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On the Trail of a Killer

By Pat Bailey

E. Coli outbreak sends scientists in search of practical solutions.

It is March, and in the broad valleys of California's central coast, millions of tiny spinach seeds are being dropped into the soil. A new growing season is stirring to life here in the nation's "salad bowl," which just a few months ago became the epicenter for a coast-to-coast outbreak of food-borne illness that touched 26 states.

In just eight weeks the devastating *E. coli* O157:H7 bacteria, transmitted via bagged fresh spinach, had sickened nearly 200 people, killing at least three and sending more than 100 to the hospital.

Lot codes on 13 samples of bagged spinach in the victims' refrigerators pointed federal and state investigators to a single lot of spinach, packaged on Tuesday, Aug. 15, during just one shift at a San Juan Bautista packing facility. With further investigation by the federal Food and Drug Administration and the California Department of Health Services, with participation from U.S. Department of Agriculture researchers, a DNA fingerprint match was made. The *E. coli* O157:H7 from case patients matched the *E. coli* strains found in a nearby stream and in the feces of cattle and wild pigs on the four implicated ranches that grew the spinach potentially associated with that lot.

It was a superb example of epidemiological detective work on the part of the state and federal agencies, but the investigation has yet to reveal just how the life-threatening *E. coli* bacteria found their way into the spinach and traveled to homes in more than half of the nation.

To complicate matters, in November and December as the pieces were beginning to fall into place on the spinach outbreak, two separate but simultaneous outbreaks of *E. coli* O157:H7 occurred, linked to two fast-food restaurant chains in the Midwest and on the East Coast. At least 150 people were sickened and the gravity of the recurring outbreaks of *E. coli* on leafy greens grew.

Looking for answers

In campus laboratories and in the fields and streams of the Central Coast region, plant scientists, food microbiologists, hydrologists and veterinary ecologists are now chasing the elusive *E. coli* bacteria, hoping to provide the data needed to assist the fresh produce industry, regulatory agencies and policymakers.

"Industry is working feverishly to arrive at standards for tighter control of all of the key risk management areas," says Trevor Suslow, a UC Davis Cooperative Extension plant scientist, who for the past decade has worked on food-safety issues related to fresh fruits and vegetables.

"While we may not have all of the data for this specific crop or region, the fields of plant pathology and microbiology have been looking at these issues for years and years, and the science is there to help guide policy decisions," he says. "What doesn't yet exist, in full, are the specific data needed for developing precise standards and measuring process controls, as well as predicting ecological behavior of these pathogens."

Following the Sept. 13 announcement of the *E. coli* outbreak, Suslow's laboratory focused its efforts on conducting research that would help both industry and government make decisions in the rapidly changing environment created by the outbreaks.

E. coli O157:H7 is one of hundreds of known *E. coli* strains. It is particularly worrisome because in humans it produces a toxin that can damage cells in the kidneys, pancreas, brain and other organs. In the worst cases it results in kidney failure and death.

Suslow's lab is investigating what the consequences would be if spinach seeds were to become contaminated with *E. coli* O157:H7. The researchers also are exploring possible technologies for disinfecting contaminated seeds.

They are studying, too, whether *E. coli* may be able to thrive at temperatures once thought to be too cold to support bacterial growth. The concern: Could *E. coli* be multiplying as the spinach is being shipped across country in refrigerated containers at suboptimal temperatures?

A third project is looking at compost and manure as a possible source for the bacterial contamination.

"One area of potential risk may be the application of manure and compost in late August and early September while crops are still being harvested in adjacent fields," Suslow says. "We want to know what the potential is for *E. coli* to drift and then survive on a nearby crop.

"If the compost has no pathogens—no bacteria or other microbes that might cause disease—it's not a problem," he says. "Unfortunately, based on recent preliminary surveys of compost, there are concerns that the composting process is not being uniformly managed to reduce the bacterial content to acceptable levels. We're trying to figure out if there is a temperature and time issue or if it's possible that the compost is being recontaminated."

In another campus laboratory, Cooperative Extension food scientist Linda Harris and colleagues are studying the behavior of *E. coli* O157:H7 on produce. She and Suslow arrived at UC Davis in 1996—the same year that serious outbreaks of *E. coli* O157:H7 were traced to unpasteurized apple juice in the United States and to radish sprouts in Japan. It was a year that brought a new emphasis on microbial food safety to both their careers.

Reporters who descended on Harris' laboratory during the recent *E. coli* outbreak expected to see bushels of spinach being put under the microscope, but hers is a research rather than a diagnostic lab. Ongoing studies include evaluating the impact of postharvest handling methods on the potential for *E. coli* O157:H7 to become internalized in lettuce tissue and determining the conditions that permit growth of the organism on cut lettuce from harvest to consumption.

When Harris entered the field of food science nearly 30 years ago, fresh produce was not seen as much of a player in the realm of food-borne disease.

"That's changed partly because we're all eating many more fresh fruits and vegetables and partly because we now have a better understanding of the microorganisms and their hosts," she says.

She notes that the focus has also shifted from just food processing to the entire food system.

"As an undergraduate student, I was told that food scientists focused on food safety issues only within the food processing 'box'—not with what went on at the farm or in the grocery store or home," she recalls. "Now we have to understand what are the inputs and what is the significance of those inputs. We have to understand the whole system, from production agriculture through to consumption. Food scientists now work with researchers from many different disciplines, from

those who have an understanding of plant and animal agriculture throughout the system to behavioral scientists."

That's where Rob Atwill, a Cooperative Extension medical ecologist comes in. A veterinarian by training, Atwill and UC Davis hydrologist Ken Tate, along with colleagues at the U.S. Department of Agriculture, recently received a $1.2 million USDA grant to track the source of *E. coli* in the Salinas Valley and neighboring counties. It's a four-year project that was proposed long before the most recent *E. coli* outbreaks.

Searching for the source

The new effort is modeled after a similar research program Atwill conducted in the Central Valley with a protozoan parasite called *Cryptosporidium*. The researchers will be looking at where *E. coli* exists in the wild, how much of it is associated with cattle and wildlife, and what kind of risk it poses to downstream surface water sources.

"It's a big detective story," says Atwill, noting that the project will require a massive collection of data.

The research team will be working with various private landowners in Salinas and possibly San Benito counties as they grapple with a microbe that is relatively rare in the environment and that may be moving between a variety of animal species that have very different diets. Much of their work is seasonal and weather-dependent.

"This is not a 9 to 5 job," Atwill says. "When a rainstorm hits, everyone jumps and runs to pull water samples from streams, canals and ditches that channel water throughout the agricultural systems that link wild and domestic animals located on rangelands with fields where produce is grown."

In addition to surface water sources, the researchers are also taking samples of lettuce and the feces of livestock and wildlife, ranging from mice to feral pigs to birds. In all, 12,000 samples will be collected, 3,000 each of animals, soil, water and crops.

Atwill is well aware that farmers, food processors and government agencies will have to be making management and policy decisions long before the four-year project is completed. "Fixes" to reduce the risk of *E. coli* contamination in leafy greens will probably be put into place long before the study is finished, simply because the consequences of another *E. coli* outbreak would be so serious.

Eventually, research should provide the data that will paint a fairly clear picture of where the *E. coli* O157:H7 is originating, how it is coming in contact with the leafy green crops and how it is multiplying to levels that are in some cases lethal to humans. If the bacterial outbreak is traced to a specific group of animals, Atwill and colleagues will work to develop management practices that would prevent the bacteria from disseminating throughout the region and contaminating fields and produce.

"We have to balance the efficacy of a beneficial management practice with its feasibility, because if we make recommendations that aren't economically feasible, it's unlikely that producers will adopt such practices," Atwill says.

Joining forces

Working quietly behind the scenes to bring industry, government and academia together to develop just such workable solutions is UC Davis' Western Institute for Food Safety and Security.

The institute currently supports the work of 10 researchers, including some of the projects conducted by Suslow and Harris.

Researchers in the institute are working to determine what mechanisms underlie the severe disease caused by some *E. coli* organisms. This information may be key to preventing future illnesses. Ironically, the institute had organized a meeting with industry and agency colleagues in Salinas to discuss contamination in leafy greens at the time the *E. coli* outbreak hit last August.

“The timing was uncanny,” says Jerry Gillespie, a veterinary pathologist who directs the institute. Three days after the outbreak was announced, farmers and food processors from the Salinas area traveled to Davis to meet with scientists and government representatives. It was an opportunity for all parties to lay their proverbial cards on the table.

“It’s very difficult for farmers and companies that have been competitors to set that competitiveness aside to solve problems, and of course the same is true of academia and government agencies,” Gillespie says.

“At the Western Institute, we try to get everyone to be very open and candid. We try to bring people together and encourage them to lower their guard,” he says. “We tell them, ‘We are here to solve the problem, and if we just rehash what a wonderful job we’re all doing, it’s not helpful because people are getting sick and dying.’”

Gillespie was encouraged to see many of his industry colleagues rise to the challenge, offering researchers access to their property and encouraging their counterparts to make sometimes-difficult decisions. He is confident that, in time, the *E. coli* problem in the Central Coast will be scientifically defined and workable solutions will be found that will dramatically lower the risk of future outbreaks.

“Every outbreak, from a research standpoint, opens new areas of investigation,” says food scientist Harris. “Historically, this outbreak will be considered a ‘tipping point’ for food safety in lettuce and leafy greens. This is an outbreak that will cause us to move forward more quickly, and progress will be made.”

Pat Bailey writes about the agricultural and veterinary sciences for UC Davis.