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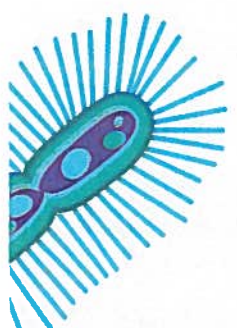
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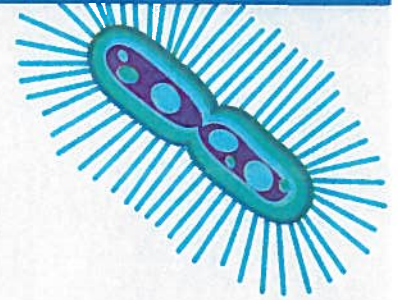
# BUSTING BACTERIA IN

by Lori Brandt

# WAST



NEPTUNE DIAGNOSTICS USES DNA-IDENTIFICATION TECHNIQUES TO ALERT V



**It helps diagnose genetic disorders, identify viruses and even match criminals to crime scenes. Medical and forensic scientists routinely rely on polymerase chain reaction (PCR), a fast and inexpensive technique that amplifies, or copies, small segments of DNA to gather information on a molecular and genetic level.**

A new startup company in Calit2's business incubator TechPortal is taking this DNA-identification technique to the wastewater industry. Called Neptune Diagnostics, the small company is developing a PCR-based product that rapidly detects and quantifies troublesome bacteria lurking in wastewater. It received a \$5,000 seed grant from the National Collegiate Inventors and Innovators Alliance, followed by a \$150,000 Small Business Innovation Research grant from the National Science Foundation to test and commercialize its product, B&F Alert.

As California enters the fourth year of an unprecedented drought, water officials are looking for alternative sources and new technologies to mitigate shortages.

# EWATER

Recycled wastewater is an integral part of the state's water supply. Indeed, Orange County's pioneering Groundwater Replenishment System (GWRS) produces up to 70 million gallons of drinkable water every day. Turning sewage into potable water, however, is rigorous and requires a multistep process involving solids separation, aeration and disinfection.

Neptune Diagnostics' B&F Alert is a diagnostic test that identifies bulking and foaming organisms early in the treatment process, before they can cause disruptions in the system. (Bulking and foaming refer to what happens to the solids, or sludge, when filamentous bacteria prevail, stopping or slowing down the solid/liquid separation process.)

When bulking and foaming occur, it's costly for wastewater treatment plants. They may incur regulatory violations and fines, or they could experience increased operating costs from the purchase of additional chemicals and higher labor. Perhaps the largest impact, though, is on energy production; most plants recover methane, a sustainable fuel, using it to generate electricity or to power motor vehicles. Foaming lowers methane production, requiring plants to draw electricity from the carbon-emitting grid. Other potential impacts include public health and economic losses that occur when beaches or recreational waters must be closed due to bacteria being discharged from wastewater plants.

*Professor Betty Olson conducts research on how microorganisms influence water quality and how to use molecular biological techniques to optimize wastewater treatment. Olson is the associate director of UCI's Water-Energy Nexus Center.*



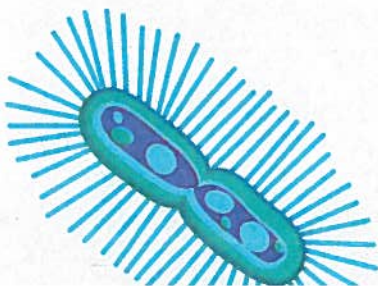
photo: Paul R. Kennedy

The idea of using PCR with wastewater was born in the lab of civil and environmental engineering professor Betty Olson, whose research expertise is in molecular techniques as well as the microbiology of drinking and wastewaters. "Right now, the way the industry diagnoses these organisms is with a microscope, and they can only be identified after they've become a problem," Olson explains. "So treatment plant operators use harsh chemicals like chlorine, which kills everything — the good and bad bacteria," she says.

When she was a doctoral student in Olson's lab, Pitiporn Asvathanagul completed a proof-of-concept study and wrote her dissertation on the idea. Today, she is an assistant professor of civil engineering and construction engineering

management at California State University, Long Beach. Her research came to the attention of Paul Merage Business School student Joseph Nadolski in 2013 during the Merage Business Plan Competition.

The B&F Alert didn't win any major prizes, but it intrigued Nadolski, a retired U.S. Army lieutenant colonel and a civil engineer who is interested in clean technology and in commercializing innovations to improve the environment. After earning an executive MBA, he figured he'd find the project's students and see if he could help. UCI's Office



of Technology Alliances told him they all had graduated. The technology was available for development, so Nadolski nabbed it and has applied for a license.

Nadolski started Neptune Diagnostics, with Olson as an adviser. He hired Xuan He, an environmental engineer, and Trisha Westerhof, a doctoral candidate in molecular biology and biochemistry. They've recruited three demonstration customers: Victor Valley Water Reclamation Authority, Santa Margarita Water District and the city of Simi Valley.

Nick Steffen, an operations supervisor at Simi Valley's wastewater treatment plant, will begin collecting samples to send weekly to Neptune Diagnostics for analysis. "Over the course of a year, we will have a good baseline of data. What we are hoping to do with Joe is to quantify specific bacteria, and when we see a certain threshold reached, we'll be able to make process control changes. We need to establish what the normal amount is, then if we see that number increase, we'll know to intervene."


B&F Alert is the company's launch product, but the platform could be used for multiple examinations. He says that they could potentially test for 96 organisms at once, and water is just the first sector. In time, they could move into agriculture and food manufacturing.

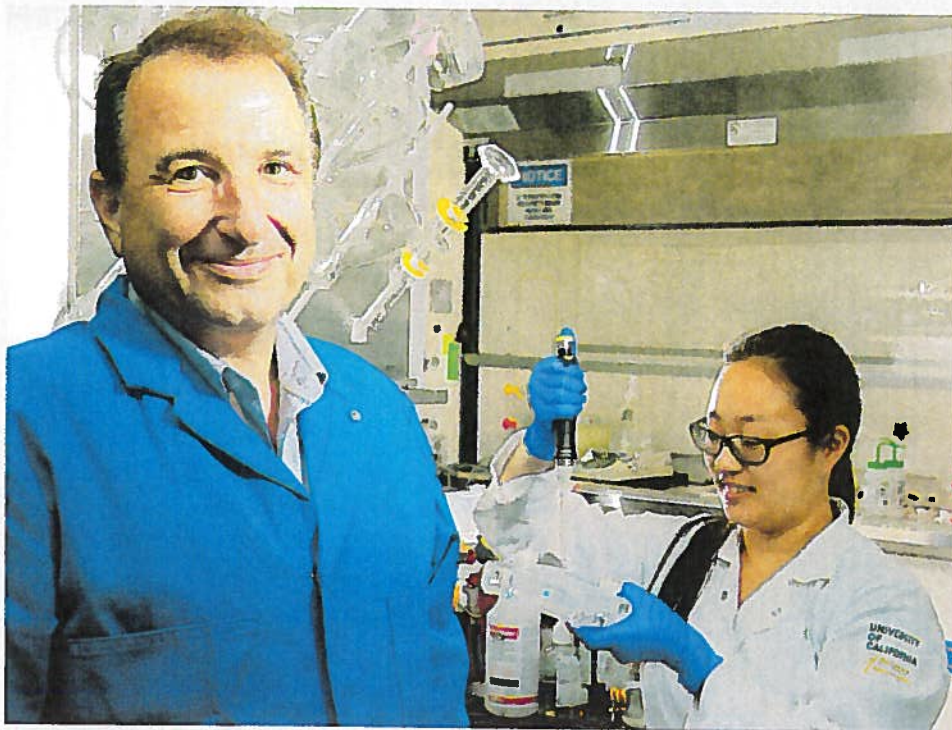
"There are a lot of choices for us. We want to see what wastewater needs first, right now," says He, who earned her master's degree in environmental engineering at UCI in 2014.

"Ours is more of a business model innovation," Nadolski explains. "This is old technology in the medical field, where they will spend a lot of money for a diagnostic test. Our goal is to get the cost point to where it could be attractive to public utilities and other industries."

"One great challenge will be to convince current wastewater operators, engineers and lab personnel of the benefits associated with the use of metagenomics (applying molecular diagnostic techniques to study DNA extracted directly from environmental samples)," says Victor Santa Cruz, a biologist with Inland Empire Utilities Agency and an informal adviser to Neptune. "In many ways, overcoming entrenched operational-engineering philosophies will be like teaching an old dog new tricks."

Nadolski agrees that the company has to consider the mindset of the wastewater operators. "They're conservative. They've been doing this for years by gut, and right now, they react. This will give them new tools allowing them to anticipate and be preventative."

"This is an early diagnostic test," reiterates Olson. "You can see the advantage of being able to diagnose something like cancer when you have just a few cells versus when you have a tumor already formed. That's what this is, an early warning signal." 



*Neptune Diagnostics CEO Joe Nadolski and graduate student researcher Trisha Westerhof are working to launch a product that would help wastewater plant operators use DNA instead of a microscope to identify problem levels of bacteria.*

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