Forage feeding to reduce pre-harvest *E. coli* populations in cattle, a review.

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Abstract

Although *E. coli* are commensal organisms that reside within the host gut, some pathogenic strains of *E. coli* can cause hemorrhagic colitis in humans. The most notable enterohemorrhagic *E. coli* (EHEC) strain is O157:H7. Cattle are asymptomatic natural reservoirs of *E. coli* O157:H7; and it has been reported that as many as 30% of all cattle are carriers of this pathogen, and in some circumstances this can be as high as 80%. Feedlot and high-producing dairy cattle are fed high grain rations in order to increase feed efficiency. Because cattle have low amylase activity, much of the starch passes to the hindgut where it is fermented. EHEC are capable of fermenting sugars released from starch breakdown in the colon, and populations of *E. coli* have been shown to be higher in grain fed cattle, and this has been correlated with *E. coli* O157:H7 shedding in barley fed cattle. When cattle were abruptly switched from a high grain (corn) diet to a forage diet, generic *E. coli* populations declined 1000-fold within 5 days and the ability of the fecal generic *E. coli* population to survive an acid shock similar to the human gastric stomach decreased. Other researchers have shown that a switch from grain to hay caused a smaller decrease in *E. coli* populations, but did not observe the same effect on gastric shock survivability. In a study that used cattle naturally infected with *E. coli* O157:H7, fewer cattle shed *E. coli* O157:H7 when switched from a feedlot ration to a forage-based diet compared to cattle continuously fed a feedlot ration. Results indicate that switching cattle from grain to forage could potentially reduce EHEC populations in cattle prior to slaughter; however the economic impact of this needs to be examined.

Key Words: *Escherichia coli*, Cattle, Forage Feeding

Abbreviations: EHEC, Enterohaemorrhagic *E. coli*; **IMS**, Immunomagnetic bead separation

Introduction

Escherichia coli is a facultative anaerobic bacterium that is a normal inhabitant of the mammalian intestinal tract (Drasar, 1974). Many *E. coli* strains are harmless or even beneficial to the host; however, some strains of *E. coli* can be pathogenic to humans and can be harbored within food animals. Although several *E. coli* strains (e.g., O111, O26) can cause hemorrhagic colitis in humans, *E. coli* O157:H7 is the best known

enterohemorrhagic *E. coli* (EHEC) strain. *Escherichia coli* O157:H7 causes over 73,000 illnesses in the United States each year and approximately 60 deaths (Mead et al., 1999). Enterohemorrhagic *E. coli* infections are estimated to cost the U.S. economy approximately $1 billion per year (USDA:ERS, 2001).

The most frequently implicated vector for *E. coli* O157:H7 outbreaks has been ground beef; and bovine-derived products have been linked to approximately 75% of outbreaks (USDA:APHIS, 1997). It is now widely accepted that ruminants are natural reservoirs of EHEC (including *E. coli* O157:H7) (Chapman et al., 1993; Rasmussen et al., 1993; Armstrong et al., 1996). Repeated outbreaks of hemorrhagic colitis linked to ground beef and/or cattle manure has firmly established the connection between cattle and *E. coli* O157:H7 in the public mind. Repeated large-scale recalls of contaminated ground beef, and the deaths of children who consumed foods contaminated by exposure to meat products have further shaken the confidence of consumers in the wholesomeness and safety of beef.

Sanitation efforts after slaughter reduce contamination of carcasses with *E. coli* O157:H7 (Elder et al., 2000), however these efforts are not necessarily enough. Approximately 30% of all cattle are asymptomatic carriers of *E. coli* O157:H7 (Elder et al., 2000). Therefore, methods that focus on reducing *E. coli* O157:H7 populations in food animals prior to entry to the food chain have the potential to reduce human illnesses. This review examines the effects of dietary manipulations on *E. coli* populations in cattle.

Enterohaemorrhagic *E. coli* as a food-borne pathogen

Escherichia coli O157:H7 was first isolated during a fatal outbreak of bloody diarrhea (hemorrhagic colitis) in 1982 caused by improperly cooked hamburger meat (Riley et al., 1983). In recent years there have been repeated outbreaks of hemorrhagic colitis in humans (Doyle et al., 1997) and in fact, *E. coli* O157:H7 infection in humans has become known as "hamburger disease", "barbecue season syndrome", or in some alarmist circles, "feedlot disease" (Martens, 2000). Even though *E. coli* O157:H7 is only one of several strains of EHEC, it is responsible for most of the large-scale outbreaks in the United States (O'Brien and Kaper, 1998).

The impact of EHEC on the human intestinal tract is profound. *Escherichia coli* O157:H7 tightly binds the bacterium to the intestinal epithelial surface and produces "attaching and effacing" lesions (Kaper et al., 1998). Enterohemorrhagic *E. coli* strains produce toxins homologous to the toxins of *Shigella dysenteriae* and are therefore also interchangeably known as Shiga Toxin-Producing or Verotoxin-Producing *E. coli* (STEC or VTEC, respectively) (Mainil, 1999). Shiga toxins inhibit protein synthesis in the intestinal epithelium producing bloody diarrhea (Su and Brandt, 1995). Shiga toxins that cross the epithelium can reach the bloodstream and eventually the kidneys and induce a condition in humans known as Hemolytic Uremic Syndrome (HUS) (Griffin, 1998). Approximately 5% of the cases of *E. coli* O157:H7 hemorrhagic colitis progress to the level of HUS which can include severe kidney damage or complete renal failure and can become life threatening, especially in children and the elderly (Mead et al., 1999). The CDC conservatively estimates 3,000 cases of HUS, and at least 60 deaths each year are attributable to *E. coli* O157:H7 in the United States (Mead et al., 1999).

Gastric shock survival

The gastric stomach serves as a barrier to intestinal colonization by pathogenic bacteria because of its low pH and enzymatic activity (Waterman and Small, 1998). However, some bacteria are able to survive gastric passage. Bacteria that are more resistant to the gastric environment therefore have a greater opportunity to survive and to ultimately colonize the intestinal tract and cause illness. The infectious dose is indicative of the virulence of pathogenic bacteria, and *E. coli* O157:H7 has an extremely low infectious dose. In one outbreak the contamination level of *E. coli* O157:H7 in uncooked hamburger meat was less than 700 cells/patty and some victims ingested very little of the (improperly) cooked meat (Griffin, 1998). In another outbreak the infectious dose of O157:H7 was less than 50 cells (Tilden et al., 1996), and for the EHEC O111, less than 1 cell/10 g of salami was sufficient to induce hemorrhagic colitis (Paton et al., 1996). Because such a low number of cells are capable of causing illness, the capacity of *E. coli* O157:H7 to survive gastric exposure directly impacts its ability to cause illness.

Recent studies have yielded conflicting results of dietary changes on the development of acid resistance of *E. coli*,

including O157:H7, and some have questioned the relevance of acid resistance (Diez-Gonzalez et al., 1998; Hovde et al., 1999). Some of this debate can be attributed to confusion over the terminology "acid resistance" which has been used interchangeably to describe both growth at acidic pH, as well as the ability to survive an acid (gastric) shock. Lin et al. (1996) suggested that a term to describe survival after an acid (gastric) shock should be "extreme acid resistance". In some cases, differences between reports can be attributed to different acid shock methodologies (e.g., differences in shock pH, length of shock time, etc.) and others can be considered artifacts of culture growth methods and recovery media (Diez-Gonzalez et al., 1998; Hovde et al., 1999; Jarvis and Russell, 2001). Because the ability to survive gastric passage is critical to the virulence of *E. coli* O157:H7, factors that impact the development of "extreme acid resistance" are important to reducing human illness and need to be elucidated through the use of a standardized acid shock methodology.

Cattle as reservoirs of enterohaemorrhagic *E. coli*

Ruminant animals are populated by a microbial consortium that allows the animal to convert cellulosic forages to high quality meat, milk or fiber (Hungate, 1966). It is well known that ruminants (both domestic and wild) can be asymptomatic reservoirs of EHEC (Wells et al., 1991; Hancock et al., 1994; Bielaszewska et al., 2000). The microbial population of the ruminant is very diverse and microbes are found throughout the reticulorumen, as well as the intestinal tract. Because the gastrointestinal tract is well-suited for microbial growth it is no surprise that the ubiquitous and adaptable *E. coli* (represented by many strains, including EHEC) lives in the gut of mammals, including cattle and humans (Drasar and Barrow, 1985).

Escherichia coli are rarely found in high numbers in the rumen of cattle (less than 10^6 cells/ml out of a population of 10^{10} cells/ml) (Wolin, 1969) and are found at concentrations from 10^2 to 10^7 cells/g feces at slaughter (Davidson and Taylor, 1978). *Escherichia coli* have rarely been considered important members of the ruminal microbial ecosystem due to the toxicity of high ruminal concentrations of VFA and competition for available nutrients (Wolin, 1969). However, this sensitivity of *E. coli* to VFA is strain specific and some *E. coli*

can grow in conditions similar to those of the rumen (Diez Gonzalez and Russell, 1997). Even though *E. coli* strains comprise a larger proportion of the intestinal microbial population (up to 1%), *E. coli* counts are highly variable and are still outnumbered by the strictly anaerobic bacterial population (Davidson and Taylor, 1978; Drasar and Barrow, 1985; Diez-Gonzalez et al., 1998). Enterohemorrhagic *E. coli* strains are very rarely the predominant strains of *E. coli* found in the rumen or intestine. Although other EHEC strains responsible for human illnesses have been isolated from cattle (e.g., O111, O136) (Hornitzky et al., 2000, Midgley et al., 1999), most surveys in cattle have primarily measured O157:H7 (Hancock et al., 1994; Hancock et al., 1998). Recently, however, Acheson (2000) has emphasized that surveys should examine the prevalence of all EHEC rather than certain species.

Cattle are not known to express receptors for Shiga toxins and do not suffer from hemorrhagic diarrhea when infected with EHEC (Pruimboom-Brees et al., 2000). Therefore it is impossible to visually identify "sick" animals. Even though cattle have been shown to be a reservoir for *E. coli* O157:H7 (Wells et al., 1991; Rasmussen et al., 1993), it has been thought that the colonization by EHEC is transient (Hancock et al., 1998). Detection of *E. coli* O157:H7 in the live animal has been hampered because studies have used enrichment culture followed by direct plate counting, which has a very low sensitivity (Buchko et al., 2000a). Even with the use of more sophisticated molecular detection methods, shedding of EHEC still appears sporadic. Estimates of *E. coli* O157:H7 and EHEC populations using molecular techniques and immunomagnetic bead separation (IMS) demonstrated an *E. coli* O157:H7 incidence of up to 100-fold greater than previously reported by use of enrichment techniques (Chapman et al., 1997a and b; Mechie et al., 1997; Buchko et al., 2000a).

While the majority of the of the epidemiological data collected on EHEC in cattle was gathered using less sensitive culture-based techniques, these studies provide important information on factors that affect the prevalence of *E. coli* O157:H7. The prevalence of *E. coli* O157:H7 in cattle was found to vary widely in several surveys, but is highly dependent on cattle age and season (USDA:APHIS, 1997; Zhao et al., 1995). Calves shed more *E. coli* O157:H7 cells and for longer periods of time than did adult cows (Zhao et al., 1995; Mechie et al., 1997). Periparturient

cows demonstrated increased fecal shedding of coliform bacteria during the period immediately before and after calving (Pelan-Mattocks et al., 2000). *Escherichia coli* O157:H7 populations in cattle vary throughout the year: as many as 80% of all feedlot cattle may be infected during the summer months, but as few as 10% may shed during the winter (Elder et al., 2000; R. O. Elder, unpublished data). This correlates with a rise in human outbreaks during each summer/early fall thus emphasizing a linkage between animal (reservoir) populations and human food-borne outbreaks. Surveys conducted throughout the United States indicated that the distribution of *E. coli* O157:H7 in cattle was not geographically linked (Dargatz et al., 1997; Hancock et al., 1997b) however this appears to contradict a report that human outbreaks are more prevalent in the northern United States (Griffin, 1998).

Escherichia coli O157:H7 colonization appears to be widespread in both beef and dairy herds and is highly variable within each animal and herd, however some herds appear to have higher shedding incidences than do others (USDA:APHIS, 1997; Hancock et al., 1998). The median percentage of *E. coli* O157:H7 positive animals in studies using traditional culture techniques was estimated at 1.7% (Jackson et al., 1998). Other culture-based studies have indicated that 4% of cattle were colonized by up to 26 different serotypes of EHEC (Schurmann et al., 2000).

Until recently, it was thought that only 1-3% of cattle were carriers of *E. coli* O157:H7. However, the use of immunomagnetic bead separation to identify *E. coli* O157:H7 in feces has steadily increased the accepted incidence value (Chapman et al., 1997a, Mechie et al., 1997). Researchers initially found that 16% of the animals tested in both beef and dairy herds were *E. coli* O157:H7 positive, and as many as 62% of dairy heifers were populated with *E. coli* O157:H7 (Mechie et al., 1997). Additional studies in Europe indicated that 18%, 32%, and 75% of dairy cows, sheep and goats, respectively (Zschöck et al., 2000), and 20% of feedlot cattle in the Czech republic were EHEC carriers (Cizek et al., 1999). In the U.S., Elder et al. (2000) demonstrated that 28% of all feedlot cattle contained *E. coli* O157:H7. More recent studies have shown that approximately 50% of feedlot cattle harbor *E. coli* O157:H7, during summer months this proportion can be as high as 80% (Keen et al., 1999; R. O. Elder, unpublished data).

These results collectively indicate that the prevalence of *E. coli* O157:H7 is much greater than was previously reported, but also indicates that processing plants effectively control the spread of *E. coli* O157:H7 after slaughter (Elder et al., 2000). However, significant levels of *E. coli* O157:H7 still enter the abattoir within the live animal and thus pose a risk to human health.

Effects of management strategies on *E. coli* populations

Several epidemiological and risk assessment studies have been performed to identify cattle management strategies associated with an increased risk of *E. coli* O157:H7 fecal shedding (Dargatz et al., 1997, Hancock et al., 1998; Herriott et al., 1998). However there have been conflicting correlations drawn because of the nature of these survey-based studies. Different dietary regimes and stages of production have been linked to *E. coli* O157:H7 shedding, however the correlations have not been great enough to lead to any new management or nutritional practices to reduce EHEC shedding.

Abrupt weaning practices have been shown to increase colonization with EHEC (Herriott et al., 1998); however, heifers older than 3 months are the most commonly colonized group of cattle (Hancock et al., 1994; Hancock et al., 1997a; Mechie et al., 1997). The only dietary practice that significantly increased the risk of EHEC shedding among heifers was feeding corn silage (Herriott et al., 1998). The use of feed additives, such as monensin and lasalocid, demonstrated a marginally significant increase of EHEC shedding by heifers (Herriott et al., 1998). In adult cows, the only dietary variable that significantly impacted EHEC shedding was the inclusion of animal by-products in the ration (Herriott et al., 1998). Other studies have found feeding whole cottonseed reduced *E. coli* O157 shedding (Hancock et al., 1994; Garber et al., 1995). Barley feeding was linked (albeit at a low correlation) to *E. coli* O157:H7 shedding (Dargatz et al., 1997); and in recent studies barley feeding was again associated with increased shedding of *E. coli* O157:H7 from experimentally-infected feedlot cattle (Buchko et al., 2000b).

Ruminal and intestinal VFA concentrations have been suggested to limit the proliferation of *E. coli* (Wolin, 1969). Feed withdrawal or starvation results in decreased VFA concentration in the rumen

and hindgut. Cattle are often transported long distances prior to slaughter and feed may be withdrawn for up to 48 h. A fasting period has been shown to increase *E. coli*, *Enterobacter* and total anaerobic bacterial populations throughout the intestinal tract (Buchko et al., 2000a; Gregory et al., 2000), increase *Salmonella* and *E. coli* populations in the rumen (Brownlie and Grau, 1967). Additionally fasting has been shown to induce "apparently *E. coli* (O157:H7) negative animals to become positive" (Kudva et al., 1995). Other studies have indicated that fasting made calves more susceptible to colonization by *E. coli* O157:H7 and demonstrated that fasted calves shed higher populations of *E. coli* O157:H7 than did calves fed normally (Cray et al., 1998). Other researchers have shown that cattle fasted for 48 h prior to slaughter contained significantly greater *E. coli* populations throughout the gut than cattle fed hay or pasture (Gregory et al., 2000). In contrast however, Harmon et al. (1999) demonstrated that fasting reduced ruminal VFA concentrations but did not significantly influence *E. coli* O157:H7 shedding.

Determination of dietary and management factors that influence EHEC shedding has been difficult using culture-based methodologies and surveys. The use of more sensitive molecular detection methods, such as IMS, in future studies may be able to elucidate subtle correlations between dietary factors and fecal shedding of EHEC. Additionally, direct, controlled experiments rather than surveys, need to be conducted to determine the impact of specific dietary modifications on intestinal EHEC populations and shedding in cattle.

Forage- versus grain-based diet effects on fecal *E. coli* populations

Finishing beef and lactating dairy cattle in the United States are often fed high grain rations in order to improve performance and animal production (Huntington, 1997). Ruminant animals evolved to eat cellulosic plant material, however the ruminal microbial population can degrade starch. Dietary starch is often enclosed by a protein (zein) matrix that protects the starch from ruminal microbial degradation and allows much of the starch to reach the intestine (Huntington, 1997). Ruminants have little pancreatic amylase activity therefore much of the dietary starch passes through the small intestine to the cecum and colon where it undergoes a secondary microbial fermentation (Huntington, 1997). Colonic and cecal

starch fermentation by bacteria (including EHEC) produces VFA that can reduce the pH of the colonic digesta and inhibit *E. coli*. However in spite of these harsh conditions, *E. coli* thrives in the intestinal tract of cattle fed high-grain rations (Allison et al., 1975; Diez-Gonzalez et al., 1998; Keen et al., 1998; Tkalcic et al., 2000; Scott et al., 2000; Stanton and Schutz, 2000).

Feeding grain to cattle has a pronounced effect on the ruminal microbial ecosystem and overall animal health (Russell and Rychlik, 2001). Studies have indicated that varying the forage to grain ratio in cattle rations can have a marked effect on shedding of *E. coli* O157:H7, but some studies have produced contradictory results (Table 1). Early studies indicated that a sudden decrease in hay intake increased *E. coli* populations in cattle feces (Brownlie and Grau, 1967). Overfeeding of cattle with grain has been shown to cause a 2-$\log_{10}$ increase in total coliform counts (Allison et al., 1975). Other studies using experimentally infected sheep found that a switch from an alfalfa pellet diet to a low quality forage diet increased *E. coli* O157:H7 shedding (Kudva et al., 1995). Kudva et al. (1997) found that sheep shifted from a 50:50 corn/alfalfa ration to poor quality grass hay shed greater populations of *E. coli* O157:H7 than animals fed the corn/alfalfa ration.

In recent research, cattle fed a 90% corn/soybean meal ration (feedlot-type ration) contained generic *E. coli* populations that were 1000-fold higher than cattle fed a 100% good-quality hay (Timothy) diet (Diez-Gonzalez et al., 1998). The *E. coli* recovered from the feces of grain-fed cattle were 1000-fold more resistant to an "extreme" acid shock that simulated passage through the human stomach than were *E. coli* from cattle fed only hay (Diez-Gonzalez et al., 1998). When cattle were abruptly switched from a 90% grain finishing ration to a 100% hay diet, fecal *E. coli* populations declined 1000-fold, and the population of *E. coli* resistant to an extreme acid shock declined more than 100,000-fold within 5 d (Diez-Gonzalez et al., 1998). Although no *E. coli* O157:H7 were specifically detected in this study, it was previously demonstrated that *E. coli* O157:H7 could grow in VFA concentrations and at pH's similar to those found in the colon of these grain-fed cattle (Diez-Gonzalez and Russell, 1997). Based on these results the authors suggested that feedlot cattle be switched from high grain diets to hay for 5 days prior to slaughter to reduce *E. coli* contamination entering the abattoir (Diez-Gonzalez et al., 1998). An

independent study examining the effect of this switch from grain to hay on cattle performance and carcass characteristics indicated that cattle fed hay during this final period had lower DMI and lost an average of 2.2 lb/head/d (Stanton and Schutz, 2000). Hot carcass weight and dressing percentage were not significantly reduced by hay feeding (Stanton and Schutz, 2000). Hay feeding did not significantly impact carcass grades, quality parameters or cause dark cutters, but did significantly reduce total coliform counts as well as generic *E. coli* counts (Stanton and Schutz, 2000), but they were not reduced as greatly as those reported by Diez-Gonzalez et al. (1998). Neither *E. coli* O157:H7 populations, nor the acid resistance of *E. coli* were measured in this study (Stanton and Schutz, 2000). In another study, cattle fed hay for 48 h prior to transport to slaughter did not lose more weight during transport than fasted or pasture fed animals (Gregory et al., 2000).

Keen et al., (1999) also found that switching cattle from grain to hay caused a decrease in body weight (approximately 1.25 lb/hd/d compared to controls). Through the use of modern molecular separation techniques, 200 cattle maintained on a grain ration were screened for natural *E. coli* O157:H7 infection and 53% were found to be positive (Keen et al., 1999). When these cattle were divided into two groups and one was fed grain and the other abruptly switched to hay, 52% of the grain-fed cattle remained *E. coli* O157:H7 positive, but only 18% of the hay-fed cattle continued to shed *E. coli* O157:H7 (Keen et al., 1999). These results again indicated that feeding hay could impact the fecal shedding of *E. coli* and potentially reduce EHEC entry into the food chain.

The proposal of such a dietary switch provoked a great deal of scientific controversy (Hancock et al., 1999; Russell and Diez-Gonzalez, 1999; Russell et al., 2000) and led to several studies that have evaluated the effect of radical dietary changes on *E. coli* populations in cattle, however these studies have also produced conflicting results (Table 1). When cattle were fed a high-concentrate diet and switched to a diet containing 50% corn silage and 50% alfalfa hay, generic *E. coli* counts decreased 0.3 log in 4 days (Jordan and McEwen, 1998). Cattle fed an 80% barley ration, fasted for 48 h and switched to 100% alfalfa silage did not exhibit any change in *E. coli* O157:H7 shedding (Buchko et al., 2000a). However, when these same forage-fed animals were again fasted for 48 h and re-fed 100% alfalfa

silage, the prevalence of *E. coli* O157:H7 shedding increased significantly (Buchko et al., 2000a).

Using cattle experimentally infected with *E. coli* O157:H7, Hovde et al. (1999) found that cattle fed hay shed *E. coli* O157:H7 longer than did grain-fed cattle (42 d vs. 4 d, respectively), but concentrations of *E. coli* O157:H7 shed were similar between dietary regimes. Generic coliform bacteria from these hay-fed cattle were significantly more sensitive to acid shock than those from grain-fed cattle (Hovde et al., 1999), but the difference in acid shock sensitivity was not as great as that found by Diez-Gonzalez et al (1998). Feeding a high-grain or -forage diet did not affect the acid resistance of *E. coli* O157:H7 isolated from these cattle; however, the researchers were unable to demonstrate sensitivity to an acid shock under their experimental conditions with an extreme shock-sensitive *E. coli* O157:H7 control strain (Hovde et al., 1999). Additionally, differences in culture methodologies make direct comparisons between the studies of Hovde et al. (1999) and Russell et al. difficult (Jarvis and Russell, 2001).

Other research groups have reported high grain or high forage diets did not affect the duration of shedding or fecal *E. coli* O157:H7 populations in experimentally inoculated calves, however the calves that consistently shed the highest concentrations of *E. coli* O157:H7 were fed a high concentrate diet (Tkalcic et al., 2000). Ruminal fluid from steers fed a high-forage diet allowed greater proliferation of *E. coli* O157:H7 in vitro than did ruminal fluid from high-grain fed steers (Tkalcic et al., 2000), possibly due to differences in VFA concentrations. Acid shock experiments indicated that *E. coli* O157:H7 incubated in ruminal fluid taken from steers fed a high-grain diet was more acid shock-resistant than *E. coli* O157:H7 cells incubated in forage-fed ruminal fluid (Tkalcic et al., 2000). The authors stated that the ability of *E. coli* O157:H7 to become acid resistant could be one factor that influences fecal shedding in cattle (Tkalcic et al., 2000).

In a recent study, it was demonstrated that switching cattle from pasture to hay for 48 h prior to slaughter significantly reduced the *E. coli* burden throughout the gut (Gregory et al., 2000). The authors found that hay feeding increased intestinal *Enterococci* populations that are capable of inhibiting *E. coli* populations (Gregory et al., 2000). However, in this study the effects of high grain versus forage diets were not examined,

only the effects of fasting vs. pasture vs. hay-feeding (Gregory et al., 2000). Based on their results, the authors concluded, "the most effective way of manipulating gastro-intestinal counts of *E. coli* was to feed hay" (Gregory et al., 2000).

Because switching feedlot cattle from grain to hay immediately prior to slaughter is not immediately practicable, feeding low-starch or high-fiber rations has been suggested as an alternative method to reduce *E. coli* O157:H7 shedding by reducing the starch load in the colon prior to slaughter (Scott et al., 2000). Fecal samples from cattle fed dry rolled corn, high-moisture corn and wet corn gluten feed did not contain different populations of generic *E. coli*, or extreme acid-resistant *E. coli* during a limit-feeding period (Scott et al., 2000). However, cattle fed wet corn gluten *ad libitum* contained significantly higher concentrations of extreme acid resistant *E. coli* than cattle fed dry-rolled or high moisture corn (Scott et al., 2000). When these cattle were abruptly switched from a finishing diet to alfalfa hay, colonic pH increased, total *E. coli* populations decreased approximately 10-fold and acid-shock resistant *E. coli* populations were reduced by over 99% (Scott et al., 2000). These authors concluded "increased colonic pH was not associated with reduced populations of acid resistant *E. coli*" but "feeding hay for a short duration can reduce acid-resistant *E. coli* populations" (Scott et al., 2000). Again, these results emphasize that dietary manipulations (e.g., hay feeding) could be a powerful method to reduce *E. coli*/EHEC populations in cattle prior to harvest.

Cleanliness of animals entering holding pens at the abattoir is an important, and often overlooked factor that can impact the incidence of food-borne illness. Dried manure on the hide and hooves has been implicated as a primary route of contamination of carcasses via removal machinery (Grau, 1987; Hancock, 1999). However, Elder et al. (2000) demonstrated that there was a direct correlation between fecal populations of *E. coli* O157:H7 and carcass contamination levels. In a study by Gregory et al. (2000), when cattle arrived at the slaughter plant the hides of cattle fed hay for 48 h prior to transport were as clean as fasted cattle, and were significantly cleaner than pasture-fed cattle (Gregory et al., 2000). Therefore these authors stated that feeding hay prior to transport to slaughter "offered the most advantages" (Gregory et al., 2000). Feeding strategies that result in cattle arriving at the abattoir with less "tag"

on the hide at the time of slaughter can greatly enhance food safety.

Implications

The United States has the safest food supply in the history of the world, however food-borne pathogenic bacteria are still significant threats to human health. Sanitation steps following slaughter effectively reduce carcass contamination with *E. coli* O157:H7, but pre-harvest intervention strategies offer avenues to reduce pathogen populations in food animals before they enter the food chain. Attempts to modify fecal shedding of *E. coli* O157:H7 through fasting and feeding poor-quality forages have been shown to increase shedding in cattle. However, abruptly switching cattle from a high grain ration to a high-quality hay-based diet has been shown to reduce generic *E. coli* and *E. coli* O157:H7 populations, but the magnitude of reduction has varied among studies. Switching all feedlot cattle in the U.S. from grain-based diets to hay prior to slaughter is not currently feasible, in spite of the potential benefits. Further research is needed to elucidate the mechanism by which forage-feeding impacts the microbial ecology of the bovine intestinal tract, including *E. coli* populations, so that economically viable dietary modifications can be devised and implemented.

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Table 1. Published reports of effects of diet on fecal *E. coli* populations

Authors	Concentrate diet	*E. coli* CFU/g feces		Forage diet	*E. coli* CFU/g feces	Log_{10} impact
Allison, 1975	Normal diet Over fed grain	8×10^6 coliforms 1×10^{10} coliforms				
Kudva, et al., 1995	100% Alfalfa pellets	Shed O157:H7 for 4 d		Sagebrush/bunchgrass	Shed O157:H7 for 15 d	
Diez-Gonzalez et al., 1998	90% Concentrate	8×10^7		100% Timothy Hay	3×10^4	-3.5
Jordan and McEwen, 1998	44% Dry corn 7% Dry Gluten 7% Distiller's Dried Grains	7×10^6		50% Corn silage 50% Alfalfa	4×10^6	-0.3
Keen et al., 1999	85% Concentrate	52% shedding O157:H7		100% Forage	18% shedding O157:H7	
Hovde et al., 1999	62% Barley/19% corn 90% Corn	7×10^6 peak (4 d of shedding)		Alfalfa or Grass Hay	7×10^6 peak (39 or 42 d shedding)	
Stanton and Schutz, 2000	85% Whole Corn	3.2×10^7		30% Millet hay 62% whole corn	1×10^6	-1.2
Scott et al., 2000	84% Dry rolled corn or 41% Dry rolled corn 45% Wet corn gluten	3×10^8 5×10^8		100% Alfalfa hay	1×10^7 9×10^6	-1.2 -1.6
Bucko et al., 2000a	80% Concentrate	5% shedding O157:H7		100% Alfalfa silage (after 48 h fast) Re-fed 100% Alfalfa silage (after 48 h fast)	5% shedding O157:H7 42% shedding O157:H7 after 5 d	