

Graduate student Annette Hynes studies the diversity of curious marine bacteria called Trichodesmium.

A most ingenious paradoxical plankton How do seemingly similar organisms coexist in the same ecological niche?

By Annette Hynes

Everybody has a unique place in the world, a job to do, a niche to fill. When you are a tiny phytoplankter, your place is in the ocean, and your job is photosynthesis. Floating in a seemingly uniform environment like the ocean, how do you stand out and find your niche amid all the other phytoplankton?

All phytoplankton basically use the same resources: light and a medley of nutrients. All else being equal, if you have two species fighting over the same resource, the stronger competitor will win and the weaker one will go extinct. For 10 different resources, you would expect 10 species to be able to coexist.

But the ocean is filled with thousands

of species of phytoplankton that coexist, though they all seem to fill the same niche —a phenomenon known as the "paradox of the plankton."

Among these paradoxical plankton is *Trichodesmium* (pronounced "trick-o-DEZmee-uhm"). The six species of *Trichodesmium* live together, yet they seem very similar.

Turning the nitrogen cycle

I started studying *Trichodesmium* in the first place because of their special talents, which make them essential to the marine ecosystem.

Trichodesmium are cyanobacteria, also known as "blue-green algae." Like other phytoplankton, these bacteria are capable of fixing carbon dioxide through photosynthesis the way plants do. No, the carbon dioxide is not broken; it is a gas that gets "fixed" into organic matter.

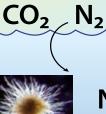
Even more impressive, *Trichodesmium* can fix nitrogen gas into a compound that other organisms can use to build their proteins and DNA. Nitrogen gas has a triple bond. It takes a lot of energy to break that bond and fix nitrogen into a different compound (see Page 33). Lightning can do it. With special enzymes to help them, a select few bacteria and archaea (another form of simple, single-celled life) can do it, too.

Most organisms get their nitrogen from a fixed source. Fish, copepods, you, and I get ours from eating food. Most phytoplankton and other single-celled organisms get theirs by absorbing waste excreted out



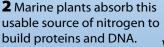
The little guys who make the ocean's nitrogen cycle go round

The marine bacteria Trichodesmium come in a variety of species (above). At left, a slick of Trichodesmium floats on the ocean surface in the western South Pacific in January 2007. Trichodesmium are essential cogs in the ecological machinery that cycles chemicals through the oceans—chemicals that sustain life and influence global climate (below).





1 Trichodesmium can convert, or "fix," carbon dioxide gas (a greenhouse gas) into organic carbon via photosynthesis, the way plants do. But the bacteria can also fix nitrogen gas into ammonia, a nitrogencontaining compound that other organisms can useproviding an important source for this essential element in the open ocean.



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5 When plankton die, some sink and decompose into usable nitrogen compounds that are recycled.

3 Zooplankton and other animals eat plants to obtain the nitrogen they need to grow.

4 Animals excrete waste containing nitrogen that is reused.

6 A portion of this debris sinks, carrying carbon to the ocean depths and drawing down concentrations of carbon dioxide from the surface. More nitrogen revs up plankton growth, which sends more carbon down. This link between nitrogen and carbon cycles ultimately influences global climate.

by animals or decayed from dead stuff.

While some places like lakes and coastal oceans have too much nitrogen (see Page 32), the open ocean is nitrogen-barren. That's why nitrogen fixers such as *Trichodes-mium* play such an important role in the marine food web.

Few critters eat *Trichodesmium* (perhaps because they are distinctively stinky). But this phytoplankter does spew out a lot of new fixed nitrogen for other bacteria and phytoplankton to use. This source of nitrogen stimulates phytoplankton growth, feeding little guys that feed the big guys and moving



Researchers aboard the R/V *Kilo Moana* collect seawater samples from Niskin bottles on a device called a rosette. The bottles can be programmed to snap shut at specific depths. When they come back on board, graduate student Annette Hynes (bottom left) filters *Trichodesmium* from the samples and freezes them in liquid nitrogen, so she can later analyze their DNA.



"Team *Tricho*" is made up of MIT/WHOI Joint Program graduate students (left to right) Annette Hynes, Elizabeth Orchard, and P. Dreux Chappell.

nitrogen and carbon up the food chain.

Eventually the large phytoplankton die and the little and big guys poop and die. Turds and carcasses clump together and sink, burying carbon at the bottom of the ocean and drawing down concentrations of carbon dioxide at the surface. This link between the nitrogen and carbon cycles can ultimately affect the global climate. But to understand how these chemical cycles work, you have to understand the creatures turning the wheels.

That's where we come in.

'Team Tricho'

At the Woods Hole Oceanographic Institution (WHOI), we call ourselves "Team *Tricho*": P. Dreux Chappell, Elizabeth Orchard, and me. To find the differences among *Trichodesmium*, we put them through all sorts of torture. Just like phytoplankton, we have to diversify to coexist.

Chappell and Orchard specialize in the ways *Trichodesmium* obtain and use nutrients such as iron and phosphorus in the oceanic desert where they thrive. Chappell focuses on iron while Orchard is devoted to phosphorus. They stress out cultures by withholding nutrients to see how they react.

I concentrate on diversity. I want to see if *Trichodesmium* have slightly different capabilities, and if that's what allows them to coexist. To do that, I have to get them to grow.

My Ph.D. co-advisor John Waterbury has a blue-green thumb. With a combination of patience, curiosity, and stubbornness, he grows in his lab many cyanobacteria that refuse to thrive for anyone else. (I tried my hand at cultivating some *Trichodesmium* from the South Pacific Ocean, but the mailman sent my new isolates to Alaska instead of Los Angeles. They didn't survive the trip.) Luckily, Waterbury has a large collection of *Trichodesmium* strains from all over the world. I'm cultivating 22 of them, representing five species, and Waterbury has many more.

Making sense of the paradox

A variety of answers could explain the plankton paradox. For one, the ocean environment is not as stable as it seems. Seasons, storms, and eddies continually change the ocean, so no one competitor is able to dominate for long. Second, a niche is not just what you do, but how, when, where, and

What do Trichodesmium smell like to you?

Trichodesmium have distinctive odors.

Annette Hynes asked colleagues how they would describe the bouquet of Trichodesmium.



Annette Hynes, Joint Program Student:

'Jalapeño oil'



Ted Kane, assistant engineer, R/V *Kilo Moana*:

'English breakfast tea'



Eric Webb, USC microbiologist:

'Don't sniff the Tricho'



P. Dreux Chappell, Joint Program Student:

'I don't smell anything'



Elizabeth Orchard, Joint Program Student:

'Old-growth forest'

under what conditions you do it. Different phytoplankton prefer distinct temperatures, depths, levels of light, colors of light, nutrient concentrations, and nutrient types.

Waterbury and I have been growing *Trichodesmium* at different temperatures to see who likes it hot. In an accidental experiment, a light went out in my incubator and some species crashed while others limped along under the low light.

I'm also going through the culture collection to describe the spectrum of pigments. Species of *Trichodesmium* have a variety of colors: brick red, camel brown, salmon pink, and sage green. The pigments responsible for this range of colors absorb different colors of light for photosynthesis. By using different parts of the light spectrum, diversely colored *Trichodesmium* might be able to share the spectrum or live where their favorite color of light is more available.

They even smell different. I have a couple of strains that smell like stinky fish, but most of them have a range of earthy but caustic smells that remind me of jalapeño oil. But smell is in the nose of the beholder (see box above).

Hunting Tricho

Trichodesmium cells are quite fragile. It's hard to get samples from deeper than 175 feet with a net because the stress makes them pop and spill their guts as you pull them up. WHOI scientists Cabell Davis and Dennis McGillicuddy used a video plankton recorder (VPR) to take pictures and count colonies of *Trichodesmium*. They found them for the

first time as deep as 450 feet, changing the way scientists thought about how deep these nitrogen-fixers are distributed.

While the VPR does an excellent job seeing colonies, it's not able to see free-living single filaments of *Trichodesmium* and it cannot distinguish species. Using Waterbury's culture collection, I found that the six *Trichodesmium* species split up into two groups that can be distinguished by their pigments and their DNA.

With this knowledge, I developed a method to identify these two groups by targeting their DNA in the open ocean. We deploy an array of Niskin sampling bottles from the side of a ship. The bottles snap shut at the depths I select. There are no messy nets, and I can catch single filaments. I filter the *Trichodesmium* from two to three gallons of water and then analyze the DNA I've caught to find out who is there and how many they are.

Once we figure out who is where and what they're doing, we can get a more detailed and global view of the ocean nitrogen cycle. Many scientists lump all the species of *Trichodesmium* together, saying "*Tricho* is *Tricho* and who the hell cares."

I care. And Team Tricho's got my back.

Annette Hynes has been supported by the J. Seward Johnson and the Arthur E. Maxwell Graduate Student Fellowship. Her research is funded by the National Science Foundation, the WHOI Ocean Life Institute, and the NSF Center for Microbial Oceanography Research and Education.



nnette Hynes started her career as a mi- \varPi crobial ecologist at a tender age. As a preschooler, she collected alfalfa and crabapples and fermented them in a large ceramic pot to stink out her parents and three sisters: Jeannette, Lynnette, and Raennette. Growing up in Oshkosh, Neb., she was fascinated by tornadoes, milkweed parachutes, and grasshoppers. She got her B.S. in biological sciences and mathematics and a teaching certificate at the University of Nebraska-Lincoln in 1998. She taught math and science in New Jersey and Kenya before coming to the MIT/WHOI Joint Program in 2003. Working with WHOI scientists Scott Doney and John Waterbury in biological oceanography, Hynes is interested in combining culturing, field, and ecological modeling techniques to answer questions about phytoplankton ecology and biogeography. When she's not popping cells or wrestling with her computer, she enjoys climbing, crocheting, and curling.