Student Cohort for Undergraduate (Marine) Bioscience Abroad

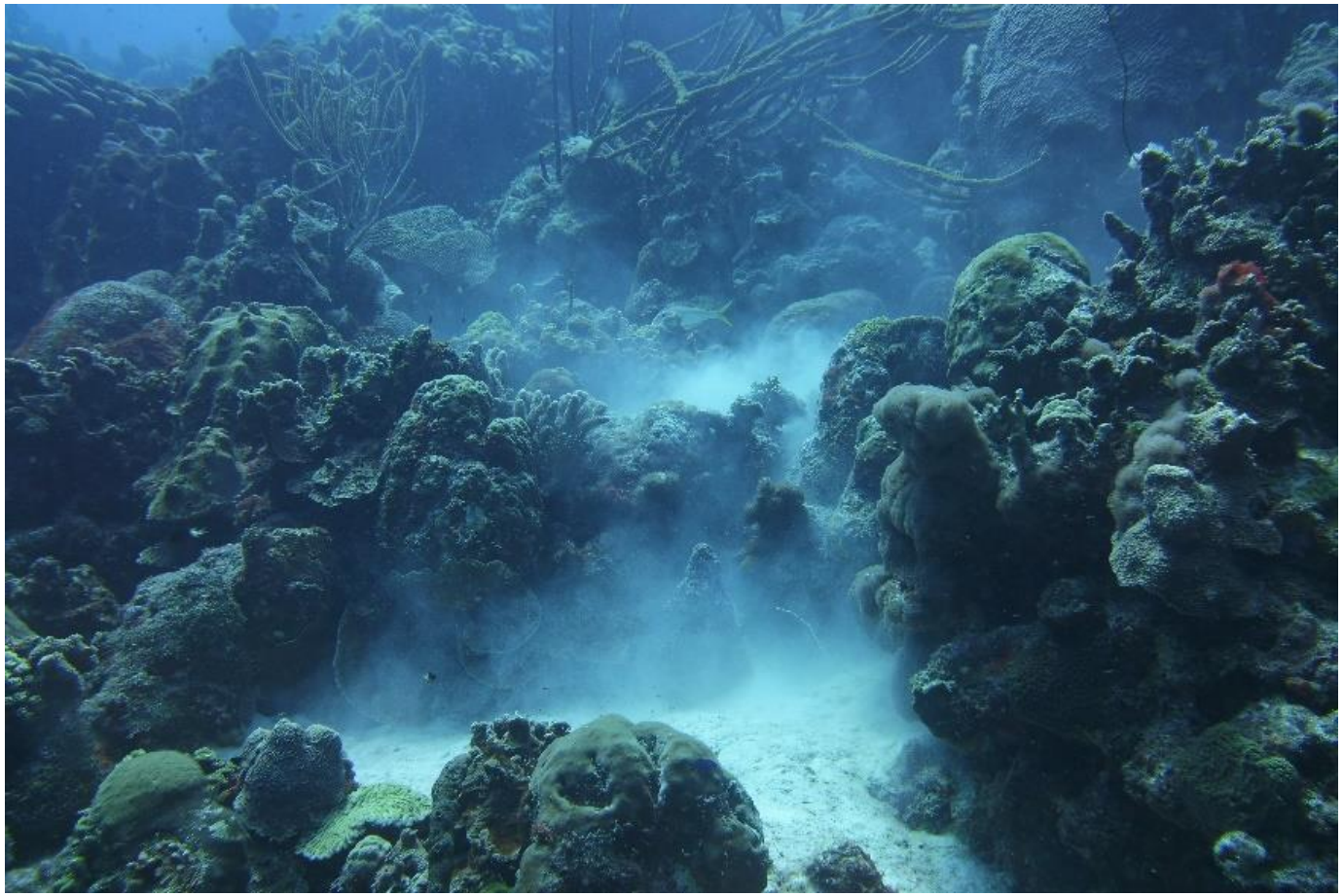
Midland College SCUBA Cohort

UT System LSAMP Conference

August 2, 2022

Midland College

Student Cohort for Undergraduate (marine) Bioscience Abroad (SCUBA)



Mission:

1) Provide undergraduate students an *international* field research experience in the Caribbean Sea
2) 2-year community college students targeted, but the opportunity is not exclusive to that group.
3) Research projects focused on aspects of coral reef ecosystems in the Caribbean Sea.

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Motivations

- We want to get students “in the water” to do science in a foreign country

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Motivations

- Coral reefs are of the densest locations of biodiversity on the planet
  - All coral reefs occupy just 0.5% of the ocean seafloor, but provide a home for up to 50% of all ocean life
- All around the world in clear, shallow (~20 ft to 150 ft), warm waters

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Coral Reefs Face Several Challenges

Natural Threats:

Oceanic tectonic shifts

Underwater volcanic activity

} El Nino / La Nina events

↓ Severe weather - Hurricanes

Coral Disease

Algae Blooms

↑

Human Activity Threats:

Pollution / Contributions to climate change

Overfishing, leading to a decrease in algae grazers

We wish to fully understand the short and long-term impacts that these threats produce on coral reef ecosystems and the mechanisms involved with these impacts

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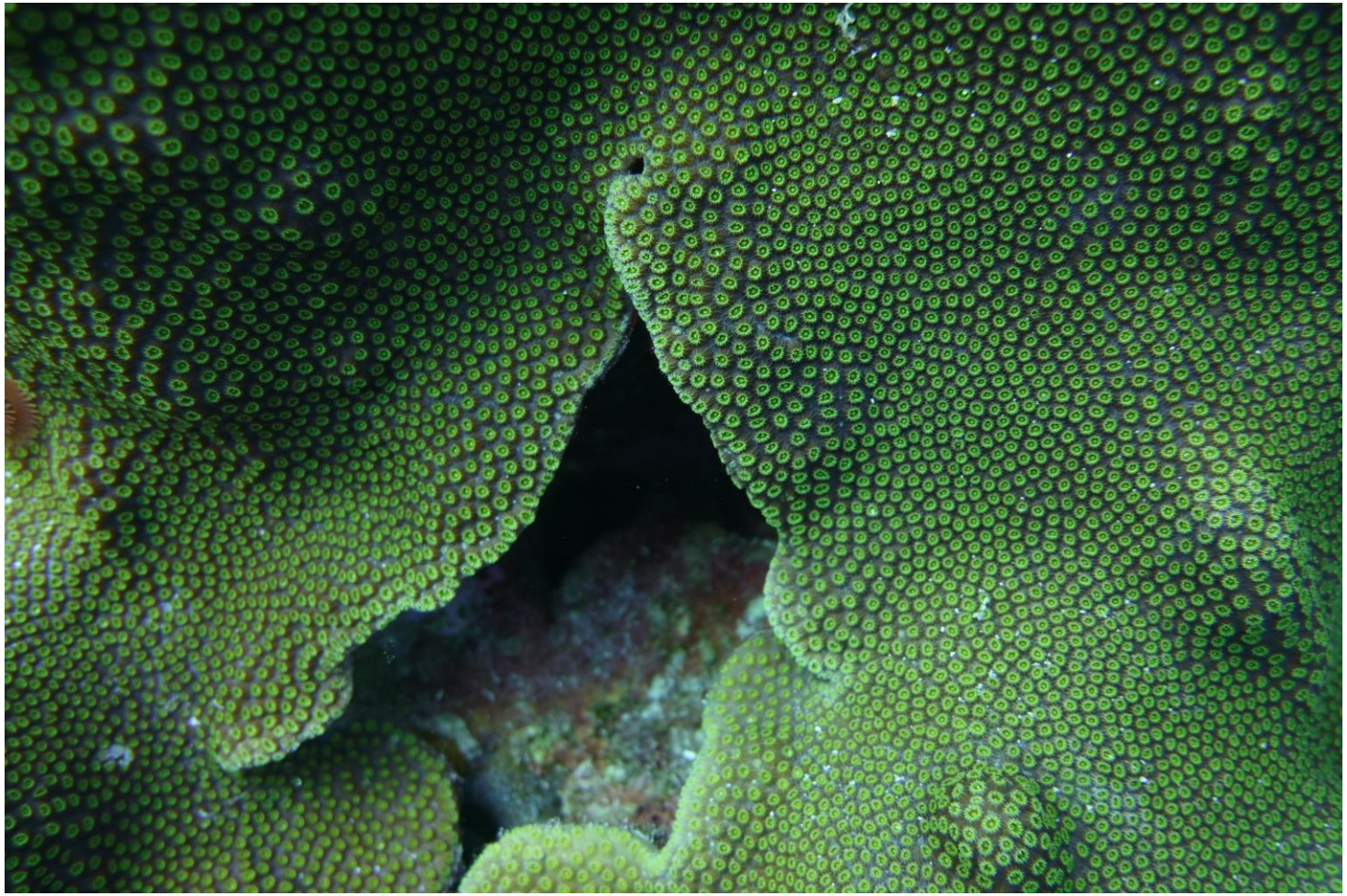
What you want to see is this:



- Lots of healthy coral
- This is a particularly great example of *Diploria strignosa*

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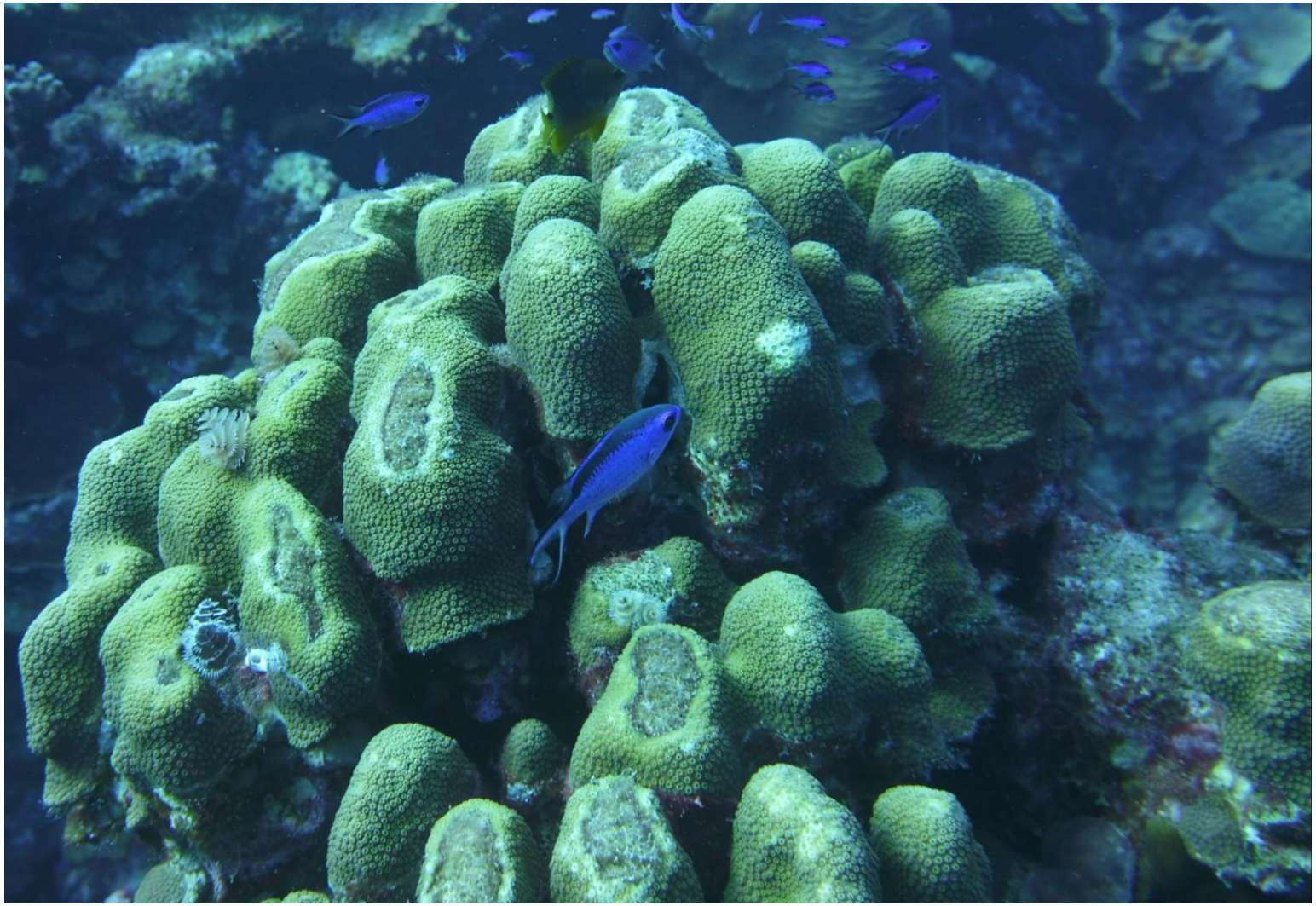
What you DON'T want to see is this:

- Diseased coral

- An example of Yellow Band Disease on an *Orbicilla anularis*



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2022 LSAMP SCUBA Student Cohort

Jennifer Hunt	Tyler Junior College
Michael Mangan	Midland College
Justin McKinney	Midland College
Jordyn Ricks	Midland College
Shaquila Sarapao	Midland College

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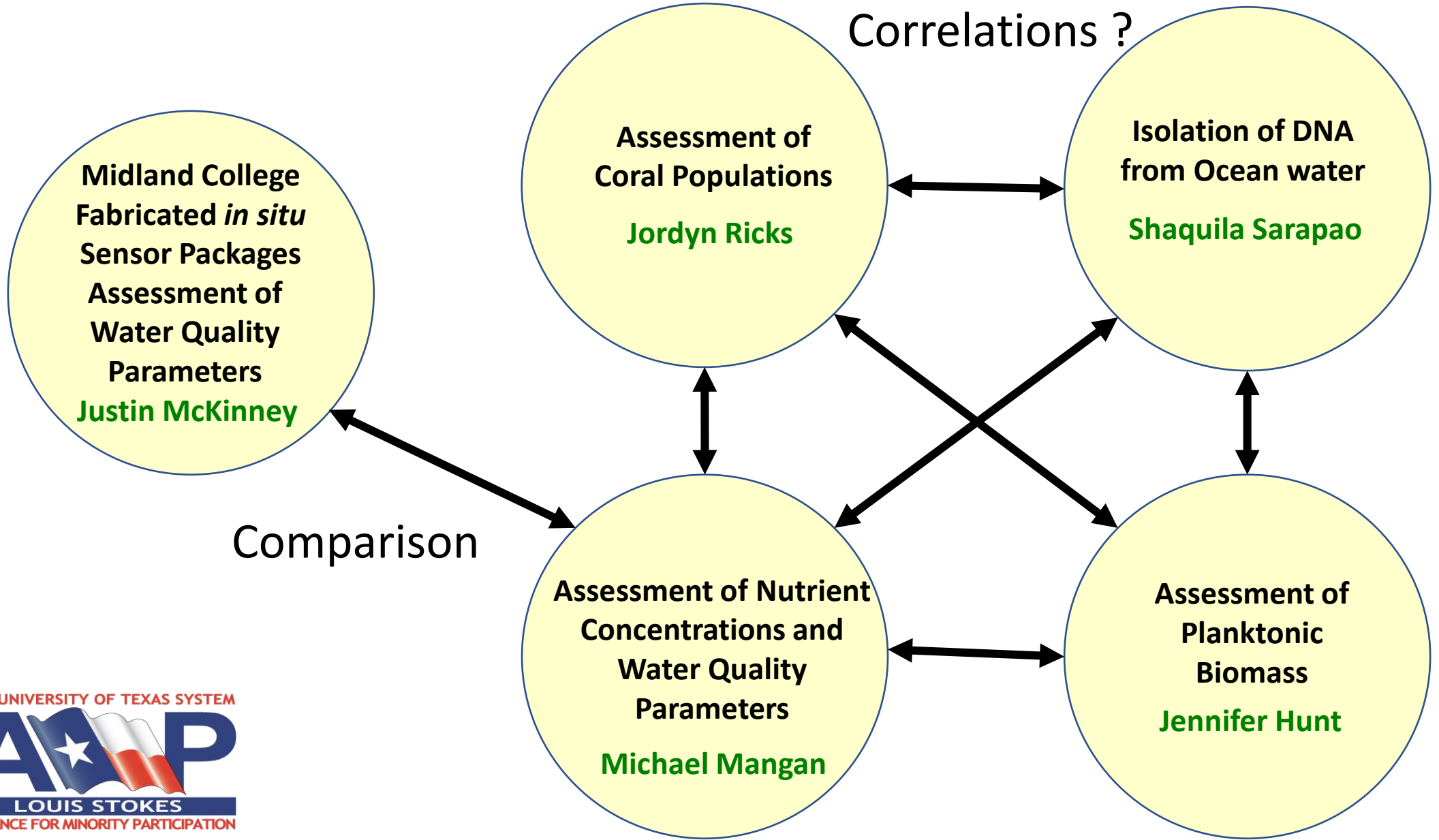
The Projects



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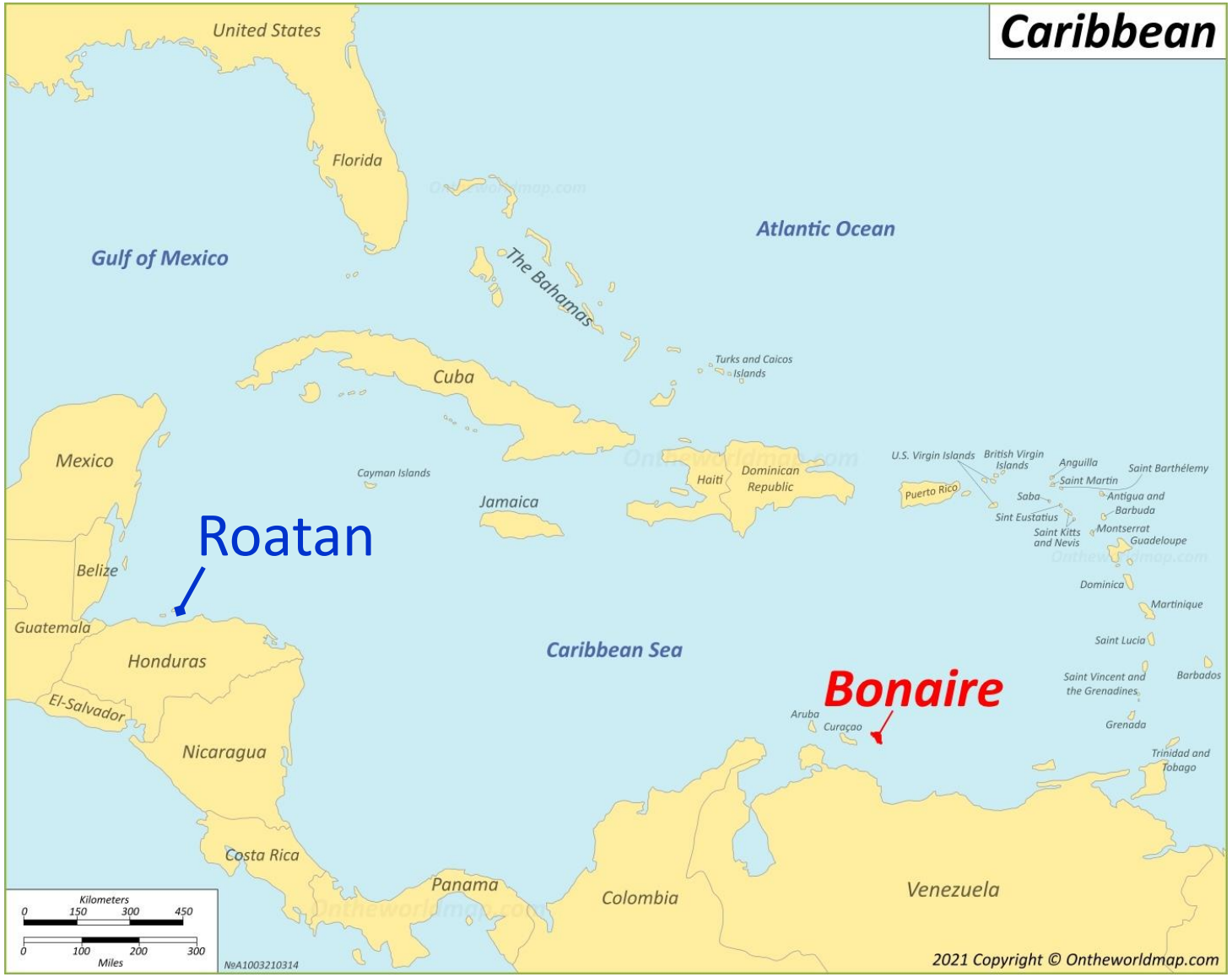
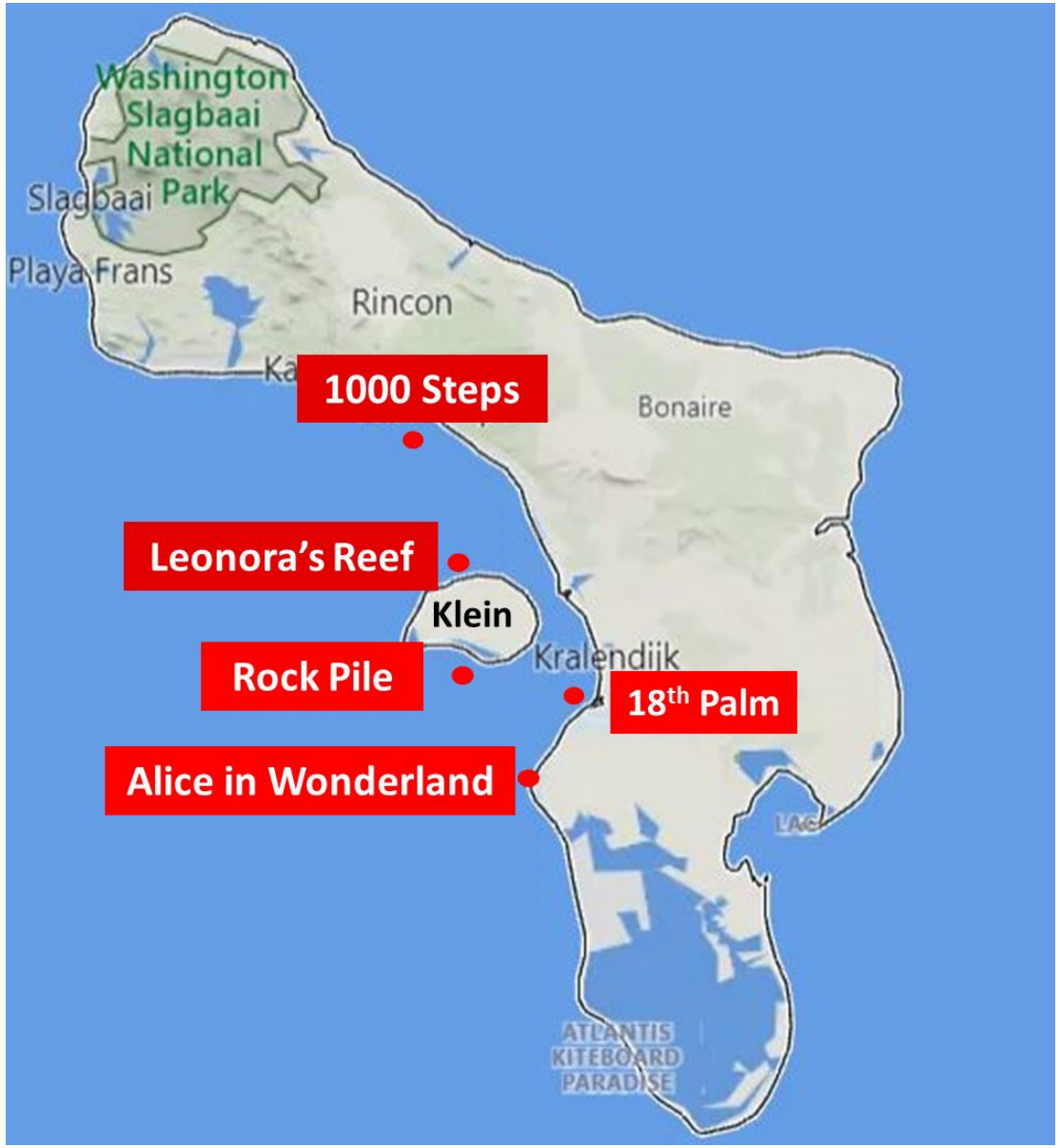
SCUBA Research Locations

- In prior years we have conducted research at Roatan, Honduras for a *longitudinal* study
- This year we have conducted research at Bonaire (Netherlands) for a *comparative* study



Bonaire Dive Sites

- Sample acquisition occurred at five dive sites
  - Rock Pile
  - Leonora's Reef
  - 1000 Steps
  - 18th Palm
  - Alice in Wonderland
- Sites were chosen to provide a geographic and developmental variety



Assessment of Coral Populations at Bonaire June 19 – July 1 2022

Jordyn Ricks

Midland College

Motivations and Current Techniques

In order to assess the overall health of a coral reef, we need to have an estimate of the coral population there

There are several accepted methodologies to survey a coral reef populations. We elected to utilize a transect belt method:

- 100 ft tape measure is laid out
- Photographers take pictures at 1 m intervals on either side of the tape measure
- Photographs uploaded to CoralNet Software

Image 2: Two Divers Use Transect Belt

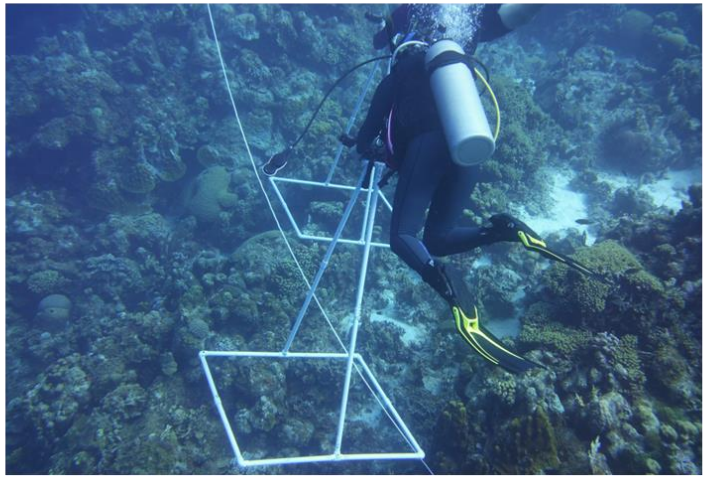
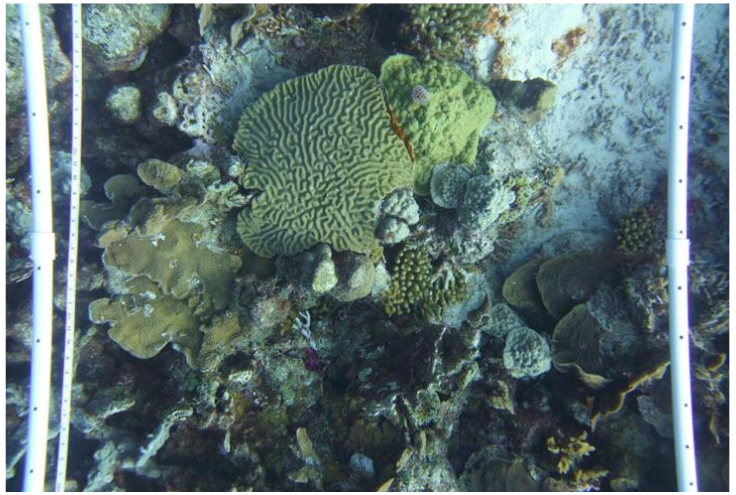


Image 3: Photograph Taken Using Transect Belt



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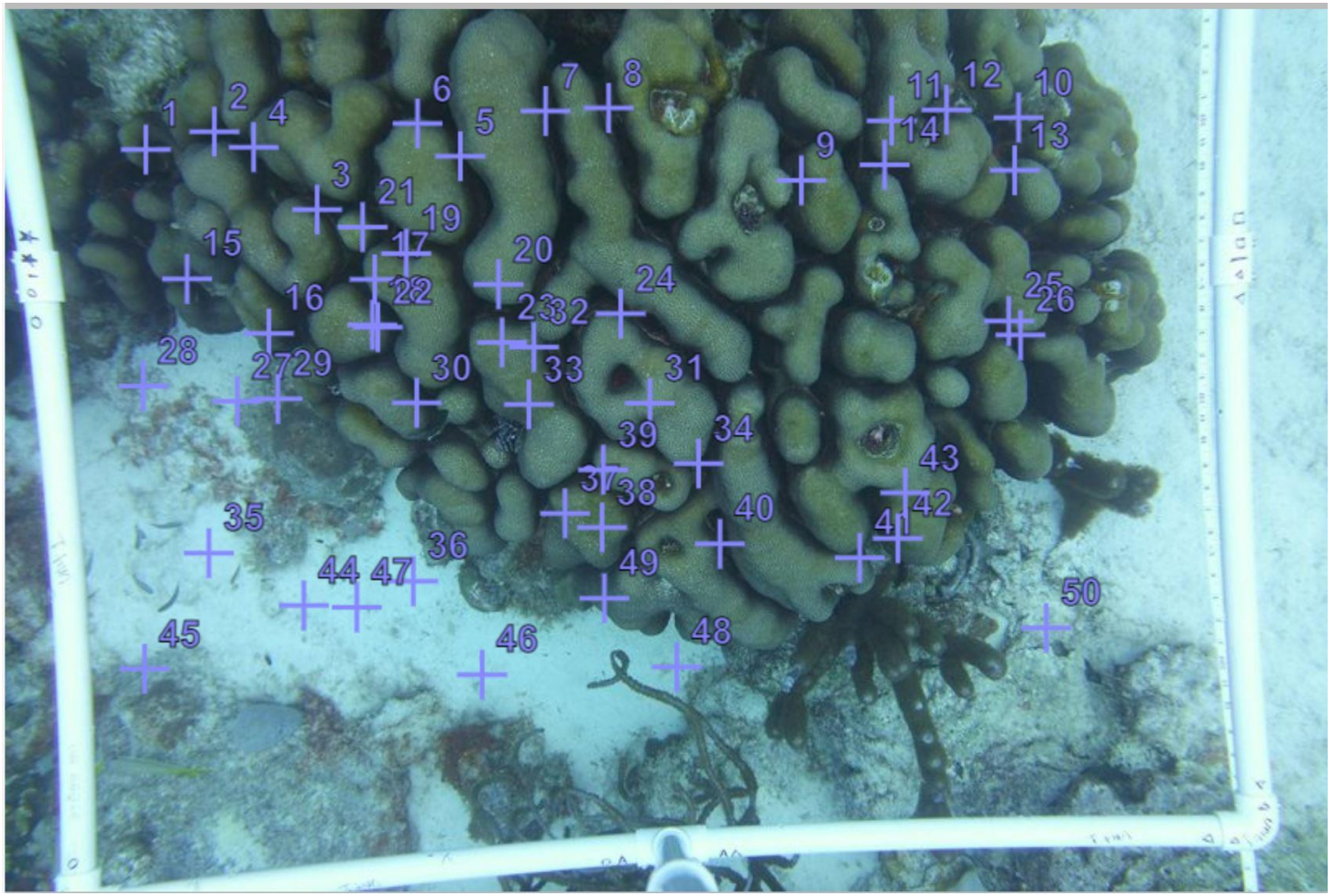
CoralNET

- CoralNet Software is an annotation program which randomly selects 50 point on the photograph
- The annotator identifies what coral species lies under the selected point
- After 25 photos have been annotated CoralNet uses machine learning to annotate the remaining photos in the set



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Results

Chart 6: Percentage of Coral Coverage at All Locations and Overall

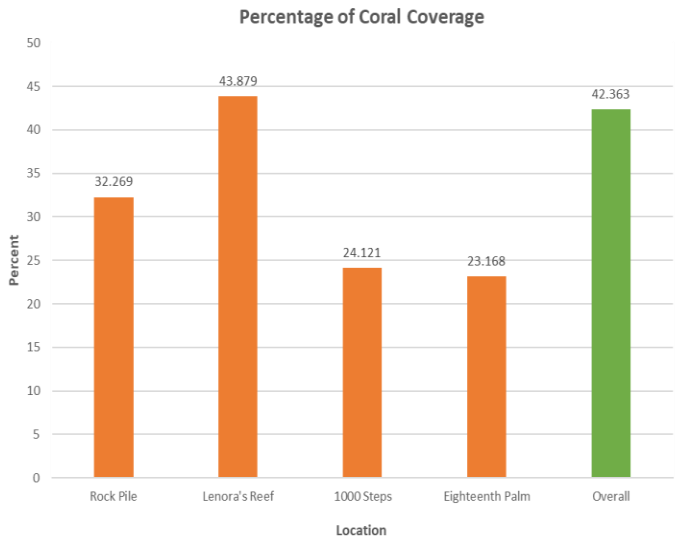


Chart 7: Percentage of Live Coral at All Locations and Overall

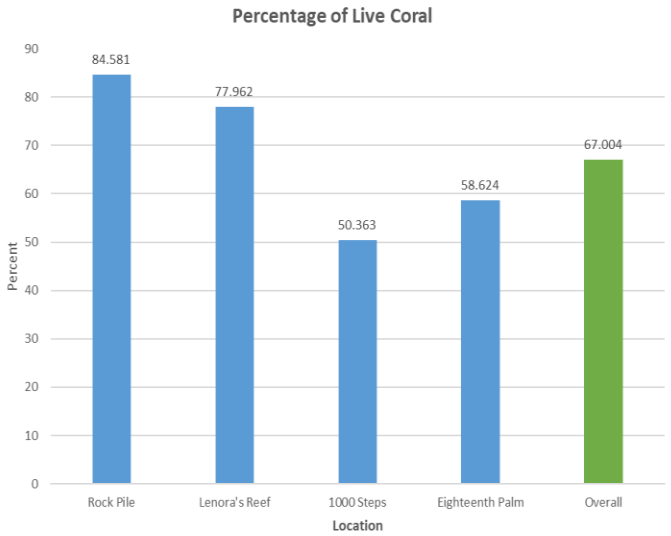


Chart 1: Individual Coral Populations from Rock Pile

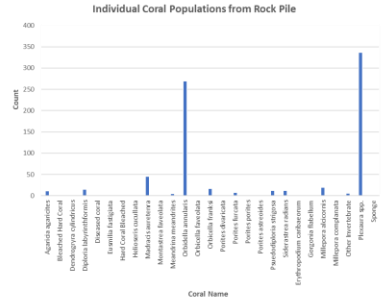


Chart 2: Individual Coral Populations from Lenora's Reef

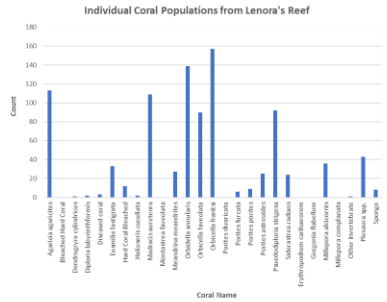


Chart 3: Individual Coral Populations from 1000 Steps

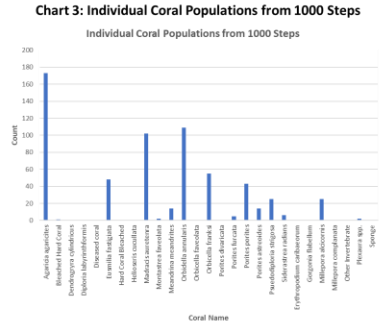


Chart 4: Individual Coral Populations from Eighteenth Palm

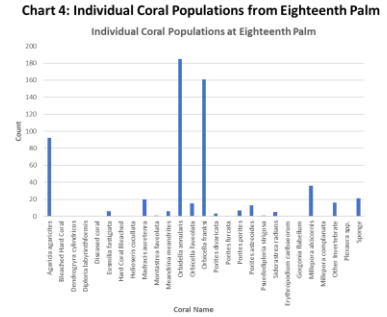
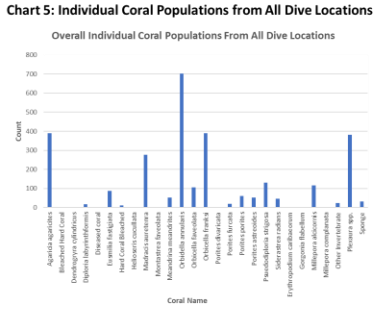


Chart 5: Individual Coral Populations from All Dive Locations



Conclusions

Conclusion

Over the four sites surveyed, the overall coverage of live coral was 42% with the percentage of live coral being 67%. The dive sites located off the shores of Klein Bonaire, Rock Pile and Lenora's Reef, showed higher rates of live coral as well as more overall coral coverage compared to the dives sites off the shores of the mainland, 1000 Steps and Eighteenth Palm, presumably because this area is heavily protected by the National Park Foundation, STINAPA. Lenora's Reef displayed the most phenotype diversity among coral species as well as the highest rate of living coral.

Assessment of Planktonic Biomass at Bonaire June 19 – July 1 2022

Jennifer Hunt

Midland College

Methodology

We wanted to compare two different methods of biomass assessments, both of which appear as accepted protocols in the literature.

Field Method

Commercial Laboratory Method
A&B Laboratories (Houston, Texas)

Ash-free, oven dried biomass **Total Carbon** = Total Inorganic Carbon (TIC) + Total Organic Carbon (TOC)

measured ↓ measured ↓

Reliable and Accurate field measurements are desirable for several reasons:

(relatively) Low cost method

Time limitation on sample integrity (can't ship via boat because it takes weeks)

Weight limitations on air transport of samples

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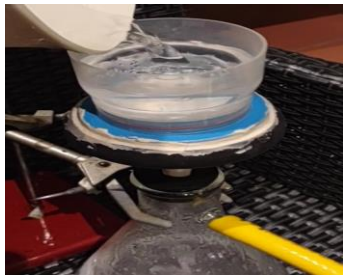
Protocol

(Constant mass) – (mass of Aluminum pan / filter disk) = **oven dried biomass**

Filter 6-gallon water sample on a glass-fiber filter disk using a vacuum pump, filter flask, and Buchner funnel



Heat solid residue on dry glass-fiber filter disk / Aluminum pan holder to 102 °C in a moisture analyzer until constant mass is achieved

Heat solid residue on dry glass-fiber filter disk / Aluminum pan holder to 550 °C in a furnace for 4 hours.

Reweigh residue/Aluminum pan/filter disk.

(mass of residue/Aluminum pan/filter disk) – (mass of Aluminum pan / filter disk) = **Ash**

Oven dried Biomass – Ash = Ash-Free oven dried biomass

Results

Rock Pile	Oven Dried Biomass (mg/L)	Ash-Free Oven Dried Biomass (mg/L)	Total Organic Carbon (mg/L)	Total Inorganic Carbon (mg/L)	Total Carbon (mg/L)
Surface	26.42	25.36			
20ft	38.92	36.9			
40ft	7.97	7.22			
60ft	24.66	24.26			

Leonora's Reef	Oven Dried Biomass (mg/L)	Ash-Free Oven Dried Biomass (mg/L)	Total Organic Carbon (mg/L)	Total Inorganic Carbon (mg/L)	Total Carbon (mg/L)
Surface	29.45	28.71			
20ft	23.25	22.76			
40ft	59.53	58.56			
60ft	23.42	22.8			

1,000 Steps	Oven Dried Biomass (mg/L)	Ash-Free Oven Dried Biomass (mg/L)	Total Organic Carbon (mg/L)	Total Inorganic Carbon (mg/L)	Total Carbon (mg/L)
Surface	31.35	30.47	1.2	22.7	23.9
20ft	135.96	121.83	1.2	21.2	22.4
40ft	73.88	71.85	1	22.5	23.5
60ft	26.24	25.49	BRL	24.3	24.3

18 Palms	Oven Dried Biomass (mg/L)	Ash-Free Oven Dried Biomass (mg/L)	Total Organic Carbon (mg/L)	Total Inorganic Carbon (mg/L)	Total Carbon (mg/L)
Surface	87.57	85.81	1.1	23.4	24.5
20ft	27.25	26.73	BRL	24.2	24.2
40ft	42.84	41.69	BRL	24.9	24.9
60ft	119.98	118.35	1	23.7	24.7

Alice & Wonderland	Oven Dried Biomass (mg/L)	Ash-Free Oven Dried Biomass (mg/L)	Total Organic Carbon (mg/L)	Total Inorganic Carbon (mg/L)	Total Carbon (mg/L)
Surface	139.26	138.56	1.1	24	25.1
20ft	24.52	24.22	1	24.3	25.3
40ft	46.49	45.83	1.2	23.7	24.9
60ft	97.83	95.28	1	24.1	25.1
		AVG = 68.86 mg/L			AVG = 24.4 mg/L

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Conclusions

- Although the two methods produce carbon measurements that are of the same order of magnitude, the ash-free oven dried biomass measurement was on average 2.8 times greater than the Total Carbon measurements
- The two methods failed the F-test, meaning that the two methods produce results that are not comparable
- Biomass measurements from both methods are consistent with those seen for healthy coral reef ecosystems

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Hunting for Pathogenic Bacteria in Ocean Waters at Bonaire June 19 – July 1 2022

Shaquila Sarapao

Midland College

Motivation

Studies revealed a consortium of bacteria are present in the interface of diseased corals, with cyanobacteria being a key player

This leads to a basic question:

How prolific are the cyanobacteria species implicated in coral disease in ocean water in general?

Protocol

1. Collect water samples
2. Concentrate samples with rapid bio-concentrator from InnovaPrep, concentrating one liter of each sample, eluting 200uL into 2mL tube

    Instrument used high-flow single-use 0.45 um hollow fiber filter tips with the "HOLLOW" protocol
3. The Qiagen DNAeasy biofilm kit and protocol were used to extract DNA from the water samples and were stored in a refrigerator until further applications
4. Upon return to the Midland College laboratory, PCR amplification was utilized to isolate marine cyanobacteria strains using the primers
5. Afterwards, the PCR products were sequenced using Thermo Fisher Scientific Invitrogen's 2% Agarose (GP) E-Gel with SYBR Safe™ and E-Gel Simple Runner. The sequences were compared to Thermo Scientific GeneRuler 1kb plus (0.5ug/uL, 50ug).

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Electrophoresis Results

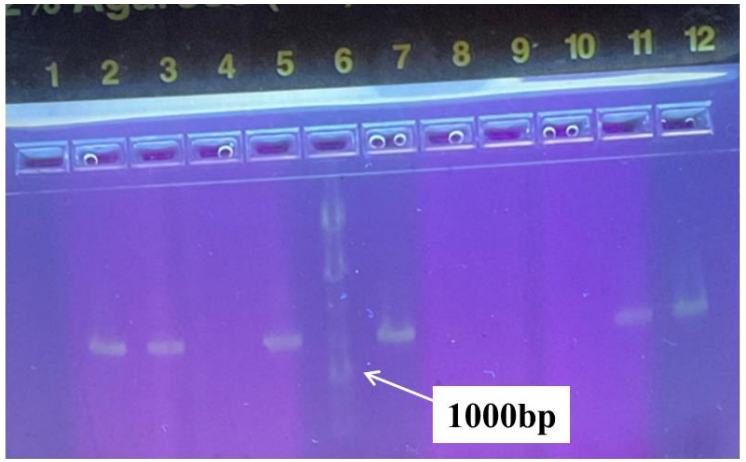

Figure 1. Gel electrophoresis results using primer 740F/1494R on samples 1-11 (left to right, column 6 being the gene ladder).

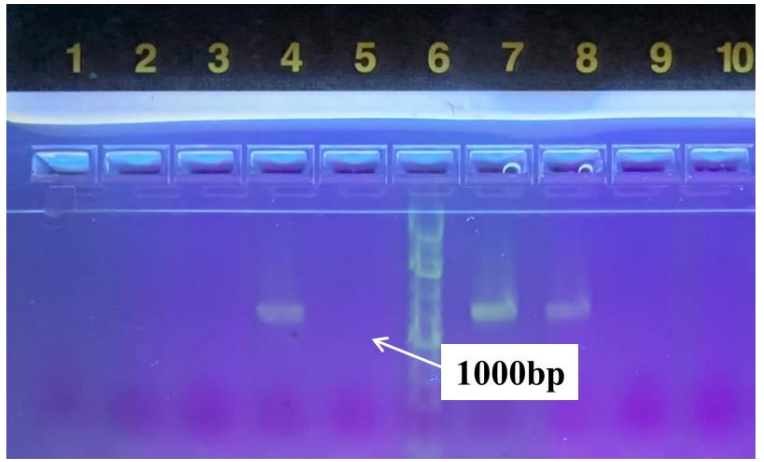

Figure 2. Gel electrophoresis results using primer 740F/1494R on samples 12-20 (left to right, column 6 being the gene ladder).

Sample	Location Depth (ft)	Primers 740F 1494R	Primers 27F 809R	Primers HEPF HEPR
1	Rock Pile 60	-	(+)	-
2	Rock Pile 40	(+)	-	-
3	Rock Pile 20	(+)	-	-
4	Rock Pile Surface	-	(+)	-
5	Leonora's Reef 60	(+)	-	-
6	Leonora's Reef 40	(+)	-	-
7	Leonora's Reef 20	-	-	-
8	Leonora's Reef Surface	-	-	-
9	A Thousand Steps 60	-	-	-
10	A Thousand Steps 40	(+)	(+)	-
11	A Thousand Steps 20	(+)	-	-
12	A Thousand Steps Surface	-	-	-
13	18th Palm 60	-	-	-
14	18th Palm 40	-	-	-
15	18th Palm 20	(+)	(+)	-
16	18th Palm Surface	-	-	-
17	Alice in Wonderland 60	(+)	-	-
18	Alice in Wonderland 40	(+)	-	-
19	Alice in Wonderland 20	-	-	-
20	Alice in Wonderland surface	-	-	-

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Conclusions

At present, the following conclusions can be made:

- Cyanobacteria is present at different depths in all of the coral reefs observed, indicating that the bacteria is widely distributed throughout the water column.
- The enzyme for the toxic microcystin/nodularin synthetase was not present in any of the coral reefs, revealing that the cyanobacteria present in the waters lack the genes to produce the toxic product.

Assessment of Water Quality Parameters and Nutrient Concentrations at Bonaire June 19 – July 1 2022

Michael Mangan

Midland College

Sonde Results

- YSI 6920-V2 sonde was used to measure ph, temperature, dissolved oxygen, and salinity at prescribed depths of 20, 40, 60 ft, and at the surface at five dive sites
- All measured variables were consistent across depths and locations

Table 1 - Bonaire Dive Sites - Sonde Field Data (6/20/22 - 6/29/22)

Dive Site (Date - Time) Sample Depth	Temp °C	Temp °F	Conductivity (µmhos/cm)	Salinity (ppt)	pH (S.U.)	Dissolved Oxygen (mg/L)
Rock Pile (6/20/22 - 1100 hrs)						
Surface	27.5	81.5	55700	36.9	8.1	6.5
20 feet	27.5	81.5	55700	36.9	8.1	6.5
40 feet	27.6	81.7	55800	37.0	8.1	6.5
Bottom: 63.0 feet	27.5	81.5	55800	37.0	8.1	6.3
Leonora's Reef (6/22/22 - 0900 hrs)						
Surface	27.5	81.5	55700	36.9	8.1	6.4
20 feet	27.5	81.5	55700	36.9	8.1	6.3
40 feet	27.4	81.3	55900	37.1	8.1	6.4
Bottom: 64.0 feet	27.4	81.3	55900	37.1	8.1	6.4
1000 Steps (6/26/22 - 1045 hrs)						
Surface	27.5	81.5	56100	37.2	8.1	6.5
20 feet	27.5	81.5	56100	37.2	8.1	6.5
40 feet	27.5	81.5	56100	37.2	8.0	6.5
Bottom: 63.6 feet	27.5	81.5	56100	37.2	8.0	6.4
18th Palm (6/27/22 - 1100 hrs)						
Surface	27.4	81.3	55800	37.0	8.0	6.6
20 feet	27.4	81.3	56200	37.3	8.0	6.5
40 feet	27.4	81.3	56200	37.3	8.0	6.5
Bottom: 63.0 feet	27.4	81.3	56200	37.3	8.0	6.4
Alice in Wonderland (6/29/22 - 0930 hrs)						
Surface	27.3	81.1	56000	37.1	8.1	6.4
20 feet	27.4	81.3	56100	37.2	8.1	6.4
40 feet	27.4	81.3	56100	37.2	8.1	6.5
Bottom: 63.0 feet	27.4	81.3	56100	37.2	8.1	6.2

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Assay Results

- Fe and P measurements were below reportable limits at each measured location
- Nitrogen concentrations demonstrated consistent decrease with depth
- Strontium and Calcium concentrations were consistent across all depths and dive sites studied

Table 3 - Bonaire Dive Sites - Calcium-Strontium Lab Data (6/26/22 - 6/29/22)

Dive Site (Date - Time) Sample Depth	Calcium (mg/L)	Strontium (mg/L)
1000 Steps (6/26/22 - 1045 hrs)		
Surface	459	8.42
20 feet	443	8.41
40 feet	442	8.37
Bottom: 63.6 feet	432	8.35
18th Palm (6/27/22 - 1100 hrs)		
Surface	439	8.25
20 feet	422	8.28
40 feet	463	8.21
Bottom: 63.0 feet	436	8.32
Alice in Wonderland (6/29/22 - 0930 hrs)		
Surface	453	8.48
20 feet	419	8.63
40 feet	475	8.33
Bottom: 63.0 feet	460	8.43

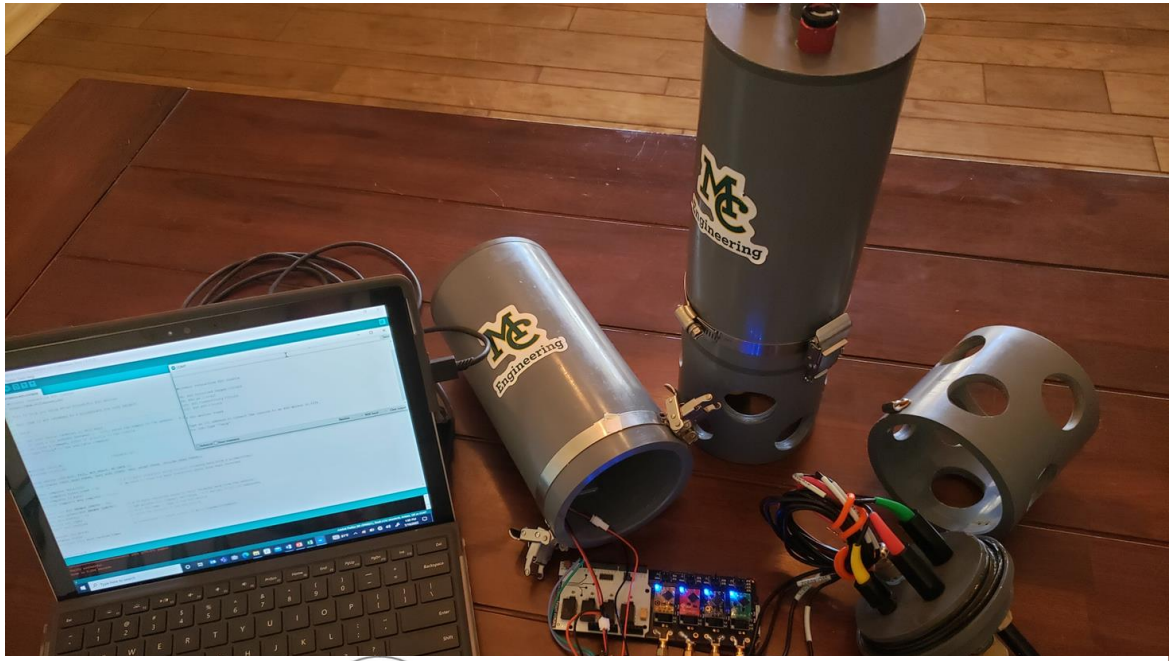
Low-Cost, Custom Fabricated, in-situ Autonomous Sensor Arrays for Monitoring Water Quality Parameters

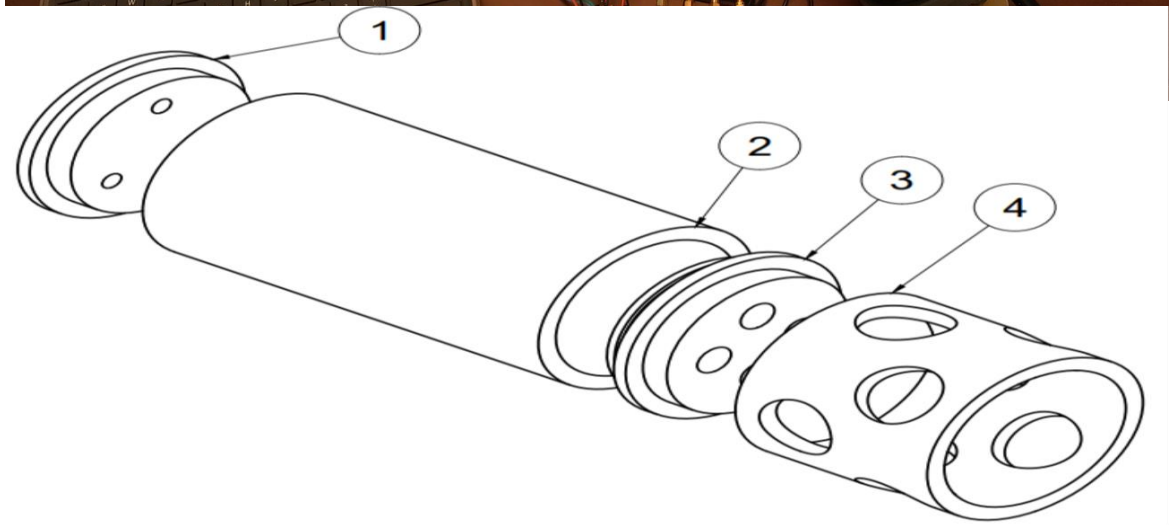
Justin McKinney

Midland College

Design and Assembly

- Used commercially available PVC, electronic and aquarium hobbyist equipment to keep costs low
- Total cost of assembly **$1,018.25**
  - Commercial equivalent retails for ~**$22,000**





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Housing and Leak Testing

- ❑ Salt water and electronics DO NOT work well together
- ❑ By pressuring a water based vessel and using probe “blanks” the housing could be tested to depths of 200 ft for 24 hours
- ❑ The housing worked!

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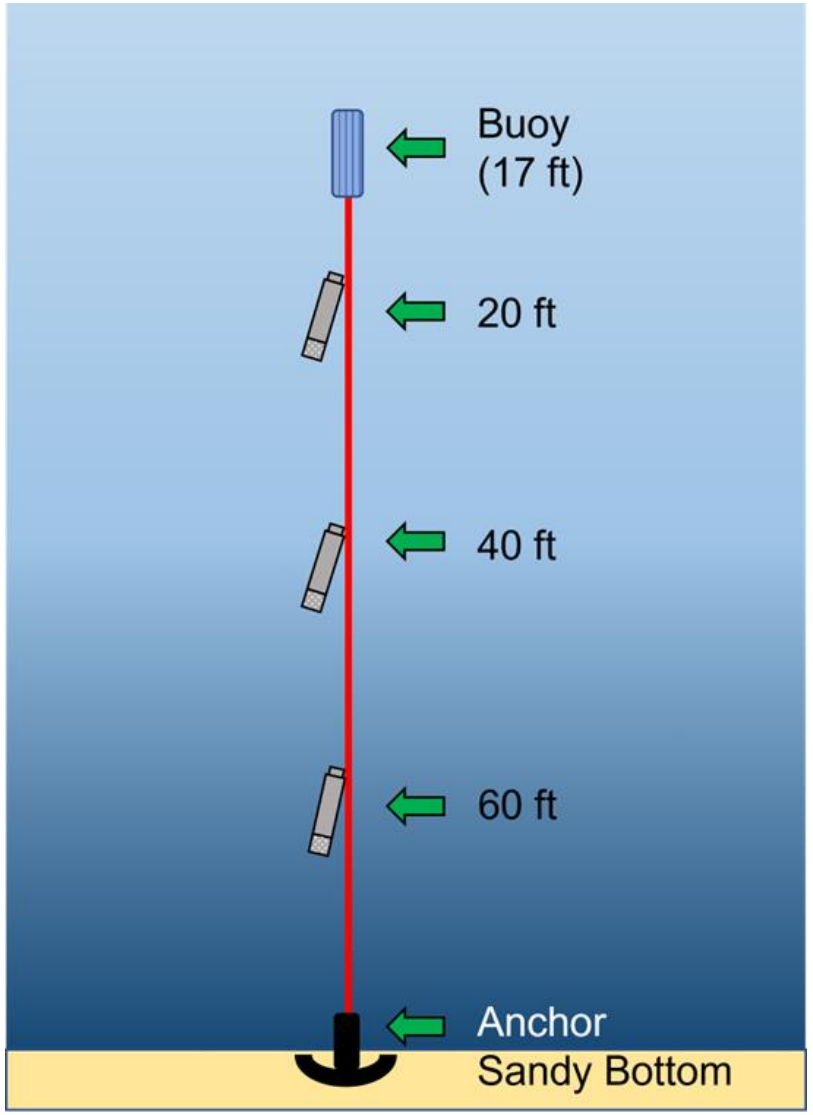
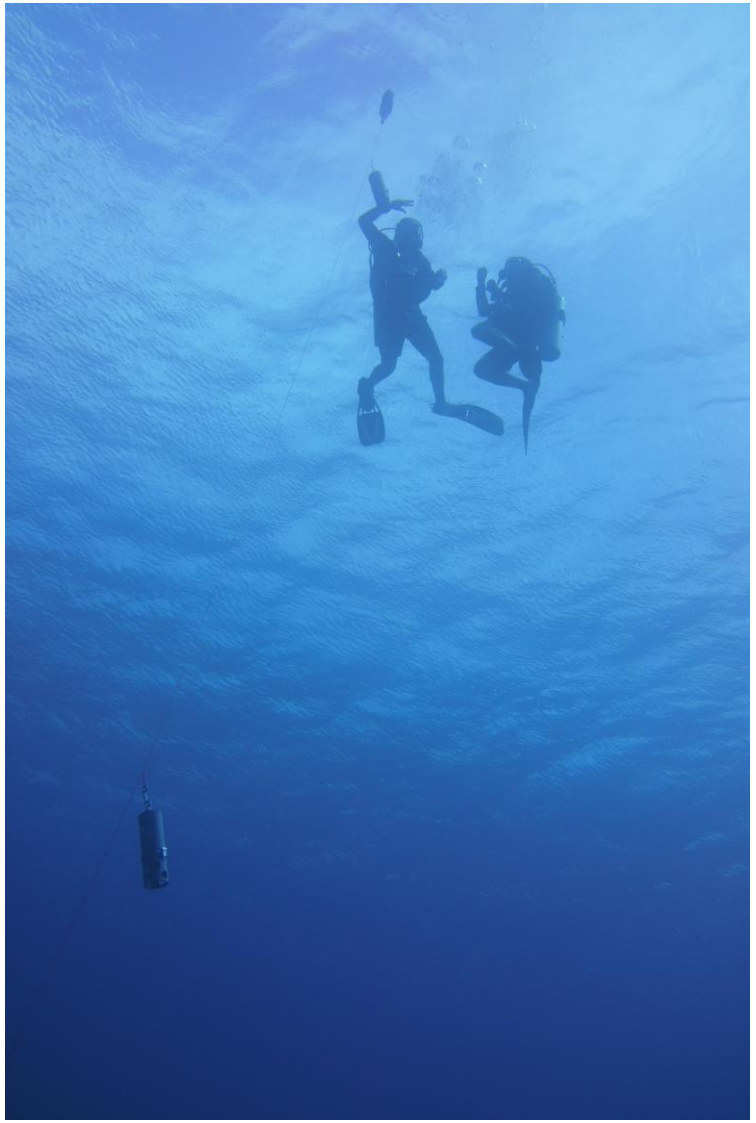
Field Testing

- Deployed three pods at 20 ft, 40 ft, and 60 ft
- Collected temperature, pH, salinity, and dissolved oxygen measurements
- Remained submerged for 24 hours at four dive sites
  - Leonora's Reef, 1000 Steps, 18th Palm, and Alice in Wonderland





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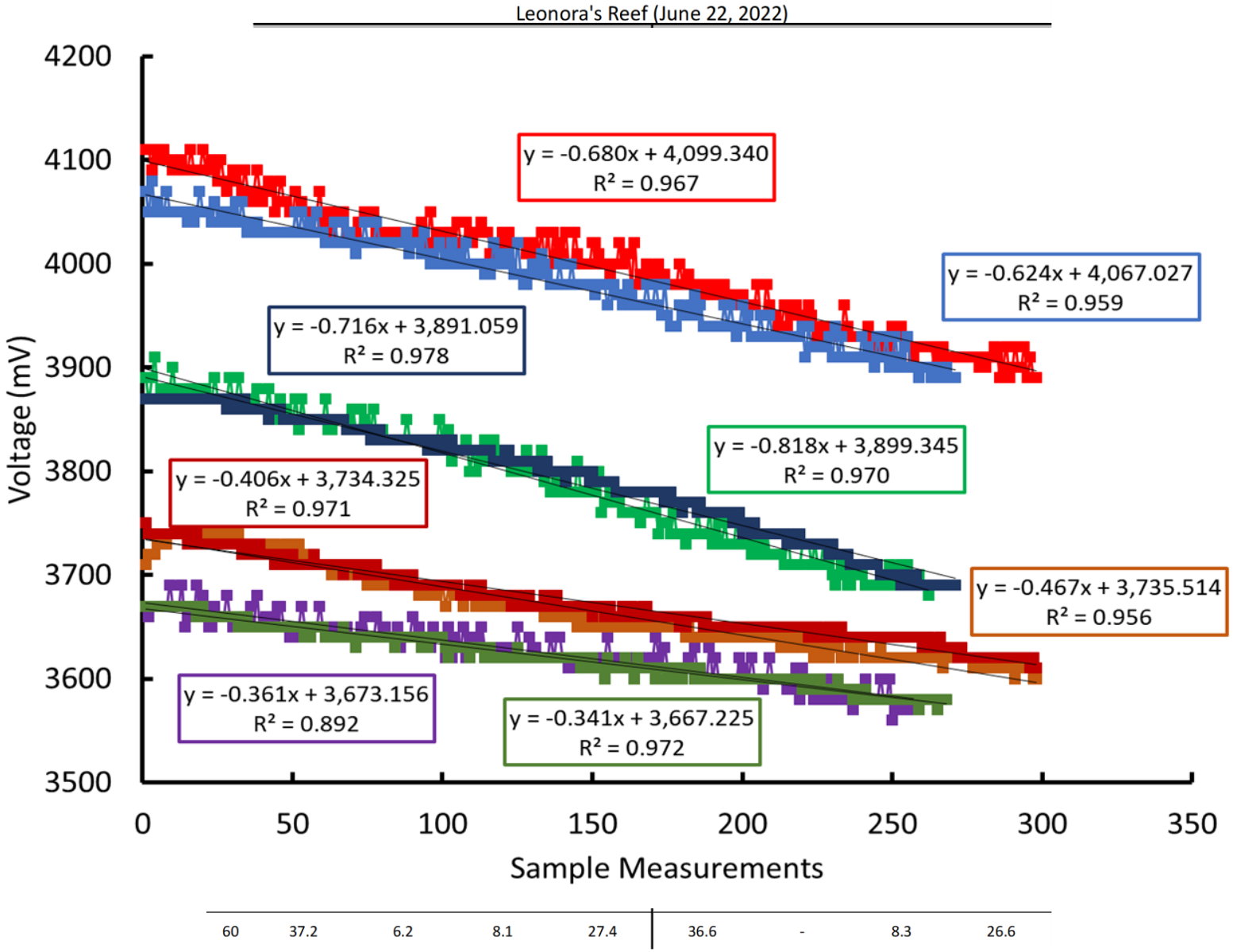
Results

- WE GOT DATA!
- Data was averaged over the 24 hour period and compared to the sonde surveys
- Battery usage was monitored and determined that it can last for five days

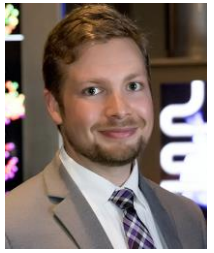


Research Mentors Who Contributed to the Effort

Assoc. Dean / Prof. Laura Mydlarz (UT-Arlington biology)

Prof. Marlana Mertens
(Midland College microbiology)

Prof. Greg Larson
(Midland College environmental biology)



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(Midland College engineering)

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Acknowledgements

National Science Foundation
UT-System LSAMP Grant

The Yarborough Foundation

Midland College

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