

MISSION

A Safer Food Supply

by Krista Weidner

“EVERY TIME I GO TO A PROFESSIONAL meeting, I hear that the United States has the safest food supply in the world,” says food scientist Stephanie Doores. “And while that is true, we all know that lapses in safety can happen. When outbreaks of foodborne illness make headlines, people get panicky about what they should eat.”

Early this year, a nationwide outbreak of salmonellosis from tainted peanut butter sickened hundreds of people, and peanut products were pulled from shelves. As the details emerged tracing the pathogen to a peanut production plant in Georgia, and an inspection by the U.S. Food and Drug Administration (FDA) revealed numerous food-safety violations within that facility, the question arose time and time again: How could this happen?

Outbreaks of foodborne illness can occur because disease-causing organisms are out there, food products are traveling longer and longer distances to reach consumers demanding variety, and the U.S. food-regulation system is not perfect.



As research into foodborne organisms continues, food scientists are working toward making our food supply safer by developing more and more accurate techniques for identifying, tracing, and eliminating pathogens.

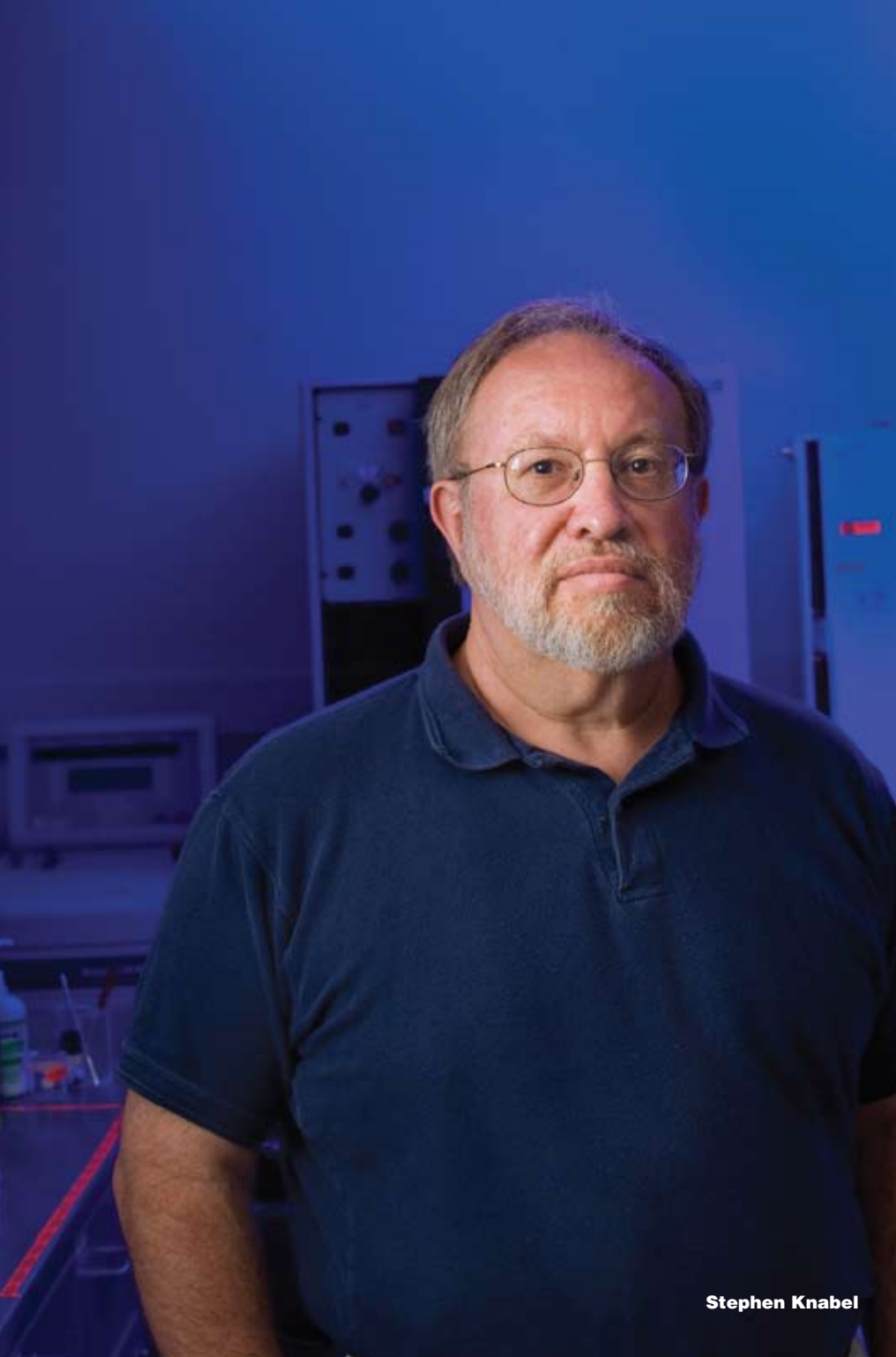
Although safety and sanitation lapses such as those in the Georgia peanut-processing plant are not the norm, they can result from miscommunication, lack of proper food-safety training, or sketchy oversight. While the U.S. Department of Agriculture (USDA) oversees meat and poultry inspection, all other foods are regulated by FDA, an organization that is “woefully underfunded and understaffed,” says Doores. “Are FDA inspectors coming into a processing plant, and if they are, how often? Different states handle regulation differently. Larger companies often employ third-party auditors to do inspections, but smaller plants might not have the resources to do that.”

Since federal, state, and even independent inspectors can't be on site continuously at every food-processing plant to catch every possible lapse, training and education play a vital role in food safety. Says food scientist Luke Laborde, “The bottom line is there are not enough



inspectors out there, and I don't think there ever will be. We in the food-safety community believe what's more effective is a systematic process in which the manufacturer is responsible for creating a plan that meets safety standards. And to some extent, we have that in place.” The Hazard Analysis Critical Control Point program, or HACCP, is currently required by USDA for meat and poultry processing plants and is required by FDA for seafood and juice products, but it is voluntary for the rest of the food industry. HACCP training, which is offered at Penn State, focuses on self-monitoring through evaluating each step of food processing, including receiving, mixing, adding ingredients, cooking, packaging, and delivering.

Bindhu Verghese, post-doctoral fellow in Food Science, works with DNA sequence alignment to better understand *Listeria monocytogenes*.



Stephen Knabel

“HACCP is about the process, not the product,” LaBorde explains. “For each step of the process, you identify the potential microbiological, biological, chemical, and physical hazards. For example, if you’re making chicken nuggets, they’re moving through a cooker at a certain speed and a certain temperature. You can’t take every chicken nugget and put a thermometer in it to see if it’s heated to 165 degrees. But based on research, we know

what time and temperature settings will generate an internal temperature high enough to kill pathogens. In other words, we figured out earlier that this product will be fine if the process is correct. So now we just have to make sure the process is working. Each processing plant develops its own HACCP plan based on its particular process and needs.”

Certified HACCP workshops use a hands-on approach, which helps food

industry workers apply what they learn to the real world. “We form teams that develop a HACCP plan for a process that simulates what participants deal with in their own facility,” says food safety extension specialist Martin Bucknavage. “By the time people leave, they have the tools to go back and develop a safety program for their facility or look at their current food safety plan in a different light. Many students leave with new insights into their own facility’s process that give them confidence to make suggestions to their management about how they could improve food safety.”

Bucknavage, LaBorde, and food scientist Catherine Cutter not only hold HACCP training for food industry employees, but they also provide follow-up help if needed. Says Cutter, “Sometimes companies ask us for help with validating that their process—whether it be heat treatment, acid wash, or any step along the way—is sufficient to meet the criteria for their HACCP plan. Since HACCP is mandated for the meat industry, and all of our instructors have experience working in that area, we can provide that help.”

One safety concern in the meat-processing industry is *Listeria monocytogenes*, a major foodborne pathogen that may be found in hotdogs, lunch meats, and raw meat. To help educate the food industry about this organism, USDA helped fund a project to develop educational materials geared toward controlling *Listeria* in very small meat and poultry plants. “We created and disseminated a series of booklets and videos, and that effort also led to a project in *Listeria* control in retail stores,” says Cutter. “Anyone cutting deli meats or making salads has the potential to introduce this organism. So we are working with retail organizations—grocery stores, delis—on safety and sanitation training.”

The retail sector is an important focus for food-safety training, Cutter says. While training programs such as ServSafe are effective, they are directed toward managers and supervisors. Retail operations such as grocery stores, convenience stores, and restaurants often have a lot of employee turnover as well as non-English-speaking workers or workers with low literacy levels. Cutter and her graduate

student, in cooperation with Virginia Tech and with USDA funding, are developing training modules for this sector of the workforce. “Let’s say you have a grocery-store deli worker with a seventh-grade reading level,” says Cutter. “You can’t pull them away from their shift, put a PowerPoint presentation up on a screen, and expect them to learn. We’re looking at ways to use pictures that tell stories, to use examples of outbreaks and how you need to prevent contamination through hand washing or wearing a clean apron. We’re working on culturally compatible training that fits their needs and gives them the tools to prevent contamination on the line.”

While food-safety training for processing plants and the retail sector is important, food safety at home also plays a vital role in combating foodborne illness. Penn State extension educators and fac-

You stumble around in the dark most of the time, and when something happens, you pounce on it.

ulty provide a variety of training programs for consumers—on campus, throughout the state, and online. “Our message is that food safety is everyone’s responsibility,” says Bucknavage. “And whether we are developing materials about home food preservation, holding a workshop on venison safety for hunters, or fielding calls about how to cook a turkey properly, our priority is to provide solid, research-based information. We have access to world-class researchers here at Penn State, and we can draw on their expertise. In that way, we’re providing a valuable service to the people of Pennsylvania.”

One of several food science extension faculty who also conduct research, Cutter specializes in quality and safety of meat products. Her current research focus, in collaboration with Doores and funded by the National Pork Board, involves identifying ultrasound techniques that will inactivate pathogens on vacuum-packaged fresh or ready-to-eat pork. Working with pork chops that have been

inoculated with *Salmonella* and ham slices inoculated with *Listeria monocytogenes*, she is searching for the ultrasound frequency that will overcome the plastic barrier and inhibit pathogens on the meat surface. “Ultrasound is a promising technique,” she says. “In the medical field, it’s been used to deliver cancer drugs to cancerous cells. For our project, we’re cautious but optimistic that we’ll soon find the right combination that will kill the pathogens while maintaining the integrity of the product.”

Listeria monocytogenes is also a research emphasis for Cutter’s colleague Stephen Knabel, whose lab work has shown a surprising “long-term survival phase” for this organism. Bacteria like *Listeria* were thought to pass through four phases during their life cycle; however, Knabel and his students recently discovered a fifth phase in *Listeria*. “No

It’s as if they have their own preservation system for long-term survival.”

Knabel believes that further research into the long-term survival phase of *Listeria* could lead to methods for controlling this foodborne pathogen in the food industry. “People have been struggling to understand how *Listeria* can survive so long in processing plants,” he says. “When it’s in this long-term survival phase it is resistant to heat and high pressure, and it’s really tough to get rid of. So this work might help explain its longevity and presence in processed foods.”

Another major research focus for Knabel is molecular epidemiology, which involves examining DNA to track the cause of an outbreak to its source. Knabel and his students do this by identifying specific strains using a method called molecular subtyping. They identify a “DNA fingerprint” specific to a strain



one had seen this before with *Listeria*,” Knabel says. “After the rod-shaped cells decline in the fourth phase, we’ve discovered that they level off and form round cells that go on surviving in a sort of dormant state. We keep incubating them, and they seem to survive indefinitely. Then, if we take those dormant cells and put them into fresh broth, they germinate immediately—you can see them swell up and start replicating.

Fenyun Liu, graduate student in Food Science, performs gel electrophoresis to check PCR amplification, an important step to determine if the correct DNA is present in samples.

of a pathogen so they can track that strain from processing plant to infected people.

Up until now, Knabel explains, most molecular subtyping has been fragment based: “You use enzymes to chop

the DNA up and then place it in a gel where pulses of electric current separate the DNA molecules by weight. The result is a banding pattern that looks like a bar code.” But that method is not as accurate or informative as looking at the entire DNA sequence, which consists of the single nucleotides A, T, G, and C. By being able to detect differences at this single nucleotide level, researchers get more accurate information about specific strains.

“But only when we have all the dots connected can we say, ‘This patient got sick from this source,’” Knabel says. “And that’s where epidemiologic concordance comes in. And that term just means that the molecular subtyping data agree with the data from conventional epidemiology. So you interview people who got sick about what they ate and when and where, then you interview a control group of similar people who did

not get sick. Then you can develop an odds ratio and say, for example, that in the group who got sick, a lot more of them ate peanut butter. The results from the case-control study strongly suggested that peanut butter caused the outbreak. If the molecular epidemiology agrees with that, the case is over. It is very difficult to refute that kind of molecular and conventional epidemiological evidence.”

Molecular epidemiology is especially useful in identifying strains of *Listeria* because the long incubation period (time between consumption and disease) associated with listeriosis makes it hard to track using conventional epidemiology. “*Listeria* can be present in someone’s body up to fifty days before they get sick. Who remembers what they ate three days ago, let alone fifty days ago? So we’ve been able to apply molecular-subtyping techniques to *Listeria* to iden-

tify first whether an outbreak is occurring, and if so, what strain is causing the outbreak. Once we identify a strain and understand where a pathogen is coming from and how it is being transmitted to foods, then we can intervene to stop the outbreak and prevent future outbreaks.

“It’s really exciting,” Knabel adds. “The DNA-based work we are doing is forensic science. It’s the same as the television show *CSI*. The only difference is that they’re dealing with human criminals, we’re dealing with bacterial criminals that actually cause far more illness and death than people do.” The DNA-sequenced-based methods that Knabel’s group have developed are now being used by the Centers for Disease Control (CDC), USDA, and the food industry to track outbreak strains of *Listeria* and prevent future foodborne disease outbreaks from this pathogen. The methods have



Edward Dudley

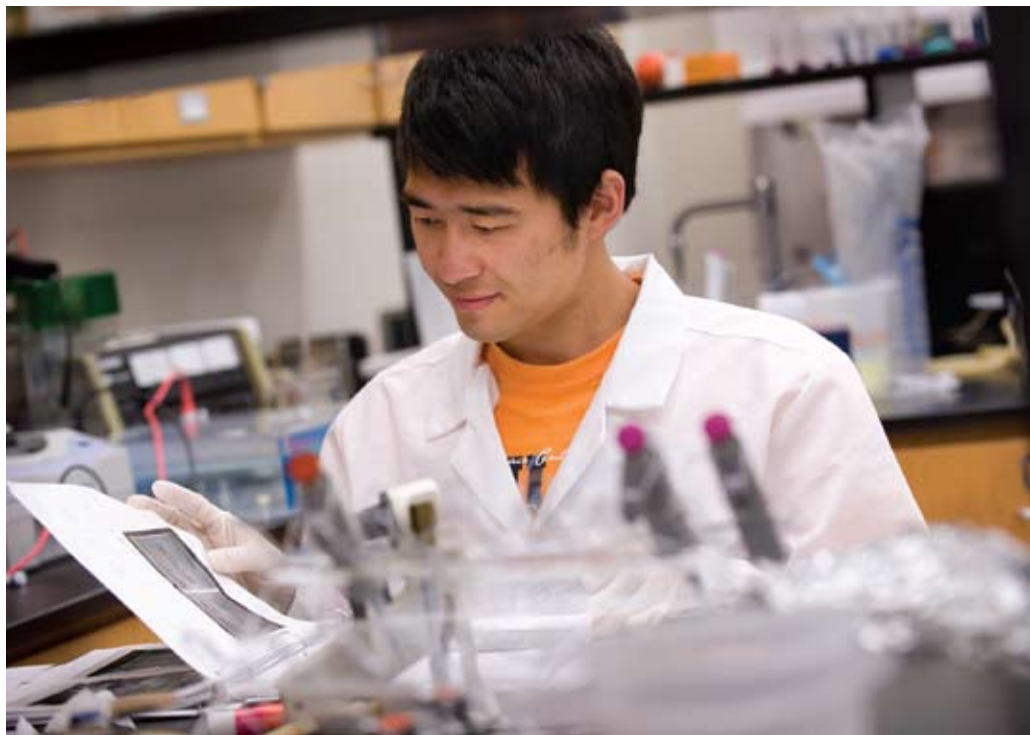
also been used to resolve major court cases that have followed foodborne illness outbreaks and recalls due to *Listeria*. In fact, “We jokingly refer to our lab as CSI: University Park,” says Knabel.

The sequence-based approach to identifying strains has worked so well with *Listeria* that Knabel is working with food scientist Edward Dudley to adapt it to track other pathogens, including *E. coli* O157:H7, a contaminant sometimes found in ground beef and fresh produce. This foodborne pathogen is blamed for about 70,000 cases of disease each year.

“This method of using sequencing to differentiate between *E. coli* strains was tried initially about seven years ago, with disappointing results,” Dudley says. “While the method worked well for *Salmonella* and *Listeria*, it wasn’t as effective with *E. coli*. So when I first came

As time passes and an organism replicates, DNA sequences change slowly, sometimes because of error while replicating, sometimes to adapt to stress for survival.

to Penn State two years ago, I asked, ‘Is that because there really are no sequence differences, or are people choosing the wrong regions of the DNA to look at?’” Dudley and his graduate students set out to identify a particular group of genes in *E. coli*—genes that are under outside pressure to change. “For example,” he explains, “*E. coli* bacteria have structures projecting from their surface called flagella, little structures that help the cells move around. Anything on the surface of bacteria is vulnerable to the pressure of the immune system, and the bacteria need to evolve quickly into something the immune system doesn’t recognize. So we came up with the idea to look at the sequences of these genes, thinking maybe they would change much faster than regions targeted previously.” One of Dudley’s graduate students is now working on



developing a subtyping method for *E. coli* O157:H7 based on finding genes that are likely to vary among strains.

Dudley is also interested in using molecular subtyping techniques to look more deeply into the evolution of pathogenic organisms and to define very specific groups, or lineages, within a population. Because researchers have a good estimate of how fast changes in DNA occur, they can trace back to the last common ancestor. “We think *E. coli* O157:H7 is about 40,000 years old, which isn’t very old for bacteria,” he says. “*Salmonella* has been around much longer. But the two organisms have the same ancestor—100 million years ago, *Salmonella* and *E. coli* were the same organism.” As time passes and an organism replicates, DNA sequences change slowly, sometimes because of error while replicating, sometimes to adapt to stress for survival. Dudley and his students are working with distinct families of *E. coli* O157:H7, hoping to learn why some families are more virulent, or disease-causing, than others.

As research into foodborne organisms continues, food scientists are working toward making our food supply safer by developing more and more accurate techniques for identifying, tracing, and

Kuangng Liu analyzes DNA fingerprints from various *E. coli* O157:H7 isolates. Liu recently graduated with a master’s degree in Food Science.

eliminating pathogens. “The way research works is a combination of hard work and luck,” says Knabel. “In science it’s called serendipity. Louis Pasteur said that ‘chance favors only the prepared mind.’ You stumble around in the dark most of the time, and when something happens, you pounce on it. My students, who are really the ones making these breakthroughs, often get frustrated because they’re trying to do something specific and then something else pops up. But a mistake can lead to another discovery. And the most interesting things are often those that are unexpected.”

Faculty and staff referenced in this article are Martin Bucknavage, food safety specialist and extension associate; Catherine Cutter, associate professor and food safety extension specialist; Stephanie Doores, associate professor of food science; Edward Dudley, assistant professor of food science; Stephen Knabel, professor of food science; and Luke LaBorde, associate professor of food science.