A Tale of an Experiment Gone Wrong, Perseverance, and Novel Discovery

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I scribble a sad face in my lab notebook beside the inscription: "Respiration Experiment #1 was a complete failure." I annotate my lab notebook like this because it's important for me to stay lighthearted at the beginning of a long research cruise. Being cheeky lifts my spirits. I also like the idea of looking back at my lab notebooks when I'm a senior scientist and remembering that failure is often the foundation of success.

It's the third night of our 2-week cruise transect from Woods Hole, Massachusetts to Bermuda aboard the R/V Knorr. She's a 279-foot research vessel built in 1969 and painted what we at the Woods Hole Oceanographic Institution call WHOI-blue, a dark shade similar to the color of the open ocean on a cloudless day. Our charge on this expedition is to better understand the chemical signals used by tiny microbes living in the ocean. In much the same way that pheromones signal attraction and repulsion in land animals, chemicals that are released by marine bacteria and tiny plant-like organisms called phytoplankton can impact the survival, reproduction, and metabolism of small organisms in the ocean. Although they're small, these organisms play a huge role in the ocean's ability to absorb carbon dioxide from the atmosphere. But what are these chemical signals? Are they in the environment? If so, what impact are they having? These are the questions that we hope to address on this voyage, assuming that we can all rise above the challenges of conducting research at sea.

I have just returned to the "01 Lab" after 20 minutes of heaving over the leeward side of the Main Deck. There's actually a scientific protocol to being seasick. When you get that metallic taste in the back of your mouth and you know for certain that you are about to succumb to your distraught equilibrium, the proper course of action is:

- 1) Get out of the interior of the ship! Fast!
- 2) Determine the direction of the wind over the bow. If you think pissing into the wind is bad...
- 3) Find your way to the railing on the leeward side of the Main Deck. Avoid on-deck incubators and crates of scientific equipment. You want to vomit into the ocean not onto someone else's experiment. Depending on how crowded the Main Deck is, this may be a very narrow piece of real estate. It's best to scope out potential places to puke at the first sign of nausea.
- 4) Go for it.
- 5) Stay outside until you're sure you're done. Stare at the horizon. Find something to lean on. Meditate. Give yourself a pep talk. Just don't go back inside until you are positive it is over.
- 6) At some point along this journey you need to find a buddy who can make sure that you aren't carried away to sea while paying penance to Poseidon.

It's this last point that sticks in my mind as I wait alone between bouts of being sick, watching the glassy black waves slip into the night. Every safety briefing that I've ever heard has contained a variation of the following sentence: "If no one witnesses you fall overboard, the odds of being rescued are non-existent. By the time we realize you're gone, it's already too late." I wonder how the twisting, nauseating pain in

my gut compares to the sinking feeling that a person overboard must get as they watch their ship sail away into the dark night, the running lights winking their final goodbyes.

I don't generally exercise my disregard for rules while at sea but a puking buddy is hard to come by at this time of night (violation of rule #6 above). It's a quarter past 2 in the morning and no other scientists are awake. The other graduate students and research technicians finished their sampling hours ago. They're down below catching what rest they can before the 6 a.m. science meeting. My advisor is no doubt in a hallucinogenic half-rest, a side effect of his answer to seasickness — the Coastguard Cocktail. It's a time-tested mixture of uppers and downers that numbs your inner ear but affords you the energy to be a productive sea-going oceanographer.

As I return to my abandoned experiment, I wish that I'd taken the boss up on that Coastguard Cocktail. I've just spent 20 minutes lying on deck waiting out my seasickness, while my precious samples were sitting on the bench top degrading in sulfuric acid. I'm particularly interested in a group of compounds called polyunsaturated aldehydes that are produced by several types phytoplankton when they experience stress. Considering that these compounds smell like rancid Chinese food, it's not hard to believe that they're toxic to a wide range of marine organisms. However, in laboratory studies some marine bacteria actually increased their growth rate when exposed to these compounds. Their ability to impact organisms in different ways is exactly why I'm interested in them. How is their production by phytoplankton affecting the other organisms in the ecosystem?

But whatever polyunsaturated aldehydes were in my samples had no doubt begun to break apart in the harsh, acidic environment while I was vomiting over the rail. I pour the rest of my incubation bottles down the sink, leaving the monotonous task of acid washing for the morning. The entire 2-day experiment has been a bust. I could blame it on the lengthy seasickness protocol, my fragility, my knack for designing far too intricate experiments and/or my disdain towards asking for help. But I reassure myself that this is just about par for the course.

Every cruise is the same: weeks of preparation, hashing out every detail of my experiments, coordinating with other scientists to accommodate everyone's goals, enduring stress dreams. The most reoccurring scientific nightmare involves me miscalculating the number of pipette tips that I'll need and then no one on the cruise is willing to spare any of theirs. And thus my lack of preparation is revealed. I'm shamed in front of my peers and superiors, and unable to complete graduate school. I drop out over and over again in my dreams. So in the days leading up to the mobilization date, I meticulously re-count the number of pipette tips, bottles of solvent, and boxes of filters needed. My advisor reminds me daily that Fisher-Scientific does not deliver to the middle of the Atlantic. Inevitably there's always some nearly forgotten piece of gear that must be thrown into a personal suitcase. The TSA must love field scientists, always carrying odd pieces of equipment in their carry-on with a socially awkward smile and a letter that easily could have been forged from their Department Head attesting to the benign nature of the suspicious luggage.

Once all of the equipment is shipped and all the scientists have made it to the port, mobilization day (MOB) begins. MOB entails forklifting the science gear onto the ship. Then I exercise my Tetris skills,

finding space for all that stuff that I told myself I needed, as thirty other scientists do the same. After everything is in its place and tied down with ratchet straps, bungee cords, and rope, we set sail.

The first few hours at sea are great. The calm coastal waters. The serene ocean views. The dolphins riding the bow. The false hope that perhaps I won't get seasick. Perhaps all my experiments will work. Perhaps Poseidon will pay me penance instead of the other way around. All are equally likely to happen. Within hours it hits. And I spend the next few days working 16 hours a day through non-stop queasiness, as I seriously contemplate a new career path and grow to revile the thought of saltines and ginger candy. Don't get me wrong – there are many wonderful aspects to being an oceanographer. They are just hard to remember when the fate of your professional life depends on your ability to conduct thoughtful science on an empty and very angry stomach.

Months removed from the tumultuous beginnings of that expedition, I sit in a loud warm instrument room on a cold March day. I scroll through chromatogram after chromatogram, searching for any sign of my compounds. I think back to the days spent transferring seawater from the ocean into clear liquid in the incubator and then into clear liquid in my extraction protocol, all along faithfully hoping that there's something within that clear liquid worth all the sweat and tears and regurgitation. Then I see it – a small but very real peak at 3.3 minutes. I check the mass spectra. Yep, 330.9 m/z: 16 carbons, 18 hydrogens, 4 nitrogens, and 4 oxygens. I cross-reference the sample number with the chart in my lab notebook. Respiration Experiment #3-Control A t=0. Eureka! This is the very first time anyone has observed polyunsaturated aldehydes on sinking particles in the ocean. This will make my dissertation! This is huge! This might even make a weekly journal – the most coveted type of publication amongst scientists!

Why is it so interesting that I've found polyunsaturated aldehydes on sinking particles? Sinking particles are made up of dead and dying phytoplankton. When those phytoplankton were alive they photosynthesized like trees, taking carbon dioxide out of the ocean and converting it into phytoplankton biomass. When carbon dioxide is removed from the ocean, carbon dioxide from the atmosphere dissolves into the ocean to replace it.

But that isn't the end of the story. After the dead and dying phytoplankton aggregate into particles, bacteria eat those particles as they sink to the bottom of the ocean. By eating the sinking particles, bacteria convert the phytoplankton biomass back into carbon dioxide which will eventually go back into the atmosphere. However, not all of the phytoplankton biomass is eaten by bacteria. A very small fraction is buried in the bottom of the ocean. This is what allows the ocean to act as a sink for carbon.

So the fate of sinking particles is important for the global balance of carbon dioxide. If more carbon is burying in the deep ocean in the form of sinking particles, the ocean can absorb more carbon dioxide from the air. But if bacteria convert more of the carbon in sinking particles to carbon dioxide before it can be buried in the ocean, less carbon dioxide from the air will be absorbed by the ocean.

Since polyunsaturated aldehydes affect the growth of bacteria, could polyunsaturated aldehydes be impacting the fate of sinking particles? If the polyunsaturated aldehydes are toxic to marine bacteria,

they would prevent the bacteria from eating the sinking particles and more phytoplankton biomass would be buried. The more phytoplankton biomass that is buried in the deep ocean the more carbon dioxide can dissolve in the ocean. This would make the ocean a larger sink for carbon. Or is it the other way around? Are the marine bacteria actually enhanced by polyunsaturated aldehydes? This would allow the bacteria to eat more phytoplankton biomass, in turn releasing more carbon dioxide back into the environment. Or do polyunsaturated aldehydes have no impact on marine bacteria, carbon dioxide, or the fate of sinking particles?

Clearly, the impact of polyunsaturated aldehydes is an open question. But we are one step closer to understanding the intricacies of our amazing ocean. Polyunsaturated aldehydes are present on sinking particles. And for now I'm the only one who knows this little morsel of information. I run down the hall to grab the graphs off the printer, eager to show them to my boss. My head begins to overflow with plans for future research cruises, new opportunities to transfer more clear liquid into other clear liquid, as I stave off seasickness and hope for a novel discovery.