

Research explores effects of marine bacteria on toxic algae

If you've ever experienced shellfish poisoning or know someone who has, you probably think twice before eating anything with a shell. Shellfish poisoning is uncomfortable at best and deadly at worst.

Of course, shellfish aren't always toxic, and those that occasionally become toxic do so for only brief periods of time. What makes some shellfish toxic, some of the time?

One culprit is simple marine algae, or *harmful algal blooms*, as scientists refer to the poisonous variety. These harmful algal blooms, which apparently have been making humans sick for as long as we've been eating shellfish, have received a lot of attention worldwide. Although scientists know a lot about their *ecophysiology*, or the relationship between their physiology and their environment, they know very little about the role marine bacteria play in the natural control of harmful algal outbreaks.

From a purely theoretical perspective, bacteria seem to have both the means and the opportunity to influence the course of algal blooms. Researchers already know, for example, that marine bacteria produce chemicals that are toxic to marine algae.



Stephen Giovannoni's Sea Grant-funded research aims to solve the puzzle of when and how harmful algal blooms affect shellfish.

Ecological theory says that a lot of bacteria are present in the ocean and most of them are not primary producers: they receive their organic carbon from phytoplankton, which picks CO₂ from the atmosphere. The question is, how does the carbon get from the phytoplankton to the bacterium?

The general theory, according to Sea Grant researcher Stephen Giovannoni, is that it's a passive process: the phytoplankton die or leak or another creature comes along and eats them, leaving little bits and pieces behind. Then the

bacteria consume the stray organic matter.

But organisms that coexist in almost any environment interact somehow, so evolution is a lot more complicated than it looks at first glance. One hypothesis is that the bacterial cells are chemically interacting with the phytoplankton to make the plankton give up their "goodies" so that the bacteria can grow.

How can this information be used to develop better methods of predicting—and maybe even controlling—harmful algal blooms?

Giovannoni, a professor in the Department of Microbiology at Oregon

State University, pursued this question in a research project funded by Oregon Sea Grant.

"Our goal," Giovannoni said, "was to identify bacteria that live in Oregon coastal seawater that might interact with phytoplankton by attaching to them or producing some kind of a compound that would affect the phytoplankton."

Giovannoni and his team of researchers first looked at naturally occurring populations of cells and asked what kinds of bacteria are found in the water when phytoplankton are blooming. These

bacteria are the most likely to be interacting with the phytoplankton. They identified a number of microbial groups that seem to correlate with phytoplankton distributions.

Then they cultured many of those organisms and tested them by putting them together to see whether the bacteria could modify the metabolism of the phytoplankton cells.

Although the team didn't observe any cases where the bacteria had an impact on how the phytoplankton grew, Giovannoni is still optimistic that his hypothesis will turn out to be correct. He and his technician, Kevin Vergin, are following up on some of the new ideas that came from their research.

What are some of those new ideas?

Giovannoni said his team now has a much better idea about what microorganisms live in the seawater off the coast. "There are a million cells per milliliter out there, and 15 years ago we had no idea what they were. We've been able to identify quite a few of them in the course of this study."

Their work has also led to improvements in the technology the scientists use to grow organisms for their research. Developed by Giovannoni and his team prior to their Sea Grant project, the technology has been patented and is being used by private industry to culture new organisms from which antibiotics, pharmaceuticals, and agrichemicals can be produced. Giovannoni said the technology

works well, but it doesn't work for all of the organisms they want to grow.

"You seem to get a lot of benthic diatoms growing, which isn't really what we wanted. We've made a change in the chemistry of the water we used for growing cells, and we're hopeful that it's going to increase the number of species we can work with in the laboratory."

If that works, Giovannoni said they'll try another idea that wasn't in the original Sea Grant project proposal—a photocytometer that deposits cells on an x-y grid. "It's a remarkable machine that sorts cells at a very high speed and produces perfect, two-dimensional grids of cells."

You could have 10,000 cells in a tube and find the one you wanted because it had an antibiotic-resistant gene.

Giovannoni and Vergin want to combine the chemistry they developed with the photocytometer and try to isolate some better examples of phytoplankton. The key to that may lie in what's called a screen.

"A lot of great molecular biology succeeded because people thought up terrific screens," Giovannoni said. "You could have 10,000 cells in a tube and find the one you wanted because it had an antibiotic-resistant gene. All you need to do is throw the antibiotic on the plate and only the one cell with the gene grows. That's called the screen. Otherwise, looking at

10,000 cells to see which one of them is antibiotic resistant would be impossible."

Giovannoni and Vergin hope to develop a screen that will let them test tens of thousands of bacterial cells randomly, out of water, and look for ones that would change the growth of phytoplankton.

"That's the tough part," Giovannoni said. "Once you get that information, then it becomes a great situation because if you've got a growing phytoplankton and a bacterium and you add them together and they interact, then all the good molecular and chemical technology that exists on campus can be used to figure out the details."

Giovannoni said that if organisms are discovered that do affect phytoplankton metabolism—cause them to leak carbon in some other way or transfer nutrients to the

bacteria—it will be very interesting to determine the mechanisms of those molecules. "Maybe some of them will be useful. There are lots of examples of situations where organisms interact chemically, where the chemical turned out to have a pharmacological use. Most don't, but some do."

Giovannoni also hopes his continuing investigation will provide fundamental insights into the coastal food web and ultimately lead to solving the puzzle of when and how harmful algal blooms affect shellfish—and the people who eat them.

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