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## Beef Cattle Salmonellosis: A Study of Oral Salmonella typhimurium and Topical Salmonella newport Inoculations

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#### Introduction

Cattle are frequently infected with salmonellae by fecaloral transmission or by being fed contaminated animal protein byproducts (40% are reported contaminated in the U.S.). Both could propagate salmonellosis in feedlots.

Research indicates that stress can induce shedding of salmonellae by asymptomatic carriers. Stress factors associated with salmonellosis include: transportation, starvation, changes in ration, overcrowding, age, pregnancy, parturition, exertion, anesthesia, surgery, intercurrent disease, and oral treatment with antibiotics and anthelmintics.

In this study, we have attempted to correlate dosage of *S. typhimurium* inoculum with disease, persistence of infection, and environmental contamination. The persistence and spread of *S. newport* placed on the skin of cattle was also studied.

#### **Procedure**

Inoculation procedures. Three groups of four steers each were inoculated orally with marker *S. typhimurium*. Groups 1, 2, and 3 were inoculated orally with 40,000,000, 7,000,000, and 1,000,000 units of *S. typhimurium*, respectively. The inoculum for each steer was placed in a gelatin capsule and administered with a balling gun. Ages of the steers were 19 mo (group 1), 8 mo (group 2), and 12 mo (group 3).

Group 2 steers were also inoculated topically with a strain of *S. newport*. Each hindfoot was placed in a plastic bag containing bovine feces inoculated with 1,100 salmonellae per lb.

Sampling procedures. Fecal samples were collected from the rectum and frozen at -4°F. At each sampling, rectal temperatures were recorded and observations of general appearance and clinical signs were noted.

In group 1, fecal samples were collected from two calves twice daily for 9 days after inoculation and then necropsied following euthanasia. The remaining two animals were sampled twice daily from 1 to 64 days, then once daily from 64 to 103 days, and, thereafter, once a day 3 days a wk (Monday, Wednesday, and Friday) from 103 to 365 days after inoculation.

In group 2, fecal samples were collected once daily for 39 days after inoculation and then once a day 3 days a wk to day 109. Rectal mucosa scrapings, using a wooden applicator stick, were collected from this group from day 5 to day 36 after inoculation. Microbiological samples of each foot were taken once a day (Monday through Friday) for 17 days after inoculation and then once a wk for two additional weeks. Foot samples were collected by scraping and swabbing the hoof walls with a sterile wooden applicator stick and then a piece of sterile gauze. On day 21 after inoculation, hair clippings from above the hoof were collected. Blood samples were taken for bacterial culture once a wk for 4 weeks.

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In group 3, fecal samples were collected once daily for 43 days after inoculation and then once a day 3 days a wk to day 68.

Ground samples of the pens, as well as feed and water samples, were taken once during clinical signs for groups 1 and 3, and four times (once a wk for the first 4 wk) for group

Tissue samples were harvested from all steers at necropsy following euthanasia. Sampling included brain, spinal cord, tonsil, various muscles, heart, lung, liver, spleen, kidney, urinary bladder, gall bladder, rumen (and contents), omasum (and contents), abomasum (and contents), duodenum (and contents), jejunum (and contents), cecum (and contents), colon (and contents), rectum (and contents), mesenteric lymph nodes, peritoneal fluid, pericardial fluid, and blood. Two steers in group 1 were necropsied 9 days after inoculation and the remaining two at approximately 1 year. Group 2 steers were necropsied 125 days after inoculation and group 3 steers were necropsied 80 days after inoculation. Tissue samples were frozen at -94°F.

Microbial analysis. Suspect colonies from all samples were identified by genus and species using a computerized microbiology system. Salmonella typhimurium isolates were also checked to verify compatibility with the marker inoculum. The salmonella isolates were also sent to the National Veterinary Services Laboratory for further verification.

#### Results

Group 1. Three steers showed severe clinical signs of diarrhea, elevated rectal temperatures (102 to 104°F), and ataxia 1 day after inoculation. The marker strain of *S. typhimurium* was found in fecal samples from two of the clinically ill steers 1 day after inoculation. The other clinically ill steer shed the marker bacteria on day 2. Fecal shedding of salmonellae persisted for 4 days in two of the steers and 6 days in the third. Clinical signs in two of the steers increased in severity and euthanasia was necessary on day 9. Salmonellae were never isolated from the feces of the steer showing no clinical signs.

Two steers were necropsied on day 9 and the marker strain of salmonella was found in the distal jejunum of both, in the proximal jejunum of one, and in the rectum of the other. Salmonella infantis was found in the urinary bladder, a mesenteric lymph node, and the caudal lumbar spinal cord of one steer. The other steer had S. infantis in contents of the middle jejunum, abomasal fluid, and the liver. This wild strain of salmonella was never isolated from fecal samples during the experiment.

Clinical signs of the surviving affected steer gradually decreased during the year; however, the steer developed signs of laminitis. Laminitis was not noted in any of the other steers (groups 1, 2, or 3). At necropsy, tissues and gastrointestinal contents of the two surviving steers were salmonella negative.

Group 2. Mild clinical signs of ataxia, slightly elevated rectal temperatures (102 to 103°F), and diarrhea were noted in all steers. Marker salmonellae were found in the fecal sample of one steer on day 4 after inoculation and in the fecal sample of another steer 13 days after inoculation.

These were the only positive fecal samples in this group. Salmonella contamination persisted between the claws for 8 days on one steer, for 3 days on another, and for 7 days on two steers. Attempts to isolate salmonellae by scraping the hoof wall or from clipped hair was unsuccessful. In all four animals, the marker strain of salmonella was found on one front foot and no positive ground samples were found.

Group 3. No clinical signs were observed. At necropsy 3 wk later, no gross lesions were noted and no salmonellae were isolated from tissues or gastrointestinal contents.

#### Conclusions

Severity of clinical signs was variable. Severity of disease appeared to be related to the infectious dosage, but individual variability was also observed.

Fecal shedding of salmonellae was also not consistent. Individual variability of both onset and duration was observed in groups 1 and 2. The fact that no fecal shedding of salmonellae was observed in group 3 suggests that there is a minimal infectious dose required to induce fecal shedding of salmonella. Long-term persistence of enteric *S. typhimurium* infection with recurrent shedding was not observed. A wild strain of salmonella, *S. infantis*, was

recovered from various tissue samples of clinically ill steers, but never recovered from rectal fecal samples. This microorganism was not recovered from fecal samples of clinically normal pen-mates.

Ground, feed, and water samplings were not reliable in evaluating fecal shedding of *S. typhimurium* in the cattle pens. In spite of the spread of *S. newport* infection between the claws of the hindfeet to the forefeet, this microorganism was never recovered from ground samples. Even during periods of known fecal shedding, salmonellae could be recovered only from one sample of damp soil at the base of

a watering unit.

It was demonstrated that active infection of the gastrointestinal tract can be present with no shedding of salmonellae in the feces. This observation suggests that isolation of salmonella from fecal material is a poor indicator of the salmonella infection status of beef cattle. Most of the time during clinical signs of salmonellosis, we were unable to isolate the organism in rectal samples or rectal mucosal scrapings. It was shown that even if fecal sampling is negative, carcass tissues may be contaminated with salmonellae and could possibly serve as a potential source of contamination to processing facilities, employees, and consumers.