Detectives Search for Antimicrobial-Resistant Organisms

Antibiotics were once called miracle drugs because they revolutionized treatment of disease, curing bacterial infections that used to lead to debilitation and, all too often, death. Not only humans, but also livestock and pets have benefited from these wonder drugs. But over the years, some bacterial pathogens have developed resistance to the antibiotics that once spelled their doom.

Agricultural Research Service scientists in Athens, Georgia, are working to learn which pathogens are resistant in livestock, determine why and how they became resistant, and find ways to turn back the tide. All the while, bacteria continue to do what they do best—adapt in order to survive.

Recently, resistance has been observed in bacteria known to cause plague and in *Staphylococcus aureas,* a common agent in wound and blood infection. When bacteria became resistant to only one or two antibiotics, there was never a concern, since pharmaceutical companies kept a steady stream of new drugs coming our way. This changed dramatically when pathogens developed resistance to more and more of the drugs. Today, there are fewer new antibiotics being developed, and we face the challenge of finding new ways to treat diseases and infections we once thought we'd vanquished.

The Chase Is Afoot

When confronted with emerging resistance to antibiotics, also called antimicrobials, several U.S. government agencies began to research and monitor the problem. In 1996, programs conducted by the U.S. Department of Agriculture (USDA), the Centers for Disease Control and Prevention (CDC), and the Food and Drug Administration (FDA) were coalesced into the National Antimicrobial Resistance Monitoring System (NARMS). USDA leads the animal arm, CDC leads the human arm, and FDA leads the retail food arm.

USDA's research component dedicated to this project is the ARS Antimicrobial Resistance Research Unit, in Athens. Led by microbiologist Paula Fedorka-Cray, the team is currently testing for and characterizing antimicrobial resistance in four bacteria: non-typhoid *Salmonella, Campylobacter,* generic *Escherichia coli,* and *Enterococcus* species. The bacterial samples tested in the animal arm of NARMS are obtained from three sources: diagnostic samples taken from sick animals in which treatment history is presumed but not confirmed, on-farm samples from healthy animals, and slaughter/processing samples from federally inspected plants. The lab's scientists isolate, test, and characterize more than 17,000 bacterial isolates per year.

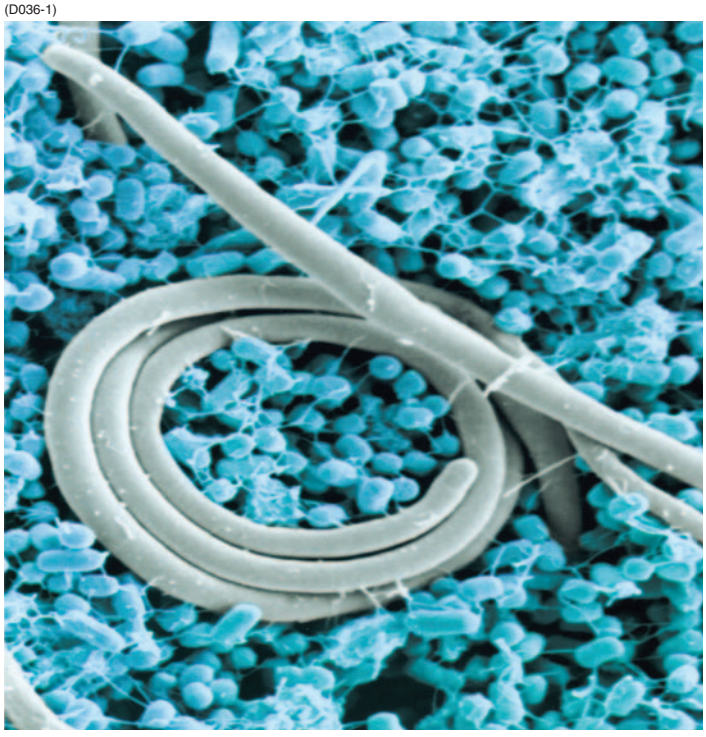
To Each Its Own

It would be simple and convenient if all bacteria reacted predictably and uniformly to treatment with antimicrobials. Unfortunately, nothing could be further from the truth. For instance, there are many different types of *Campylobacter*. Two of the most common types that cause illness in humans are *C. jejuni* and *C. coli*. Each responds differently to antimicrobials, and *C. coli* appears to become resistant to them faster than *C. jejuni.* Both have demonstrated resistance to one of the newest classes of antimicrobials—fluoroquinolones. Used in human medicine since the 1980s, fluoroquinolones were approved for use in chickens in 1995. Since then, microbiologist Mark Englen of the Athens team has been studying and tracking *Campylobacter* resistance to these drugs.

Another potentially harmful bacterium, *Salmonella,* has more than 2,400 different serotypes, and each one appears to develop resistance to antimicrobials at a different rate. One type of *Salmonella* that most often sickens both humans and other animals is *S. typhimurium*. It appears to easily acquire resistance to multiple antimicrobials. Some strains are now resistant to 13 of 17 antimicrobials tested. Of all *Salmonella* types tested from

Cells of *Salmonella enteritidis* change shape as they grow. This scanning electron micrograph shows a mixture of small cells with filaments and very large cells that lack filaments. Small cells arise only during certain growth stages and efficiently contaminate eggs when the time is right. Magnified about 5,400x. Photo by P.J. Guard-Petter, digital colorization by Stephen Ausmus.

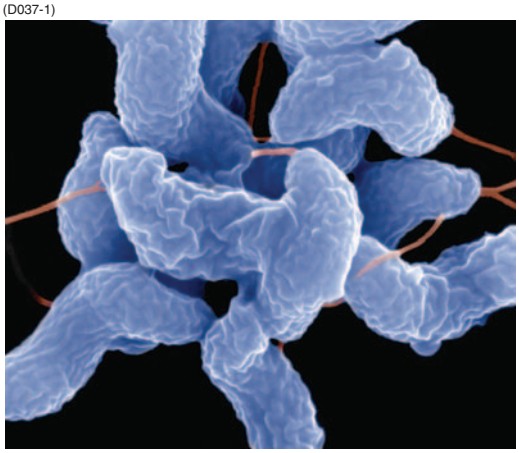
(D036-1)



1997 to 2003, the rate of single-drug resistance has remained relatively stable at 9.5 percent of the samples. But *Salmonella* that are resistant to more than five drugs rose from 11 percent to 20 percent. And those that are resistant to more than 10 drugs rose from a scant 0.8 percent to almost 6 percent.

"The problem is quite clear," says Fedorka-Cray. "More than 35 percent display resistance to one or more antibiotics. Unfortunately, there is a finite number of antibiotics, and *Salmonella* continues to alter to give itself the greatest chance of survival." Jonathan Frye, a microbiologist who studies resistant *Salmonella* in Athens, has developed new molecular technology that may provide a more accurate analysis of resistance and the genetics behind acquisition of resistance.

(D037-1)



***Campylobacter,* magnified about 35,000x. Photo by De Wood, digital colorization by Stephen Ausmus.**

Tangled Web of Transmission

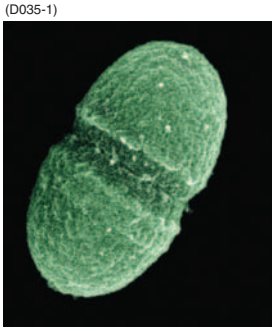
Controlling resistance starts with understanding how it develops and how bacteria move throughout the ecosystem.

"Antimicrobial use in livestock production can result in resistant bacteria that can be passed to humans via the food chain, and our team is studying how and with what frequency this occurs," says Fedorka-Cray. Meat can also become contaminated during processing, and fruits and vegetables can pick up bacteria from the manure used to fertilize them.

"Resistance can also develop when a human or a pet is prescribed an antimicrobial—especially when it's used inappropriately or improperly. Since any use of antimicrobials may result in selection of resistant bacteria, these drugs should only be used when necessary," Fedorka-Cray says. And, she adds, "Since food can be a transmission vehicle, it is important for people to handle foods correctly, cook food thoroughly, and always thoroughly wash their hands and food-preparation areas." Still, some bacteria harbor resistance even though they haven't been exposed to manmade antimicrobial drugs, she says. This is called "intrinsic resistance."

Fedorka-Cray's group has developed the nation's largest descriptive database of resistant populations of bacteria recovered from animals over time. "The data help us determine the probability that resistance will occur or be maintained if antimicrobials are used," she says. "When you reduce use of antimicrobials, you may reduce prevalence of resistant organisms. On the other hand, some swine studies have shown that eliminating antimicrobials does not eliminate resistant bacteria. This may mean that bacteria have become permanently resistant to some antimicrobials as a survival tool." Scientists face many such challenges as they try to determine how to decrease numbers of resistant bacteria. Microbiologist Charlene Jackson of Athens is focusing her research on *Enterococcus* species, bacteria commonly found in nature but particularly problematic in hospitals.

(D035-1)



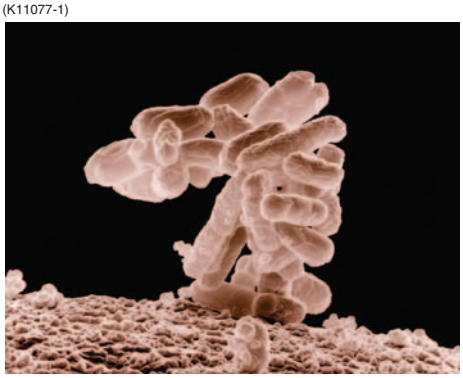
***Enterococcus faecalis,* magnified about 33,000x. Photo by Nathan Shankar, University of Oklahoma, digital colorization by Stephen Ausmus.**

Changes in antimicrobial use in food-animal production are being made. "Until recently, the pattern of antimicrobial use on farms changed very little. Now, veterinarians—as well as physicians and those in related fields—are reassessing how and when they use antimicrobials. Today's production systems may not need the same level of antimicrobial use or may be able to eliminate the use of certain antimicrobials altogether," says Fedorka-Cray. "The good news is that we have never reached a situation where an entire bacterial population isolated from animals is resistant to all known antimicrobials."—By **Sharon Durham,** ARS.

This research is part of Food Safety (Animal & Plant Products), an ARS National Program (#108) described on the World Wide Web at www.nps.ars.usda.gov.

Paula Fedorka-Cray is in the USDA-ARS Antimicrobial Resistance Research Unit, Richard B. Russell Research Center, 950 College Station Rd., Athens, GA 30605; phone (706) 546-3305, fax (706) 546-3066, e-mail pcray@saa.ars.usda.gov. ◆

(K11077-1)



Low-temperature electron micrograph of a cluster of *E. coli* bacteria, magnified about 4,000x. Each individual bacterium is oblong shaped. Photo by Eric Erbe, digital colorization by Christopher Pooley.