PHOTO BY DR. ZHANG LU, NANJING INSTITUTE OF GEOGRAPHY AND LIMNOLOGY

Quick—what's polluting the water?

BY ANGELA SPIVEY

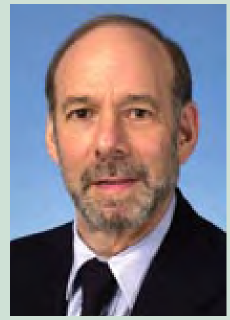
UNC School of Public Health researchers develop faster, better ways to warn when our waterways pose a danger to people or fish

At least a couple things can make a river or ocean unfit. For people, it's microbes — such as disease-causing bacteria, viruses, and protozoan parasites — that contaminate water when a sewer pipe breaks or a hog-waste lagoon fails. For fish, the danger can lie in nutrients, such as nitrogen, which are washed into waterways when wastewater is pumped in or runs off from fertilized land. Excess nutrients can cause overgrowth of algae. Algal blooms can upset the water's balance, causing dangerously low oxygen levels in some parts of the water.

For both these problems—microbial pathogen contamination and excess nutrients—Carolina School of Public Health researchers have been developing better, quicker ways to know when water is polluted.

On Monday, July 1, 2005, the thing you hope never happens did: a main sewer line from Wilmington, N.C., ruptured, spilling several million gallons of raw sewage into Hewletts Creek, a small tributary of the Cape Fear River. Swimming in water contaminated with sewage can cause diarrhea, abdominal cramps and skin infections. The state closed all the waters between the Wrightsville Beach bridge and the Intracoastal Waterway near Peden Point: no fishing, shellfishing or swimming.

To advise the state, scientists at UNC-Wilmington sampled the waters and performed tests for bacteria that are found in sewage, particularly *E. coli*. They also sent some bottom sediment samples to Dr. Mark Sobsey, Kenan Distinguished University Professor of environmental sciences and engineering at Carolina's School of Public Health and director of the School's Environmental Virology and Microbiology Laboratory. Sobsey tested the sediment for coliphages, which are viruses that infect *E. coli*. Coliphage levels are an indicator of fecal contamination and are good predictors of the presence of human enteric viruses, such as noroviruses and hepatitis A. Testing sediments was important since contaminants can persist in sediments, re-contaminating surrounding waters when disturbed by swimmers and changing weather conditions.



Dr. Mark Sobsey

By July 4, the creek and surrounding waters were still closed. "The state and the local authorities were under tremendous pressure to open the beaches back up," Sobsey says. But he and his colleagues advised them to wait until coliphage levels were back to normal.

How long would that take? "We said, 'We'll have to keep measuring,'" Sobsey recalls.

The problem was, whenever the scientists brought a sample into the lab, with available

PHOTO BY RICK DOVE/WWW.DOVEIMAGING.COM

Aimee Trombley (right), age 6, of Carolina Pines, N.C., goes for a run along the south shore of the Neuse River below Carolina Pines.

Dr. Hans Paerl (previous page) samples a blue-green algae (cyanobacteria) bloom on Lake Taihu, in China, where he conducted research in July 2007. This lake has been impacted by toxic cyanobacterial blooms caused by excessive nutrient deposits from wastewater, industry and agriculture.

tests, it took a full day to get results—another full day of waiting. In the end, the state didn't lift the swimming and fishing advisory until July 10.

Wouldn't it have been great if scientists could sample water in the morning, then have an answer by afternoon?

When such an accident happens again, it's likely they will. Carolina School of Public Health researchers have had some promising successes developing quicker tests to detect fecal contamination.

Rapid DNA Detection

Rachel Noble, associate professor at the UNC-Chapel Hill Institute of Marine Sciences with an adjunct appointment in the Department of Environmental Sciences and Engineering at Carolina's School of Public Health, has developed rapid tests that detect the DNA of two different kinds of bacteria found in fecal matter—*Enterococcus* and *E. coli*. Both tests can be completed in less than two hours. "That means you could go out to the beach, take a water sample at 7 a.m., and by 9 a.m., you could close that beach with a warning sign if indeed it should be closed," says Noble, who is also on faculty at the UNC Institute for the Environment and directs the Institute's Morehead City (N.C.) Field Site.

Right now the U.S. Environmental Protection Agency (EPA) is evaluating Noble's *Enterococcus* test for approval to use in marine waters. The *E. coli* test isn't far behind, she says.

PHOTO BY LINDA KASTLEMAN

David Love, a 2007 doctoral graduate from the School's Department of Environmental Sciences and Engineering, conducts a laboratory experiment. Love has developed a rapid fecal indicator test that can be used to monitor the microbiological quality of drinking, recreational and shellfishing waters.

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Noble's tests not only are quicker; they also are less expensive than some rapid assays in development. They avoid using a particular chemistry technique that carries a very high licensing fee. A rapid test isn't much good if the state of North Carolina, for instance, can't afford to use it. "Most water quality agencies operate on very small budgets," Noble says. "They have their hands tied as far as how much they can afford to spend."

Noble is also at work on a rapid test for a bacterium that is native to North Carolina's estuarine and coastal waters—*Vibrio vulnificus*. "This organism has caused deadly wound infections in people who are immunocompromised, such as people who are susceptible to infection, like people with diabetes," Noble says. "There have been some deaths in North Carolina as a result."

The test would be especially useful in waters used for shellfish harvesting. The bacterium can cause disease when people eat infected shellfish or swim in infected waters with an open wound.

A sudden success

Back in Sobsey's lab, David Love, a 2007 doctoral graduate in environmental sciences and engineering, has made a breakthrough in developing a three-hour test to detect coliphages (the viral pathogens that infect *E. coli*).

Before this success, Love had spent nine months looking for a DNA or RNA test for coliphages, without making much headway. Sobsey tells the story. "David came to me and said, 'I'm now thinking I should look at proteins—antigens. Has anybody tried what's known as particle or latex agglutination tests for this?'" Love knew that such tests are used in medical diagnostic labs to detect viruses in human fecal specimens.

PHOTOS BY THE NASA/GSFC/LARC/JPL, MISR TEAM

Satellite images such as these of the North Carolina coast (left) and Pamlico Sound (right), can tell researchers a lot. Satellites combined with bioptical sensors can provide data about the color of water, which can be used to tell scientists about water quality. UNC researchers are working with colleagues to use this technology to identify harmful algal blooms and clarity problems that might adversely affect seagrass, fish and shellfish habitats.

Sobsey told Love the idea was intriguing and had not been tried before. He suggested Love test it in the lab right away.

"Within two weeks, he had positive results: conceptual proof this would work," Sobsey says.

Coliphages are good candidates for a rapid test, because they're viruses which multiply quickly. "One virus makes thousands to tens of thousands within an hour," Sobsey says. But the challenge was developing a way to detect the viral growth almost immediately.

Love's test uses antibodies (immune proteins that attach to the coliphages) that are attached to latex beads. When the antibody-labeled beads are added to a water sample, if coliphages are present in the water, the antibodies (on the plastic beads) will stick to the coliphages. A positive result is easily visible as the plastic beads clump together in just a few seconds. The visual read-out and simple methods make this a good candidate for further development as a field-portable kit.

Love's success came just in time for him to use the research this summer in two studies of beach water quality and swimmer health in Orange County, California, and Fairhope, Alabama, and in his doctoral dissertation, which he defended in April 2007. The initial paper describing the new method was published in the July 2007 issue of *Applied and Environmental Microbiology*, one of the top journals in the field. It can be found online at *http://aem.asm.org/cgi/content/full/73/13/4110*.

Seeing safe water

Dr. Hans Paerl, Kenan Distinguished University Professor of marine and environmental sciences at UNC's Institute of Marine Sciences in Morehead City, N.C., and a joint professor in the Department of Environmental Sciences and Engineering, has long worked to improve sampling and other methods to continually monitor water conditions, so that when water problems occur, officials can intervene quickly.

Paerl co-directs FerryMon (*www.ferrymon.org*), an automated water-quality monitoring system aboard the North Carolina Department of Transportation ferries that cross the Neuse Estuary and Pamlico Sound. FerryMon has been monitoring these bodies of water 365 days a year since 2000. "The ferries are out there every day, intercepting the waters as they move down the Neuse and the Pamlico, and they can give us highly informative data that would not be obtainable with weekly or monthly monitoring programs," Paerl says. "If you suspect there is a problem—for instance, reports of people having rashes or diarrhea after having been in the water—FerryMon would be our early-warning tool to immediately collect space-time intensive samples and analyze them for potentially harmful explosive growths or "blooms" of algae and

UNC researchers are working with colleagues to combine water sampling data with satellite and aircraft data to develop graphic indicators of water quality that could provide rapid warnings of potential problems due to algal blooms.

PHOTO BY ALAN JOYNER/UNC-CHAPEL HILL INSTITUTE OF MARINE SCIENCES

PHOTO BY DR. HANS PAERL

other water quality indicators that might adversely affect human health."

Paerl also is working with colleagues to combine water sampling data with satellite and aircraft data to develop graphic indicators of water quality that could provide rapid warnings of potential problems due to algal blooms. Doing this requires that Paerl, who's an aquatic ecologist, work with scientists who understand and interpret remote sensing images generated by optical sensors. "Collaborations with others outside your field make the whole greater than the sum of its parts," Paerl says. "We're trying to put our resources together with our partner at the EPA, Ross Lunetta, to address issues such as harmful algal blooms, problems with optimal transparency and clarity problems that might adversely affect seagrass, fish and shellfish habitats."

Satellites use bioptical sensors that provide data about the color of the water, which can be used to tell scientists about the water's quality. The color of water isn't random; it's determined in large part by its quality.

TIP: Join a community group to help restore a stream or clean a beach. For more information, visit: *www.nrcs.usda.gov/technical/stream_restoration.*

Chlorophyll, the main pigment in plants and algae, turns the water green. Dissolved organic matter and sediments in the water turn it brown. In the Pamlico, for instance, a NASA satellite produces a snapshot of the water's transparency and clarity every few days. FerryMon provides daily measurements of water quality to calibrate satellite images. A computer can use mathematical algorithms to correlate both sets of data. What emerges are images whose colors correspond to various levels of water quality. So by checking the latest satellite images, researchers could instantly "see" today's water quality.

"We can look at color with a satellite and then use our indicators to calibrate it so we can scale it up for the entire system," Paerl says. "It's called groundtruthing: using real data from our monitoring programs to allow the optical sensors to be scaled up to apply to the entire estuary."

Paerl is seeking funding to try to do that. Because such a tool could provide an early-warning system, he says, "it would be very useful for looking at harmful algal blooms such as red tides or blooms produced by toxic blue-green algae—being able to spot them before they cause serious problems, or warn the public of potential health problems." ■

PHOTO BY RICK DOVE

Dr. Hans Paerl (above, left) filters algae samples incubated with nutrient additions to determine the effects nutrients have on algal bloom formation and growth rates. The samples were taken from Florida's St. Johns River, which periodically experiences algal blooms.

A ferryboat crossing North Carolina's Pamlico Sound (above, left) not only transports people and cars but also gathers water samples as part of FerryMon (*www.ferrymon.org*), an automated water quality system co-directed by Dr. Hans Paerl, UNC Kenan Distinguished University Professor of marine and environmental sciences.

A pelican swoops in for a landing on North Carolina's Neuse River (left).