

Harmful Algal Blooms: Biosensors Provide New Ways of Detecting and Monitoring Growing Threat in Coastal Waters

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This red tide, or *Noctiluca miliaris* bloom, was sighted 1 April 1996 in the Arabian Gulf during a Naval Oceanographic Office cruise on the USNS Kane. Photograph: US Navy.

Deep in the southwest of Scotland ripples a pool of moss-green water. Soulseat Loch, as this small lake in the United Kingdom is known, glows with unearthly green water—green because pixies, or elves, once washed their clothes in the loch. Or so claim local legends.

But scientists who have studied the loch's water, like Geoffrey Codd of Scotland's University of Dundee, believe there's another explanation for its green color: a bloom of blue-green algae. "At certain times," explains Codd, "Soulseat Loch's waters are covered with a 'scum' of green material. This floating raft is an overgrowth of blue-green algae named *Oscillatoria agardhii*. Blooms of these algae can make the water toxic to people—or animals—who drink it."

That's exactly what happened in 1994, when six cattle died from drinking the green water of Soulseat Loch. The cows exhibited signs of rapid-onset disease, according to the farmer who owned them, "and in fact were subsequently found to have partially broken-down filaments of *O. agardhii*, which was toxic to the animals' livers, in their rumens," says Codd. Soulseat Loch, local farmers maintain, has a history of annual blooms of blue-green algae.

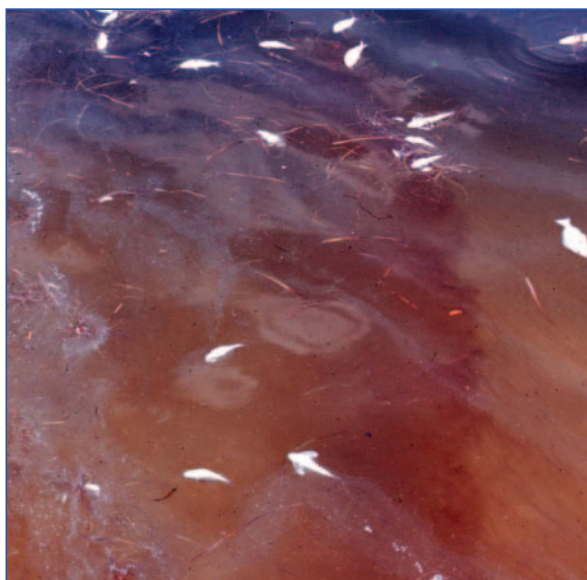
Awareness of these blooms may extend far back in human history. In AD 1175, clerics who lived on the loch's shore were most likely familiar with algal blooms: The monks named their residence *Monasterium Viridis Stagni*, or Monastery of the Green Loch.

What happened in the intervening centuries to the loch's sometimes-green waters? Any historical "data" on such blooms were lost in the monks' departure from the area and the monastery's eventual fall into ruins by the late 1600s. Says Codd, "It's interesting to speculate whether the life of this monastery might have ended in part because of increasing blue-green algal blooms in the loch and resulting poor drinking water there, but that's something we'll probably never find out."

Whether in freshwater bodies like Souleseat Loch or coastal waters around the United States, Japan, Mexico, and many other nations, blooms of so-called harmful algae are on the rise. "The questions now," Codd says, "are why is this happening, and what can be done about it? And how can we develop an early-warning system, so we're aware of the presence of harmful algae as they're forming a bloom, rather than after a bloom has become full-fledged? In situ and other 'biosensors' now being designed may provide new insights into an age-old problem that today has become a major coastal issue."

Harmful algal blooms are a story almost as old as time. The Bible's pages contain what may be the earliest explanation for blooms of harmful algae. In the Revelation of St. John the Divine, seven angels emptied vials of the wrath of God upon the earth. "The second angel," reads one passage, "poured out his vial upon the sea; and it became as the blood of a dead man, and every living soul died in the sea." Even in biblical times, it seems, humans were aware of harmful algal blooms, also called "red tides" for the blood-red color they often stain seawater.

Bloom-producing dinoflagellates have been found far back in the fossil record. Cyanobacteria, which produce rafts of floating green filaments, may have been the first photosynthetic forms of life on Earth. Spanning millions of years, the



Dead fish floating in water discolored by a red tide.

Photograph: Karen A. Steidinger.

tale of harmful algal blooms is an act in three parts: the players, the plot, the resolution.

The Players

"Throughout the world's coastal oceans, observations of harmful algal blooms, also called HABs, are being reported more and more often," according to Kevin Sellner, a HAB scientist and director of the Chesapeake Research Consortium in Edgewater, Maryland. "These events are accompanied by severe impacts to coastal resources, local economies, and public health." Some species recur in the same geographic regions each year, says Sellner, "while others are episodic, leading to the unexpected deaths of local fish, shellfish,

mammals, and birds." However, only about 50 of the thousands of known algal species produce the toxins responsible for these deaths. Other algae have indirect effects on organisms by changing local environmental conditions.

In general, explains Sellner, most harmful algal blooms are caused by microscopic species of algae. "A bloom occurs when an alga rapidly increases in numbers to the extent that it dominates the local planktonic or benthic community. Such high abundance can result from growth caused by a response to a particular stimulus," such as excess nutrients in the water or another environmental condition (a change in water temperature, for example); it can also



A clean-up crew in Tampa Bay, Florida, surrounded by dead fish during a red tide. Photograph: Karen A. Steidinger.

Weather Report
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Seabird Invasion Hits Coastal Homes

He Got The Bird—Or Was It Vice Versa



Thousands Of Birds
 Floundering In Streets

Did Lights Bring Birds Ashore



Agency Okays
 SL Park Pact
 With Shaffer

University May Use Cabrillo
 Facilities To Open Its Doors

The headlines that inspired Alfred Hitchcock's movie *The Birds*. Reprinted with permission of the Santa Cruz Sentinel.

result from the physical concentration of a species in a certain area because of patterns in water circulation.

All HABs once were referred to as "red tides," Sellner says, "but the description has become a misnomer because not all HABs are red." Some may be brown, yellow, or green, and some may not change the color of the water at all. Blooms of cyanobacteria, like those in Souleat Loch, are visible as floating green scum; species in the genus *Aureococcus* or *Aureoumbra* turn coastal waters chocolate brown; and the dinoflagellates *Alexandrium* and *Karenia* cause traditional red tides. However, the chlorophyll-free, fish-killing dinoflagellate *Pfiesteria piscicida*, subject of Rodney Barker's best-selling book *And the Waters Turned to Blood*, is invisible to the naked eye.

The Plot

It's a scene straight out of Alfred Hitchcock's movie *The Birds*. In the summer of 1961, shearwaters, oceanic birds that feed on anchovies, flew not over the ocean but inland toward the city of Capitola, California, dying by the dozens as they crashed into buildings and cars. "The anchovies in waters off Santa Cruz were poisoned by a toxin from harmful algae,

a bloom of a species named *Pseudo-nitzschia australis*," according to David Garrison, an oceanographer at the University of California—Santa Cruz who first recognized the possible connection in the early 1990s. "The planktivorous fish had eaten the algae, which produce the toxin domoic acid, and the birds in turn had eaten the fish." Domoic acid, a neurotoxin, affected the birds' ability to fly.

Alfred Hitchcock, who lived in nearby Scotts Valley, California, at the time of the incident, used it as the basis for his classic thriller about a coastal town terrorized by deranged birds. Less than two years after the 1961 Capitola seabird deaths, *The Birds* hit movie theaters across the country.

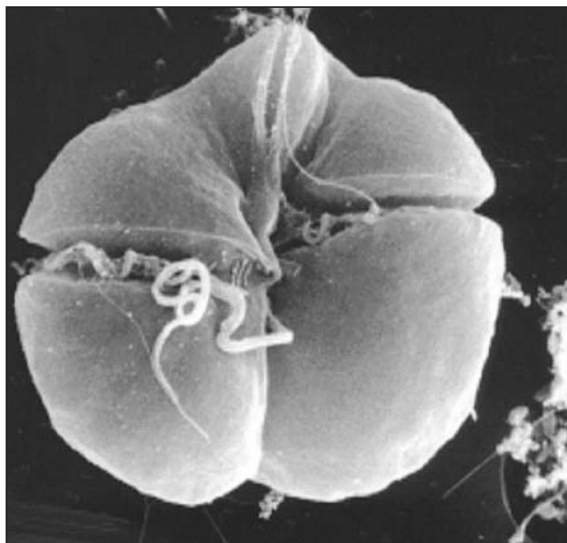
The detrimental effects of harmful algal blooms, according to biologist Donald Anderson of the Woods Hole Oceanographic Institution in Massachusetts, can range from cell and tissue damage to the death of an animal. "Usually it's the production of toxins by these algae that kills other organisms," Anderson says, "but effects can also be the result of localized anoxic conditions created by the bloom. A bloom can affect the entire coastal ecosystem, from

phytoplankton to zooplankton, to fish and shellfish, to people." In recent years, more than 50 endangered Florida manatees and several endangered northern right whales have succumbed to toxins produced by harmful algal blooms.

Among the best-known resulting human afflictions are ciguatera fish poisoning, neurotoxic shellfish poisoning, paralytic shellfish poisoning, diarrhetic shellfish poisoning, and amnesic shellfish poisoning. Cyanobacterial blooms produce liver toxins as well as nerve toxins. In February 1996, patients at a hospital in Caruaru, Brazil, died from cyanobacterial toxin poisoning when the hospital's water filtration system failed to screen out a blue-green algal bloom.

Blue-green algae are a recurring problem in Lake Champlain, Vermont, according to Suzanne Levine, a biologist at the University of Vermont in Burlington. "Bloom detection is critical," says Levine, "because of the danger posed to human swimmers and pets like dogs who contract 'swimmer's itch' from the toxin, but especially because many northeastern US lakes, like Champlain, are important sources of drinking water."

Exposure to toxins produced by harmful algal blooms can include gastro-



Scanning electron micrograph of *Karenia brevis*.
Micrograph: Florida Marine Research Institute.

intestinal, neurological, cardiovascular, and hepatic (liver) symptoms. In marine and estuarine waters, fish and shellfish are central to the pathways of these illnesses from algae to humans. The toxins concentrate in fish and shellfish that ingest the harmful algae, and people (and marine mammals) become sick when they eat these animals.

Some harmful algae produce toxins with no ill effects on humans, says Sellner, but devastating effects on fish. The species *Heterosigma akashiwo* makes a toxin lethal to fish, resulting in threats to penned finfish in aquaculture operations.

How are these algal villains getting away with increasing instances of their crimes? Are humans somehow involved in the plot, a plot that costs the US economy more than \$50 million a year?

To find out, Congressman Vernon Ehlers (R-Michigan) and the subcommittee he chairs, the Environment, Technology, and Standards subcommittee of the US House of Representatives, recently approved legislation addressing harmful algal blooms and hypoxia in coastal waters. "This legislation will provide an updated research framework for addressing a nationwide problem," says Ehlers. "The programs in this bill will improve our ability to understand and predict harmful algal bloom events, and ensure the participation of local man-

agers in developing research plans so that the results can be fully utilized by everyone concerned with these important issues."

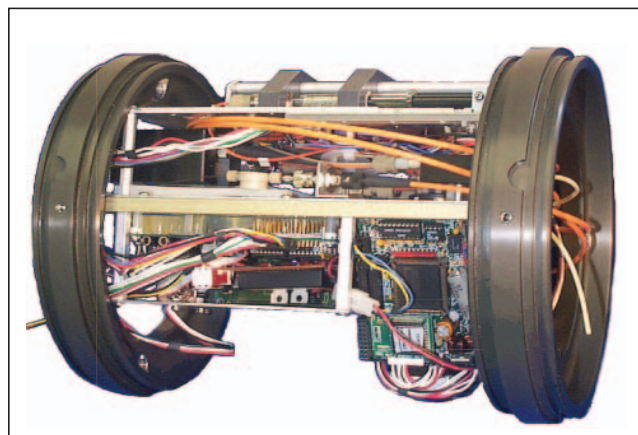
The answer to the question of whether people are contributing to the problem is a resounding yes, believes scientist Karen Steidinger of the Florida Marine Research Institute in St. Petersburg, part of the Florida Fish and Wildlife Conservation Commission. "In certain places," she says, "there are strong relationships between harmful algal blooms and human-caused nutrient enrichment, the Gulf of Mexico being among the most problematic

areas." High nutrient loads from agricultural activities upstream in the Mississippi River, and the resulting excess nutrients that flow out and into the Gulf of Mexico, in turn fuel growth of algae like *Karenia brevis*, a species named for Steidinger.

In a situation similar to that of the Gulf of Mexico, blooms of *Pfiesteria* along the US East Coast probably resulted from nutrient-laden runoff from hog farms and sewage discharge. Nutrient enrichment from human activities also has severely affected Japan's Inland Sea and Hong Kong's harbor, sites where massive blooms of harmful algae have occurred.

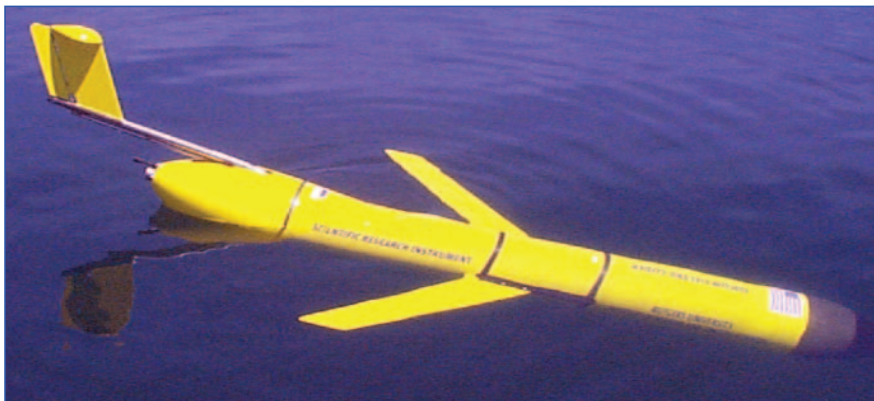
In addition to the problem of excess nutrients in coastal waters, introduction of exotic species through exchange of ships' ballast water and through aquaculture activities may be the source of some harmful algal blooms. "Increased pollution, habitat degradation, alterations in natural water flows, and climate change also are all leading contenders for the reason...harmful algal blooms have increased in recent decades," Steidinger states.

Finally, marine animals like zooplankton, the grazers of the seas, may be involved. Evidence exists that so-called brown tides in Rhode Island's Narragansett Bay, for example, were caused when reduced grazing by zooplankton allowed algae blooms to develop.



The inner workings of the recently deployed miniature, in-water optical phytoplankton discriminator. This configuration will serve as the standard design for AUV (autonomous underwater vehicle) installation.

Photograph: Gary Kirkpatrick.



The Slocum Glider (Webb Research Corporation), fitted with an optical phytoplankton discriminator (OPD), beginning its first open-water mission off the coast of New Jersey, 20 May 2003. The OPD payload is housed in the vehicle's midsection where the wings attach. Water intakes are located below the trailing edge of the wings. Photograph: Gary Kirkpatrick.

The Resolution

"To sense this world of waters known to the creatures of the sea," wrote Rachel Carson in *Lost Woods: The Discovered Writing of Rachel Carson*, "we must shed our human perceptions of length and breadth and time and place, and enter vicariously into a universe of all-pervading water."

That's exactly what oceanographers studying harmful algal blooms are trying to do, says Tim Orsi, a scientist at the National Coastal Data Development Center (NCDDC) in Stennis Space Center, Mississippi. NCDDC is part of the US National Oceanic and Atmospheric Administration (NOAA).

"With their ingenious ways of infiltrating coastal waters, it's a big challenge to develop an advance warning system for HABs," says Orsi. "We still have a long way to go in our basic understanding of the species responsible for blooms, and how they fit into the overall marine ecosystem. However, we're making significant progress toward the much-needed ability to predict bloom events, thanks to new technologies now coming on-line. These technologies will allow us to be 'in the water' monitoring conditions 24 hours a day, seven days a week."

Orsi is coordinator of a Gulf of Mexico-wide project called HABSOS, or Harmful Algal Blooms Observing System. "The goal of HABSOS," explains Orsi, "is to collect data from far-flung sources—university researchers; federal,

state, and local governments; and industries like fishing and aquaculture—and make these data compatible so a user can easily see patterns in time and space. This information will allow us to determine when and where a bloom is occurring and, eventually, when and where a bloom might occur—before it happens."

University researchers and government officials, which include US Environmental Protection Agency scientists and state government officials in Mississippi, Alabama, Florida, and Louisiana, as well as those in other nations (such as Mexico), are involved in HABSOS. They plan to meld data from satellite observations of ocean color (as measured by concentrations of chlorophyll) and information from a network of buoys outfitted with instruments that track currents, water temperature, and salinity to come up with answers. "We'd like to design sensors that collect biological information, as well," adds Orsi. "These sensors might consist of things like probes placed on buoys; the probes would be capable of determining what algal species are in the water column and whether they're producing toxins." Adds Russ Beard, chief scientist at NCDDC, "We're on the cusp of exciting, and somewhat futuristic, instrumentation."

A related NOAA effort, MERHAB (Monitoring and Event Response for Harmful Algal Blooms), and an inter-agency project, ECOHAB (Ecology and

Oceanography of Harmful Algal Blooms), are also working to transfer state-of-the-art HAB research into management tools for detection of blooms by officials in local and regional jurisdictions.

Scientists involved in HABSOS, MERHAB, and other such programs gathered at a March 2002 workshop sponsored by the Alliance for Coastal Technologies (ACT), headquartered at the Chesapeake Biological Laboratory (CBL) in Solomons, Maryland. More than 50 participants discussed new ways of monitoring and detecting harmful algal blooms with novel biosensors.

"Many of the technologies recently developed," explains Mario Tamburri, ACT chief scientist, "have remained research tools, used in ongoing, science-based experiments and programs. As such, these technologies usually aren't available or incorporated into the coastal monitoring programs of states, counties, and localities. This transition from a specific use in individual-scientist, coordinated research to larger, routine coastal monitoring programs was the main reason for convening the workshop."

Funded by NOAA's Coastal Services Center in Charleston, South Carolina, the workshop brought together scientists who had developed HAB research tools, state management officials responsible for assessing living resource and coastal ecosystem health, and industry representatives interested in providing commercial products for private and public users.

"In the last five years, tremendous technological advances have been made in the detection of HAB species and toxins as a result of research projects in academic and federal laboratories," says Ken Tenore, director of CBL. Areas in which these advances have happened include "molecular-based" detection, in situ and hand-held sensor platforms, detection of specific pigments, detection of toxins, and in vitro bioassays.

Molecular-based detection of individual HAB species, states the workshop report by the University of Maryland Center for Environmental Science (UMCES Technical Report Series, TS-374-02-CBL), has progressed extremely rapidly. There are now species-specific

probes for numerous taxa common in US waters and in those of other countries. Such biosensors are based on either nucleic acid probes that detect intracellular genetic signatures or antibodies that target cell surface antigens. "Some of these techniques," says Sellner, "are starting to be used in routine monitoring programs, which has resulted in a much faster assessment of potential problems than does conventional microscopy." Examples include detection of the brown-tide organism *Aureococcus anophagefferens* in Long Island Sound and identification of *P. piscicida* in the mid-Atlantic region.

In addition to laboratory-based application of molecular probes, says Tamburri, "a considerable effort is under way to adapt these methods for use on both in situ and handheld sensors." The deployment of in-water sensors for individual HAB species is being tested with an instrument called the Environmental Sample Processor (ESP), a prototype developed by biologist Chris Scholin of the Monterey Bay Aquarium Research Institute. Using a watertight submerged system with software-controlled pumping, filtering, and sample processing, the ESP is capable of autonomously conducting assays and sending the digitized data to a land-based laboratory via radio modem. The ESP also has sample archiving capabilities, thus providing material for toxin testing, microscopic examination, and additional genetic analyses. Molecular-probe-based sensors are also being developed by scientists at the Alfred Wegener Institute in Bremerhaven, Germany, for use with handheld DNA microchip readers. "Such sensors," adds Sellner, "show good potential for use in both laboratory and field-based detection of HAB species within the next couple of years."

The detection of specific pigments associated with HAB species also has proven valuable for assessment of blooms. For example, satellite-based detection of *K. brevis* blooms off western Florida is now possible, using a species-specific algorithm. Suzanne Levine is working on a related way of determining the extent of blue-green algal blooms in Lake Champlain using satellite images

that reflect light at certain wavelengths when phycocyanin, a pigment unique to cyanobacteria, is present.

"In addition to the detection of HAB species," Tamburri says, "detection of the toxins produced by these organisms is of critical importance to HAB monitoring programs." HAB toxin detection and quantification is easily done in the laboratory, Sellner adds, with sensitive analytical techniques like high performance liquid chromatography and mass spectrometry. "What would take this an important step further is to apply these methods to in situ platforms." Deployment of underwater mass spectrometers is being tested by the Center for Ocean Technology at the University of South Florida. Scientists there are working on ways of configuring the instruments for algal toxin detection.

In addition to analytical approaches, continues the ACT workshop report, "HAB toxins and HAB-produced compounds can be detected with various in vitro bioassays, including those based on a toxin's activity (cell-based assays and receptor-binding assays), and those relying on the structure of a toxin molecule (immunoassays)." An inability to maintain receptors or cells under the conditions experienced on in-water platforms, however, "makes in situ deployment of these assays unlikely in the near future," workshop participants concluded. But research on how to configure immunoassays for the ESP in situ platform continues.

Among the more unusual ways of detecting and monitoring harmful algal blooms has been designed by marine scientist Gary Kirkpatrick of the Mote Marine Laboratory in Sarasota, Florida. Kirkpatrick is testing robotic undersea gliders—autonomous underwater vehicles, or AUVs—carrying miniature sensors that detect cells of *K. brevis* and other species and collect data on water temperature and salinity. The data are beamed back to a lab via satellite. The glider then wings off on a new course, searching for other likely HAB-affected areas. Kirkpatrick's goal is to develop a way to alert coastal regions well ahead of time that a red tide is imminent.

Then there's a sensor that makes toxin-containing waters glow. Roger Leblanc at the University of Miami has found a substance that lights up only in water in which an algal toxin called saxitoxin is present. Saxitoxin is among the most potent of the toxins produced by HABs. Leblanc has discovered molecules, called crown ethers, that recognize saxitoxin but ignore nontoxic compounds in coastal waters. Crown ethers emit a fluorescent compound that glows more brightly in ultraviolet light when saxitoxin is present. The scientist hopes to incorporate these molecules into a monitoring device that could be used in field research as well as inshore laboratories.

"The goal of all these sensors," says Sellner, "is to be in the water with the algae continuously. It's the only way we'll better understand harmful algal blooms, why they happen,...and what we might be able to do about them."

Epilogue

Today's sophisticated new technologies for detecting and monitoring HABs notwithstanding, there may be a place in science for a bit of folklore.

To the people who live around meres, or small shallow lakes, in northwest England, a dependable way of forecasting the end of a blue-green algal bloom is to watch for "the breaking of the meres," explains Geoffrey Codd. "Blue-green algae in these meres form bundles and coils of colonies, becoming a mat of green virtually overnight and completely obscuring the water's surface. But they disappear just as quickly, first cleaving neatly in two and creating something of a pathway across the mere, in which open waters once again can be seen."

For centuries, the breaking of the meres has been a magical event to those who live in northwest England. Local residents believe that an unseen force reaches out to end the scourge.

Science can't tell us, at least at this time, just how it is that the meres return to their original state—in the blink of an eye. Perhaps biosensors can provide the answer.

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