Testing of Probiotic Bacteria for the Elimination of Escherichia coli O157:H7 in Cattle

Final Report

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Objectives of the Research Proposal. The overall objective of this project was to determine the efficacy of probiotic lactic acid bacteria (LAB) fed to cattle as a daily feed supplement with respect to the following areas: 1) shedding of *E. coli* O157:H7 by the live animal; 2) contamination of the carcasses during slaughter; and 3) effects on body weight gain and feed intake.

This study consisted of two phases:

- Cattle feeding trials. Cattle were monitored for fecal shedding of *E. coli* O157:H7 in a university feedlot environment. Groups of cattle were identified as those shedding *E. coli* O157:H7 just before probiotic supplementation and those not shedding. Cattle were divided into three groups. Two groups received two separate probiotic supplements and the other received the probiotic carrier only and served as a control. Body weight gain and feed intake by replicate pens of cattle on each treatment were measured during the probiotic supplementation period.
- 2) Carcass safety. The hides and carcasses of the cattle in the feeding trials were examined at points during the slaughter process for the presence of *E. coli* O157:H7.

Experimental Procedures

Cattle. One hundred eighty-five (185) steers of British breeding (primarily Angus, Hereford, and Angus x Hereford) were purchased through Caprock Industries, Inc. at auction in Pratt, KS and transported to the Texas Tech University Burnett Center. The cattle had been purchased on 5/31/01 and held on hay and water until shipment. Their pay weight was 804 lb. Cattle arrived at the Burnett Center at approximately 1045, at which time them were unloaded and processed. Processing included: 1) individual body weight (BW) measurement; 2) uniquely numbered ear tag in the left ear; 3) vaccination with UltraChoice 7 (Pfizer Animal Health; Lot No. S900024 – Exp. 07/05/01 and Lot No. S903222B – Exp. 09/10/01); 4) vaccination with Bovishield 4 + Lepto (Pfizer Animal Health; Ser. No. SNA019266/A013371 – Exp 10/01/02; Ser. No. SNA018354/A019248 – Exp. 09/11/02); and 5) treatment down the back line with Dectomax (Pfizer Animal Health; Lot No. K9T04911 – Exp. 07/02). Processing began at approximately 1200 was completed at approximately 1530. Steers were sorted to 37 concrete, partially slotted floor pens with five steers per pen and offered 10 lb/steer of a

65% concentrate starter diet. On 6/6/01, all cattle switched to a 70% concentrate diet, and the switch to an 80% concentrate diet occurred on 6/11/01.

All cattle were weighed on 6/12/01 to initiate the preliminary phase of the experiment. At this time, each steer was implanted with Revalor S (Intervet; Lot No. 321 - Exp. Nov. 2002) in the right ear. Pen assignments were retained from the original sort that had occurred at the time of arrival processing. On 6/16/01, all cattle were switched to the final 90% concentrate diet (Table 1). Each steer was weighed on 7/31/01 (49 d on feed), and an individual fecal sample was obtained to test for shedding of *E. coli* 0157:H7.

Treatment and Pen Assignments. Because shedding of E. coli 0157:H7 was limited to a smaller fraction of the cattle than originally anticipated (only 25 steers were shedding E. coli O157-H7 in the feces on 7/31/01), the cattle were not sorted into shedding and non-shedding groups as originally planned. Rather, a randomized complete block design with three treatments (Control, and two Lactobacillus acidophilus cultures – NPC 747 and NPC 750) was used with cattle blocked by BW without respect to shedding. To assign cattle to the pens and treatments, the BW data for the 185 steers obtained on 7/31/01 were entered into a Microsoft Excel[®] spreadsheet. Five steers (one with injured foot, one with an injured shoulder, one that did not gain BW for the first 49 d, and the two steers of lightest BW among the remaining cattle) were designated as extra cattle. The data for the 180 selected steers were then sorted in ascending order by BW. The first 15 steers of lightest BW were designated as Block 1, continuing through blocking groups of 15 steers each to the 15 steers of heaviest BW, which were designated as Block 12. Within each block, a sequence of three randomly selected integers numbered 1, 2, and 3 was assigned to the steers, starting with the lightest three steers and proceeding through the heaviest three steers in a block of 15 steers. This process was continued until each of the 180 steers had been assigned a The three treatments (Control, NPC 747, and NPC 750) were random number. arbitrarily assigned to the numbers 1 through 3 (1 = Control, 2 = NPC 747, and 3 = NPC 750). Blocks were assigned to three contiguous pens in the Burnett Center, beginning with Pens 23 through 25 for Block 1, Pens 82 through 84 for Block 2, Pens 85 through 87 for Block 3, and continuing to Pens 112 through 114 for Block 12. Within each set of three contiguous pens in a block, treatments were assigned randomly to pens by the use of 12 sets of three randomly selected integers (numbered 1, 2, and 3), with the first pen in the block assigned to the corresponding treatment code of the first randomly selected integer, proceeding to the third pen, which was assigned to the corresponding treatment code of the third randomly selected integer. Pen and treatment designations were input to the spreadsheet, which was then sorted by block and pen number.

Starting at approximately 0700 on 8/7/01, the cattle were sorted to their assigned pens determined by the computer assignment described for 8/6/01. However, because a bolt sheared on one of the four load cells at the working chute, all the cattle could not be weighed. As a result, the cattle were sorted to their new pens, and the initial BW measurement was postponed until 8/14/01. After being moved to their new pens, the cattle were fed as they had been previously. The sorting process and movement to new

pens was completed at approximately 1100. Repairs to the load cell by personnel from B and C Supply Company were initiated as soon as possible after sorting was completed.

Each steer was weighed on 8/14/01 to begin the study. Weighing started at approximately 0700 and was completed by approximately 0930. Treatments (control and the two *Lactobacillus acidophilus* cultures) were applied when the cattle were fed after they were returned to their pens.

Experimental Design. The experimental design was a randomized complete block, with pen as the experimental unit (12 pens per each of the three treatments with five steers per pen for a total of 180 steers). The three treatments were as follows:

- **Control** standard TTU Burnett Center 90% concentrate diet with carrier (lactose) only mixed in water and added to the diet at the time of feeding;
- **NP 747** standard TTU Burnett Center 90% concentrate diet with 1 x 10⁹ CFU *Lactobacillus acidophilus* Strain NPC 747 mixed in water and added to the diet at the time of feeding;
- **NP 750** standard TTU Burnett Center 90% concentrate diet with 1 x 10⁹ CFU *Lactobacillus acidophilus* Strain NPC 750 mixed in water and added to the diet at the time of feeding.

Each treatment culture (lactose for the control treatment) was prepackaged in aluminum foil packets by personnel of Nutrition Physiology Corp., Indianapolis, IN. Each aluminum packet had a colored dot that corresponded to the treatment codes of Green = Control, Red = NPC 747, and Yellow = NPC 750. The contents of one packet were sufficient to supply the desired dose of microbial culture for the 12 pens of cattle on each treatment. The contents of the packet for each treatment were mixed with 2.5 L of distilled water in a plastic sprinkler can, after which the contents of the sprinkler can were poured onto the diet as it mixed in a Rotomix 84-8 mixer/delivery unit (described in a subsequent section). Three sprinkler cans were used, each with a colored tape wrapped on the nozzle of the sprinkler can that corresponded to the color code for each of the three treatments.

Experimental Diets. Ingredient composition of the 90% concentrate diet fed during the experiment is shown in Table 1. These data reflect adjustments for the average dry matter (DM) matter content of feed ingredients for the period during which microbial culture treatments were applied. Each diet contained the same intermediate premix (Table 1), which supplied protein, various minerals and vitamins, Rumensin (30 g/ton, DM basis), and Tylan (8 g/ton, DM basis).

Management, Feeding Procedures, Weighing Procedures, and Carcass Data Collection. Standard procedures at the Burnett Center were used throughout the experiment. The three treatment diets were mixed in a 45-cubic foot capacity Marion

paddle mixer. The Burnett Center feed milling system is operated by a computercontrolled batching system. A printout of the weight of each dietary ingredient was recorded on a daily ingredient usage output from the computerized batching system. Once the total amount of feed for a given treatment was mixed, the mixed batch was released from the Marion paddle mixer and delivered by a drag-chain conveyer to a Rotomix 84-8 delivery system. After feed was delivered, and the mixer unit for the Rotomix 84-8 unit was operating, the contents of the sprinkler can for a given treatment were poured onto the diet. After mixing for approximately 4 to 5 min, the quantity of feed allotted to each of the 12 pens within treatment was then weighed to the nearest 1 lb by use of the load cells and indicator on the Rotomix 84-8 unit. Feeding order of treatment diets throughout the experiment was Control, NPC 747, and NPC 750. Clean-out of the Rotomix 84-8 was monitored closely to avoid cross-contamination of diets, and at least one batch of a diet from another experiment (equivalent to the Control diet) was mixed between the NPC 747 and NPC 750 treatment diets.

Dry matter determinations on ingredients used in the experimental diets were made every 2 wk throughout the experiment. These ingredient DM values were used to calculate the DM percentage of each dietary ingredient during the experiment. In addition, samples of mixed feed delivered to feed bunks were taken weekly throughout the experiment. These bunk sample DM values were used to compute average DM intake (DMI) by the cattle in each pen. Samples of feed taken from the bunk were composited for the entire preliminary period and for each interval in which cattle were weighed (typically 28-d intervals) after the microbial culture treatments were initiated. Composited feed samples were ground to pass a 2-mm screen in a Wiley mill and analyzed for DM, ash, crude protein (CP), acid detergent fiber (ADF), Ca, and P (AOAC, 1990).

Each feed bunk of the 36 pens was evaluated visually at approximately 0700 to 0730 daily. The quantity of feed remaining in each bunk was estimated, and the suggested daily allotment of feed for each pen was recorded. This bunk-reading process was designed to allow for little or no accumulation of unconsumed feed (0 to 1 lb per pen). A computer printout of the intake by each pen for the previous 3-d period was available each morning to assist with bunk reading. Pens of cattle that maintained a given level of feed intake for a 3-d period were challenged to consume a higher level (0.4 lb/steer challenge). Each challenge level was maintained for a 3-d period when the pen accepted the challenge and consumed all the feed offered. The ultimate goal of the challenge process was to ensure that the cattle were consuming the maximum quantity of feed possible. Feed bunks were cleaned, and unconsumed feed was weighed (Ohaus electronic scale, ± 0.1 lb) at intervals corresponding to intermediate weigh dates throughout the trial. Dry matter content of these bunk weighback samples was determined in a forced-air oven by drying overnight (typically 20 h) at 100°C. All weights for DM determinations were obtained on an Ohaus electronic balance (± 0.1 g). The DMI by each pen was calculated by multiplying the DM content of the delivered feed by the total feed delivery to each pen, with correction for the DM of any feed weighed back from each pen.

All BW measurements taken during the experiment were obtained using a single-animal scale (C & S Single-Animal Squeeze Chute set on four load cells). The scale was calibrated with 1,000 lb of certified weights (Texas Dept. of Agriculture) on the day before or day of each scheduled weigh day. Intermediate BW measurements were taken every 28 d after initiation of the feeding of the microbial cultures to assess performance of the cattle and shedding of *E. coli* 0157:H7 on a regular basis. Fecal grab samples were collected at the time of regularly scheduled BW measurements, at 14-d intervals in between regularly scheduled BW measurements, and at the time cattle were shipped to slaughter. Because the cattle were blocked by BW, blocks were deemed to have reached sufficient finish to grade USDA Choice after different days on feed. All cattle in the study were shipped for slaughter to the Excel Corp. facility in Plainview, TX. Steers in Blocks 11 and 12 were shipped on 9/28/01. Steers in Blocks 9 and 10 were shipped on 10/8/01, those in Blocks 5 through 8 were shipped on 10/17/01, those in Bocks 3 and 4 were shipped on 11/6/01, and steers in Blocks 1 and 2 were shipped on 11/30/01. All the original 180 steers that were assigned to three treatment groups completed the study. Personnel of the Texas Tech University Meat Laboratory collected carcass data at the Excel Corp. facility. Measurements included hot carcass weight, longissimus muscle area, marbling score, percentage of kidney, pelvic, and heart (KPH) fat, fat thickness measured between the 12th and 13th ribs, USDA yield grade, and USDA quality grade.

Statistical Analyses. All performance and carcass data were analyzed with pen as the experimental unit. A complete randomized block design was employed, and computations were made with the GLM procedure of SAS (1987). Pen means for daily gain (ADG) and average daily DMI were included in the data file, and feed:gain ratio was computed as the quotient of daily DMI divided by ADG. The effect of treatment and block were included in the model for pen-based data. Carcass data were entered on an individual animal basis and analyzed with a model that included effects of treatment, block, and block x treatment. Block x treatment was specified as the error term for testing treatment effects with carcass data. Two orthogonal contrasts were used to test treatment effects: 1) Control vs the average of the NPC 747 and NPC 750 treatments; and 2) NPC 747 vs NPC 750. Carcass quality grade data were analyzed by Chi-square procedures (SAS, 1987) using individual animal data.

Microbiological Measurements Monitored. Fecal samples were taken directly from the rectum of the animal initially and every 28 d thereafter until they received probiotic supplementation. Following probiotic supplementation, fecal samples were taken every 14 d. A newly developed assay for the detection of *E. coli* O157:H7 was used to detect the organism in this study (modification of Laegreid et al., 1999). This method is very sensitive and will detect *E. coli* O157:H7 when numbers are very low. The method was originally developed at the Meat Animal Research Center in Clay Center, NE. Ninety millileters of GN-VCC broth were inoculated with 10 g of feces (GN-VCC is GN broth with 8 µg/mL of vancomycin, 50 ng/mL of cefixime and 10 µg/mL of cefsulodin) and incubated for 6 h at 37° C. *E. coli* cells were subjected to immunomagnetic separation by mixing 1 mL of the culture above with 20 µl of Dynal O157 beads for 30 min at room temperature. Beads were washed three times in PBS-Tween 20 and 50 µL of the bead-

bacteria mixture were spread on to CT-SMAC plates (SMAC containing 50 ng/mL of cefixime and 2.5 μ g/mL of tellurite) and streaked for isolation. Plates were incubated overnight at 37^oC. Three sorbitol-negative colonies were picked and streaked for isolation on CT-SMAC (to verify purity of colony selection). Plates were incubated overnight at 37^oC. A single colony from the CT-SMAC plate above was selected and inoculated into MacConkey agar, Fluorocult agar, and MacConkey broth. Broth was incubated overnight at 37^oC. MUG-negative, lactose-positive colonies were selected, and indole, TSI, and VP tests were conducted on selected colonies. Colonies that were indole-positive, A/A, or K/A plus gas and VP-negative were boiled (using cells from the MacConkey broth above) and tested for the O157 antigen using a Remel latex agglutination kit. Colonies were sub-cultured on to blood agar. The H7 agglutination and API 20 tests were conducted on O157-positive cells for confirmation. Final confirmation will be made by PCR analysis on a Dupont BAX system.

Post-Harvest Presence of E. coli O157:H7. As noted previously, the cattle were slaughtered in a commercial processing facility at the end of the study (average of 70 d of probiotic administration). Samples were collected from the hide, intestine, hot carcasses (before any intervention treatment), and chilled carcasses. These samples were examined for the presence of *E. coli* O157:H7.

<u>Hide sampling.</u> Immediately after stunning, 2 in x 2 in sterile gauze pads saturated with sterile distilled water were used to sample 450 cm² of the ventral brisket. Samples were placed into 20 mL of sterile 2% Brilliant Green Bile and held for transport to the laboratory. Samples were processed using the same method described for feces.

<u>Intestinal sampling.</u> Large intestinal tissue specimens will be collected from the cattle before complete evisceration. Grab samples directly from the rectum were taken and analyzed as previously described.

<u>Carcass sampling.</u> Carcasses were sampled before chilling and any intervention treatments. A sterile Speci-Sponge was hydrated with sterile Butterfield's phosphate diluent. Sponges were used to sample the midline of the carcass (brisket, plate) on-line, before interventions.

Results and Discussion

Incidence of E. coli O157;H7. Upon arrival at the feedlot all animals were tested for the presence of *E. coli* O157:H7 in fecal grab samples. Only three of the animals had detectable amounts of the pathogen upon arrival (Figure 1). Just before probiotic supplementation, 24 of the animals had detectable amounts of the pathogen in fecal grab samples. These animals were distributed equally among the three treatment groups. The total incidence of the pathogen varied from 18 to 19% during the midfeeding period to less than 10% near slaughter (Figure 1). These data are similar to incidence levels previously reported by NAHMS (USDA, 2000).

Just 14 d after initiating treatment, significant (P < .05) differences were observed among the three treatment groups. At this sampling time 56.6% of the control animals were positive, whereas only 20% of the animals fed the NPC 747 sample and 11% of those fed the NPC 750 probiotic were positive. Comparing the data based on a positive pen basis, significant (P < .05) differences were also observed. Forty-one percent of the pens receiving the NPC 750 treatment had at least one positive animal, which was significantly (P < .05) the percentage of pens in cattle receiving NPC 747 (66% with at least one positive sample). The control pens had a significantly higher (P < .05) shedding rate than both the NPC 750 and NPC 747 pens, with 83% of the pens having at least one positive sample.

The animals were sampled again 28 d after treatment, with differences once again observed for both animal samples and pen samples. Of individual control animals, 43% were positive, with 83% of the pens having at least one positive animal. The cattle receiving probiotic treatments had significantly (P < .05) less shedding. Of those receiving the NPC 750 treatment, 15% of the individual animals were positive, whereas 50% of pens had at least one positive animal. Of those receiving the NPC 747 treatment, 8.3% of the animals were positive, with 41.6% of the pens containing at least one positive.

On d 42, there were significant (P < .05) differences among treatments for the individual animal samples but for the pen data. Ten percent of the animals fed the NPC 747 probiotic were positive, whereas 20% of those fed NPC 750 were positive. In contrast, 58% of the control animals were positive. The control pens had 33.3% of the pens having at least one positive, whereas both probiotic treatments had 41.6% positive, but pen-based shedding rates were not statistically different.

The animals were sampled once more before the pre-slaughter sample was taken just before the animals were shipped to slaughter. The animals receiving the NPC 747 treatment had significantly (P < .05) less detectable *E. coli*, with only 3.3% of the animals testing positive. The animals receiving the NPC 750 probiotic and those in the control group were not significantly different, with 15% and 20% shedding, respectively.

Fecal samples taken in the slaughter plant indicated that only a very small number of animals were shedding just before slaughter. Only 3.3% of the animals receiving the NPC 747 treatment were positive, whereas 6.6% of those receiving the NPC 750 treatment were positive, and 10% of the control animals were positive. When these data are correlated back to the original pen in which the animal was housed, there were significant differences (P < .05) among control pens and treatment pens. Both the probiotic treatments resulted in 16% of the pens containing at least one animal that was positive, whereas 33% of the control pens had at least one positive sample.

Averaging over all sampling times, 61.67% of the control animals shed the pathogen at some point during the feeding period. There were no significant differences between the control and NPC 750 treatment, with 61.67% and 51.67% of the animals shedding at least once during the study, respectively. The percentage of animals in the NPC 747

treatment was significantly less than for the other two treatments, with only 35% shedding at any one time during the study.

Performance Data. Analyzed nutrient content of the 90% concentrate diet fed during the treatment period was generally in close agreement with formulated values. Averaged over the treatment feeding period, the diet contained (DM basis) 12.69% CP, 4.5% ash, 8.82% ADF, 0.56% Ca, and 0.30% P.

By chance in the allotment process, initial BW differed slightly among the three treatments (Table 2), with control cattle being approximately 8 lb heavier ($P \le 0.013$) than those in the NPC 747 and NPC 750 treatments at the time the treatments were initiated. However, final BW (average of 70 d on feed) and final BW calculated from hot carcass weight divided by the average dressing percent (adjusted final BW) did not differ among the three treatments. The calculation of final BW from hot carcass weight divided by a common dressing percent was done in an effort to decrease the affect that differences in gastrointestinal tract fill might have on the final BW data.

Average daily gain (Table 2) did not differ among treatments (P > 0.10), but cattle in both the Lactobacillus culture groups had numerically higher ADG for the first 28 d of the treatment feeding period. In addition, cattle in the NPC 747 and NPC 750 treatments tended ($P \le 0.144$) to have greater adjusted ADG (calculated from adjusted final BW) for the overall study than control cattle. Dry matter intake did not differ among treatments (P > 0.10) for the first 28 d or for the overall treatment period. Similarly, feed:gain ratio did not differ (P > 0.10) for the first 28 d or for the overall period; however, feed:gain ratio based on adjusted ADG tended ($P \le 0.067$) to be improved for the two probiotic culture treatments than for the control. Results of previous research at the Burnett Center (Galyean et al., 2000) indicated that adding live cultures of Lactobacillus acidophilus Strain 45 and(or) Strain 51 plus Propionibacterium freudenreichii (PF-24) increased ADG 2.2 to 5.4% by growing finishing steers compared with a control diet. On average, for the three microbial culture treatments used in that study, ADG was increased 4.3% (P < 0.06) relative to the control treatment. Microbial cultures also increased daily DMI slightly above that of the control treatment, but differences were not statistically significant. Feed:gain ratio was numerically improved for probiotic treatments, but calculated NEm and NEg values for the treatment diets suggested that cattle on the microbial culture diets converted DMI to gain at approximately the same efficiency as control cattle. Results of Galyean et al. (2000) are not directly comparable with the present results because of differences in strains of Lactobacillus and the feeding of Propionibacterium, but both studies suggest potentially positive effects on performance with addition of live microbial cultures to the feed.

Carcass Data. Carcass data are shown in Table 3. No major differences were noted among the three treatments for carcass data. Control steers tended ($P \le 0.072$) to have a slightly smaller longissimus muscle area than cattle in the NPC 747 and NPC 750 treatments, and steers in the NPC 750 treatment tended ($P \le 0.09$) to have a lower marbling score than those in the NPC 747 treatment. With the exception of hot carcass weight, which was greater for cattle fed probiotic cultures than for control cattle,

Galyean et al. (2000) reported that carcass characteristics were not greatly affected by feeding various *Lactobacillus* cultures plus *Propionibacterium freudenreichii* PF-24 to finishing steers.

Summary and Conclusions

Based on these observations, supplementing cattle with certain probiotic cultures (two strains of *Lactobacillus acidophilus;* NPC 747 and NPC 750) decreases the incidence of *E. coli* O157:H7 in the feces of finishing beef cattle. Initially both cultures resulted in significant (P < .05) reductions in incidence. At the end of the study, the NPC 747 treatment was the most effective in reducing the incidence of *E. coli* O157:H7.

Under the condition of this experiment, feeding the NPC 747 and NPC 750 probiotic cultures tended to improve feed:gain ratio when final body weight was calculated from hot carcass weight and the overall average dressing percent. Carcass characteristics were minimally affected by feeding the two cultures. Based on these results, feeding such microbial cultures to decrease fecal shedding of *E. coli* 0157:H7 should either have no effect of slightly improve performance by finishing beef steers.

Literature Cited

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Table 1. Ingredient composition (%, DM basis) of the 90% concentrate experimental diet

Percentage of DM						
Alfalfa hay, ground	4.96					
Cottonseed hulls	5.04					
Steam-flaked corn	64.60					
Dry-rolled corn	10.15					
Cottonseed meal	4.82					
Molasses	4.12					
Fat (yellow grease)	2.93					
Urea	0.90					
Premix ^a	2.48					

^aComposition of the premix was as follows: cottonseed meal = 23.9733; high-calcium limestone = 42.1053; dicalcium phosphate = 1.0363; potassium chloride = 8.0000; magnesium oxide = 3.5587; ammonium sulfate = 6.6667; salt = 12.0000; cobalt carbonate = 0.0017; copper sulfate = 0.1572; iron sulfate = 0.1333; EDDI = 0.0025; manganese oxide = 0.2667; selenium premix, .2% Se = 0.1000; zinc sulfate = 0.8251; vitamin A, 650,000 IU/g = 0.0122; vitamin E, 275 IU/g = 0.1260; Rumensin, 80 mg/lb = 0.6750; and Tylan, 40 mg/lb = 0.3600. Concentrations by the certain ingredients are on a 90% DM basis.

		Treatment ^a			Contrast ^c	
Item	Control (C)	NPC 747	NPC 750	SE ^b	C vs others	747 vs 750
Initial BW. Ib	1.037.2	1.029.1	1.029.6	2.38	0.013	0.895
Final BW, Ib	1.277.7	1.271.5	1,283.4	6.90	0.971	0.235
Adjusted final BW, Ib ^d	1,272.1	1,276.4	1,283.4	7.69	0.418	0.525
Daily gain, lb						
d 0 to 28	4.07	4.28	4.33	0.150	0.207	0.782
d 0 to end ^e	3.50	3.49	3.65	0.101	0.569	0.278
Adjusted 0 to end ^d	3.44	3.58	3.69	0.106	0.144	0.462
Daily DMI, lb/steer						
d 0 to 28	20.02	20.29	20.36	0.264	0.352	0.846
d 0 to end ^e	20.54	20.32	20.85	0.249	0.875	0.146
Feed:gain						
d 0 to 28	4.94	4.84	4.73	0.133	0.359	0.587
d 0 to end ^e	5.94	5.92	5.76	0.116	0.483	0.319
Adjusted d 0 to end ^d	6.01	5.75	5.66	0.127	0.067	0.634

Table 2. Effects of live cultures of Lactobacillus acidophilus Strains NPC 747 and NPC 750 on performance by finishing beef steers

^aControl = standard TTU Burnett Center 90% concentrate diet with carrier (lactose) only mixed in water and added to the diet at the time of feeding; NPOC 747 = Control + 1 x 10^9 CFU *Lactobacillus acidophilus* Strain NPC 747 per animal; NPC 750 = Control + 1 x 10^9 CFU *Lactobacillus acidophilus* Strain NPC 750 per animal. Average days on feed = 70.

^bPooled standard error of treatment means, n = 12 pens per treatment.

^cObserved significance level for orthogonal contrasts.

^dAdjusted final BW was calculated as follows: (Hot carcass weight/average dress of 62.41%). Adjusted daily gain was calculated as follows: (Adjusted final BW – initial BW)/days on feed. Adjusted feed:gain was the ratio of daily DMI and adjusted daily gain.

	Treatment ^a				Contrast ^c		
Item	Control (C)	NPC 747	NPC 750	SE ^b	C vs others	747 vs 750	
Hot carcass wt, lb	793.9	796.6	801.0	4.80	0.418	0.525	
Dressing percent	62.14	62.65	62.43	0.227	0.164	0.486	
LM area, sq. in. ^d	12.97	13.43	13.24	0.160	0.072	0.426	
Fat thickness, in.	0.47	0.47	0.50	0.016	0.458	0.139	
KPH, % ^e	1.98	1.94	2.01	0.028	0.810	0.106	
Yield grade	3.17	3.03	3.20	0.072	0.560	0.107	
Marbling score ^f	428.7	422.7	403.7	7.57	0.109	0.090	
Choice, % ^g	60.00	53.33	46.67	-	-	-	
Select, %	40.00	46.67	53.33	-	-	-	

Table 3. Effects of live cultures of *Lactobacillus acidophilus* Strains NPC 747 and NPC 750 on carcass characteristics of finishing beef steers

^aControl = standard TTU Burnett Center 90% concentrate diet with carrier (lactose) only mixed in water and added to the diet at the time of feeding; NPOC 747 = Control + 1 x 10^9 CFU *Lactobacillus acidophilus* Strain NPC 747 per animal; NPC 750 = Control + 1 x 10^9 CFU *Lactobacillus acidophilus* Strain NPC 750 per animal. Average days on feed = 70.

^bPooled standard error of treatment means, n = 12 pens per treatment.

^cObserved significance level for orthogonal contrasts.

^dLM = longissimus muscle.

^eKPH = kidney, pelvic, and heart fat.

 $^{f}300 = Slight^{0}; 400 = Small^{0}; 500 = Modest^{0}.$

^gDistribution of Choice and Select + Standard carcasses did not differ among treatments (P > 0.34).

Figure 1. Incidence of *E. coli* O157:H7 in Beef Feedlot Cattle Housed at the Texas Tech University-Burnett Center; June through November 2001.



Days After Probiotic Supplementation

Figure 2. Incidence of *E. coli* O157:H7 in 180 Cattle (60/treatment) After Treatment with a Competitive Exclusion (CE) Product





Figure 3. Incidence of *E. coli* O157:H7 in Feedlot Pens (12/Treatment) After Treatment with a Competitive Exclusion (CE) Product

Figure 4. Percentage of Animals Shedding at Least One Time After Probiotic Supplementation in Feedlot Cattle at the Texas Tech University Burnett Center

