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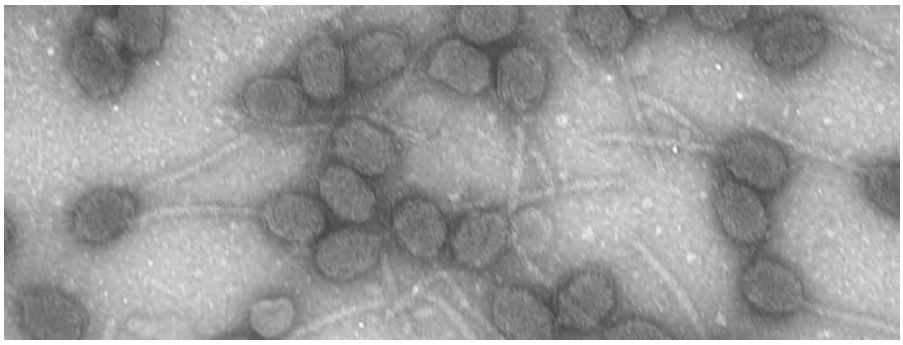

Oysters and clams and bacteriophages (oh my!)

As Americans continue to increase seafood consumption in their diets, detection of contaminated shellfish is a growing concern for public health. Rigorous food safety testing is particularly important for bivalve mollusks (which include oysters, clams, and mussels), because they are commonly consumed raw or partially cooked. According to the CDC, between 1998 and 2008 there were an estimated 280,000 cases of foodborne disease attributed to contaminated mollusks.

Current tests to detect contamination in mollusks are slow and laborious. They require isolation and culture of potentially contaminating bacteria, which takes several days. But a team of Cornell Veterinary Medicine and Food Science researchers led by Professor Hélène Marquis in the Department of Microbiology and Immunology has received a new grant to develop a much faster diagnostic test for the detection of virulent pathogens that infect bivalve mollusks. The research is being funded through the Northeastern Regional Aquaculture Center at the University of Maryland, which receives support from the USDA.

Mollusks are filter feeders, meaning they obtain food and nutrients by pumping seawater through their gills and filtering out small organisms. A a result, their digestive system (which is one of the parts we eat) is populated by thousands of bacteria and viruses from the environment wher they live. While most of these microbes are harmless to humans, contamination of oysters with a virulent strain of bacteria – known as *Vibrio parahaemolyticus, or V. parahaemolyticus* for short – can make people sick if consumed in contaminated seafood. Dr. Marquis and her team pla to engineer a diagnostic test to detect this harmful pathogen using a fluorescent marker to visualize the infection.

"The problem is that food-borne disease spreads with raw seafood," explains Dr. Marquis. The risk for seafood contamination depends in part on environmental factors, including tide levels and temperature. Levels of contamination increase when the tide is low or water temperature is warm. *V. parahaemolyticus* is a bacteria strain that is ubiquitous in sea water, and is responsible for over 4,500 cases of food poisoning in the U.S. each year according to CDC reports.

A key benefit of this new diagnostic will be the ability to detect *V. parahaemolyticus* infection at multiple stages of oyster production. A quick turnaround time could tell a farmer when the best time to harvest is to keep bacteria populations low. It could also be used as a cost-effective way for government agencies or seafood distributors to monitor food contamination. Most importantly, it will be much faster than current methods. Dr. Marquis says her new test will use "a simple and rapid fluorescent marker that could give results within hours."

Although Dr. Marquis was primarily trained as a bacteriologist, she diverted her research focus to study the microbiology of fish and aquatic animals when she took over direction of the Aquatic Animal Health Program at the college in 2014. During her sabbatical at the University of Toronto in 2015, she also began to study small viruses that infect bacteria – known as bacteriophages. It was then that Dr. Marquis realized that the bacteriophages she was studying could be utilized in a diagnostic tool for oyster contamination.

Oysters harvested from Savannah, GA, like those Dr. Marquis and her team will use for this study. Photo credit: Josh Hallett, Creative Commons

The test will capitalize on bacteriophages that specifically infect virulent *V. parahaemolyticus* strains. These bacteriophages will be engineered with DNA that encodes "instructions" for infected bacteria cells, directing them to produce fluorescent protein molecules that generate light when viewed under a portable illuminator or spectrophotometer. If the oysters are infected with *V. parahaemolyticus*, water samples collected from the inside the oyster shells will glow green after just a few hours of incubation – a far cry from the span of days it would take to detect the same infection using current methods.

To complete the study, Dr. Marquis and her team will collect water samples and oysters from different areas on the Atlantic coast – where most oysters in U.S. supermarkets come from. They will also receive virulent bacteria strains from clinical cases studied by collaborators at the University of New Hampshire. The main challenge of the study is to isolate bacteriophages that only infect virulent strains of bacteria, so that the test will not record false positives.

The end goal of the study will include testing the protocol in the field. Funding from the project will allow aquaculture specialists from Cornell Cooperative Extension to work with oyster farmers tc integrate the new strategy into current testing procedures. If all goes well, Dr. Marquis' new test will assure rapid detection of contaminated mollusks, and the rest of us will continue to enjoy oysters from the raw bar.

– Erin Nicklow, PhD candidate in Molecular Medicine

Featured Image: Bacteriophages purified by Dr. Marquis, imaged with electron microscopy. Photo credit: Hélène Marquis

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