Letters in Applied Microbiology ISSN 0266-8254

ORIGINAL ARTICLE

Prevalence of *Escherichia coli* O157:H7 and *Salmonella* in beef steers consuming different forage diets*

M.L. Looper[1], T.S. Edrington[2], R. Flores[3], C.F. Rosenkrans Jr[3], M.E. Nihsen[3] and G.E. Aiken[4]

1 USDA-ARS, Dale Bumpers Small Farms Research Center, Booneville, AR, USA
2 USDA-ARS, Food and Feed Safety Research Unit, College Station, TX, USA
3 Department of Animal Science, University of Arkansas, Fayetteville, AR, USA
4 USDA-ARS, Forage Animal Production Research Unit, Lexington, KY, USA

Keywords
anthelmintic, beef cattle, *Escherichia coli* O157:H7, *Salmonella*, tall fescue.

Correspondence
M.L. Looper, USDA-ARS, Dale Bumpers Small Farms Research Center, 6883 South State Hwy 23, Booneville, AR 72927, USA. E-mail: mlooper@spa.ars.usda.gov

*Names are necessary to report factually on available data; however, the USDA does not guarantee or warrant the standard of the product, and the use of the name by the USDA implies no approval of the product to the exclusion of others that also may be suitable.

Present address
M.E. Nihsen, Montefiore's Institute for Reproductive Medicine and Health, 141 South Central Ave. Suite 201, Hartsdale, NY 10530, USA.

2005/1077: received 19 September 2005, revised 9 December 2005 and accepted 20 December 2005

doi:10.1111/j.1472-765X.2006.01888.x

Abstract

Aims: To compare the prevalence of faecal shedding of *Escherichia coli* O157:H7 and *Salmonella* in growing beef cattle consuming various forages.
Methods and Results: In Experiment I, faecal samples were collected from steers grazing either endophyte-infected (E+) tall fescue or common bermudagrass (CB). Steers grazing E+ tall fescue were confined to a dry-lot pen and fed CB hay *ad libitum* for 10 days. In Exp. II, faecal samples were collected from steers grazing either E+ or novel endophyte-infected (NE) tall fescue and treated with one of two anthelmintics: ivermectin (I) or fenbendazole (F). In Exp. I, prevalence of *E. coli* O157:H7 was less in E+ tall fescue steers fed CB hay than steers grazing CB. More I-treated steers shed *Salmonella* than F-treated steers at 42-day postanthelmintic treatment but shedding of *Salmonella* was similar between anthelmintics at day 63 in Exp. II.
Conclusions: Faecal shedding of pathogenic bacteria was not affected by grazing E+ tall fescue. Alterations of forage diets may influence the prevalence of *E. coli* O157:H7, and anthelmintic treatment could affect faecal shedding of *Salmonella*.
Significance and Impact of the Study: Knowledge of factors that influence shedding of pathogenic bacteria in cattle is necessary to develop on-farm intervention strategies aimed at reducing pathogen shedding.

Introduction

Limited studies with grazing systems suggest pasture-based cattle are infected with *Escherichia coli* O157:H7 (Laegreid *et al.* 1999; Riley *et al.* 2003; Dunn *et al.* 2004) and *Salmonella* (Looper *et al.* 2003; Fossler *et al.* 2005); it is probable that some of these animals could shed bacteria during the feedyard phase and at harvest.

Stress may predispose cattle to be more susceptible to opportunistic bacteria such as *E. coli* O157:H7 and *Salmonella* (Fitzgerald *et al.* 2003; Looper *et al.* 2005). Cattle grazing endophyte-infected (E+) tall fescue, a cool-season forage found throughout the Southeastern US, are exposed to ergot alkaloids, which cause several disorders, collectively characterized as fescue toxicosis (Hoveland *et al.* 1983; Paterson *et al.* 1995). A novel endophyte-infected (NE) tall fescue variety has been developed (West *et al.* 1998; Sleper *et al.* 2002) that is ergopeptine-deficient and does not reduce animal performance (Nihsen *et al.* 2004). Studies comparing the prevalence of faecal shed-

ding of foodborne pathogens from cattle consuming different forage diets are limited. Objectives of these studies were to compare the prevalence of faecal shedding of *E. coli* O157:H7 and *Salmonella* in growing beef cattle consuming E+ tall fescue, NE tall fescue or common bermudagrass (CB).

Materials and methods

Animal models and collection of faecal samples

All animal procedures used in this study were approved by the committee for animal welfare at the USDA-ARS, Dale Bumpers Small Farms Research Center in Booneville, AR, USA. In Experiment I, faecal samples were collected on days 1, 64, and 95 from crossbred (≤1/4 *Bos indicus*), yearling steers (initial body weight = 260 ± 4 kg) grazing either E+ tall fescue (*Festuca arundinacea* Schreb; $n = 36$) or CB [*Cynodon dactylon* (L.) Pers; $n = 32$] paddocks (30 March to 2 July). Faecal samples were collected when body weights of steers were recorded to minimize animal handling and stress, and to maximize labour resources. All steers were treated with an anthelmintic (Dectomax, Pfizer Animal Health, New York, NY, USA; 200 μg kg^{-1} of body weight) for control of internal parasites at the initiation of the experiment. On day 85, steers grazing E+ tall fescue paddocks were confined to a dry-lot pen and fed CB hay *ad libitum* for 10 days. Faecal samples were collected at the end of the dry-lot phase (day 95). Steer weights at the initiation and termination of the experiment were used to determine average daily gain (ADG).

Experiment II included collection of faecal samples on days 42 and 63 from crossbred Angus steers (initial body weight = 314 ± 4 kg) grazing E+ ($n = 18$) or NE tall fescue ($n = 12$) for 63 days (17 June to 19 August). As in Exp. I, faecal samples were collected when body weights of steers were recorded to minimize animal handling and stress, and to maximize labour resources. Steers grazing each forage type were treated every 21 days with either ivermectin (I; Ivomec, Merial Limited, Iselin, NJ, USA; 0·2 mg kg^{-1} of body weight) or fenbendazole (F; Safeguard, Intervet, Inc., Millsboro, DE, USA; 15 mg kg^{-1} of body weight) for control of internal parasites. Both forage treatments also received a mineral containing fenbendazole (Vigortone Ag Products, Hiawatha, IA, USA; 1 mg kg^{-1} of body weight). Steer weights at the initiation and termination of the experiment were used to calculate ADG.

Blood serum samples were collected (Exp. I = 1 June; Exp. II = 8 July) from steers for quantification of prolactin (Bernard *et al.* 1993) to assess fescue toxicosis. Intra- and interassay coefficient of variation (CV) were 2% and 5% respectively. In both experiments, approximately 30 g of faecal material was obtained via the rectum using a separate sterile veterinary palpation sleeve for each animal. Faecal samples were placed in Whirlpacks™ (Modesto, CA, USA), packed on ice, and shipped to the USDA-ARS, Food and Feed Safety Research Laboratory, College Station, TX, USA, to determine the prevalence of *E. coli* O157:H7 and *Salmonella*.

Bacterial culture and isolation

An immunomagnetic separation (IMS) technique was used to isolate *E. coli* O157:H7. Faeces (10 g) were enriched in 90 ml of Gram-negative broth containing vancomycin (8 μg ml^{-1}), cefixime (0·5 μg ml^{-1}), and cefsoludin (10 μg ml^{-1}). Following incubation for 6 h at 37°C, 20 μl of anti-*E. coli* O157:H7 antibody-labelled paramagnetic beads (Neogen Corp., Lansing, MI, USA) were added to 1 ml volumes of the above enrichments, mixed and washed thrice in 1 ml of phosphate-buffered saline (PBS) with 0·05% Tween 20 (Keen and Elder 2002). Fifty microlitres of the resulting suspension was plated on CHROMagar™ O157 (DRG International, Mountain Side, NJ, USA) plates [containing potassium tellurite (2·5 μg ml^{-1})] and incubated overnight at 37°C. Pink colonies exhibiting *E. coli* O157:H7 colony morphology were resuspended in PBS and confirmed as *E. coli* O157:H7 using the Reveal® microbial screening test (Neogen Corp., Lansing, MI, USA).

Salmonella was cultured by enriching approximately 10 g of faecal material in 90 ml of tetrathionate broth for 24 h at 37°C. The enrichment (200 μl) was added to 5 ml of Rapport–Vassilidis R10 broth and incubated for 24 h at 42°C before plating on brilliant green agar supplemented with novobiocin (25 μg ml^{-1}). Colonies exhibiting *Salmonella* morphology were confirmed biochemically using lysine agar and triple sugar iron agar tests. *Salmonella*-positive samples were confirmed by slide agglutination using SM-O antiserum poly A-I and V-I, and group C1 factors.

Statistical analyses

Chi-squared analysis, using the FREQ procedure of SAS (SAS Inst., Inc., Cary, NC, USA), was used to determine the influence of forage treatment on the frequency of positive *E. coli* O157:H7 samples in Exp. I. In Exp. II, effects of forage and anthelmintic treatment, and their interaction on the frequency of positive *Salmonella* samples were analysed with the CATMOD procedure of SAS. The effect of forage treatment, faecal shedding of pathogenic bacteria, and their interaction with ADG for both experiments was analysed by ANOVA using the MIXED procedure of SAS. Data from replicate samplings were pooled for analysis.

Results

Experiment I

Overall, the percentage of samples positive for *E. coli* O157:H7 was 5·4% (11 of 204 samples). The percentage of steers ($n = 68$) positive for *E. coli* O157:H7 on at least one occasion was 16·2% (11 of 68 steers). Overall percentage of samples positive for *Salmonella* was 0·5% (one of 204 samples); the single positive sample was detected on day 1. The percentage of steers ($n = 68$) positive for *Salmonella* on at least one occasion was 1·5% (one of 68 steers); consequently, statistical analysis to determine effects of forage treatment on faecal shedding of *Salmonella* was not performed. Faecal shedding of *E. coli* O157:H7 was not detected from steers on either forage treatment at the initiation (day 1) of the experiment. Forage treatment did not influence ($P > 0·10$) the incidence of faecal shedding of *E. coli* O157:H7 in steers grazing E+ tall fescue (5·6%) or CB (6·3%) on day 64. Because of the low prevalence of both *E. coli* O157:H7 (four of 136 samples) and *Salmonella* (one of 136 samples) from steers on days 1 and 64, faecal samples were not collected on day 85 prior to switching steers that were grazing E+ tall fescue paddocks to CB hay. On day 95, no *E. coli* O157:H7 was detected in the faeces of E+ tall fescue steers fed CB hay; however, 22% of steers grazing CB shed *E. coli* O157:H7 ($P < 0·05$). Concentrations of prolactin were decreased ($P < 0·05$) in steers grazing E+ ($10·1 \pm 3·6$ ng ml^{-1}) compared with steers grazing CB ($170·9 \pm 16·3$ ng ml^{-1}).

Average daily gain was not different ($P > 0·10$) between steers shedding *E. coli* O157:H7 at least once during the experiment ($0·88 \pm 0·08$ kg d^{-1}) and steers not shedding *E. coli* O157:H7 ($0·99 \pm 0·05$ kg d^{-1}). The forage treatment × faecal shedding of *E. coli* O157:H7 interaction was not significant ($P > 0·10$); however, E+ tall fescue steers fed CB hay had decreased ($P < 0·05$) ADG for the entire experiment (95 days) compared with steers grazing CB ($0·69 \pm 0·07$ *vs* $1·18 \pm 0·09$ kg d^{-1} for E+/CB hay and CB steers respectively).

Experiment II

Culture for *E. coli* O157:H7 could not be performed for 11 steers on day 63 because of insufficient faecal sample. Overall percentage of samples positive for *E. coli* O157:H7 was 2·0% (one of 49 samples); the positive sample was detected on day 42. The percentage of steers ($n = 30$) positive for *E. coli* O157:H7 on at least one occasion was 3·3% (one of 30 steers). Because of limited detection of *E. coli* O157:H7, statistical analysis was not conducted on the effects of forage and anthelmintic treatment on faecal shedding of *E. coli* O157:H7. Culture for *Salmonella* could not be performed for two steers on day 42 because of insufficient collection of faeces. Overall percentage of samples positive for *Salmonella* was 37·9% (22 of 58 samples). The percentage of steers ($n = 30$) positive for *Salmonella* on at least one occasion was 73·3% (22 of 30 steers). No nematode eggs were observed in either anthelmintic treatment on days 21 or 42 of grazing. A forage × anthelmintic treatment interaction for faecal shedding was not observed ($P > 0·10$). Faecal shedding of *Salmonella* was not affected ($P > 0·10$) by forage treatment. Detectable faecal shedding of *Salmonella* was reduced ($P < 0·05$) in F-treated steers (60%) than I-treated steers (92%) on day 42 (Table 1). However, prevalence of faecal shedding of *Salmonella* on day 63 was not different ($P > 0·10$) between anthelmintic treatments and averaged 3·3%. Concentrations of prolactin were decreased ($P < 0·05$) in E+ tall fescue steers ($3·1 \pm 0·5$ ng ml^{-1}) compared with NE tall fescue steers ($50·2 \pm 9·1$ ng ml^{-1}).

Average daily gain was affected ($P < 0·001$) by forage treatment; steers grazing E+ tall fescue gained an average of $0·80 \pm 0·06$ kg day^{-1} while NE-steers gained $1·29 \pm 0·08$ kg day^{-1}. A faecal shedding × type of anthelmintic interaction tended ($P = 0·08$) to influence ADG. Steers treated with fenbendazole and not shedding *Salmonella* had reduced ADG ($0·93 \pm 0·1$ kg day^{-1}) compared with I-treated steers not shedding *Salmonella* ($1·2 \pm 0·1$ kg day^{-1}).

Table 1 Percentage of steers shedding *Salmonella* in the faeces on day 42 grazing endophyte-infected (E+) or novel endophyte-infected (NE) tall fescue and treated with either ivermectin (I) or fenbendazole (F) (Exp. II)

	Forage treatment						
	E+		NE		*P*-value		
Variable	Ivermectin	Fenbendazole	Ivermectin	Fenbendazole	F	A	F × A
Salmonella (%)	100 (7/7)	56 (5/9)	83 (5/6)	67 (4/6)	0·99	0·05	0·21

F, forage treatment (E+ or NE); A, anthelmintic treatment (I or F); F × A, forage × anthelmintic interaction; Number of observations are given in parentheses.

Discussion

Consumption of E+ tall fescue by growing beef cattle in the current study did not influence faecal shedding of either pathogen. Cattle grazing E+ tall fescue have increased body temperature during summer months, reduced reproductive performance and growth rate, and decreased milk production (Hoveland *et al.* 1983; Paterson *et al.* 1995). Ergot alkaloids produced by the fungus *Neotyphodium coenophialum* (Hill *et al.* 1994; Glenn *et al.* 1996) in E+ tall fescue decrease concentrations of prolactin via dopaminergic activity (Schillo *et al.* 1988; Porter and Thompson 1992). Reduced concentrations of prolactin were observed in steers grazing E+ tall fescue in both experiments compared with steers grazing CB and NE tall fescue. Recently, Looper *et al.* (2005) reported faecal shedding of *E. coli* O157:H7 was increased in ewes experimentally inoculated with *E. coli* O157:H7 and fed an E+ tall fescue seed diet for 7 days compared with *E. coli* O157:H7-inoculated ewes fed an endophyte-free (E−) tall fescue seed diet. Shedding of *E. coli* O157:H7 in naturally infected animals is highly sporadic, with the possibility of an animal testing positive at one sample collection and negative at the subsequent collection (Callaway *et al.* 2004), and may explain variations among these studies. Length of exposure to the toxic effects of E+ tall fescue, differences in concentrations of ergot alkaloids in E+ tall fescue responsible for the toxicosis effects, physical attributes of forage *vs* concentrate diets, and (or) the possible differences in the pattern of faecal shedding of cattle and sheep may also have influenced the results among studies.

Steers grazing E+ tall fescue and fed CB hay for 10 days were less likely to shed *E. coli* O157:H7 than steers grazing CB in Exp. I. These preliminary data should be interpreted with some caution. Faecal samples were not collected on day 85 prior to switching E+ tall fescue steers to a hay diet because of the low prevalence of both *E. coli* O157:H7 (four of 136 samples) and *Salmonella* (one of 136 samples) on the two previous collection dates. A major goal of this pilot project was to establish baseline prevalence levels of *E. coli* O157:H7 and *Salmonella* in cattle consuming E+ tall fescue as these data are non-existent. Our sampling strategy may not have fully elucidated the effects of feeding hay on the prevalence of *E. coli* O157:H7 because of the sporadic shedding of *E. coli* O157:H7 in naturally infected animals (Callaway *et al.* 2004). However, numerous studies have suggested that altering the forage-to-grain ratio in cattle diets can influence faecal shedding of *E. coli* O157:H7, although results have not always been conclusive (Callaway *et al.* 2003). Diez-Gonzalez *et al.* (1998) reported generic *E. coli* from the faeces of grain-fed cattle was 100-fold higher and 1000-fold more resistant to acid shock compared with *E. coli* from the faeces of hay-fed cattle. Generic *E. coli* populations were reduced 1000-fold in cattle switched from a concentrate diet to a hay diet (Diez-Gonzalez *et al.* 1998). Faecal *E. coli* was reduced in cattle fed hay for 48 h prior to slaughter compared with cattle grazing pasture or fasted (Gregory *et al.* 2000). However, Buchko *et al.* (2000) reported an increase in faecal shedding of *E. coli* O157:H7 from cattle fasted and then re-fed an all-forage diet. Steers fed hay diets and inoculated with *E. coli* O157:H7 shed the pathogen longer (average = 39–42 days) than steers inoculated with *E. coli* O157:H7 and fed concentrate diets (average = 4 days; Hovde *et al.* 1999). Acid resistance of *E. coli* O157:H7 was similar among diets.

In Exp. II, faecal shedding of *Salmonella* was reduced in steers treated with fenbendazole when compared with steers treated with ivermectin at day 42; however, prevalence of *Salmonella* was not different between anthelmintic treatments at day 63. Parasitic infestations can induce stress in animals, and stress may result in increased faecal shedding of *E. coli* O157:H7 and *Salmonella* (Fitzgerald *et al.* 2003; Looper *et al.* 2005). Parasite-induced stress in the current study is unlikely as faecal nematode eggs were not detected in steers from either anthelmintic treatment. The dramatic reduction in the prevalence of faecal shedding of *Salmonella* between day 42 (75%) and day 63 (3·3%) also may have influenced the current findings. Prevalence of *Salmonella* was variable for dairy cattle in the summer and ranged from 15% to 92% (Edrington *et al.* 2004). Further research with an increased frequency of faecal collections to fully investigate anthlemintic treatment on pathogenic bacteria shedding is warranted.

Average daily gain of steers was not influenced by shedding *E. coli* O157:H7 or *Salmonella* in either of the two experiments. Presence of *E. coli* O157:H7 and *Salmonella* in mature ruminants (Hancock *et al.* 1994; Bialaszewska *et al.* 2000) usually is asymptomatic while *Salmonella* can be pathogenic in young calves (Rings 1985). Cattle on both forage types, independent of anthelmintic treatment, gained weight during the experiment. The tendency for I-treated steers not shedding *Salmonella* to have greater ADG than F-treated steers that did not shed *Salmonella* is probably because of the limited number of observations in these treatment groups. The main effect of anthelmintic was not significant ($P = 0{\cdot}17$). Not surprisingly, steers consuming E+ tall fescue had decreased ADG in both experiments compared with steers grazing CB or NE forages. The decrease in ADG in Exp. I was a result of steers grazing E+ tall fescue for 85 days and not because of switching these steers to CB hay for 10 days. Numerous studies have reported reduced performance in ruminants consuming E+ tall fescue

(Hoveland *et al.* 1983; Paterson *et al.* 1995; Looper *et al.* 2005).

Results of these preliminary experiments help establish baseline prevalence levels of *E. coli* O157:H7 and *Salmonella* in beef cattle grazing different forages, especially E+ tall fescue. Further, these experiments demonstrate the variability of pathogen shedding in grazing beef cattle and the complexity of pathogen control at the farm level. Stressful conditions generally associated with consumption of E+ tall fescue did not influence faecal shedding of *E. coli* O157:H7 or *Salmonella*. Knowledge of factors that influence the shedding of pathogenic bacteria in cattle is necessary to identify the time(s) when pathogen control may be most effective.

Acknowledgements

The authors gratefully acknowledge L. Huddleston, S. Tabler and B. Woolley, USDA-ARS, for technical assistance and daily animal management.

References

Bernard, J.K., Chestnut, A.B., Erickson, B.H. and Kelly, F.M. (1993) Effects of prepartum consumption of endophyte-infected tall fescue on serum prolactin and subsequent milk production. *J Dairy Sci* **76**, 1928–1933.

Bialaszewska, M., Schmidt, H., Liesegang, A., Prager, R., Rabsch, W., Tschape, H., Cizek, A., Janda, J. *et al.* (2000) Cattle can be a reservoir of sorbitol-fermenting shiga toxin-producing *Escherichia coli* O157:H7-strains and a source of human diseases. *J Clin Microbiol* **38**, 3470–3473.

Buchko, S.J., Holley, R.A., Olson, W.O., Gannon, V.P.J. and Veira, D.M. (2000) The effect of fasting and diet on fecal shedding of *Escherichia coli* O157:H7 by cattle. *Can J Anim Sci* **80**, 741–744.

Callaway, T.R., Elder, R.O., Keen, J.E., Anderson, R.C. and Nisbet, D.J. (2003) Forage feeding to reduce pre-harvest *E. coli* populations in cattle, a review. *J Dairy Sci* **86**, 852–860.

Callaway, T.R., Anderson, R.C., Edrington, T.S., Genovese, K.J., Bischoff, K.M., Poole, T.L., Jung, Y.S., Harvey, R.B. *et al.* (2004) What are we doing about *Escherichia coli* O157:H7 in cattle? *J Anim Sci* **82**(E. Suppl.), E93–E99.

Diez-Gonzalez, F., Callaway, T.R., Kizoulis, M.G. and Russell, J.B. (1998) Grain feeding and the dissemination of acid-resistant *Escherichia coli* from cattle. *Science* **281**, 1666–1668.

Dunn, J.R., Keen, J.E., Del Vecchio, R., Wittum, T.E. and Thompson, R.A. (2004) *Escherichia coli* O157:H7 in a cohort of weaned, preconditioned range beef calves. *J Food Prot* **67**, 2391–2396.

Edrington, T.S., Hume, M.E., Looper, M.L., Schultz, C.L., Fitzgerald, A.C., Callaway, T.R., Genovese, K.J., Bischoff, K.M. *et al.* (2004) Variation in the faecal shedding of *Salmonella* and *E. coli* O157:H7 in lactating dairy cattle and examination of *Salmonella* genotypes using pulsed-field gel electrophoresis. *Lett Appl Microbiol* **38**, 366–372.

Fitzgerald, A.C., Edrington, T.S., Looper, M.L., Callaway, T.R., Genovese, K.J., Bischoff, K.M., McReynolds, J.L., Thomas, J.D. *et al.* (2003) Antimicrobial susceptibility and factors affecting the shedding of *E. coli* O157:H7 and *Salmonella* in dairy cattle. *Lett Appl Microbiol* **37**, 392–398.

Fossler, C.P., Wells, S.J., Kaneene, J.B., Ruegg, P.L., Warnick, L.D., Bender, J.B., Eberly, L.E., Godden, S.M. *et al.* (2005) Herd-level factors associated with isolation of *Salmonella* in a multi-state study of conventional and organic dairy farms. I. *Salmonella* shedding in cows. *Prev Vet Med* **70**, 257–277.

Glenn, A.E., Bacon, C.W., Price, R. and Hanlin, R.T. (1996) Molecular physiology of *Acremonium* and its taxonomic implications. *Mycology* **88**, 369–383.

Gregory, N.G., Jacobson, L.H., Nagle, T.A., Muirhead, R.W. and Leroux, G.J. (2000) Effect of preslaughter feeding system on weight loss, gut bacteria, and the physico-chemical properties of digesta in cattle. *N Z J Agric Res* **43**, 351–361.

Hancock, D.D., Besser, T.E., Kinsel, M.L., Tarr, P.I., Rice, D.H. and Paros, M.G. (1994) The prevalence of *Escherichia coli* O157:H7 in dairy and beef cattle in Washington state. *Epidemiol Infect* **113**, 199–207.

Hill, N.S., Thompson, F.N., Daive, D.L. and Stuedemann, J.A. (1994) Antibody binding of circulating ergot alkaloids in cattle grazing tall fescue. *Am J Vet Res* **55**, 419–424.

Hovde, C.J., Austin, P.R., Cloud, K.A., Williams, C.J. and Hunt, C.W. (1999) Effect of cattle diet on *Escherichia coli* O157:H7 acid resistance. *Appl Environ Microbiol* **65**, 3233–3235.

Hoveland, C.S., Schmidt, S.P., King, C.C. Jr, Odom, J.W., Clark, E.M., McGuire, J.A., Smith, L.A., Grimes, H.W. *et al.* (1983) Steer performance and association of *Acremonium coenophialum* fungal endophyte on tall fescue pasture. *Agron J* **75**, 821–824.

Keen, J.E. and Elder, R.O. (2002) Isolation of shiga-toxigenic *Escherichia coli* O157 from hide surfaces and the oral cavity of finished beef feedlot cattle. *J Am Vet Med Assoc* **220**, 756–763.

Laegreid, W.W., Elder, R.O. and Keen, J.E. (1999) Prevalence of *Escherichia coli* O157:H7 in range beef calves at weaning. *Epidemiol Infect* **123**, 291–298.

Looper, M.L., Rosenkrans, C.F. Jr, Aiken, G.E. and Edrington, T.S. (2003) *Escherichia coli* and *Salmonella* in beef cattle grazing tall fescue. *Ark Agric Exp Stat Rep* **509**, 58–60.

Looper, M.L., Edrington, T.S., Rosenkrans, C.F. Jr, Burke, J.M., Flores, R., Callaway, T.R. and Aiken, G.E. (2005) Effects of feeding endophyte-infected tall fescue seed to sheep experimentally infected with *Escherichia coli* O157:H7. *Proc West Sec Am Soc Anim Sci* **56**, 213–216.

Nihsen, M.E., Piper, E.L., West, C.P., Crawford, R.J. Jr, Denard, T.M., Johnson, Z.B., Roberts, C.A., Spiers, D.A. *et al.* (2004) Growth rate and physiology of steers grazing tall fescue inoculated with novel endophytes. *J Anim Sci* **82**, 878–883.

Paterson, J., Forcherio, C., Larson, B., Samford, M. and Kerley, M. (1995) The effects of fescue toxicosis on beef cattle productivity. *J Anim Sci* **73**, 889–898.

Porter, J.K. and Thompson, F.N. Jr (1992) Effects of fescue toxicosis on reproduction in livestock. *J Anim Sci* **70**, 1594–1603.

Riley, D.G., Gray, J.T., Loneragan, G.H., Barling, K.S. and Chase, C.C. Jr (2003) *Escherichia coli* O157:H7 prevalence in fecal samples of cattle from a southeastern beef cow-calf herd. *J Food Prot* **66**, 1778–1782.

Rings, D.M. (1985) Salmonellosis in calves. *Vet Clin North Am Food Anim Pract* **1**, 529–539.

Schillo, K.K., Leshin, L.S., Boling, J.A. and Gay, N. (1988) Effects of endophyte-infected fescue on concentrations of prolactin in blood sera and the anterior pituitary and concentrations of dopamine and dopamine metabolites in brains of steers. *J Anim Sci* **66**, 713–718.

Sleper, D.A., Mayland, H.F., Crawford, R.J. Jr, Shewmaker, G.E. and Massie, M.D. (2002) Registration of HiMag tall fescue germplasm. *Crop Sci* **42**, 318–319.

West, C.P., Marlett, M.L., McConnell, M.E., Piper, E.L. and King, T.J. (1998) Novel endophyte technology: selection of the fungus. In *Molecular and Cellular Technologies for Forage Improvement* ed. Brummer, E.C., Hill, N.S. and Roberts, C.A. pp. 105–115. Madison, WI: Crop Science Society of America.

