The Use of an Experimental Vaccine in Gestating Beef Cows to Reduce the Shedding of *Escherichia coli* O157:H7 in the Newborn Calf

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**ABSTRACT**

Beef cows in the last trimester of pregnancy were used to determine if vaccinating against *Escherichia coli* O157:H7 would increase antibody titers in the serum and also result in the transfer of these antibodies to the neonatal calf. Seventy-one cows were vaccinated 30 d before parturition with an experimental vaccine and then commingled with 66 non-vaccinated cows. Cow fecal and venous blood samples were collected at trial initiation and again about 14 d after parturition. Calf feces and serum were collected at about 14 d after parturition and 60 d later. The serum was analyzed for antibody titers to *E. coli* O157:H7, and the prevalence of *E. coli* O157:H7 in feces was determined by the Barkocy-Galgagher procedure. Initial cow antibody titers to *E. coli* O157:H7 were not different (P = 0.50) between treatments, but by parturition, the antibody titers for *E. coli* O157:H7 in vaccinated cows were 11 times higher (P < 0.001) than for control cows (917 vs. 83). The serum titers for calves suckling vaccinated cows were higher (P < 0.001) than control calves (1,485 vs.135) at about 14 d after calving. By 60 d, titer levels were still higher (P < 0.001) for calves suckling vaccinated cows. Initial fecal *E. coli* O157:H7 concentrations for cows were negative for both treatments and remained low. There were no differences in fecal *E. coli* O157:H7 at 60 d postpartum among calves; less than 5% of calves were shedding. Results suggest that vaccinating gestating cows for *E. coli* O157:H7 resulted in elevated antibody titers, and these antibodies transferred to the calf.

**Key words:** *Escherichia coli* O157:H7, antibodies, colostrum, cattle

**INTRODUCTION**

*Escherichia coli* O157:H7 is one of several food borne pathogens present in the current food industry. *Escherichia coli* O157:H7 is commonly associated with hemorrhagic colitis in humans and has few therapeutic alternatives. Poor prognoses for severe sequelae have led to intensive research targeting elimination or reduction of fecal shedding (Kudva et al., 1999; Lahti et al., 2003). Post-harvest interventions have dealt with controlling food borne pathogens in what industry has deemed a “multiple hurdle approach,” to reduce pathogen contamination primarily in ground beef (Smith et al., 2000; Ransom et al., 2001). More emphasis has been placed on carcasses contaminated by feces primarily occurring during the slaughter process. The Federal Register (2002) reported 1) that 5 multi-state studies showed the apparent prevalence in breeding herds containing one or more infected with *E. coli* O157:H7 was 24%, 61%, 75%, 87%, and 100%; 2) 3 multi-state studies reported the apparent prevalence in feedlots containing one or more cattle infected with *E. coli* O157:H7 was 63%, 100%, and 100%; and 3) Elder et al. (2000) found that 28% of fecal samples from fed cattle were positive for *E. coli* O157:H7. Similarly, Smith et al. (2001) reported that the fecal prevalence of *E. coli* O157:H7 in fed cattle was 23%.
The beef industry has focused on pre-harvest methods to reduce the incidence of *E. coli* O157:H7 (Elder et al., 2000; Barkocy-Gallagher et al., 2001). Rice et al. (2003) found that the introduction of one animal, which was shedding *E. coli* O157:H7 at high rates quickly infected other cohorts. The beef industry is researching potential interventions at various stages of production (LeJeune and Wetzel et al., 2007). Multiple hurdle interventions have had a positive impact on significantly reducing food-borne pathogens entering the food supply if the levels of these bacteria allowed into the packing plant were limited (Elder et al., 2000). Dean-Nystrom et al. (1997) reported that an experimental vaccine interfered with *E. coli* colonization within the gut of the calf. The objective of this research was to determine if an experimental *E. coli* O157:H7 vaccine given to the gestating beef cow would result in increased antibody titers and if titers atating beef cow would result in in-

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### MATERIALS AND METHODS

#### Cow Management

This experiment was conducted at the USDA-Agriculture Research Service Fort Keogh Livestock and Range Research Laboratory at Miles City, MT. One hundred thirty-seven gestating beef cows in the last trimester of pregnancy were used for this study. Cows were weighed and randomly assigned to 1 of 2 treatments (control vs. vaccinated) approximately 30 d before the start of expected calving (Table 1). Cows were previously synchronized and artificially inseminated and were expected to calve within a 30-d period starting the end of March. The average weight of the cows approximately 30 d before calving was 567 kg. Seventy-one of the gestating cows were vaccinated with an experimental vaccine, developed by Fort Dodge Animal Health Laboratories, which was designed to prevent the attachment of *E. coli* O157 to the intestinal wall (Cormick et al., 2002). These cows were then commingled with 66 non-vaccinated control cows. Cows remained in calving pens for the first 15 d after parturition and then were placed on native range pasture after parturition.

#### Sample Collection and Analysis

At the prescribed sampling time, 10 mL of whole blood were collected from the tail vein of cows and jugular vein of calves. Samples were placed on ice and transported to the Montana State University laboratory where they were centrifuged at 447 \( \times \) g (2,000 rpm) for 20 min and the serum was separated for analysis. The analyses for antibody titers of *E. coli* O157:H7 were conducted as for a blind study by Fort Dodge Animal Health Laboratories (Fort Dodge, IA). Serum titers against *E. coli* O157:H7 were analyzed using an ELISA (Widiasih et al., 2004). Briefly, samples were dissolved in 1 mg/mL in PBS and then diluted to 1:100 with the PBS for a final concentration of 10 \( \mu \)g/mL (Cray and Moon, 1995). Microtiter plates were coated with diluted lipopolysaccharide solution. The samples were placed through a series of similar steps of washing and coating the plates. Results were then read by a spectrophotometer (Molecular Devices, Union City, CA) at 405 nm, approximately 10 to 30 min after the addition of a substrate solution. The results were expressed according to the final dilution factor on the plate (Fort Dodge Animal Health Laboratories, 2004).

#### Recovery of *E. coli* O157:H7

The prevalence of *E. coli* O157:H7 in feces from cows and newborn calves was determined by collecting a fecal sample via a rectoanal mucosal swab (RAMS; Rice et al., 2003). The swab sample was collected from cows on about d 30 (before parturition) and about 9 to 14 d after parturition. The calves were also swabbed at approximately 14 d after parturition and again at about 60 d. Due to the lack of facilities at branding, cows were not sampled at 60 d. Fecal samples were obtained by inserting the sterile foam-tipped applicator approximately 2 to 8 cm (Grauke et al., 2002; Naylor et al., 2003) into the anus, and by using a rapid in-and-out motion, the entire mucosal surface of the rectoanal junction was swabbed (Rice et al., 2003). The swab samples were submitted to a commercial laboratory (Food Safety Net, San Antonio, TX) for analysis for *E. coli* O157 using

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### Table 1. Experimental design and samples collected from cows and calves to determine if vaccinating gestating cows with an experimental *Escherichia coli* O157:H7 vaccine would transfer antibodies to the newborn calf via the colostrum

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Samples collected</th>
<th>Samples analyzed for</th>
</tr>
</thead>
<tbody>
<tr>
<td>66 Pregnant control cows</td>
<td>Cows: Blood and fecal samples at −30 d parturition and −14 d after calving</td>
<td>Blood: <em>E. coli</em> O157:H7 antibody titers</td>
</tr>
<tr>
<td>(not vaccinated)</td>
<td>Calves: Blood and fecal samples within 14 d of calving and again at −60 d</td>
<td>Feces: prevalence of <em>E. coli</em> O157:H7 shedding and enumeration of positive samples</td>
</tr>
<tr>
<td>71 Pregnant vaccinated cows</td>
<td></td>
<td></td>
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<tr>
<td>(vaccinated 30 and 14 d before calving)</td>
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</tbody>
</table>
Figure 1. Effect of vaccinating gestating cows with an experimental vaccine against *Escherichia coli* O157:H7 on antibody titers and subsequent titer levels in the neonatal calf.

The procedure of Barkocy-Gallagher et al. (2002).

**Statistical Analysis**

Data were analyzed using the GLM procedure (SAS Institute, Cary, NC, 2000). A comparison of control and vaccinated cows and corresponding calves for *E. coli* O157:H7 prevalence was measured by Chi-square analyses. The serum titer levels were analyzed by one-way ANOVA.

**RESULTS AND DISCUSSION**

Initial cow titer levels before vaccination revealed no differences (*P* < 0.50) between treatments for *E. coli* O157:H7 antibody (Figure 1). Similarly, there were no differences in fecal shedding rates of *E. coli* O157:H7 at about 30 d before parturition. However, by about 14 d after calving, the vaccinated cows had 11 times higher serum titer levels than that of the control group (917 vs. 83). The fecal samples for the approximate 14-d sample again were similar (*P* > 0.10) between treatment groups.

Calf serum titer levels for *E. coli* O157:H7 14 d postparturition showed a 10-fold difference (*P* < 0.001) between treatments (135 vs. 1,485 for control calves vs. calves that suckled previously vaccinated cows, respectively). After about 60 d, calves from vaccinated cows had a 7-fold (*P* < 0.001) higher titer level compared with the control calves.

Calf fecal samples did not show *E. coli* O157:H7 presence at 14 d after parturition. Similarly, no differences were detected between treatments at 60 d with only 3% of calves infected with *E. coli* O157:H7.

A low level of shedding may have been due to the cool spring weather or the high percentage of native grass and roughage consumption, which could prevent *E. coli* O157:H7 from shedding in greater numbers (FDA, 2002; Barkocy-Gallagher et al., 2003, Callaway et al., 2003). Previous studies have revealed that *E. coli* O157:H7 was pathogenic for neonatal calves (Dean-Nystrom et al., 1997; Widiasih et al., 2004). In these studies, calves less than 36-h old developed diarrhea and enterocolitis with attaching and effacing lesions. Other studies have shown that weaned (ruminating) calves dosed with *E. coli* O157:H7 remained clinically healthy, with no evidence of fever or diarrhea (Zhao et al., 1998, Gyles, 2007), compared with the newborn calf.

**IMPLICATIONS**

Results of this experiment showed that vaccinating the late gestating cow with an experimental vaccine against *E. coli* O157:H7 resulted in increased antibody titers in cows and calves. A partial reduction in the shedding of *E. coli* O157:H7 will be useful in controlling the contamination of meat.

**LITERATURE CITED**


Use of vaccine to reduce Escherichia coli O157:H7 in cattle


