the cuvette holder we tested different methods of draining the solution. Using different sized holes through the kickboard we concluded that a 1/16th whole allowed the solution of dye and water to leak out just slow enough for us to get a light reading. We then realized that if we tried to make the same size whole through the cuvette base then our cuvette housing would flood and ruin our light sensor. We came to the conclusion that we had to cut the bottom of it out and stick the cuvette directly into the kickboard, to avoid leaking. In the case of our cuvette housing linking we had to protect our circuit board by mounting it to two pieces of plastic drilled into the kickboard. This elevated the circuit board and would protect it in the case of our cuvette housing leaking. Once we knew the size of our cuvette holder and figured out how our circuit board was going to fit on the kick board, we then needed



to find a cover for the entire sensor. Using a plastic bin we were able to create a water resistant housing that covered the circuit board and the cuvette housing.

We then had to adjust the location of our water and dye pumps. There were pre-cut holes in the original design of the kick boar to which we put our water pump through so that it could dangle in the water. As for the dye pump, we taped the reservoir to the kickboard on the outside of the housing due to a lack of space inside the housing. By cutting holes into the top of our plastic cover we fed the water and dye tubes through the cover and then into the CAD designed whole in the top of our cuvette housing.

The fritzing diagram consists of two water pumps, two water pump electrical switches that trigger the water pumps, a microcontroller, energy source/battery, two LEDs (white in the end), light sensor, and an external RTC.



Implementation

The sampling scheme of the instrument involves multiple steps. To receive one sample from the environment, it will take about 52.5 seconds for the instrument to run one cycle. For better and more accurate results, two more samples should be taken. All three samples will take about 107.5 seconds overall to measure, not including the draining of the cuvette of the last sample back into the environment. If we were to deploy the instrument over a period of time, our sensor would sample once per day at least, or up to four times per day in synch with the tidal movement. The composition of water does not change frequently over the period of a one day cycle, but in waters that are affected by tidal movement can bring in different water compositions to the sensor. Measuring up to four cycles of three samples each per day will give the best picture of pH compositions of different bodies of water.

Depending on the amount of cycles that are measured by the instrument, the sensor could be deployed up to ten days before needing to be received once more at shore. If the instrument is to cycle only once per day, the sensor can stay out to a maximum of ten days deployed in the water. If the instrument is to cycle four times per day, the sensor can only stay deployed for a maximum of two days. These cycle limitations come from our largest limiter of the container that holds our indicator. The container is constructed by a 20 fluid oz. plastic water bottle that has been cut at the top for the placement of the pump for the indicator. The top 1.5 inches of another 20 fluid oz. plastic bottle is cut and placed on top of the original plastic bottle. Around 6 ml of cabbage juice indicator is pumped through the pump and into the cuvette each sample which means overall, the pump can only run around 30 times before air bubbles appear in the pump, which can skew our absorbance output from the lights in the cuvette.

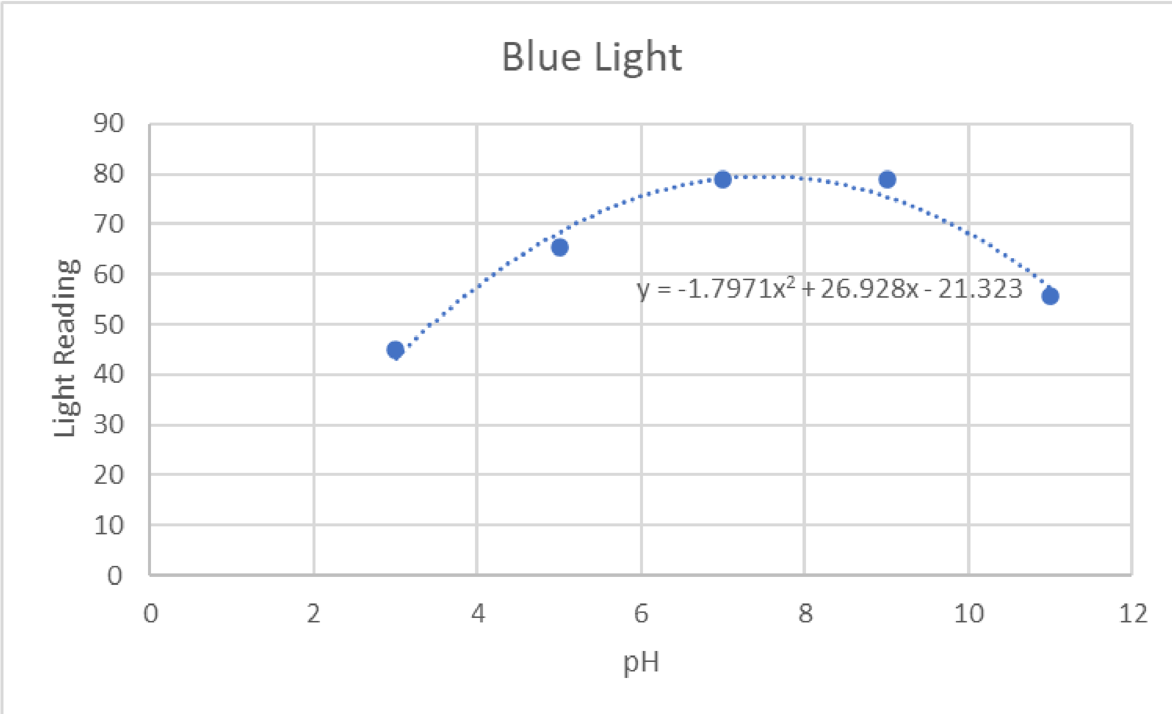
If we were to have a larger container that works with the instrument constructed, then the limitations of sampling would fall onto our power supply. Currently we have a battery that produces 9900 mAh (3.7 volts), and with this size battery, we could gather 128,432 samples when fully charged. The memory of the microcontroller within our system can hold significantly more samples, making the power source the second limiter within our system.

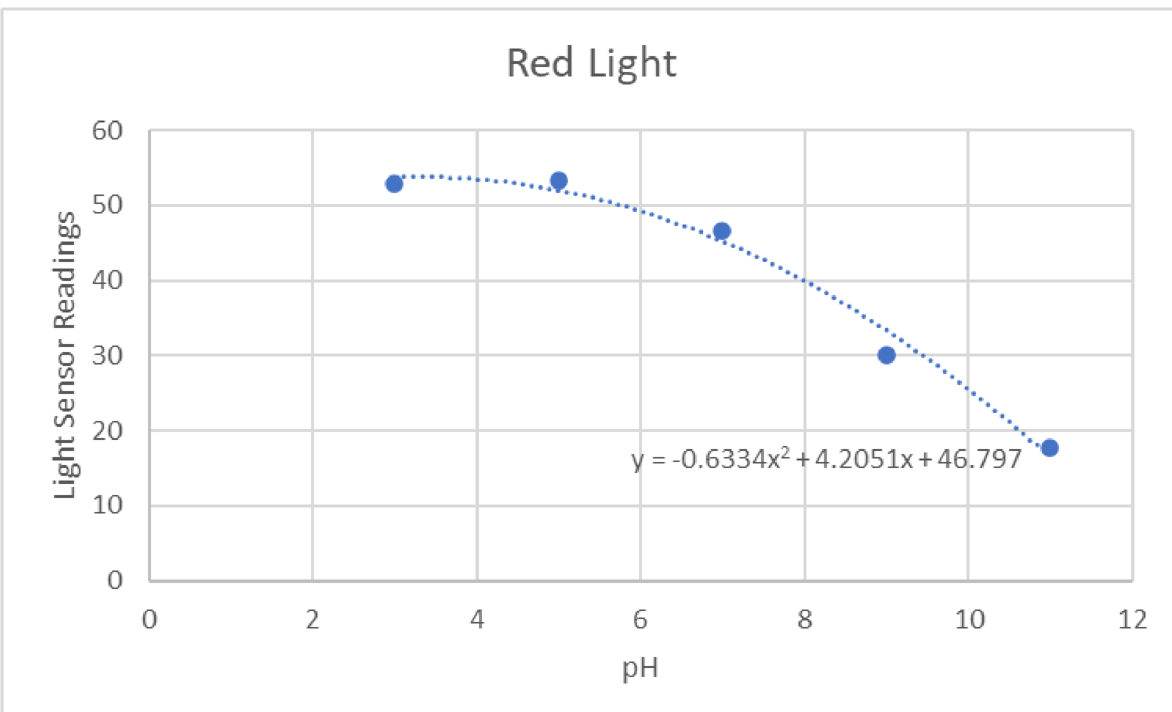
The deployment locations that we chose were Shilshole Marina and Portage Bay. Both of these locations hold brackish water with Shilshole holding saltier brackish water and Portage Bay holding fresher brackish water. Both have concentrations of salt and fresh water within them, making them both brackish. Shilshole is connected to Puget Sound, which is a salt water body connected to the Pacific ocean with freshwater outputs around the perimeter. Portage Bay is part of the connecting channel of water between Lake Union and Lake Washington, both freshwater lakes. With the Ballard Locks that lets ships travel to and from Lake Union to Puget Sound, there is salt water within Lake Union and into the channel where Portage Bay stands. This concentration would be significantly less that what we would see at Shilshole Marina, but still making the water brackish. Both Shilshole Marina and Portage Bay receive runoff from human interference that can also affect the brackish levels of each body of water.

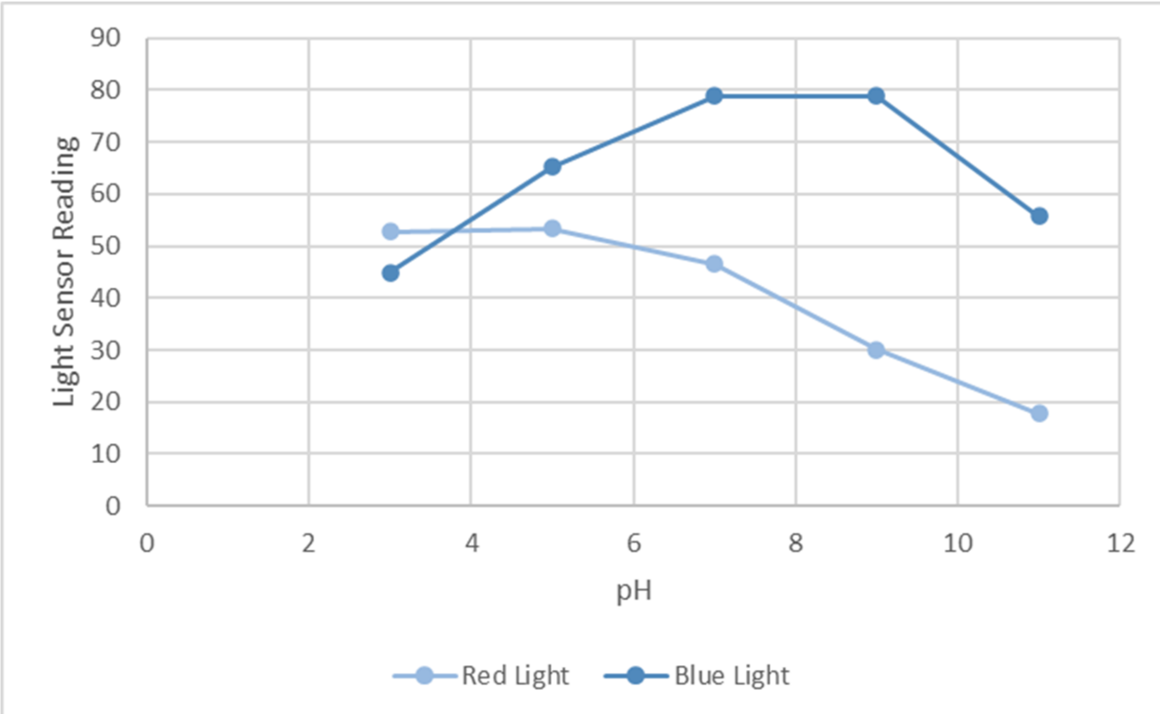
We chose these bodies of water to look at the differences in pH levels between saltier water and fresher water. We were unable to go to Shilshole Marina because of time constraints but we were successful in deploying into Portage bay.

Deliverables

We were able to get light readings from both lights, red and blue, and calibrate the readings to make a calibration curve for each light. We used the pre-made pH solutions of 3,5,7,9,11 to get light readings in order to calibrate the sensor. The graphs below are our calibration curves.


Blue Light
Light Reading
90
80
70
60
50
40
30
20
10
0
0
2
4
6
8
10
12
pH
y = -1.7971x² + 26.928x - 21.323


Red Light
Light Sensor Readings
60
50
40
30
20
10
0
0
2
4
6
8
10
12
pH
y = -0.6334x² + 4.2051x + 46.797



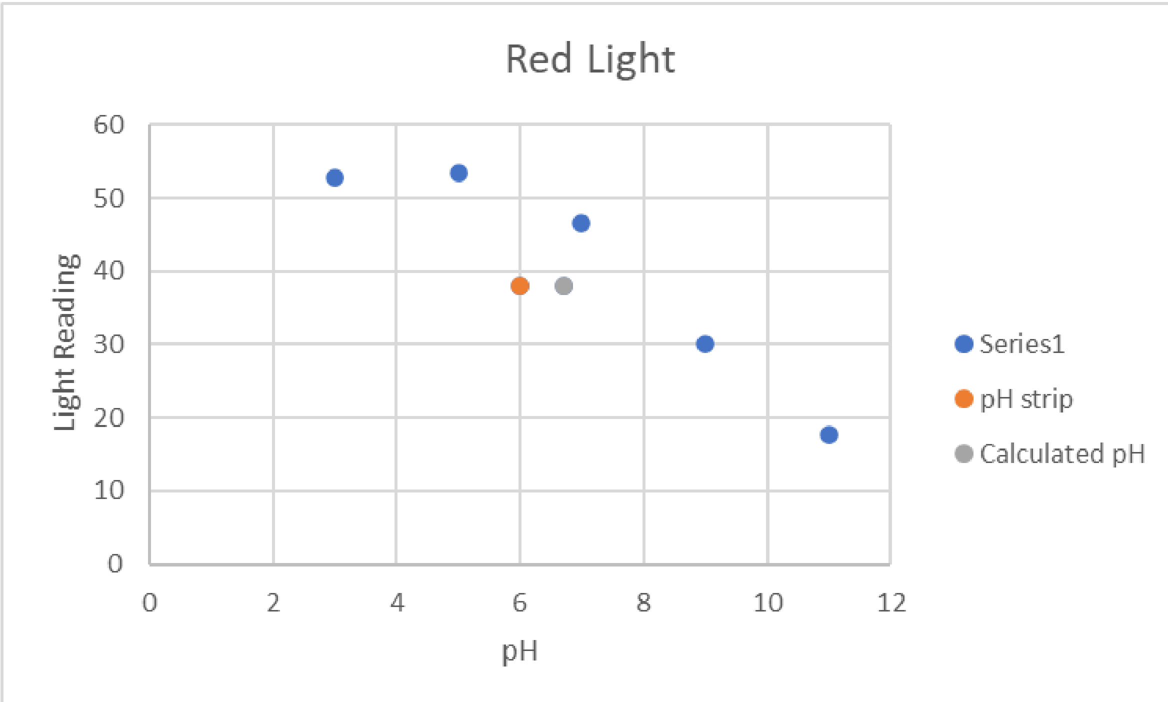
1	Time	Raw Data(	Raw Data(	Luminosity		
2	2021-12-1	12	0	48.96	Blue	3
3	2021-12-1	19	3	57.4464	Red	3
4	2021-12-1	12	0	48.96	Blue	3
5	2021-12-1	20	4	54.8352	Red	3
6	2021-12-1	11	0	44.88	Blue	3
7	2021-12-1	19	4	50.7552	Red	3
8	2021-12-1	15	0	61.2	Blue	5
9	2021-12-1	18	3	53.3664	Red	5
10	2021-12-1	17	0	69.36	Blue	5
11	2021-12-1	18	3	53.3664	Red	5
12	2021-12-1	16	0	65.28	Blue	5
13	2021-12-1	18	3	53.3664	Red	5
14	2021-12-1	19	0	77.52	Blue	7
15	2021-12-1	17	3	49.2864	Red	7
16	2021-12-1	20	0	81.6	Blue	7
17	2021-12-1	16	3	45.2064	Red	7
18	2021-12-1	19	0	77.52	Blue	7
19	2021-12-1	16	3	45.2064	Red	7
20	2021-12-1	19	0	77.52	Blue	9
21	2021-12-1	11	2	31.4976	Red	9
22	2021-12-1	19	0	77.52	Blue	9
23	2021-12-1	11	2	31.4976	Red	9
24	2021-12-1	20	0	81.6	Blue	9
25	2021-12-1	10	2	27.4176	Red	9
26	2021-12-1	14	0	57.12	Blue	11
27	2021-12-1	9	2	23.3376	Red	11
28	2021-12-1	13	0	53.04	Blue	11
29	2021-12-1	6	1	17.7888	Red	11
30	2021-12-1	14	0	57.12	Blue	11
31	2021-12-1	3	0	12.24	Red	11
32	2021-12-1	18	0	73.44	Blue	Seawater
33	2021-12-1	14	3	37.0464	Red	Seawater
34	2021-12-1	16	0	65.28	Blue	Seawater
35	2021-12-1	20	4	54.8352	Red	Seawater
36	2021-12-1	17	0	69.36	Blue	Seawater
37	2021-12-1	7	1	21.8688	Red	Seawater

The data above was from our bench test when we calibrated the sensor and measured the pH of a pre-made seawater sample. The far-right column is the pH or the type of sample, the next one to the left tells us which light the reading was from, and the next one to the left gives us the actual light reading.

2021-12-6	21	1	78.9888	Blue
2021-12-6	21	4	58.9152	Red
2021-12-6	20	1	74.9088	Blue
2021-12-6	22	5	56.304	Red
2021-12-6	19	1	70.8288	Blue
2021-12-6	22	5	56.304	Red

The data above is from our field deployment. The light readings for the red light were not on our calibration curve, so we used the data from the run before. The previous run only took one reading per light, so we used one value rather than an average of three. We believe that the pump's tubes were pushed too far into the chamber, which caused the liquid to overflow as the tubes took up more volume.

We found the standard deviation of the red light to be 13.473 and blue 3.331. We can see in the graphs below how the blue light was more accurate when the pH values we measured via the sensor and pH strips were plotted on the calibration graphs. The pH strip and sensor values are from the pre-made seawater sample we took. The pH test strip's accuracy was roughly+/- .1, which was important because we based our expected pH values on the strips.



Our results gave us calibration curves that were similar to those in the papers we read and what we originally calibrated using the light kit. We were able to get different values between the lights for different pH samples. Our deployment in Portage Bay was successful, as was the bench test. In the end, we were able to collect light readings for both red and blue lights for the pH's 3,5,7,9,11, pre-made seawater, and water from portage bay.

Water sample	pH measured by sensor	pH measured by strips
Bench test pre-made	6.7	6
Deployment in Portage Bay	6.3	7

Conclusions and Design Improvements

Our final output was a bench-tested and calibrated pH sensor that could detect changes in pH to precision of about 0.7, which was calculated from data and knowledge of pH strip precision. However, we did not collect field data from all the locations we wanted to in order to disprove our null hypothesis. We collected data from Portage Bay, our fresher test location, but not Shilshole Marina, our saltier deployment location. So, we would have to test our design in our second field location and collect real data to prove or disprove our original hypothesis.

Our current system could use improvements in waterproofing, weather durability, and accuracy to make it a practical and reliable instrument. It would not be able to withstand deployments unmonitored by humans due to its fragility and lack of waterproofing, however, it is capable of procuring data samples without direct human instruction, so with the proper batteries, a larger dye reservoir, and waterproofing it could potentially be field deployable for a prolonged period of time. This could be achieved by potentially making the buoyant body larger and more stable using weights, waterproofing the boundaries between our cover and base, waterproofing the boundary between the cuvette and base, and sealing off the electronics in a completely waterproof location rather than just a raised plate above the kickboard. Having a larger body would also allow us to have a larger dye reservoir, which would enable us to leave the sensor in the environment for a longer period of time. We could also make our base more durable by using a material other than foam, which would also be more environmentally friendly because it won't degrade into microplastics.

Some sources of error for our sensor were that we didn't rinse our cuvette between readings, so there may have been contamination between samples. Although it wouldn't be a large change because all of the measurements were taken in the same location, if we were to change locations it might have an impact, so changing our code to run water through the cuvette prior to taking each measurement would help solve this problem. We did also have some light leak in from the environment due to our pump tubes and LEDs being translucent, so having black tubes and sealing off the backs of our LEDs with paint or tape would create a darker chamber to give us more accurate readings. The dye we used could have also been a source of error because it does not contain any preservatives and therefore degrades quicker than a dye that is more harmful to the environment, so there may have been some natural change in the color of the dye between when we calibrated it and when we actually took measurements with it.

A more accurate light sensor would also help us take more accurate measurements because we would be able to calibrate our dye more precisely, which would then give us more accurate pH readings and create a better sensor overall. If we could get a standard pH sensor that doesn't rely on a calibration curve or light measurement proxy, we would be able to bypass that problem in the first place, however, many commercially available pH sensors have quirks like the value for the pH’s drifting and changing over time and not aligning with their calibrations. Others have operational inconveniences like needing to stay in one position over time and any movement could affect the pH outputs. These quicks and inconveniences would potentially make them also difficult to deploy and leave unaccompanied for long periods of time.

Overall, we successfully designed, built, and deployed our very own pH sensor that measured the pH of water samples to a precision of 0.7 units. Although we did not deploy in all of the locations we wanted to, and therefore could not prove or disprove our null hypothesis, we can continue to build upon and improve our current design to be able to deploy in more locations in the future with a higher level of accuracy and durability.

References

Carter, B. R., J. A. Radich, H. L. Doyle, and A. G. Dickson. 2013. An automated system for spectrophotometric seawater pH measurements: Automated spectrophotometric pH measurement. Limnol. Oceanogr. Methods **11**: 16–27. doi:10.4319/lom.2013.11.16

Richard A. Feely, Simone R. Alin, Jan Newton, Christopher L. Sabine, Mark Warner, Allan Devol, Christopher Krembs, Carol Maloy (2010) The combined effects of ocean acidification, mixing, and respiration on pH and carbonate saturation in an urbanized estuary, Estuarine, Coastal and Shelf Science, Volume 88, Issue 4, 2010, Pages 442-449, ISSN 0272-7714, https://doi.org/10.1016/j.ecss.2010.05.004.

Sasha K. Seroy, Hanis Zulmuthi & Daniel Grünbaum (2020) Connecting chemistry concepts with environmental context using student-built pH sensors, Journal of Geoscience Education, 68:4, 334-344, DOI: 10.1080/10899995.2019.1702868

Yang B., M.C. Patsavas, R.H. Byrne, M. Jian (2014) Seawater pH measurements in the field: A DIY photometer with 0.01 unit pH accuracy, Marine Chemistry, Volume 160, 2014, Pages 75-81, ISSN 0304-4203, https://doi.org/10.1016/j.marchem.2014.01.005.

Appendix

User guide

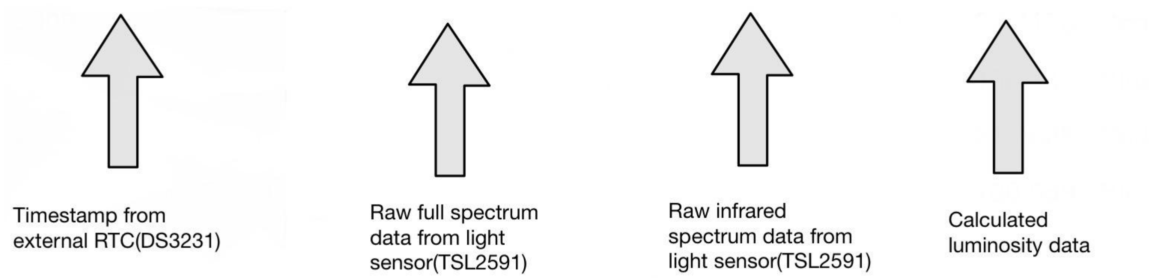
- Deployment
  - Refill dye reservoir and power on the sensor.
  - Place the instrument in a targeted water body.
  - Make sure the water pump is completely submerged in the water.
- Operation
  - Connect to the microcontroller via wireless access point.
  - Load files needed for operation with “import pH”.
  - Start testing cycle with “pH.start(number of consecutive testrun)”.

```
# Sample Commands
import pH # load file for running test cycle
pH.start(1) # start testing cycle and run the pH test once
```

Explanation of data structure output by device/system

Data from each consecutive testrun will be saved to an onboard file named “pH_data.csv”. Raw inferred, raw visible spectrum data and calculated luminosity data will be saved.

Time	Raw Data(Full Spectrum)	Raw Data(Infrared)	Luminosity	
2021-12-1-17-41-42	15	3	41.1264	Red
2021-12-1-17-41-44	29	0	118.32	Blue
2021-12-1-17-43-7	14	3	37.0464	Red
2021-12-1-17-43-9	32	0	130.56	Blue
2021-12-1-17-44-59	4	1	9.6288	Red
2021-12-1-17-45-1	23	0	93.84	Blue



Place to access code

Code and electrical design were added to a public Github repository and accessible to everyone. (https://github.com/CNA0086/cabbage-juice-pH-tester)

Other specs

Parts List with ID and URLS for parts (but not cost)

3V relay electrical switch (2)
3V submersible pump (2)
White LED (2)
Plastic colored filters (Red & Blue)
Light sensor TSL2591
Rubber tubes
3D printed housing
3D printed reservoir for dye
Buoyant platform
ESP8266 Microcontroller
External RTC DS3231