**Animal issues associated with Escherichia coli O157:H7**

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Since *Escherichia coli* O157:H7 was first recognized in 1982 as a human pathogen, considerable progress has been made in elucidating principal vehicles of transmission. Cattle have been identified as a major source of *E coli* O157:H7 infection of humans, with as many as 1 in 4 animals at slaughter shedding the pathogen in feces during the summer months. Case-control studies of sporadic cases of *E coli* O157:H7 infection in the United States, Canada, and Europe have identified eating undercooked ground beef, visiting farms, and handling animals on the farm as principal risk factors for infection. Cattle manure, of which an estimated 1.2 billion tons are produced annually in the United States, appears to be a principal source of the *E coli* O157:H7 problem. Animals, water, and food that contact cattle manure are potential vehicles of *E coli* O157:H7. An effective control program to substantially reduce *E coli* O157:H7 infections will require the implementation of intervention strategies throughout the food continuum, from farm to table. Promising intervention measures at the farm include competitive exclusion bacteria, bacteriophage, and targeted animal management practices addressing common points of contamination. Innovative intervention treatments are under development for use by food processors; however, most treatments have limitations that restrict their use to specific types of foods. For example, irradiation can create major off odors and flavors in foods that contain more than 10% fat. Consumers also have a role in implementing intervention controls in food handling and preparation. Unfortunately, many consumers eat high-risk foods, improperly handle and store foods, and ignore warnings regarding foods known to be unsafe. We all have a role in reducing the risk of foodborne illness, including *E coli* O157:H7 infections, but clearly more needs to be done on the farm, including validating proposed and developing innovative on-farm control measures.

**Public Health Concerns**

Estimates by the Centers for Disease Control and Prevention (CDC) indicate that enterohemorrhagic *E coli* (EHEC) serotype O157:H7 is responsible for approximately 62,300 cases of foodborne infection annually in the United States. These estimates include hospitalizations and 52 deaths, which are largely associated with cases of pediatric hemolytic uremic syndrome (HUS), a leading cause of renal failure in children. Sporadic cases in adults are less likely to be diagnosed and reported, because the gastrointestinal disease can be mild. The CDC, using the FoodNet surveillance system, reported an overall incidence of 2.1 to 2.8 cases/100,000 persons from 1996 to 1999. Although enterohemorrhagic disease occurred in most of the confirmed cases, HUS was more likely to develop in female children (61% of cases) around 4 years of age. In the United States, the major serotype of EHEC is O157:H7, although other serotypes can be associated with enterohemorrhagic disease. *Escherichia coli* O157:H7 can be cultured from only about 40% of fecal samples of patients that have signs of enterohemorrhagic disease, although approximately 60% of HUS cases can be confirmed by bacteriologic culture. Toxin activity can be detected in 76% of fecal samples from HUS patients; however, < 25% of samples are usually tested.

Undercooked ground beef is the most commonly identified vehicle associated with outbreaks of *E coli* O157:H7 infection, and the bacteria have been cultured from 28% of cattle at slaughterhouses in the Midwest during the summer months. However, some *E coli* O157 isolates may not contain the virulence factors necessary for human disease, and their importance on public health is not yet clear. Although undercooked ground beef is the vehicle most often associated with human disease, other foods such as salami, sushi, ice cream, milk, cheese curds, unpasteurized apple cider and juice, lettuce, and alfalfa sprouts have also been implicated in outbreaks of illness. Contaminated water in lakes, ponds, and swimming pools has also been a major vehicle of outbreaks. Several outbreaks involving children have been reported.
following a visit to a county fair, farm, or petting zoo following contact with carrier animals. Handling calves is a common source of human infection, as calves are colonized by *Escherichia coli* O157:H7. Although no cases have been definitively confirmed in humans, dogs have been implicated in a few sporadic infections in children. Person-to-person transmission has also been reported as a source of infection in child-care centers.

**Current Issues Associated with Animals**

Cattle, including dairy animal production systems, are important sources of *Escherichia coli* O157:H7. *Escherichia coli* O157:H7 is a transient member of the normal flora of cows and has been only exceptionally associated with clinical disease in neonatal calves. The prevalence of *Escherichia coli* O157:H7 has been reported to be from <1 to 5% in numerous studies in the United States, Canada, Australia, Norway, and Finland; however, studies using more sensitive methods for detection now report rates of *Escherichia coli* O157:H7 fecal contamination as high as 13 to 28% of animals. The mean duration that an individual animal has positive culture results is 30 days, but the range in duration can vary from a few days to a year. This variation has been attributed to many factors such as diet, drinking water contamination, competing microflora, immune response, age, breed, environment, and season. Warm weather (summer) is a high-turnover period with a high turnover of cattle are often being replaced by new and different housing conditions, and season. Warm weather (summer) is a high-turnover period with a high turnover of fecal *Shiga toxin-producing Escherichia coli* (STEC) isolates like *Escherichia coli* O157:H7 from the general herd and not solely from the dam. Many STEC strains cycle with a high turnover through individual cattle, though predominant strains exist collectively on a farm and can be specific to that farm. Calves at weaning are of the age group that most exist collectively on a farm and can be specific to that farm, and a child that developed bloody diarrhea after the infection. Although in this study the cattle and the goat had negative test results, the limited number of samples taken and the fact the agent is shed intermittently does not rule out cattle as the source of the pathogens. In a study of the sources of *Escherichia coli* O157:H7 in feedlots and farms in the northwestern United States, 65 dog samples and 33 cat samples were obtained and analyzed for the presence of *Escherichia coli* O157:H7. Results for all cat samples were negative, and 3.1% of the dog samples tested positive. The probable source of the bacterium for the dogs on these farms could have been the cattle directly or the water troughs. In a comparison study using a newly developed phage typing scheme for *Escherichia coli* O157:H7, it was determined that an STEC strain isolated from a dog was of the same phage type as *Escherichia coli* O157:H7 isolates obtained from humans. Two additional reports, both from the United Kingdom, associating *Escherichia coli* O157:H7 with dogs have been reported through Pro-Med and the FS Net. The Pro-Med report describes a geriatric, double incontinent dog that shed *Escherichia coli* O157:H7 in its feces. The dog’s diet consisted of dry food and occasional food scraps. The FS Net report described results of an outbreak investigation in which 90 fecal samples from dogs were obtained from 4 different beaches, and 7.8% of the samples were contaminated with *Escherichia coli* O157:H7. The role of dogs in transmitting *Escherichia coli* O157:H7 to humans should be considered as a potential risk factor for infection.

**Antimicrobial Resistance**

Compared with other foodborne pathogens, antimicrobial resistance in *Escherichia coli* O157 is generally low (0.8 to 8%) and limited to a few antimicrobials, tetracycline, streptomycin, sulfamethoxazole, and trimethoprim. Antimicrobial resistance in *Escherichia coli* O157:H7 appears to be attributable to the acquisition of conjugal R plasmids, possibly from the microflora of its ruminant host. Although epidemiologic studies suggest that administering antimicrobials to animals is at least partially responsible for the emergence of antimicrobial resistance in important foodborne pathogens, there is little direct experimental evidence to indicate the frequency of these events. We do not know why antimicrobial resistance has been slow to develop, despite the existing reservoir for drug resistance genes in other *Escherichia coli* and *Salmonella* spp colonizing various animal species, including cattle. Acquisition of antimicrobial resistant plasmids can result in an ecological disadvantage for the recipient of plasmids if it has to compete with the resident microflora. We do not know whether R plasmids might also present *Escherichia coli* O157:H7
with similar hardship in the rumen or elsewhere in the gastrointestinal tract. It is also possible that the O157 virulence plasmid prohibits the establishment of certain R plasmids that belong to the same incompatibility group as the virulence plasmid, preventing coexistence of both plasmids. A limited repertoire of R plasmids belonging to incompatibility groups compatible with the O157 virulence plasmid may explain the slow and limited emergence of resistance in E coli O157:H7.

Pathogenesis of EHEC

Enterohemorrhagic E coli are characterized by the presence of Shiga toxin (Stx) genes, locus for enterocyte effacement (LEE), and a large molecular weight plasmid that encodes for a hemolysin. These 3 virulence factors are present in most E coli associated with bloody diarrhea and HUS in humans.

The LEE is a large cluster of genes that collectively are responsible for the intimate attachment of the bacterium to the apical membrane of the enterocyte and subsequent destruction or effacement of the microvilli. The intimate attachment of the bacterial cell to the epithelium is attributed to the adhesin, intimin, and Tir, a bacterial protein, which is inserted into the host membrane and serves as the receptor for intimin. Both factors are part of LEE in enteropathogenic E coli (EPEC) and EHEC. Intimin appears to be an essential component in initiating attachment, colonization, and the subsequent pathologic changes that follow infection with EPEC and EHEC. Outside of its role in the disease process in human infections, it is uncertain whether intimin has a role in the colonization of healthy cattle.

In addition to LEE, E coli O157:H7 possesses a large molecular weight plasmid that contains several putative virulence genes, including a pore-forming hemolysin. Virulence plasmids are common features of pathogenic E coli, encoding toxins, adhesins, and other factors necessary for colonization, survival, and ability to cause disease in its animal host. The contribution of any of these plasmid-associated factors to disease is currently unknown.

Escherichia coli O157:H7 and other EHEC O157:H7. Feces and E coli O157:H7 contain similar amino acid sequences to Shigella toxin, hence the commonly used nomenclature for this E coli toxin in the literature, Shiga toxin or vero toxin. Among EHEC, there are 2 major serotypes produce a toxin similar in its amino acid sequence to Shigella toxin, hence the commonly used nomenclature for this E coli toxin in the literature, Shiga toxin or vero toxin. Among EHEC, there are 2 major

Stx genes. Deoxyribonucleic acid damaging agents activate the dormant, toxigenic bacteriophages of E coli O157:H7 from its latent state. The virus particle may be activated from its latent state in its E coli host as a response to signals present in the gastrointestinal tract. Once released, this virus can infect susceptible E coli hosts, creating new toxin-producing cells. In addition to releasing the virion from its quiescent state in E coli, DNA-damaging agents can also induce expression and release of toxin, consequences of which may be devastating to its animal host. Therefore, antimicrobials that cause DNA damage in the bacterial cell (ie, quinolones) would be contraindicated for treatment of E coli infection.

On-Farm Intervention Strategies

Escherichia coli O157:H7 is a transient inhabitant of the gastrointestinal tract of ruminants and other mammals. Cattle and sheep feces serve as sources for contamination of food products and water sources. Escherichia coli O157:H7 is widely distributed in cattle populations throughout the world. Prevalence of infection for individual cattle varies from 0 to 28% and herd incidence varies from 0 to 75%, with more recent surveys using more sensitive assays identifying that the prevalence of E coli O157:H7 is considerably higher than reported previously. In a prevalence study of US dairies, E coli O157:H7-positive fecal samples were found in 75% of herds tested over a 6-month period. In a survey of 120 fecal samples from each of 100 feedlots in 13 states, 63% of feedlots tested positive for E coli O157:H7. A large survey of 10,415 fecal samples of US beef cattle at feedlots during October 1999 through September 2000 in 12 leading cattle feeding states revealed E coli O157 in 11% of samples, with the highest prevalence (19.9%) during September and the lowest (3.3%) during February.

Escherichia coli O157:H7 is not pathogenic in cattle and is shed in the feces of healthy cattle. Escherichia coli O157:H7 does not invade the gastrointestinal tract, and adherence to the mucosa does not appear to be a prerequisite for fecal shedding. Fecal shedding is transient in cattle, often lasting 1 to 3 months or less, but E coli O157:H7 can persist on individual farms for up to 2 years. Escherichia coli O157:H7 can be isolated from other sources on the farm, such as water, horses, sheep, milk filters, and stable flies. Fecal shedding is more prevalent in the United States and Canada during the summer months and is more prevalent in Britain in the spring and fall. Also, fecal shedding varies among different classes of animals. Weaned heifers between 3 months of age and breeding age are more likely to shed E coli O157:H7 in feces than adult cattle or younger calves. Increased fecal shedding is associated with weaning and with the first month of lactation in dairy herds, and culled dairy cattle have a higher prevalence than has been previously reported. Contaminated water troughs, particularly those that are allowed to develop sediments, provide an environment for survival, proliferation, and horizontal spread of E coli O157:H7. Feces and E coli O157:H7 contaminated water can contaminate pastures and crops where it can survive and serve as a source for contamination of feed. In addition, E coli O157:H7 proliferates to extremely high populations in moist silage.
Prevalence of *E. coli* O157:H7 has been associated with some management practices on individual farms. However, cause and effect relationships between specific management practice and increased fecal shedding have not been established.

The effects of different feeding practices were addressed in several prospective studies. Growth of *E. coli* and *Salmonella* spp increases in the rumen of sheep and cattle from which feed has been withheld. However, studies in which calves were experimentally inoculated with *E. coli* O157:H7 and subsequently had feed withheld failed to have an increase in fecal shedding. Results of studies addressing the effects of feeding different amounts of concentrate and roughage, or feeds from different plant sources, are contradictory. Given that fecal shedding of *E. coli* O157:H7 is transient in cattle, any feed change or feeding practice or stress that disturbs the normal flora of the intestine could promote transient colonization with *E. coli* O157:H7 resulting in increased shedding. This may explain the association of increased shedding with the onset of lactation, weaning, and cattle recently placed on feed at feedlots. Undercooked beef products, unpasteurized milk and dairy products, and contaminated water are all potential vehicles for human infection with *E. coli* O157:H7. Therefore, efforts to decrease fecal shedding of *E. coli* O157:H7 in cattle must be considered as an important part in any plan to reduce the incidence of human infection. However, a transient, nonpathogenic, enteric bacterium that is not even host specific for cattle does not lend itself to conventional control measures. At present, there are few recommendations that can be made with confidence. The widespread prevalence of *E. coli* O157:H7 on farms and the transient nature of shedding make preslaughter testing and trace back impractical. Vaccination is not likely to succeed because mucosal colonization is not a requirement for shedding, and prior exposure does not prevent subsequent shedding. Strategies to alter or stabilize the microflora to reduce shedding of *E. coli* O157:H7 or coliforms in general are currently under investigation. Strategies to alter the rumen and intestinal microflora to exclude establishment of the transient *E. coli*, even if only temporary, may prove to be beneficial if shedding were reduced prior to slaughter or if horizontal spread among cattle were reduced. Competitive exclusion using a mixture of *E. coli* isolated from cattle feces reduces shedding in cattle experimentally inoculated with *E. coli* O157:H7, as does administration of bacteriophages that lyse *E. coli* O157:H7. A combination of substrates from feedstuffs, along with the proper indigenous microflora, can produce antimicrobial plant compounds that are toxic to coliforms. For example, *Prevotella* spp and *Bacteroides* spp hydrolyze plant esculin to aglycones. These aglycones can inhibit growth of *E. coli* O157:H7. Improved overall sanitation on the farm to reduce fecal contamination of water troughs, other water supplies, and fecal soiling of hides should reduce environmental contamination and contamination of beef products at slaughter.

**Conclusion**

Present estimates indicate more than 1.2 billion tons of cattle manure are produced annually in the United States. Cattle manure is an important source of *E. coli* O157:H7 that is carried in the gastrointestinal tract of cattle and shed in their feces. Cattle feces can contaminate food when used as a soil amendment to fertilize crops, when introduced into water used to irrigate crops, when cattle graze near fields of food crops, when milk is obtained from dairy cows, and when intestinal contents or fecal-contaminated hides contact carcasses during slaughter operations. Because *E. coli* O157:H7 is not an invasive pathogen, contamination of meat is principally from contact with feces from cattle. In addition, many recent outbreaks of *E. coli* O157:H7 infections resulted from children handling cattle on a farm or contact with manure at a farm or fair. Exposure to *E. coli* O157:H7-contaminated food and water and direct contact with *E. coli* O157:H7-contaminated manure are important public health problems. The veterinary community should play a leadership role in finding ways to reduce the risk of human exposure to *E. coli* O157:H7 from animal sources.

**References**


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