

NUTRITION: *Original Research*

# Evaluation of an experimental bio-nutrient feedlot premix on health, growth performance, carcass merit, and prevalence and severity of liver abscesses in feedlot cattle managed under a natural feeding protocol\*

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## ABSTRACT

**Objective:** This research evaluates the efficacy of an experimental diet premix on growth performance, carcass traits, liver abscesses, lung lesions, and health of naturally raised feedlot cattle.

**Materials and Methods:** A randomized complete block design trial was conducted at a commercial feedlot using 32 pens of steers and heifers (initial BW = 382 kg) and fed an average of 223 d. Antimicrobials, medicated feed additives, or growth-promoting implants were not administered. Treatments included (1) standard diets (CON) and (2) standard diets including a premix (TST) providing lysine and glutamic acid complexes of zinc and manganese, cobalt pectinate, organic iodine (ethylenediamine dihydroiodide), and rumen protected folate.

**Results and Discussion:** Mortality rate was not affected ( $P = 0.68$ ) by treatment. Removal for respiratory issues and the natural program fallout rate were greater ( $P < 0.01$ ) for TST cattle compared with CON. Control cattle had greater DMI, final BW, ADG, and carcass weight ( $P \leq 0.05$ ) compared with TST. Control cattle had greater G:F than TST cattle when deads and fallouts were included ( $P = 0.02$ ), but not when deads and fallouts were excluded ( $P = 0.92$ ). Percentage carcasses qualifying for each USDA quality grade category did not differ between treatments ( $P \geq 0.07$ ). No treatment differences were ob-

served for the percentage of abscessed livers ( $P = 0.62$ ) or for the percentage of cattle with lung lesions ( $P = 0.51$ ).

**Implications and Applications:** The TST diet premix evaluated did not improve growth performance, carcass merit, or feedlot health attributes under the conditions of this study.

**Key words:** beef cattle, liver abscess, pulmonary lesion, chelated mineral, bovine respiratory disease

## INTRODUCTION

Conventionally raised feedlot cattle are often administered antimicrobials for control of bovine respiratory disease and various other maladies. Despite their effectiveness, use of antimicrobials in livestock production has received increasing scrutiny by the public in recent years, as issues regarding ethics and antimicrobial resistance have come to the forefront. A growing number of consumers are purchasing beef raised under natural feeding protocols.

Dietary amino acid chelated or complexed trace minerals may improve feedlot cattle performance and health (Greene et al., 1988; George et al., 1997; Chirase and Greene, 2001; Spears and Kegley, 2002; Dorton et al., 2006). For example, the role of zinc and the importance of maintaining integrity of intestinal epithelium relative to health has been well documented (Wapnir, 2000; Ulluwishewa et al., 2011; Chase, 2018). Increased bioavailability of complexed minerals (Spears, 2003) may decrease bacterial escape from the rumen by strengthening epithelial tight junctions, thereby lessening liver abscess incidence. Although Wagner et al. (2008) reported no difference in liver abscesses from feeding zinc methionine, Reinhardt et al. (2019) and Lundy et al. (2017) observed a reduction in liver abscesses in steers fed zinc-amino acid complex.

It is widely accepted that B-vitamin requirements of ruminant animals with a functional rumen are met by mi-

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crobial synthesis; however, the demands of modern feedlot production, such as increased concentrate and reduced roughage feeding, resulting in less-than-optimal rumen fermentation, could increase the need for B-vitamin supplementation. Folic acid is a B-complex vitamin that serves as a donor and acceptor of single carbon units (Lucock, 2000), which is essential for cell division and protein synthesis (Combs, 2012). Folic acid deficiency in humans and mice is associated with liver damage and disease (Pogribny et al., 2013; Sid et al., 2017; Xia et al., 2018). Deters et al. (2021) demonstrated a tendency for reduced incidence of liver abscesses when steers were supplemented with 30 or 60 mg of rumen protected folic acid per head daily.

Nonantibiotic mitigation strategies are needed to manage the challenges presented to growing-finishing cattle fed under natural feeding protocols. The objectives of this study were to evaluate the effects of a non-pharmacological, nutrient-based diet premix on feedlot health and growth performance, carcass merit, incidence and severity of liver abscesses, and pulmonary lesions in cattle fed under a natural feeding protocol.

## MATERIALS AND METHODS

This protocol was approved by the Research Committee of Five Rivers Cattle Feeding, Johnstown, Colorado. The study was conducted according to the Five Rivers Cattle Feeding standard operating procedures for humane handling. These standard operating procedures were developed in accordance with the principles and guidelines presented by FASS (2010).

### Cattle Population and Processing Protocol

Crossbred yearling steers and heifers from multiple sources in California, Texas, Colorado, and Kansas were received at the study site, a commercial feedlot in north-central Colorado, over a 15-d period from May 9 through 23, 2019. All cattle were assigned to the Aspen Ridge Natural Beef branded program (JBS Foods USA, Greeley, CO), in which no medicinal feed additives (ionophores, tylosin, melengestrol acetate, or  $\beta$  agonists) or anabolic implants were used. In addition, cattle requiring therapeutic antibiotic treatment were removed from the program. At initial processing, all cattle were weighed and received a bovine rhinotracheitis and bovine viral diarrhea vaccine that included a *Mannheimia haemolytica* toxoid (Pyramid 3 + Presponse; Boehringer Ingelheim Animal Health); a bovine rhinotracheitis, parainfluenza 3, and respiratory syncytial virus vaccine (Inforce 3; Zoetis); a *Clostridium* toxoid (Ultrachoice 7; Zoetis); ivermectin (Noromectin 1%; Norbrook Inc.); fenbendazole (Safeguard 10%; Merck Animal Health); electronic identification tags; and duplicate lot tags (one in each ear). Heifers were checked for pregnancy status and steers checked for intact testicles. Pregnant heifers and intact males were not enrolled in the study. Body weight was not accounted for during the randomization process.

### Experimental Design and Blocking

The experiment was conducted as a randomized complete block design. Blocking factors included sex and receiving group, which was established based on date of arrival and source stratified within arrival date. Each block consisted of 2 pens, 1 pen for each of 2 treatments. As individual cattle from each source exited the squeeze chute following processing, they were placed in 1 of 2 pens within each block on an alternating basis. If space remained in the pens for a given block, cattle from the next source processed on the same day were placed in the pens on an alternating basis until the pens were filled. Thus, sources of cattle included in each block were minimized. A total of 16 complete blocks were used, with 10 blocks consisting entirely of steers and 6 blocks consisting entirely of heifers. Pen stocking rates averaged 277 cattle (range: 249–282) and were within 6 cattle for each pen within a block. A total of 8,635 cattle were initially enrolled, and 7,678 cattle finished the study.

### Dietary Treatments

Treatments evaluated in the study were the standard trace mineral program typically used at the study site (CON), consisting of inorganic sources for the primary trace metals of interest (Co, Cu, I, Mn, Se, and Zn) versus the standard trace mineral program plus an experimental trace mineral program (TST), providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and rumen protected folic acid (RP-folate; Zinpro Corporation). The TST trace mineral treatment was supplemented in addition to the sources of trace mineral provided in the CON diet. The TST trace mineral supplement was fed at a rate of 3.5 g/animal per day, providing 30 mg/animal per day of folate, and was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists). Trace mineral sources used for CON, and thus included in the TST diet, were zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide. During the experiment, it was discovered that the folate used for this study did not meet the desired product standards and was likely subject to greater rumen degradation than anticipated. The extent to which dietary folate escaped rumen degradation is not known.

### Cattle Diets and Management

Cattle were managed and fed in a similar manner across all study pens. A 3-diet system using receiving (low concentrate, high forage, 1.10 Mcal of NEg/kg of DM), transition (moderate concentrate, 1.28 Mcal of NEg/kg of DM), and finishing diets (high concentrate, low forage, 1.52 Mcal of NEg/kg of DM) was fed. Test premix was included in the

receiving, transition, and finishing diets for cattle assigned to the TST treatment. Pens were fed twice daily, with the first feeding in the morning and the second feeding in the afternoon. A “clean bunk” approach to feeding management, where the amounts of residual feed remaining in the feed bunk each morning were minimized while still allowing cattle to express ad libitum intake, was used. Minimal feed refusals were observed, and, on the day of the observed refusal, feed delivered to the bunk was reduced by approximately twice the observed refusal amount, to force cattle to clean their bunk the following day. Basal diets for both CON and TST treatment groups were similar, consisting of steam-flaked corn, steam-flaked wheat, corn silage, whey de-lactose permeate, dried distillers grains, alfalfa hay, wheat hay, triticale hay, corn oil, and a liquid supplement containing vitamins and trace minerals. Diets were formulated to meet or exceed NASEM (2016) feeding recommendations (Table 1). Diets were mixed fresh daily using a stationary auger mixer (Harsh International Inc.) and transported to the pens via commercial feed trucks (Harsh International Inc.). Diet samples were collected daily for DM determination, and weekly samples of each trial diet were collected for determination of chemical composition at a commercial laboratory (ServiTech Laboratories, Hastings, NE).

Cattle were fed in soil-surfaced pens with concrete feeding aprons, and all pens within block provided similar surface areas and linear feed bunk and water trough space. All pens within block were in a similar location in the feedlot and faced the same direction. Treatments were assigned randomly to pens within each block. Animal health personnel were blinded to treatment. Study cattle were observed daily by pen riders, between 0600 and 1000 h, with a single pen rider examining both pens within a block whenever possible, to minimize bias. Moribund cattle were treated between 0900 and 1400 h, and all cattle selected for treatment and removed from the pens within a block were treated at the same hospital facility.

Cattle removed from a pen for the treatment of bovine respiratory disease that had a rectal temperature greater than 40°C or that exhibited moderate to severe signs of depression or weakness were administered tildipirosin (Zuprevo, Merck Animal Health) at a dose of 4 mg/kg of BW (1.0 mL/100 lb BW) subcutaneously. Treatments were administered according to Beef Quality Assurance guidelines (NCBA, 2019). Standard feedlot protocols were implemented for the treatment of digestive disease, lameness, or other maladies unrelated to bovine respiratory disease, and were consistent for cattle across experimental treatments. Due to the specifications of the natural beef program, any animal treated with an antibiotic was weighed and was re-lotted to a non-trial lot. The weights and intakes of these fallout cattle were no longer tracked after they were re-lotted. Cattle were sold for salvage following antibiotic withdrawal if removed from the study and if full recovery from disease symptoms was deemed

**Table 1.** Dry matter ingredient and chemical composition for finishing diets averaged over feeding period

Item	Control	Test
Ingredient, % DM basis		
Steam-flaked corn	43.2	43.2
Steam-flaked wheat	23.9	23.9
Corn silage	10.2	10.2
Wheat or triticale hay	3.9	3.9
Dried distillers grains	9.2	9.2
Whey delactose permeate	2.5	2.5
Supplement <sup>1</sup>	3.9	3.9
Vegetable oil	3.2	3.2
Test premix <sup>2</sup>	0	+
Chemical composition <sup>3</sup>		
Crude protein, %	14.6	14.3
NPN, %	2.9	3.0
Crude fat, %	6.1	6.6
NDF, %	17.0	17.0
Ca, %	0.51	0.52
P, %	0.48	0.47
K, %	0.98	0.97
Na, %	0.20	0.20
S, %	0.19	0.20
Cu, mg/kg	15.3	14.8
Zn, mg/kg	93.0	122.2
Mn, mg/kg	49.8	64.7
Se, mg/kg	0.33	0.36
Co, mg/kg	0.20	0.60
I, mg/kg	0.89	3.33

<sup>1</sup>Supplements for the control and test groups were identical, providing NPN, macro-minerals, vitamins A and E, and inorganic trace minerals from zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide.

<sup>2</sup>Test premix was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists) and was fed at the rate of 3.5 g/animal per day, providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and 30 mg/animal per day rumen protected folic acid (RP-folate; Zinpro Corporation).

<sup>3</sup>As determined by ServiTech Laboratories (Hastings, NE).

unlikely. Mortalities were subject to postmortem examination by a licensed veterinarian or trained feedlot employee.

Cattle were fed an average of 222.6 d (range = 186 to 258 d). Pens within each of the blocks were marketed at equal days on feed according to feedlot standard operating procedures determining the appropriate degree of finish for the all-natural branded program. Cattle were pen-weighted the morning of shipment for harvest, and feed and water were not withheld from the cattle before weighing. Al-

though sequential pens within harvest block were shipped, shipment order for treatments within harvest blocks was randomly determined because assignment of treatment to pen within block was random. All cattle were harvested at a commercial abattoir (JBS Foods USA; Greeley, CO) between November 15, 2019, and February 10, 2020. All pens in each harvest block were shipped together on the same day.

### **Feedlot Performance**

Growth performance data were collected by pen and included unshrunk initial BW, final BW (4% pencil shrink included), ADG, G:F, and DMI. Average daily gain and G:F were calculated on the bases of both deads and fallouts included and deads and fallouts excluded. For deads and fallouts included, ADG was computed by subtracting the total starting weight for all cattle in the pen from the total sale weight of non-fallout cattle and dividing the result by the total days on feed by all cattle for the pen. For deads and fallouts excluded, ADG was calculated by subtracting the average starting weight for the pen from the average sale weight of non-fallouts and dividing the result by the average days on feed for the pen.

### **Carcass Merit**

Carcass data were collected using an in-house camera system, USDA graders, and an independent third-party contractor (Diamond T Livestock Services, Inc.; Alliance, NE), where one person verified the kill order of individual animals by scanning the radio frequency identification tags or recording individual ear tag numbers, and another trained technician scored and recorded incidence and severity of liver abscesses and other liver pathology. Carcass metrics included DP, hot carcass weight (HCW), LM area, KPH, fat depth at 12th to 13th rib (FT), YG, marbling score (400 = Small<sup>0</sup>, 500 = Modest<sup>0</sup>, 600 = Moderate<sup>0</sup>, and 700 = Slightly abundant<sup>0</sup>), and QG. For QG and YG, the percentage of carcasses in each pen qualifying for each QG and YG category was used as the response variable for analysis.

### **Liver Abscesses**

Assessment of liver abscesses was conducted by a trained technician on the day of harvest. Livers were observed for the presence, number, and severity of abscesses and the presence of liver cirrhosis, flukes, and telangiectasis. Livers were scored using the Elanco Liver Check System and classified into 4 categories, including the percentage of livers qualifying for the A<sup>-</sup> (1 or 2 small abscesses), A (2 to 4 small active abscesses), and A<sup>+</sup> (1 or more large active abscesses) categories plus the percentage of livers showing any liver abscesses regardless of severity. The percentage of livers from each pen falling into each category was used as the response variable for the analysis.

### **Pulmonary Lesions**

Lungs of harvested cattle were also observed at the abattoir on the day of harvest by a trained technician. Evaluation of pulmonary lesions was performed according to the system described by Tennant et al. (2014), and lungs were classified into 5 categories, including normal (no visible lesions or pathology), mild ( $\leq 5\%$  of the lung surface exhibited lesions and consolidation due to previous or current pneumonia), moderate (5 to 15% of the lung surface exhibited lesions and consolidation attributable to previous or current pneumonia), moderately severe (15 to 50% lung surface exhibited lesions or consolidation due to previous or current pneumonia), or severe ( $> 50\%$  of the lung surface was consolidated due to previous or current pneumonia, visible signs of adherence of the lung to the pleural cavity, or a significant percentage of the lung missing). The moderately severe and severe pulmonary lesion categories were combined into a single category for the analysis. Percentage of animals from each pen falling into each category was used as the response variable for analysis. Assessment of pulmonary lesions was challenging, particularly for the first 7 blocks of cattle shipped, due to inability to place a technician in a proper vantage point along the viscera belt to allow for adequate observation of the lungs. For the remaining 9 blocks, a better vantage point was present; however, because of the speed along which lungs passed the observation point and the inability to manipulate each lung for a more thorough assessment, only lesions readily visible from a surface perspective could be scored. Thus, pulmonary lesion data were recorded for only 9 of 16 study blocks and accounted only for lesions that were readily visible on the surface of the lung.

### **Statistical Analysis**

Analysis was performed using ANOVA of linear mixed-effects regression models fitted by the lmer function in R (R Foundation for Statistical Computing, version 4.0.0, April 2020 release date). Each model included fixed effects of treatment, sex, and the interaction between treatment and sex. Block was included in each model as a random effect. All terms were left in each model regardless of significance. Pairwise comparisons of estimated marginal means were performed using the emmeans function. Pen ( $n = 16$  for most analyses,  $n = 9$  for lung lesion analysis) was treated as the experimental unit. Significance was declared at  $P < 0.05$ , and tendencies were declared at  $0.05 \leq P \leq 0.10$ .

## **RESULTS AND DISCUSSION**

### **Feedlot Health and Growth Performance**

The effects of treatment on feedlot health are summarized in Table 2. Treatment by sex interactions ( $P \geq 0.15$ ) and the effects of sex were not significant ( $P \geq 0.19$ ) for

**Table 2.** The effects of dietary premix treatment on feedlot health

Item <sup>1</sup>	Control <sup>2</sup>	Test <sup>3</sup>	P-value	SEM
Number of pen replicates	16	16	—	—
Total cattle enrolled	4,425	4,431	—	—
Cattle sold from original lot	3,937	3,826	—	—
Mortality and fallout rate, <sup>4</sup> %	11.02	13.68	0.01	1.5
Mortality rate, %	2.42	2.28	0.68	0.3
Respiratory, %	0.27	0.18	0.44	0.1
Atypical interstitial pneumonia, %	0.09	0.11	—	—
Bloat, %	1.42	1.20	0.37	0.2
Digestive, %	0.02	0.09	—	—
Other, %	0.61	0.70	0.53	0.1
Fallout rate, <sup>4</sup> %	8.61	11.40	0.01	1.6
Respiratory, %	8.45	11.10	0.01	1.6
Buller, %	0.02	0.02	—	—
Other, %	0.14	0.27	0.18	0.1
Pulled and not removed from study, <sup>5</sup> %	2.94	3.88	0.03	0.4
Respiratory, %	1.88	2.32	0.20	0.3
Digestive, %	0.11	0.27	0.13	0.1
Other, %	0.95	1.29	0.19	0.2
Total pulled, <sup>6</sup> %	11.54	15.28	0.01	1.7
Respiratory, %	10.33	13.42	0.01	1.6
Other, %	1.22	1.85	0.10	0.2

<sup>1</sup>Sex ( $P \geq 0.19$ ) and treatment by sex interactions ( $P \geq 0.15$ ) were not significant. Therefore, only main effects of treatment are shown.

<sup>2</sup>Dietary supplements for the control and test groups were identical, providing NPN, macro-minerals, vitamins A and E, and inorganic trace minerals from zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide.

<sup>3</sup>Test premix was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists) and was fed at the rate of 3.5 g/animal per day, providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and 30 mg/animal per day of rumen protected folic acid (RP-folate; Zinpro Corporation).

<sup>4</sup>Fallout rate is the percentage of initial cattle enrolled in a pen that were removed from the branded program due to failure to recover from various maladies or due to antibiotics administered to aid in recovery.

<sup>5</sup>These cattle were temporarily removed from the pen and subjected to closer examination of health status. They were either not treated or treated without the administration of antibiotics and returned to the pen.

<sup>6</sup>Fallout rate plus cattle pulled and not removed from the study.

any of the health parameters evaluated; therefore, only the main effects of treatment are described. Total dead and all-natural branded program fallout rates were 11.02 and 13.68% for CON and TST cattle, respectively ( $P \leq 0.01$ ). Total mortality rate ( $P = 0.68$ ) and mortalities due to respiratory ( $P = 0.44$ ), bloat ( $P = 0.37$ ), or other causes ( $P = 0.53$ ) were not affected by treatment. Treatment differences in pull rate (cattle removed from the pen and evaluated more closely for disease symptoms) were influenced by treatment. Cattle pulled from the pen and removed from the all-natural branded program, and hence the study, was 11.40% for the TST and 8.61% for the CON cattle ( $P \leq 0.01$ ). Most fallouts were due to respiratory

problems (11.10 and 8.45% of initial enrollments for TST and CON cattle, respectively). Total pull and respiratory pull rates were greater ( $P \leq 0.01$ ) for TST cattle compared with CON (15.28 and 13.42% vs. 11.54 and 10.33%, respectively).

Growth performance results are described in Table 3. Most interactions between treatment and sex were not significant ( $P \geq 0.12$ ) for feedlot growth performance variables. Initial BW averaged 378.5 kg and did not differ between treatments ( $P = 0.82$ ) or for heifers versus steers ( $P = 0.18$ ). Daily DMI was lesser ( $P = 0.03$ ) for TST compared with CON cattle (10.0 and 10.2 kg, respectively) and was not different for steers compared with heifers ( $P$

**Table 3.** The effects of dietary premix treatment and sex on growth performance

Item	Treatment			Sex			P-value		
	Control <sup>1</sup>	Test <sup>2</sup>	SEM <sup>3</sup>	Steer	Heifer	SEM <sup>4</sup>	Treatment	Sex	Treatment × sex
Initial BW, kg	378	379	8.1	390	367	12.9	0.82	0.18	0.90
DMI, kg	10.2	10.0	0.06	10.2	10.0	0.09	0.03	0.31	0.99
Deads and fallouts out <sup>5</sup>									
Final BW, <sup>6</sup> kg	653	650	2.9	668	635	4.3	0.05	≤0.01	0.12
ADG, kg	1.25	1.23	0.018	1.22	1.26	0.027	0.03	0.32	0.07
G:F <sup>7</sup>	0.122	0.122	0.001	0.120	0.125	0.002	0.92	0.09	0.01
Deads and fallouts in <sup>8</sup>									
Final BW, <sup>9</sup> kg	623	616	4.2	638	601	6.1	0.03	≤0.01	0.67
ADG, kg	0.99	0.91	0.045	0.95	0.95	0.068	≤0.01	0.95	0.93
G:F	0.097	0.091	0.004	0.093	0.095	0.007	0.02	0.84	0.91

<sup>1</sup>Dietary supplements for the control and test groups were identical, providing NPN, macro-minerals, vitamins A and E, and inorganic trace minerals from zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide.

<sup>2</sup>Test premix was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists) and was fed at the rate of 3.5 g/animal-per day, providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and 30 mg/animal-per day of rumen protected folic acid (RP-folate; Zinpro Corporation).

<sup>3</sup>Pens = 16.

<sup>4</sup>Pens = 10 for steers and 6 for heifers. Largest SEM shown.

<sup>5</sup>(Average sale weight of non-fallouts – average starting weight of all cattle enrolled)/average days on feed.

<sup>6</sup>Average gross weight of non-fallouts sold, less 4%, representing an industry standard shrink.

<sup>7</sup>For steers, G:F was 0.170 and 0.120 for control compared with test treatment. For heifers, G:F was 0.126 and 0.124 for control compared with test treatment, respectively.

<sup>8</sup>(Total sale weight of non-fallouts – total starting weight of all cattle enrolled)/total head days for all cattle.

<sup>9</sup>Average of the total gross weight of all non-fallouts sold plus removal weight for fallouts, less 4%, representing a standard industry shrink.

= 0.31). Final BW was greater ( $P \leq 0.05$ ) for CON than for TST cattle and lesser ( $P \leq 0.01$ ) for heifers compared with steers on the bases of both deads and fallouts included (623 vs. 616 and 602 vs. 638 kg, respectively) and deads and fallouts excluded (653 vs. 650 and 635 vs. 668 kg, respectively). Control cattle had greater ( $P \leq 0.05$ ) ADG than TST cattle on the bases of both deads and fallouts included (0.99 vs. 0.91 kg) and deads and fallouts excluded (1.25 vs. 1.23 kg). The effect of sex on ADG was not significant ( $P \geq 0.32$ ). Control cattle had greater ( $P = 0.02$ ) G:F than TST cattle when analyzed on a deads and fallouts included basis, but this difference did not exist ( $P = 0.92$ ) when deads and fallouts were excluded. The effect of sex on G:F was not significant when expressed on a deads and fallouts included basis ( $P = 0.84$ ) and tended to be significant when deads and fallouts were excluded ( $P = 0.09$ ). However, the sex by treatment interaction for G:F on a deads and fallouts excluded basis was significant ( $P = 0.01$ ), indicating that the effect of treatment on G:F was greater in steers (0.120 and 0.170 for TST and CON, respectively) than in heifers (0.124 and 0.126 for TST and CON, respectively).

### Carcass Merit

The effects of treatment on carcass merit are described in Table 4. Control cattle had greater HCW (409 and 406 kg, respectively;  $P = 0.03$ ), FT (1.73 and 1.70 cm, respectively;  $P = 0.02$ ), and yield grade calculated from carcass measurements (3.49 and 3.44, respectively;  $P = 0.02$ ) than TST cattle. Steers had greater ( $P \leq 0.01$ ) HCW (419 and 396 kg, respectively) and calculated yield grade (3.65 and 3.28, respectively) and lesser ( $P \leq 0.01$ ) FT (1.64 and 1.79 cm, respectively) and LM area (83.8 and 90.3 cm<sup>2</sup>, respectively) compared with heifers. Fat thickness observed for these cattle was greater than what is generally seen for conventionally raised cattle. These cattle were fed according to standards for a branded all-natural program. This means no growth-promoting implants or  $\beta$ -agonists were used, premiums are placed on achieving high quality grades, and discounts for excessive yield grade are minimal. These factors contribute to the apparent added fatness for these cattle. The effects of treatment ( $P = 0.21$ ) and sex ( $P = 0.57$ ) on DP were not significant; however, a significant treatment by sex interaction ( $P \leq 0.01$ ) sug-

**Table 4.** The effects of dietary premix treatment and sex on carcass merit

Item	Treatment			Sex			P-value		
	Control <sup>1</sup>	Test <sup>2</sup>	SEM <sup>3</sup>	Steer	Heifer	SEM <sup>4</sup>	Treatment	Sex	Interaction
Hot carcass weight, kg	409	406	1.4	419	396	2.1	0.03	≤0.01	0.36
Dressing percentage <sup>5</sup>	62.35	62.28	0.002	62.41	62.41	0.003	0.21	0.57	≤0.01
Fat depth, 12th rib, cm	1.73	1.70	0.025	1.64	1.79	0.051	0.02	≤0.01	0.14
LM area, cm <sup>2</sup>	87.0	87.1	0.45	83.8	90.3	0.68	0.84	≤0.01	0.20
Calculated yield grade <sup>6</sup>	3.49	3.44	0.03	3.65	3.28	0.05	0.02	≤0.01	0.82
Marbling score <sup>7</sup>	638	633	7.7	635	635	11.8	0.21	0.97	0.28
USDA QG distribution <sup>8</sup>									
High Prime, %	0.6	0.7	0.2	0.7	0.6	0.2	0.78	0.91	0.82
Average Prime, %	5.6	4.6	1.0	5.4	4.8	1.3	0.07	0.73	0.36
Low Prime, %	28.0	27.9	2.5	27.6	28.3	3.8	0.90	0.90	0.90
High Choice, %	23.8	22.2	2.0	22.2	23.8	3.1	0.15	0.68	0.88
Average Choice, %	26.5	28.3	1.6	26.8	28.0	2.4	0.10	0.71	0.79
Low Choice, %	10.5	11.5	1.7	12.2	9.9	2.3	0.21	0.47	0.23
Select, %	1.1	1.0	0.2	0.8	1.4	0.1	0.71	0.16	0.89
Standard, %	0.1	0.1	0.1	0.1	0.1	0.1	0.70	0.38	0.72
Missing QG data, %	3.8	3.7		4.2	3.1				
USDA YG distribution <sup>8</sup>									
YG 1 and 2, %	6.5	7.3	1.0	7.0	6.8	1.4	0.17	0.95	0.78
YG 3, <sup>9</sup> %	65.5	63.1	3.1	68.1	60.3	4.9	0.06	0.21	≤0.01
YG 4 and 5, <sup>10</sup> %	26.5	27.8	3.6	23.3	31.4	6.0	0.24	0.26	≤0.01
Missing YG data, %	1.5	1.8		1.6	1.5				

<sup>1</sup>Dietary supplements for the control and test groups were identical, providing NPN, macro-minerals, vitamins A and E, and inorganic trace minerals from zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide.

<sup>2</sup>Test premix was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists) and was fed at the rate of 3.5 g/animal per day, providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and 30 mg/animal-per day of rumen protected folic acid (RP-folate; Zinpro Corporation).

<sup>3</sup>Pens = 16.

<sup>4</sup>Pens = 10 for steers and 6 for heifers. Largest SEM shown.

<sup>5</sup>For steers, dressing percentage was 62.53 and 62.30 for control and test treatments, respectively. For heifers, dressing percentage was 62.16 and 62.25 for control and test treatments, respectively.

<sup>6</sup>Calculated from carcass measurements.

<sup>7</sup>Marbling score units, 600 = moderate<sup>00</sup>; 700 = slightly abundant<sup>00</sup>.

<sup>8</sup>Percentage of individual carcasses within each pen qualifying for each category.

<sup>9</sup>For steers, the percentages of YG 3 carcasses were 66.6 and 69.6 for control and test treatments, respectively. For heifers, the percentages of YG 3 carcasses were 64.4 and 56.1 for the control and test treatments, respectively.

<sup>10</sup>For steers, the percentages of YG 4 and 5 carcasses were 25.4 and 21.3 for control and test treatments, respectively. For heifers, the percentages of YG 4 and 5 carcasses were 27.7 and 35.4 for the control and test treatments, respectively.

gested that DP was greater for CON compared with TST steers (62.53 and 62.30, respectively) but lesser for CON compared with TST heifers (62.16 and 62.25, respectively).

Percentage carcasses grading low Choice or greater exceeded 95%. The main effects of treatment or sex on carcass quality grade were not significant ( $P \geq 0.07$ ). However, the sex by treatment interaction was significant ( $P \leq 0.01$ ) for percentage carcasses qualifying for the YG 3 and YG 4 and 5 categories, suggesting that for steers the percentage YG 3 carcasses was greater and the percentage

YG 4 and 5 carcasses was lesser for TST compared with CON treatments, but in heifers the percentage YG 3 carcasses was lesser and the percentage YG 4 and 5 carcasses was greater for TST compared with CON treatments. No other differences were found in carcass metrics between treatments or for steers compared with heifers ( $P > 0.05$ ).

The experimental, non-pharmacological, nutrient-based diet premix did not improve cattle health, feedlot performance, or carcass merit in the current study. In fact, in several instances TST cattle experienced poorer performance than control cattle. This observation might result

from energy expenditures to eliminate excess trace mineral from the body. As shown in Table 1, the primary differences between dietary treatments were the concentration and source of trace minerals and the inclusion of 30 mg/animal per day of folate from the TST premix. Concentrations of Cu and Se in the CON and TST diets were similar and averaged 15.3 and 0.33 compared with 14.8 and 0.36 mg/kg of DM, respectively. Concentrations of Zn, Mn, Co, and I were greater in the TST compared with CON diets, averaging 122.2, 64.7, 0.60, and 3.33 compared with 93.0, 49.8, 0.20, and 0.89 mg/kg of DM, respectively. These concentrations exceed NASEM (2016) trace mineral requirements for growing and finishing cattle of 10, 30, 20, 0.10, 0.15, and 0.50 mg/kg DM for Cu, Zn, Mn, Se, Co, and I, respectively.

Supplementation of trace minerals that exceed NASEM (2016) recommendations in diets for growing and finishing cattle is common in the feedlot industry (Galyean and Gleghorn, 2001; Vasconcelos and Galyean, 2007; Samuelson et al., 2016). Berrett et al. (2015) compared a non-supplemented basal diet to NASEM (2000) trace mineral standards and industry dietary trace mineral supplementation strategies as described by Vasconcelos and Galyean (2007). No treatment differences in feedlot performance or carcass merit were observed, indicating that the basal diet adequately met trace mineral requirements for yearling feedlot steers. Other studies examining the effects of dietary Cu and Zn concentration and source on feedlot performance have shown positive effects (Nunnery et al., 1996; Ward and Spears, 1997; Engle et al., 2000b; Malcolm-Callis et al., 2000; Lee et al., 2002; Spears and Kegley, 2002), negative effects (Engle and Spears, 2000a), or no effect (Greene et al., 1988; Rust and Schlegel, 1993; Engle and Spears, 2000b, 2001; Engle et al., 2000a; Malcolm-Callis et al., 2000).

### Liver Abscess Incidence and Severity

Treatment effects on liver abscess incidence and severity are presented in Table 5. Approximately 42% of all livers were abscessed. Treatment differences were not observed for the percentage of livers with no abscesses ( $P = 0.62$ ), A<sup>+</sup> abscesses ( $P = 0.14$ ), A abscesses ( $P = 0.88$ ), or A<sup>-</sup> abscesses ( $P = 0.63$ ). No significant differences were observed for sex for all liver abscesses ( $P = 0.32$ ), A<sup>+</sup> liver abscesses ( $P = 0.82$ ), A liver abscesses ( $P = 0.72$ ), or A<sup>-</sup> liver abscesses ( $P = 0.18$ ). No significant interaction was found between sex and treatment ( $P > 0.10$ ) for any of the response variables.

Deters et al. (2021) reported on the results of a dose response trial with rumen protected folate where increases in carcass weight and dressing percentage and a reduction in overall as well as A<sup>+</sup> liver abscess incidence were observed when fed at 3 mg/kg of diet DM. Rumen protected folate has also been reported to improve growth and performance in Holstein steers (La et al., 2019). Unfortunately, midway through the current feeding study,

**Table 5.** Effects of treatment on liver abscess prevalence and severity

Category <sup>1</sup>	Percent of livers observed		P-value	SEM <sup>4</sup>
	Control <sup>2</sup>	Test <sup>3</sup>		
A <sup>-</sup> abscesses <sup>5</sup>	11.5	12.0	0.63	1.35
Heifers	13.3	13.8	0.78	2.14
Steers	9.8	10.3	0.68	1.66
A abscesses <sup>6</sup>	11.7	11.5	0.88	0.76
Heifers	12.3	11.4	0.54	1.20
Steers	11.1	11.7	0.59	0.93
A <sup>+</sup> abscesses <sup>7</sup>	19.6	18.3	0.14	1.09
Heifers	20.2	18.2	0.16	1.73
Steers	19.0	18.4	0.54	1.34
All abscesses <sup>8</sup>	42.8	41.9	0.62	2.34
Heifers	45.8	43.3	0.44	3.71
Steers	39.9	40.4	0.84	2.87

<sup>1</sup>Liver abscess evaluations performed using Elanco Liver Check System. A<sup>-</sup> = percentage of livers showing 1 or 2 small abscesses; A = percentage of livers showing 2–4 small active abscesses; A<sup>+</sup> = percentage of livers showing 1 or more large active abscesses; All = percentage of livers showing any liver abscesses, regardless of severity.

<sup>2</sup>Dietary supplements for the control and test groups were identical, providing NPN, macro-minerals, vitamins A and E, and inorganic trace minerals from zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide.

<sup>3</sup>Test premix was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists) and was fed at the rate of 3.5 g/animal-per day, providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and 30 mg/animal-per day of rumen protected folic acid (RP-folate; Zinpro Corporation).

<sup>4</sup>Pens = 10 for steers and 6 for heifers.

<sup>5</sup>Sex,  $P = 0.18$ ; treatment  $\times$  sex,  $P = 0.98$ .

<sup>6</sup>Sex,  $P = 0.72$ ; treatment  $\times$  sex,  $P = 0.42$ .

<sup>7</sup>Sex,  $P = 0.81$ ; treatment  $\times$  sex,  $P = 0.45$ .

<sup>8</sup>Sex,  $P = 0.32$ ; treatment  $\times$  sex,  $P = 0.46$ .

notice was received that the rumen protected folate being used was found to not meet product specifications, which may have had a role in the failure to repeat the results of Deters et al. (2021). The effects of this error in production are unclear but should be noted in any interpretation of the results from the current study.

Results from this study showed that the TST premix did not control liver abscess incidence or severity in natural-fed cattle. Reinhardt et al. (2019) reported a reduction in liver abscess incidence in cattle fed either Zinpro, zinc methionine, or Availa Zn (Zinpro Corporation). In the

present study, supplemental Zn was provided to cattle for the first time using a new product (ProPath Zn; Zinpro Corporation) in large part due its suitability for being administered through a water-flush micro-ingredient machine, thus precluding the necessity for a separate dry supplement.

Prevention and control of liver abscesses remains a challenge. For cattle fed conventional feedlot diets, tylosin or other in-feed antimicrobials can be used to manage the incidence and severity of liver abscesses (Brown et al., 1973, 1975; Potter et al., 1985; Baba et al., 1989; Rogers et al., 1995; Nagaraja and Lechtenberg, 2007; Weinroth et al., 2019). For cattle enrolled in programs that do not allow these antimicrobial interventions, liver abscesses can be a significant issue affecting animal health, well-being, and productivity as well as presenting challenges at the packing plant. Increasing scrutiny surrounding use of antimicrobials, particularly feed-grade antimicrobials fed at subtherapeutic levels, has led to renewed interest in development of alternative methods to control liver abscesses.

Liver abscesses can be regarded as a pathology resulting from feeding diets providing increased concentrations of ruminally fermentable starch from feed grains. The economic realities of the cattle feeding industry continue to favor grain feeding and disincentivize feeding roughage. Although effective for reducing liver abscesses, increasing dietary roughage inclusion increases production costs and inefficiencies in feed manufacturing (Bartle and Preston, 1991; Birkelo et al., 1991; Bartle et al., 1994). Increasing roughage particle size has been shown to increase rumination time (Gentry et al., 2016) and may help manage liver abscesses. However, increases in roughage particle size may cause feed milling issues for feedlots using stationary batch mixing systems requiring finished feed storage in overhead bins. At present, it is unlikely that roughage particle size or diet concentrations will substantially increase in feedlot diets, thereby requiring development of alternative, nutrition-based methods as potential avenues for liver abscess control in both conventionally fed feedlot cattle as well as those managed under a similar protocol to those in the present research.

The results of this study may be viewed in the wider context of challenges in demonstrating success in controlling liver abscesses using non-antimicrobial, non-pharmacological feed additives. *Saccharomyces cerevisiae* fermentation products (Huebner et al., 2019), antioxidants (Müller et al., 2018), and essential oils (Meyer et al., 2009) have all been investigated and have shown limited efficacy for control of liver abscesses. Vyas et al. (2015) reported a decrease in time that rumen pH was below 6.2 but no effect on acidosis through supplementation of malic acid. It may be that these products have no long-acting effect on rumen health or that the rumenitis-liver abscess complex is difficult to control without more conservative approaches to feeding, thus demonstrating the difficulty in controlling ruminal lesions and associated liver abscesses.

## Pulmonary Lesion Incidence and Severity

The effects of treatment on pulmonary lesions are summarized in Table 6. Approximately 35% of the cattle within each pen, among the 9 blocks evaluated, had lungs with surface lesions. Treatment differences were not observed for percentage of cattle exhibiting mild ( $P = 0.64$ ), moderate ( $P = 0.86$ ), or severe ( $P = 0.30$ ) surface pulmonary lesions at slaughter. The combined percentage of cattle observed with any lung lesions likewise was not different between treatments ( $P = 0.51$ ) or between sexes ( $P = 0.39$ ), with no interaction evident between sex and treatment observed ( $P = 0.29$ ) for this category. However, an interaction was observed between sex and treatment in the severe category ( $P < 0.01$ ). There was a tendency ( $P = 0.05$ ) for more heifers than steers in the TST treatment to be in the severe category. Within heifers, more cattle in the TST treatment fell in the severe category than those in the CON treatment ( $P < 0.01$ ). However, more steers fell in the severe category for the CON compared with the TST treatment ( $P = 0.01$ ). Overall, the effect of sex was not found to be significant in the severe category ( $P = 0.30$ ).

The overall prevalence of total pulmonary lesions observed in this study was approximately 35%, as compared with a similar value of 33% reported by Rezac et al. (2014). Rezac et al. (2014) likewise reported prevalence of mild and severe pulmonary lesions of 22.5% and 9.8%, respectively, of cattle scored. Overall mean values for pulmonary lesions designated as either mild or severe in this study were approximately 5% and 12%, respectively. A somewhat comparable incidence of pulmonary lesions between this study and that of Rezac et al. (2014) is surprising, considering that, in the current study, cattle treated for respiratory disease were removed from the study. For the current study, cattle with lung lesions likely represent undiagnosed respiratory issues, substantiating the findings of Gardner et al. (1999), White and Renter (2009), and Leruste et al. (2012), that a sizable percentage of feedlot cattle have an ongoing challenge with undiagnosed or sub-clinical respiratory disease throughout the feeding period.

## APPLICATIONS

The formulation and manufacturing process used for the TST premix used in this study did not result in a product that improves health, feedlot performance, and carcass merit, or that reduces the incidence and severity of liver abscesses or pulmonary lesions. However, as an initial pilot study, the lack of a more positive response from the TST premix is not altogether unexpected. Given the novelty of the individual components, as well as the total premix, these results indicate the need for further research. It is not known whether the lack of a positive response observed for the TST premix was the result of suboptimum dose and timing for feeding of the experimen-

**Table 6.** Effects of treatment on pulmonary lesion prevalence and severity

Category <sup>1</sup>	Percent of all lungs observed		P-value	SEM <sup>4</sup>
	Control <sup>2</sup>	Test <sup>3</sup>		
Mild <sup>5</sup>	4.6	5.5	0.64	1.69
Heifers	5.7	2.3	0.33	2.76
Steers	3.4	8.8	0.05	1.95
Moderate <sup>6</sup>	17.8	18.2	0.86	2.20
Heifers	21.9	18.0	0.33	3.59
Steers	13.7	18.5	0.11	2.54
Severe <sup>7</sup>	11.3	12.4	0.30	1.94
Heifers	11.1	16.9	0.01	3.17
Steers	11.6	8.0	0.01	2.24
Any lesions <sup>8</sup>	33.7	36.2	0.51	3.73
Heifers	38.7	37.1	0.80	6.09
Steers	28.7	35.2	0.16	4.30

<sup>1</sup>Evaluations performed using the system described by Tennant et al. (2014): Mild ( $\leq 5\%$  of the lung surface exhibited lesions and consolidation due to previous or current pneumonia); moderate (5–15% of the lung surface exhibited lesions and consolidation attributable to previous or current pneumonia); severe ( $>15\%$  of the lung surface was consolidated due to previous or current pneumonia, had visible signs of adherence of the lung to the pleural cavity, or a significant percentage of the lung was missing).

<sup>2</sup>Dietary supplements for the control and test groups were identical, providing NPN, macro-minerals, vitamins A and E, and inorganic trace minerals from zinc sulfate, manganese sulfate, copper sulfate, cobalt carbonate, sodium selenite, and ethylenediamine dihydroiodide.

<sup>3</sup>Test premix was added to the diet through a water-flush micro-ingredient machine (Comco Systems and Automation Specialists) and was fed at the rate of 3.5 g/animal-per day, providing lysine and glutamate amino acid complexes of Zn and Mn, complexed Co in the form of cobalt pectinate (ProPath; Zinpro Corporation), additional I from potassium iodine and ethylenediamine dihydroiodide, and 30 mg/animal-per day of rumen protected folic acid (RP-folate; Zinpro Corporation).

<sup>4</sup>Pens = 10 for steers and 6 for heifers.

<sup>5</sup>Sex,  $P = 0.47$ ; treatment  $\times$  sex,  $P = 0.06$ .

<sup>6</sup>Sex,  $P = 0.33$ ; treatment  $\times$  sex,  $P = 0.10$ .

<sup>7</sup>Sex,  $P = 0.30$ ; treatment  $\times$  sex,  $P < 0.01$ . For steers, severe abscess rates were 8.0 and 11.6 for the test and control treatments, respectively. For heifers, severe abscess rates were 16.9 and 11.1 for the test and control treatments, respectively.

<sup>8</sup>Sex,  $P = 0.39$ ; treatment  $\times$  sex,  $P = 0.29$ .

tal product components, or of manufacturing problems associated with ramping up premix production practices to produce the amounts of product required for commercial application. The complexity and diversity of current beef

feedlot production and marketing systems place additional challenges on the commercial development of alternative feed additive or other products that can meet ever-changing consumer perceptions and demands while still providing evidence-based solutions for issues that affect cattle health, well-being, productivity, and, ultimately, profitability to the producer and a safe and high-value product for the consumer.

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