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## Diving into Aquatic Microbial Ecology and Evolution with Anne Thompson

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Welcome to PDXPLORES, a Portland State Research podcast featuring scholarship innovations and discoveries, pushing the boundaries of knowledge, practice and what is possible for the benefit of our communities and the world.

My name's Ann Thompson. I'm an assistant professor in biology at Portland State University.

The work of our research group looks at bacteria or microbes that live in the ocean and how their work in the ocean drives major nutrient and energy flow in the earth system. And our work is funded by the National Science Foundation as well as recently through the Simon's Foundation.

So my research studies microbes or bacteria that live in the ocean. And one of the things we're really interested in is where do they go? Who eats them? What is their death process or their mortality process? And this is really important because we need to know where their carbon and energy is going in food webs. So it's important for us to know if they're lysed by viruses, so all of their nutrients and guts kind of just get spread out to the seawater, or are they eaten by bigger things? And so knowing that helps us understand and model and predict how the number of cells that are present support different ecosystems.

We know several of the different pathways that these cells die or who eats them, so viruses, lice them, kinda break them apart. Protists or other single cells actually engulf them and can turn them into food that way. We also know that there are small crustaceans, like Copapods that can predate on them or attack them and eat them. But there's another type of animal that we refer to as gelatinous grazers that are much larger organisms, and they have, basically, a mesh within them and it's made of kind of a gel-like material. And this allows them to act like vacuum cleaners basically in the ocean so they can go around pumping seawater onto this mucus mesh, filtering out all the particles, and then basically eating those particles. So these are the mucus mesh grazers of the gelatinous grazers. And up till this point, they've really been underestimated in the ocean because they're so delicate. They've not been well counted with traditional methods like nets and things like that. And so now that we know that there are actually a lot more out there than we thought, we're trying to understand what role they play in these mortality processes. So, in removing our bacteria from the water.

So we have several different ways of going about this work. One of them is using scuba based approaches. So these animals are super delicate. They're basically like just a gelatinous bag that's really supported by the seawater around them. So when you

collect them with a net, you collapse them. They could disintegrate in the net and you might not be able to recognize them much less experiment with a live organism. So one of the techniques we use, and this is really led by my collaborator, Kelly Sutherland at University of Oregon, is a scuba based approach. So Kelly's team that leads this part, does scuba in the open ocean. We capture these animals in jars, keep them intact, keep them happy, and then we're able to do experiments with them live and see which types of cells they're eating.

We can also do some experiments with cultured grazers, and there's really only one kind that is in culture. We refer to them as appies or appendicularians, and they're pretty small, and some groups around the world have figured out how to keep them happy in a lab setting. So one of the things we're planning to do in the next few years is develop a culture of these appendicularians here at Portland State so we can really do some of these controlled experiments in the lab with students at Portland State as opposed to these very technical approaches like the scuba. So we can couple this cultured approach as well as this field scuba approach.

And a third approach is to work on a ship. And there are a few of these gelatinous grazers that are a little bit less delicate. For example, pyrosomes. So we can collect those in nets and we've done that work off the Oregon coast and are able to study how those more robust gelatinous grazers can feed on our bacteria.

So we study the interaction between microbes and these gelatinous grazers in a few different field sites. So one of our field sites is here off the Oregon coast, and so we have lots of microbes that are taken care of, all kinds of important jobs for the food webs there, and they're grazed upon by these gelatinous grazers. That's a really great study system. It's close to home. And we've been lucky to collaborate with folks at Oregon State University and University of Oregon on those projects. We also have worked in the Gulf Stream, so we've been able to access the Gulf Stream from a field station in West Palm Beach, Florida. And this gives us access to the Gulf Stream water, which is really warm and representative of the large area of the ocean. This very warm, open ocean setting.

And our third field site is off the coast of Hawaii, so off the Kona Coast. And this is a fantastic site for studying some of these gelatinous grazers. They're really abundant. The ocean conditions are great, and there's also a great community of recreational divers there. So my collaborator on the project, Kelly Sutherland, is in touch with them and we can kind of know what types of animals are there before we go.

So the scuba approach is one that was developed many, many years ago in the '70's and has not been used as much recently. But my collaborator is really an advocate for studying these gelatinous animals in CTU, so in their natural habitat. And that's really the best way to see how they handle fluids, see what their body form is, and then from our perspective, see how they're really feeding on different types of cells. So we're using a well-established technique, the scuba technique that was pioneered many, many years ago in the '70's and is really still powerful today to understand these animals and their interaction with the microbes that surround them.

Marine microbes are really cool and really powerful. They're very simple organisms. They have small genomes. They're very small cells, but what they're really good at is taking inorganic nutrients from their surroundings in the seawater and energy from the sun and converting that into biomass. And then they can be the food through these grazing or predation interactions to larger and larger types of organisms. So they're really analogous to say grass in a terrestrial system, grass on land, they are the base of the food web, the foundation of the food web. That first step in getting the sun's energy into biomass that can then fuel the rest of the ecosystem. So that's what they do in the ocean. They also cycle all kinds of different nutrients that are important to other bacteria and other microbes in the ocean, including carbon, which we know is really important in the atmosphere and in global climate change.

So there is no one process for mortality. We tend to think of it as a combination of lots of different processes. So there's some of them that are much better studied than others, and we know from modeling work as well as experiments that we're still missing some of the mortality. So there are some sources of mortality that we haven't been able to measure. And that's one of the roles I hope our work can play is looking at these gelatinous animals that we know are ubiquitous in the oceans all over the world, and trying to understand what role they have in microbial mortality. There are viruses, there are other single cells, there are small crustaceans, and now we know that there are these gelatinous animals that can contribute to that mortality process as well.

The process for this work starts in the field, or really the ocean as we say. So my collaborator, Kelly and her team dive down and they look for these animals, and they're very practiced at identifying these animals and seeing them. So they'll capture them in a jar, which they'll then bring up to me on the surface. At the dive boat, and that's when I start my incubation of these animals. So they're surrounded by microbes, lots of different types of microbes. And so I sample the water around them on a time course for about an hour or two hours. And that's really where the experiment ends in the field. So we pack up our samples, we freeze them, we bring them home, and when we're back

in the lab, we extract the DNA from the guts of the animals, but also the seawater that surrounded the animals. And then using different molecular techniques we can see which microbes are there and how many of them there are. And then if we compare our different time points across the feeding incubation or just look in the guts of the animals, we can see how the types of microorganisms and their abundances changed in the experiment. Then we can determine which microbes were eaten and how quickly, and then we can compare our different types of gelatinous grazers to each other.

The one thing we've been surprised by again and again is how one of the microbes we study, called *prochlorococcus*. It's kind of a mouthful, so we call it *pro* affectionately, but one of the things we found was that these gelatinous animals just are not consuming it very quickly. And this is a big deal because this is the most abundant photosynthetic cell on the planet. And it's a very small cell, and it's been thought that it's small size is a strategy it can use to avoid predation. But so far most of its predators have been either viruses, which are smaller than it is or other single cells. But now we're kind of adding this additional layer where these large gelatinous grazers that are taking up other bigger cells like diatoms, but they're leaving these very small cells like *prochlorococcus* and it's kind of other counterpart called SAR 11.So these very small cells, it looks like are really not removed from seawater to the same extent as the larger microbes. And this process is selectivity has a big impact on the composition of the microbial communities and knowing that the different types of microbes do different jobs in the ocean, it also impacts how the nutrients and energy flow in the ocean and by extension on the entire earth system.

We study several different taxa of these gelatinous grazers. One of them is called Salps. This is a pretty big organism. They're, I don't know, as big as sometimes even as big as a Ziploc bag or something like that, like a small sandwich size Ziploc bag, and they look like a bag too. They're clear and gelatinous in the water. Another type of gelatinous grazer is called a pteropod. It's kind of like a snail with wings. It's a very beautiful, beautiful organism, and it makes a mucus mesh as well, but it's very different. It's kind of like a bubble and it sediments the particles. We study a few other gelatinous grazers. One is an appendicularian, which is the one that I mentioned that can be successfully cultured. It's adorable. It looks like a tiny little tadpole, and it basically inflates a mucus bubble, which it uses to capture it's microbial prey. There are doliolids, which one of my students, Melissa Steinman, is studying. They're much like salps, but smaller and they don't swim quite as quickly as the salps. And then finally, pyrosomes are another type of gelatinous grazer, and they're really interesting because they're colonial, so they have lots of different individuals all linked together in these bigger colonies that can propel themselves up and down in the water column. So, all these different taxa have in

common that they use this mucus mesh to filter particles and trying to understand how they differ from each other, which microbes they take out of the water, how quickly and what the impact of that interaction is on, on global processes.

When we start looking at the DNA of the microbes and the DNA from the guts of our grazers, one of the first things we do is look at the microbial diversity that's in those two compartments. And what we see is there's a lot of diversity in the seawater, and that's really consistent with our understanding of the seawater with lots of tiny little micro habitats. But the diversity in the guts of the grazers is much, much less. We think this is because we have these really specially paired host, which is the grazer and microbial symbiomes potentially. So it's kind of a lower diversity situation. But we also know that these organisms are pulling microbes from the seawater. So we're interested to see that their diversity isn't not simply a reflection of the seawater, but that is very specific and kind of tuned in to the host or the ISTs grazer.

In one of our studies in West Palm Beach, we not only sampled the guts of the salps and the seawater that surrounds them, but we were also able to sample their fecal pellets. And these are really carbon rich particles that tend to sink to the bottom of the ocean. And we were curious which microbes were present in those fecal pellets that maybe didn't get digested by the salp. But what we found is that most of the microbes in the fecal pellets were this type of microbe called Vibrio. And Vibrio is known to be a very fast swimmer and very good at taking advantage of rich organic carbon sources. So this is some of the first evidence that within an hour of being produced, Vibrio is colonizing and perhaps feeding upon these fecal pellets, preventing that carbon from sinking down to the bottom of the ocean and basically digesting or recycling that carbon in the upper ocean, making it available to Vibrio, to the predators of Vibrio and to other microbes that live in the surface ocean. So this was really interesting to see how quickly this compact carbon source is colonized and where that carbon could be going.

What we found when we looked at the microbial prey that could be in the guts was that it wasn't just one thing. So these animals are not super selective. They're not just coming to the buffet and saying, "I only eat pasta, or I only eat meatballs," but they're saying,"I'll take a little bit of everything."And we see that both of these taxa do take a little bit of everything, but they do have preferences as well. So we could see that, for example, the salps seem to like larger cells like *synechococcus*, which is a cyanobacterium and did not pick up some of the smaller cells like *prochlorococcus* and like SAR-11, which are really abundant cells, but clearly not as valuable to the grazer as some of these much larger cells. What our work and the work of other research groups are showing is that these mucus mesh grazers really do have an important role in shaping microbial communities in the ocean. So they've been overlooked for a long time. Because we didn't know how to measure them properly. We didn't really know how abundant they were, how ubiquitous they were, how they could respond very quickly to blooms of phytoplankton. But now that we know how important they are in marine systems, we have contributed this understanding of their feeding on all types of different microbes. And that is one of many mortality sources for these microbes, but a really important one and one that we should take into account when we're trying to create forecasts of how microbial systems will change with different perturbations, like heat waves in the ocean or climate change, and how the entire food web or marine food web would change with those conditions. So this is one process of many, and it's the complex process, but it's important for us to be able to quantify it and recognize it, and our work is starting to show that.

One of the important things is that these are relatively large organisms, these grazers, and so they're kind of skipping many of the steps of a traditional food chain or food web. They're really taking that foundation of the food web and using it for food and kind of shooting that carbon and energy up to higher tropic levels. So, it kind of speeds how quickly much larger organisms can use some of that energy generated by microbes if we can reduce the number of steps in the food web. These taxa, like appendicularians, like salps, they're really feeding on microbial production and making that carbon available for much, much larger organisms. So it really makes the connection between microbes, between the environment, between the chemical environment to higher tropic levels much shorter and much more sensitive too. So it could mean that food webs are more stable, but it could also mean that they can respond more quickly when the oceanic conditions change.

I started studying microbes in the ocean because I was really interested in microbial life and how tiny, tiny organisms could have such a big impact on the earth's system as well as the human system through medicine. And I did my PhD in microbial oceanography, so I studied one particular type of cyanobacteria and how it lives and how it gets through its day. And one of the people who I knew at that time as a graduate student was another graduate student, Kelly Sutherland. And much later, after we had left our PhD programs and gone on to start our careers in different ways, we reconnected and realized that her interest in gelatinous organisms and my interest in microbes actually had this link because the gelatinous grazers, the gelatinous organisms were potentially feeding on these cells. So we started talking and were able to get funded through the National Science Foundation, and that's just been a really fun and productive collaboration.

Some of these types of experiments are really challenging. For example, the scuba based approach. And so one of the things that I'd like to do in the future is be able to bring some of these organisms, for example, these appendicularians, so the appies ,to culture here at Portland State. I'm really excited to be able to do that with the Simon's Foundation grant. And this will give students here at Portland State a chance to interact with the microbes and with these organisms in a way that they haven't been able to do before. So instead of needing decades of training to be able to do scientific diving, we'll be able to do some training in several hours in a lab setting and have students be able to work with organisms that really represent the real ocean environment and be able to make contributions to science. So that's one of the future things that I'd like to do with these projects and with this funding.

My name is Anne Thompson, and as a microbial oceanographer, I lead a group of scientists whose goal is to discover how tiny cells in the ocean shape the nutrient and energy cycles of the earth system.

Thank you for listening to PDXPLORES. If you liked what you heard on this episode, please rate and follow the show anywhere you get your podcasts.