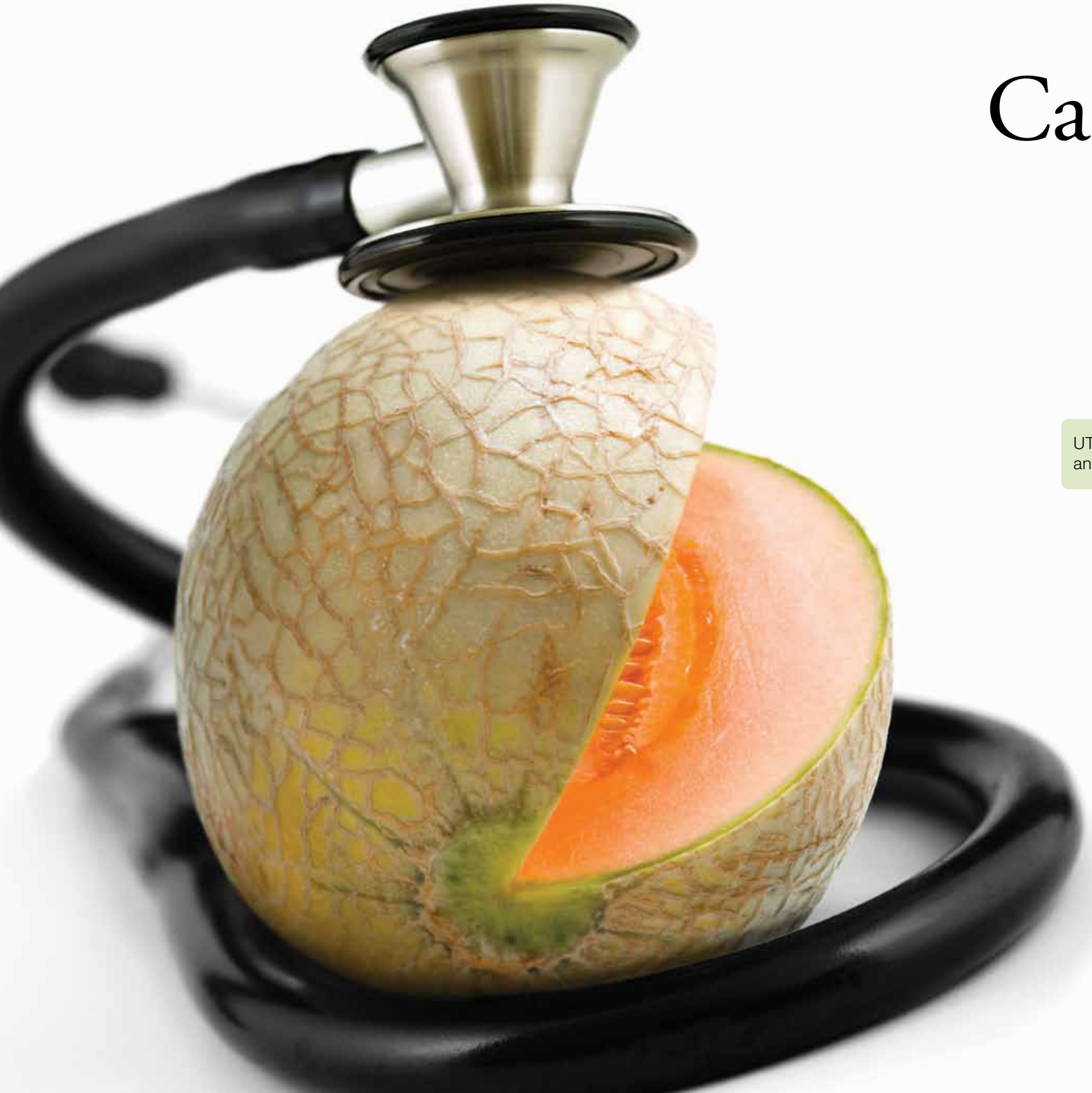


Can Foodborne Illnesses be Stopped?

UT Southwestern researchers are at the forefront of understanding and preventing the spread of dangerous bacteria in food.

You're looking for fresh produce at the grocery when a text comes across your smartphone. Holding the device away from your 3-year-old, you read a frantic message from your mother: "CANTALOUPES DANGEROUS!!! DO NOT BUY CANTALOUPES!!!" ● Turns out the media are reporting a Listeria outbreak. In addition to her text message, your mother called five times. She's panicking because her grandchild, your toddler, happens to love cantaloupe. ● The Centers for Disease Control and Prevention (CDC) estimates that each year foodborne illnesses sicken roughly one in six Americans, kill 3,000 and cause 128,000 to be hospitalized. The most commonly recognized bacterial pathogens are *Campylobacter jejuni* (*C. jejuni*), *Salmonella* and *Escherichia coli* (*E. coli*). ● After contaminated food is consumed, the bacterium incubates in the host before revealing itself through illness symptoms. During incubation the bacterium passes through the stomach into the intestine, attaching itself to the cells lining the intestinal walls.

By Erin Prather Stafford

> “Disease caused by foodborne pathogens can be anything from mild diarrhea in some individuals to more serious hemorrhagic diarrhea, kidney failure and stroke-like symptoms. The bacteria can even be fatal, especially for young children, the elderly and those with compromised immune systems.”
>
> *—Dr. Vanessa Sperandio*

The pathogen then multiplies, killing cells by stealing their nutrients. Some of the bacteria stay in the intestine, while others produce toxins that then are absorbed into the host’s bloodstream or even body tissues.

Many pathogens responsible for food-borne illness cause similar symptoms such as diarrhea, abdominal cramps and nausea. There is so much overlap that it can be difficult to identify what specific bacterium is causing illness without laboratory tests to identify the pathogen unless there is a recognized outbreak. Most cases are mild and can be treated by increasing fluid intake. But not all reactions are minor, and death is a tragic possibility.

“These bacteria are very potent; it only takes a few to get someone severely sick,” said Dr. Vanessa Sperandio, professor of microbiology and biochemistry at UT Southwestern Medical Center. “Disease caused by foodborne pathogens can be anything from mild diarrhea in some individuals to more serious hemorrhagic diarrhea, kidney failure and stroke-like symptoms. The bacteria can even be fatal, especially for young children, the elderly and those with compromised immune systems.”

Many antibiotics have become ineffective against foodborne pathogens and can potentially cause more harm by increasing the amount of released toxins. Instead of focusing on the bugs after they cause illness, UT Southwestern researchers are investigating how to neutralize the pathogens so they never have a chance to wreak havoc on the human body.

Exploiting Cell-To-Cell Signaling

Dr. Sperandio always has been fascinated with bacteria/host interactions. Much of her research has focused on *E. coli*, specifically the lethal enterohemorrhagic E. coli (EHEC) strain. The bacterium is primarily transmitted to humans through consumption of beef. However fecal contamination of water and other foods, as well as cross contamination during food preparation, also can cause disease. Recent *E. coli* outbreaks have been linked to beef, cheese, cookie dough, poultry, Romaine lettuce, spinach and sprouts.

Dr. Sperandio and her team are investigating two tactics to fight the pathogen. The first involves exploiting its cell-to-cell signaling to stop EHEC from colonizing in the intestines. Her lab has successfully identified two receptors required by the bacteria for infection of the host.

In 2006 she found the receptor QseC, a protein in the EHEC’s membrane. After a person ingests the bacterium, it travels peacefully through the digestive tract until reaching the intestine. Once there, the QseC receptor communicates with the human stress hormones epinephrine and norepinephrine, a signaling cascade begins and EHEC colonizes the intestine and causes disease.

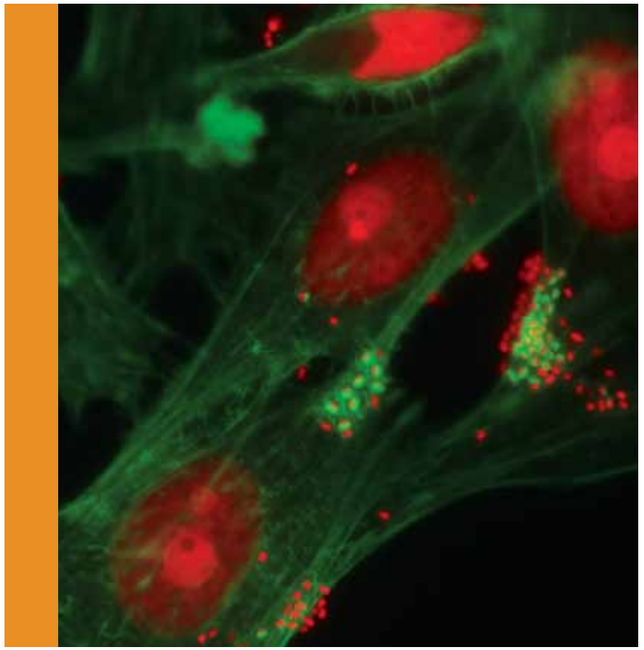
Searching UT Southwestern’s library of 150,000 small molecules, Dr. Sperandio un-covered LED209. Nontoxic to mammal cells, LED209 has proven successful in preventing QseC from initiating the virulence cascade and allowing EHEC to colonize.

She and research colleagues also are targeting EHEC in live cattle. An estimated 70 percent to 80 percent of herds in the U.S. carry the bacteria. Although deadly to humans, EHEC is a natural part of cattle’s normal gastrointestinal flora. It harbors a gene called *SdiA*, which makes the *SdiA* protein. The *SdiA* protein senses a chemical made by microbes in the animal’s rumen, the first of a cow’s four stomachs, which serves as a large fermentation chamber. Detecting this signal allows EHEC to pass through the rumen and colonize the recto-anal junction.

Dr. Sperandio’s lab injected two types of EHEC into the rumens of eight grain-fed adult cows. One mutant version lacked *SdiA* and could not detect the signal in the rumen. Another strain produced an enzyme that destroyed the chemicals in the rumen sensed by *SdiA*.

“If there’s no signal, then there’s no acid resistance, a requirement for the pathogen to make it to the recto-anal junction,” Dr. Sperandio said. “Everybody had thought that this type of signaling occurred naturally in the gastrointestinal tract of mammals. Our findings suggest we might be able to target this system to prevent food contamination from cattle. We can render EHEC harmless before it reaches humans.”

Dr. Sperandio also found that colonization diminished significantly when EHEC was unable to sense the rumen chemicals. The process prevented the bacteria from moving on through the stomach and colonizing. Her next step is to assess what happens to cows fed a grass-based, rather than grain-based, diet.



E coli, shown forming lesions on host cells, is primarily transmitted to people by eating beef.

Stopping Attachment

Bacterial pathogens face a number of challenges when trying to connect with human cells. Evading immune recognition, invading cell tissue and modulating cellular signaling are just a few tasks they must accomplish to ensure survival. Dr. Kim Orth, professor of molecular biology and biochemistry, is investigating whether cell damage caused by pathogens can be prevented by disrupting their initial attachment to host cells.

She and her team looked to the pathogen *Vibrio parahaemolyticus (V. para)* for answers. The bacterium, found in every ocean, can cause food poisoning through consumption of raw or undercooked shellfish, particularly oysters. Previous work by Dr. Orth had found that *V. para* kills its host's cells by causing them to burst, providing the bacteria with food used to fuel proliferation. To accomplish this nutrient robbery, the bacteria must successfully attach to the membranes of host cells.

"Bacteria are dependent on that union," said Dr. Orth, a W.W. Caruth Jr. Scholar in Medical Research. "We suspect many bacterial pathogens actually use a common mechanism to establish that first contact. Our goals are to understand this mechanism and then exploit it to develop therapeutic tools that completely shut down the pathogens' ability to colonize cells."

In investigating *V. para*, Dr. Orth's laboratory found that the protein MAM7 enables a wide range of pathogens (including *E. coli*, *Salmonella* and *Cholera*) to bind to host cells during the early stages of infection. When researchers deleted MAM7, *V. para* had difficulty binding to host cells and efficiently killing them.

They also engineered a harmless strain of *E. coli* to express MAM7. Exposing human cells to this harmless form of the bacteria caused human cells to become more resilient to V. para and other types of pathogenic bacteria. Dr. Orth is optimistic that the protein eventually could help prevent foodborne illness.

"It would be amazing to give susceptible children in pathogen-plagued areas a bit of yogurt containing some form of MAM7 and have it decrease their risk for serious complications," she said. "Bacteria have been evolving for a very long time. If we can figure out how they work, it could have an incredible impact on everyone in the world."

> "Bacteria have been evolving for a very long time. If we can figure out how they work, it could have an incredible impact on everyone in the world."
>
> —*Dr. Kim Orth*

> "It's known that as people get older they become more susceptible to infectious diseases and also that autophagy decreases."
>
> —*Dr. Beth Levine*

Autophagy Fights Back

Sometimes the answers to stopping bacterial pathogens are found in the cell's own natural process. Autophagy is the way cells eliminate their own unwanted or damaged parts. It is a highly regulated and completely normal process by which they remain healthy by performing "routine housekeeping" and "garbage disposal."

Dr. Beth Levine, director of the Center for Autophagy Research and professor of internal medicine and microbiology, has determined that autophagy prevents *Salmonella* bacteria from becoming successful intracellular pathogens. She and her team studied the effects of *Salmonella* infections in two organisms they had genetically engineered to lack active autophagy genes.

In both cases, the animals with inactive autophagy genes fared far worse than those with active ones. Rather than being targeted for elimination, the bacterium was able to invade the host cells, where it started replicating.

Dr. Levine's findings also suggest that decreases in autophagy – such as those that occur in the elderly and in certain patients with Crohn's disease – may lead to abnormalities in the way the intestinal tract deals with bacterial infections.

"It's known that as people get older they become more susceptible to infectious diseases and also that autophagy decreases," said Dr. Levine, who holds the Charles Cameron Sprague Distinguished Chair in Biomedical Science. "We've found that signaling pathways that extend life and protect against bacterial invaders do so by triggering autophagy. This suggests that therapeutic strategies to increase autophagy may be effective in defeating harmful bacteria that can enter inside cells."

It's unclear why older people become more susceptible to infections, but research has shown that autophagy declines with age. Dr. Levine, a Howard Hughes Medical Institute Investigator at UT Southwestern, suggests that by reversing or regulating this process, researchers could make the elderly and others with weakened immune systems more resistant to infections.

Her lab is now looking into the efficacy of a new autophagy-inducing molecule for treating a number of intracellular bacterial infections including Salmonellosis, tuberculosis, tularemia and Listeriosis.

"Unlike in humans, C. jejuni does not invade chicken intestinal epithelial cells and damage these cells. Also, the immune systems of chickens likely deal with the bacterium differently. Chickens are a great model for investigating how disease by C. jejuni is prevented."
—Dr. David Hendrixson

Immobilizing Colonization and Mobility

Dr. David Hendrixson, associate professor of microbiology, is conducting research aimed at ridding the world of one of the most common and severe forms of diarrheal disease in humans. *Campylobacter jejuni* (*C. jejuni*) is a bacterial pathogen that colonizes the gastrointestinal tracts of humans and animals, particularly chickens and other types of poultry.

It often is spread among flocks in feces and infects humans through their consumption of contaminated poultry. *C. jejuni* has also sickened consumers of other meats and unpasteurized milk. The bacterium is harmless in livestock but it can cause mild to severe diarrhea when passed to humans.

"The chicken gastrointestinal tract has adapted to the presence of *C. jejuni*," Dr. Hendrixson said. "Unlike in humans, C. jejuni does not invade chicken intestinal epithelial cells and damage these cells. Also, the immune systems of chickens likely deal with the bacterium differently. Both of these factors contribute to the reasons why chickens don't become ill due to C. jejuni infections. Chickens are a great model for investigating how disease by *C. jejuni* is prevented."

Dr. Hendrixson's lab is studying the genes the bacterium needs to live within an avian host in hopes that this understanding may lead to strategies to eradicate it from chickens and thus decrease the amount of the bacterium in human food.

He and his UT Southwestern colleagues have identified many factors *C. jejuni* requires for the colonization of poultry and are analyzing how these factors benefit bacteria growth and might be targets for future vaccines.

Dr. Hendrixson also is focusing on the genes involved in the function and regulation of the flagellum *C. jejuni* produces, which allows the bug to propel itself throughout a host's gastrointestinal tract. Flagellum is a tail-like projection that protrudes from the pathogen and can move it toward various nutrients for growth. Bacteria create only a certain number of flagella and at very specific locations.

"Scientists are still unsure how *C. jejuni*, or similar bacteria, know to create an exact number of flagella and at specific locations," said Dr. Hendrixson. "We want to understand these spacial and numerical parameters, because if we know how *C. jejuni* builds its flagellum, we may be able to disrupt the process and immobilize the pathogen."

Protecting the Kitchen

The global incidence of foodborne disease is difficult to estimate. As international shipping in the food industry continues to expand, so does the possibility of bacterial pathogens crossing borders.

There are steps consumers can take to try to keep a market trip from leading to a stay in the hospital. The CDC reports that raw meat and poultry, raw eggs, unpasteurized milk and raw shellfish are the foods most likely to be contaminated. Lona Sandon, assistant professor of clinical nutrition and a spokeswoman for the American Dietetic Association, stresses that all meat, poultry and eggs should be cooked thoroughly. She recommends using a thermometer to measure the internal temperature of meat to ensure it is cooked sufficiently to kill bacteria. Most meats should be cooked to 165 degrees and leftovers also need to be heated to this temperature.

"It's also important to avoid cross-contamination of one food with another," said Ms. Sandon. "Wash hands, utensils and cutting boards after they have been in contact with raw meat or poultry and before you touch another food. Place the cooked meat on a separate dish, rather than one that has held raw meat. And be sure to refrigerate leftovers promptly. Bacteria can grow quickly at room temperature."

Regarding vegetables and fruits, Ms. Sandon says washing these foods with running water is the best way to remove unwanted pathogens. She warns that the cut surface on fruits and vegetables is a prime environment for bacteria growth. It also is important for consumers to be mindful of slicing the foods on clean cutting surfaces.

Yet despite such precautionary measures everyone, no matter their location in the world, is susceptible to illness caused by foodborne bugs. And the clock is ticking as more and more of these bacteria are becoming immune to the effects of antibiotics. Over time bacteria have adapted to make human cells work to their advantage. UT Southwestern researchers are determined to turn the tables on them.

"Wash hands, utensils and cutting boards after they have been in contact with raw meat or poultry and before you touch another food. Place the cooked meat on a separate dish, rather than one that has held raw meat. And be sure to refrigerate leftovers promptly. Bacteria can grow quickly at room temperature."
—Lona Sandon