

Supplementing Micro-Aid to optimize health and performance of receiving cattle¹

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INTRODUCTION

Feedlot receiving is one of the most critical phases within the beef production cycle when cattle are exposed to a multitude of stress and health challenges that directly impact their immunocompetence and productivity. These include road transport, commingling with different animals, and exposure to novel diets and environments (Cooke, 2017). Accordingly, incidence of bovine respiratory diseases (BRD) is elevated during the initial 30 d of feedlot receiving, with clinical symptoms observed in up to 60% of receiving cattle despite efforts associated with stress minimization and vaccination against BRD pathogens (Cooke, 2017).

Prophylactic medication with feed-grade antimicrobials is often effective in mitigating BRD incidence during feedlot receiving (Wilson et al., 2017). However, with increased regulations regarding the use of feed-grade antimicrobials in livestock systems (US Food and Drug Administration, 2015), alternative dietary strategies that enhance immune function of receiving cattle are warranted. These include the use of nonantibiotic feed ingredients with immunomodulatory properties, such as Micro-Aid (DPI

Global; Porterville, CA). This ingredient is manufactured from purified extract of *Yucca schidigera* plant and contains saponins, which have ruminal and systemic immunological benefits (Moreau et al., 2002). Therefore, we hypothesized that Micro-Aid supplementation improves cattle immunocompetence and productivity during feedlot receiving. To investigate this hypothesis, this experiment evaluated the effects of supplementing Micro-Aid on performance, health, and physiological responses of receiving cattle.

MATERIALS AND METHODS

This experiment was conducted at the Oregon State University—Eastern Oregon Agricultural Research Center (Burns station). All animals were cared for in accordance with acceptable practices, and experimental protocols reviewed and approved by the Oregon State University, Institutional Animal Care and Use Committee (#4973).

Animals and Treatments

One hundred and five recently weaned Angus × Hereford calves (75 steers and 30 heifers) were purchased from a commercial auction yard (Producers Livestock Marketing Association; Vale, OR). Calves originated from eight cow-calf operations located in Eastern Oregon and Western Idaho. On the day of purchase (day –2; 1800 h), calves were loaded into a commercial livestock trailer (Legend 50' cattle liner; Barrett

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LLC; Purcell, OK) at the auction yard and transported for 800 km (12 h) to stimulate the stress of a long haul (Cooke, 2017). On day -1 of the experiment (0600 h), calves were unloaded at the Eastern Oregon Agricultural Research Center, arrival shrunk body weight (BW) was recorded, and calves were maintained as a single group with free choice hay, water, and mineral supplement for 24 h.

On day 0 of the experiment, calves were ranked according to sex, source, and shrunk BW and allocated to a 21-pen drylot (five calves/pen, being one or two heifers per pen), in a manner that pens had equivalent initial shrunk BW and calves from at least three different sources to stimulate the stress of comingling. Pens were assigned to receive total mixed ration (TMR) and one of three treatments (as-fed basis): 1) 1 g/calf daily of Micro-Aid (M1; $n = 7$), 2) 2 g/calf daily of Micro-Aid (M2; $n = 7$), or 3) no Micro-Aid supplementation (CON; $n = 7$). Calves had free choice access to water and TMR (Table 1), which was offered twice daily (0800 and 1300 h) from days 0 to 59. Micro-Aid was mixed with soybean meal (1.25 kg/pen) and top-dressed daily into the morning TMR feeding of M1 and M2 pens. Soybean meal was also top-dressed into the morning TMR feeding of CON pens (1.25 kg/pen), without the addition of Micro-Aid. On days 0 and 21, calves were vaccinated and administered anthelmintic as described by Lippolis et al. (2017).

Sampling

Samples of TMR ingredients were collected weekly, pooled across all weeks, and analyzed for nutrient content by a commercial laboratory (Dairy One Forage Laboratory, Ithaca, NY) as in Lippolis et al. (2017). Nutrient profile of TMR is described in Table 1. Full BW was recorded on days 0, 2, 6, 10, 14, 21, 28, 34, 45, and 59. Shrunk BW (after 16 h of water and feed withdrawal) was also collected on day 60 for average daily gain (ADG) calculation, using shrunk BW on day 1 as initial BW. Blood samples were collected from all calves, concurrently with full BW evaluation into commercial blood collection tubes (Vacutainer, 10 mL; Becton Dickinson, Franklin Lakes, NJ) containing no additive or containing freeze-dried sodium heparin for serum and plasma collection, respectively. Intake of TMR (DM basis) was evaluated daily from days 0 to 59 from each pen by collecting and weighing offered and nonconsumed TMR. Daily TMR intake of each pen was divided by the number of calves within each pen and expressed as kg

per calf/day. Total BW gain and TMR intake of each pen were used for feed efficiency (G:F) calculation. Calves were observed daily for BRD signs according to the DART system (Zoetis, Florham Park, NJ) and received antimicrobial treatment if diagnosed with BRD as in Lippolis et al. (2017).

Laboratorial Analysis

After collection, all blood samples were placed immediately on ice, centrifuged ($2,500 \times g$ for 30 min; 4°C) for plasma or serum harvest and stored at -80°C on the same day of collection. Samples collected from days 0 to 28 were analyzed for plasma cortisol (Immulite 1000; Siemens Medical Solutions Diagnostics, Los Angeles, CA), serum nonesterified fatty acids (NEFA; colorimetric kit HR Series NEFA—2; Wako Pure Chemical Industries Ltd. USA, Richmond, VA), and plasma haptoglobin concentrations (Lippolis et al., 2017), given that these responses return to baseline levels in receiving cattle within 4 wk after feedlot entry (Cooke, 2017). Plasma samples collected on days 0, 14, 28, 45, and 59 were analyzed for insulin and insulin-like growth factor (IGF)-I concentrations (Immulite 1000; Siemens Medical Solutions Diagnostics) to metabolically assess calf nutritional status throughout the experimental period (Lippolis et al., 2017). Plasma samples collected from days 0 to 14 were analyzed for plasma tumor necrosis- α (TNF α ; bovine TNF-alpha ELISA kit #ELB-TNF α -1; RayBiotech, Inc., Norcross, GA), as cytokines are expected to return to baseline levels within 2 wk after feedlot entry (Cooke, 2017). The intra-assay and interassay CV were, respectively,

Table 1. Ingredient composition and nutrient profile of TMR offered during the experiment (days 0 to 59)^a

Item	A	B	C	D
Ingredient (% DM basis)				
Grass hay	74.5	58.2	37.0	33.7
Cracked corn	17.5	35.0	54.6	58.2
Soybean meal	7.2	6.0	7.7	7.4
Mineral mix ^b	0.80	0.80	0.70	0.70
Nutrient profile (DM basis)				
Net energy for maintenance (Mcal/kg)	1.38	1.55	1.76	1.80
Net energy for growth (Mcal/kg)	0.80	0.95	1.14	1.17
Neutral detergent fiber, %	46.8	39.3	29.5	27.9
Crude protein, %	13.7	13.1	13.6	13.5

^aA = days 0 to 8; B = days 9 to 19; C = days 20 to 33; and D = days 34 to 59. Calves had free-choice access to the TMR and water throughout the experimental period.

^bCattleman's Choice (Permix Nutrition Systems, Nampa, ID).

1.95% and 8.6% for haptoglobin, 4.3% and 5.2% for NEFA, and 4.7% and 6.3% for TNF α . Plasma cortisol, insulin, and IGF-I concentrations were analyzed within a single assay. The intra-assay CV was, respectively, 4.2% for cortisol, 1.7% for insulin, 0.9% for IGF-I.

Statistical Analysis

Pen was considered the experimental unit for all analyses. Quantitative data were analyzed using the MIXED procedure of SAS (SAS Institute Inc., Cary, NC), whereas binary data were analyzed using the GLIMMIX procedure of SAS (SAS Institute Inc.). All data were analyzed using Satterthwaite approximation to determine the denominator df for tests of fixed effects, with pen (treatment) and calf (pen) as random variables, but for TMR intake and G:F that used pen (treatment) as the random variable. Model statements contained the effects of treatment, in addition to day and the resultant interaction for repeated measures, and calf sex as an independent variable. Blood variables were analyzed using results from day 0 as covariate. The specified term for all repeated statements was day, with pen (treatment) as subject for TMR intake and calf (pen) as subject for all other analyses. The covariance structure used was first-order autoregressive, which provided the smallest Akaike information criterion, and hence, the best fit for all variables analyzed. Results are reported as least square means and separated using PDIF. Significance was set at $P \leq 0.05$ and tendencies at $P > 0.05$ and ≤ 0.10 .

RESULTS

As designed, initial BW (day -1) was similar ($P = 0.97$) among treatments (Table 2). Average daily gain during the experiment was greater ($P = 0.03$) in M2 vs. M1 and CON calves and similar ($P = 0.95$) between M1 and CON calves (Table 2; main treatment effect, $P = 0.05$). However, no treatment effects were detected ($P \geq 0.49$) final shrunk BW (day 60; Table 2) or full BW measurements (data not shown). No treatment effects were detected for TMR intake ($P = 0.52$) during the experiment (Table 2), whereas G:F was greater ($P \leq 0.05$) in M2 vs. M1 and CON calves and similar ($P = 0.40$) between M1 and CON calves (Table 2; main treatment effect, $P = 0.04$).

No treatment effects were detected for concentrations of plasma cortisol, haptoglobin, TNF α , insulin, and IGF-I, as well as serum NEFA (Table 3). Day

Table 2. Performance parameters of beef calves supplemented or not (CON; $n = 7$) with Micro-Aid (DPI Global) at (as-fed basis) 1 g/calf daily (M1; $n = 7$) or 2 g/calf daily (M2; $n = 7$) during feedlot receiving (days 0 to 59)*

Item	CON	M1	M2	SEM	<i>P</i> value
Initial BW (day 1; kg)	220	220	221	4	0.97
Final BW (day 60; kg)	307	307	315	4	0.49
Average daily gain (kg/day)	1.42 ^b	1.42 ^b	1.53 ^a	0.03	0.05
Feed intake (kg/day)	7.16	7.33	7.35	0.13	0.52
Feed efficiency (g/kg)	204 ^b	200 ^b	213 ^a	3	0.04

*Within rows, values with different superscripts differ ($P \leq 0.05$).

effects were detected ($P \leq 0.05$) for all of these variables (Table 4). No treatment effects were detected ($P = 0.39$) for incidence of BRD symptoms (Table 3), which was mainly observed during the initial 21 d of receiving. However, the number of antimicrobial treatments required per calf diagnosed with BRD symptoms to recover from sickness was greater ($P \leq 0.01$) in CON vs. M1 and M2 and similar ($P = 0.60$) between M1 and M2 calves (Table 3). No incidence of mortality was observed during the experiment.

DISCUSSION

Calves utilized in this experiment were considered high risk, given that their prior management and health history were not fully known (Wilson et al., 2017). Moreover, cattle experienced the stress of weaning, auction, transportation, commingling, vaccination, and feedlot entry within a 72-h period, and the combination of these stressors impacts cattle immunocompetence and performance (Cooke, 2017). Accordingly, day effects observed for plasma cortisol, TNF α , and haptoglobin (Table 4) corroborate that calves experienced adrenocortical and acute-phase protein responses elicited by transport, vaccination, and feedlot entry (Lippolis et al., 2017). Collectively, these stress-induced inflammatory processes are linked with the BRD complex in receiving cattle (Cooke, 2017), supporting the substantial incidence of BRD observed in the present experiment (Table 3), which is comparable with research efforts conducted at commercial receiving yards (Snowder et al., 2006).

Micro-Aid supplementation increased calf ADG when included at 2 g/calf daily (M2) but not when included at the lower dose (1 g/calf daily; M1). This outcome should be primarily attributed to increased G:F feed efficiency in M2 cattle, given that TMR intake during the experiment was similar across treatments. Similar concentrations of plasma and serum variables among CON, M1, and

Table 3. Physiological and morbidity parameters from beef calves supplemented or not (CON; $n = 7$) with Micro-Aid (DPI Global) at (as-fed basis) 1 g/calf daily (M1; $n = 7$) or 2 g/calf daily (M2; $n = 7$) during feedlot receiving (days 0 to 59)^a

Item	CON	M1	M2	SEM	<i>P</i> value
Physiological variables ^b					
Plasma cortisol, ng/mL	23.6	21.6	21.7	1.7	0.63
Plasma insulin, pmol/L	29.8	29.1	33.3	3.0	0.55
Plasma IGF-I, ng/mL	159	155	149	7	0.42
Plasma haptoglobin, mg/mL	0.37	0.40	0.37	0.05	0.82
Plasma TNFa, ng/mL	0.14	0.24	0.35	0.13	0.46
Serum NEFA, μ Eq/L	0.25	0.25	0.23	0.02	0.67
Morbidity variables ^c					
Incidence of BRD signs, %	42.9	60.0	54.3	8.8	0.39
Number of antimicrobial treatments required	1.40 ^a	1.05 ^b	1.10 ^b	0.08	0.01
Mortality, %	0.0	0.0	0.0	–	–

^aWithin rows, values with different superscripts differ ($P \leq 0.05$).

^bBlood samples were collected on days 0, 2, 6, 10, 14, 21, 28, 34, 45, and 59 of the experiment. Data were analyzed using results from day 0 as independent covariate.

^cCalves were observed daily for BRD signs according to the DART system (Zoetis) and received antimicrobial treatment as in Lippolis et al. (2017).

M2 (Table 2) indicate that none of the experimental treatments modulated the physiological, metabolic, and acute-phase responses typically associated with feedlot receiving (Cooke, 2017). Therefore, plasma and serum variables evaluated herein failed to elucidate biological mechanisms by which M2 supplementation benefited performance of receiving cattle; perhaps, these occurred without substantial impacts on systemic inflammatory and metabolic responses.

Incidence of BRD was similar among treatments (Table 3) and thus did not contribute to treatment differences reported for ADG and G:F (Schneider et al., 2009). In turn, calves supplemented with Micro-Aid (M1 and M2) and diagnosed with BRD

symptoms required less antimicrobial treatments to recover from sickness compared with nonsupplemented cohorts (Table 3). These outcomes partially support our hypothesis that Micro-Aid supplementation improves cattle immunocompetence during feedlot receiving (Moreau et al., 2002) but also do not fully explain the increased performance of M2 cattle, given that such health benefits were observed for both M1 and M2 treatments. Yet, research has shown that saponins have immunostimulatory properties in mammals, including enhanced antibody and lymphocyte response to antigens (Shi et al., 2004). Therefore, it is plausible to speculate that Micro-Aid supplementation enhanced the ability of cattle to recover from BRD upon antimicrobial administration, although research is still warranted to elucidate the immunological benefits of this feed ingredient to beef cattle.

Alternatively, the M2 treatment may have improved cattle receiving performance by enhancing rumen fermentation and diet utilization. More specifically, McMurphy et al. (2014) reported that Micro-Aid supplementation at 2 g/day to beef steers improved rumen DM and neutral detergent fiber digestibility by decreasing rumen particulate passage rate, given the increased rumen fluid viscosity from the foam-forming characteristics of saponins. These authors also reported that Micro-Aid supplementation increased flow of microbial protein to the small intestine and associated this outcome to increased diet digestibility in the rumen and reduction of ruminal protozoa population that scavenge bacteria. Others have also reported that

Table 4. Concentrations of plasma cortisol (ng/mL), insulin (pmol/L), IGF-I (ng/mL), haptoglobin (mg/mL), TNFa (ng/mL), and NEFA (μ Eq/L) in beef calves during feedlot receiving (days 0 to 59)*

Day	Cortisol	Insulin	IGF-I	Haptoglobin	TNFa	NEFA
0	29.2 ^a	28.8 ^c	82.6 ^d	0.26 ^e	0.12 ^b	0.42 ^a
2	20.3 ^c	–	–	0.37 ^{cd}	0.18 ^b	0.36 ^b
6	20.9 ^c	–	–	0.48 ^{ab}	0.33 ^a	0.31 ^c
10	20.2 ^c	–	–	0.52 ^a	0.22 ^{ab}	0.22 ^d
14	20.3 ^c	26.9 ^c	131 ^c	0.41 ^{bc}	0.14 ^b	0.19 ^c
21	23.8 ^b	–	–	0.31 ^{de}	–	0.19 ^c
28	23.7 ^b	37.6 ^a	162 ^b	0.31 ^{de}	–	0.18 ^c
45	–	30.3 ^{bc}	165 ^b	–	–	–
59	–	34.5 ^{ab}	175 ^a	–	–	–
SEM	1.6	2.7	4.8	0.05	0.07	0.01
<i>P</i> value	0.01	<0.01	<0.01	<0.03	0.05	<0.01

*Within columns, values with different superscripts differ ($P \leq 0.05$).

dietary saponins shift ruminal fermentation toward propionate production due to decreased protozoa numbers (Hristov et al., 1999). Although ruminal parameters were not evaluated herein, published research suggests that G:F was enhanced in M2 cattle due to ruminal benefits of Micro-Aid supplementation at 2 g/calf daily (McMurphy et al., 2014).

IMPLICATIONS

This experimental model fully represented the stress and health challenges that commercial feeder cattle experience during feedlot receiving, resulting in substantial BRD incidence and morbidity. Supplementing Micro-Aid at 2 g/calf daily increased feedlot receiving ADG due to enhanced G:F compared with nonsupplemented cattle, whereas the same outcome was not observed when Micro-Aid was supplemented at 1 g/calf daily. Moreover, supplementing Micro-Aid did not prevent the incidence of BRD, but both doses reduced the need for antimicrobial treatments to heal calves diagnosed with BRD symptoms. Collectively, these results suggest that Micro-Aid should be supplemented at 2 g/calf daily to benefit performance and immunocompetence of feedlot receiving cattle

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