

## Stocking rate and monensin supplemental level effects on growth performance of beef cattle consuming warm-season grasses<sup>1</sup>

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**ABSTRACT:** The objective of this study was to evaluate the effects of monensin supplementation on animals receiving warm-season grass with limited supplementation. In Exp. 1, treatments were a factorial combination of 2 stocking rates (1.2 and 1.7 animal unit [AU] [500 kg BW]/ha) and supplementation with monensin (200 mg/d) or control (no monensin) distributed in a complete randomized design with 3 replicates. Thirty Angus × Brahman crossbred heifers (*Bos taurus* × *Bos indicus*) with initial BW of 343 ± 8 kg were randomly allocated into 12 bahiagrass (*Paspalum notatum*) pastures and supplemented with 0.4 kg DM of concentrate (14% CP and 78% TDN) daily for 86 d. Herbage mass (HM) and nutritive value evaluations were conducted every 14 d, and heifers were weighed every 28 d. There was no effect ( $P \geq 0.97$ ) of monensin on HM, herbage allowance (HA), and ADG; however, animals receiving monensin had greater ( $P = 0.03$ ) plasma urea nitrogen (PUN) concentrations. The stocking rate treatments had similar HM in June ( $P = 0.20$ ) and July ( $P = 0.18$ ), but the higher stocking rate decreased ( $P < 0.01$ ) HM and HA during August and September. Average daily gain was greater ( $P < 0.01$ ) for the pastures with the lower stocking rate in August but not different in July and

September ( $P \geq 0.15$ ). Gain per hectare tended to be greater on pastures with the higher stocking rate ( $P \leq 0.06$ ). In Exp. 2, treatments were 3 levels of monensin (125, 250, and 375 mg/animal per day) and control (no monensin) tested in a 4 × 4 Latin square with a 10-d adaptation period followed by 5 d of rumen fluid collection and total DMI evaluation. Blood samples were collected on d 4 and 5 of the collection period. Ground stargrass (*Cynodon nlemfuensis*) hay (11.0% CP and 52% in vitro digestible organic matter) was offered daily. The steers received the same supplementation regimen as in Exp. 1. Total DMI was not different among treatments ( $P = 0.64$ ). There was a linear increase ( $P \leq 0.01$ ) in propionate and a tendency for decreased acetate ( $P \leq 0.09$ ) concentrations in the rumen with increasing levels of monensin; however, there was no effect ( $P \geq 0.19$ ) of monensin levels on ruminal pH and ruminal concentrations of butyrate and ammonia. In addition, there was no effect ( $P \geq 0.73$ ) of monensin levels on plasma concentrations of glucose, insulin, IGF-1, and PUN. In summary, monensin supplementation effects were not detected at either stocking rate and may not be effective in increasing performance of beef cattle grazing low-quality warm-season grasses with limited supplementation.

**Key words:** beef cattle, monensin, ruminal fermentation parameters, supplementation, warm-season grass

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J. Anim. Sci. 2015.93:3682–3689  
doi:10.2527/jas2015-8913

### INTRODUCTION

Warm-season grasses are the main forages for beef cattle production in tropical and subtropical areas. Due to seasonal variation in herbage mass (HM) and nutritive value, concentrate supplementation is typically used to maintain the productivity of beef cattle grazing warm-season grasses.

<sup>1</sup>Financial support for this study was provided by Elanco Animal Health and Florida Agricultural Experiment Station.

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Received January 13, 2015.

Accepted April 28, 2015.

Ionophores have been shown to induce positive responses in growth and feed efficiency in ruminants receiving diets with high levels of concentrate (Duffield et al., 2012). However, the effectiveness of ionophores for beef cattle grazing warm-season forages in extensive grazing systems with low-quality forage is not consistently reported. Rowe et al. (1991) suggested that in animals grazing low-quality pastures, while maintaining or losing weight, there is less likelihood of a positive response to ionophores, which may be attributed to a reduction in forage DMI (Ellis et al., 1984). Stocking rate is the most important decision in grazing management because it affects herbage allowance (HA), intake, and animal performance (Sollenberger et al., 2012); however, the effects of monensin on the performance of cattle grazing low-quality warm-season grasses at different stocking rates are not known.

Increasing levels of monensin effectively enhanced the performance of feedlot beef cattle receiving diets with greater levels of concentrate (Duffield et al., 2012). However, there is limited information on the effects of different levels of monensin on beef cattle consuming warm-season grasses with a limited amount of concentrate supplementation.

This study tested the hypothesis that heifers grazing low-quality warm-season pastures with a lower stocking rate and greater HA may respond positively to monensin supplementation compared with animals on pastures with a higher stocking rate. In addition, the effect of increasing levels of monensin on animal performance was tested. The objectives of this study were 1) to test the effects of stocking rates on the performance of heifers receiving monensin and 2) to determine the effects of increasing levels of monensin supplementation on ruminal and blood parameters when cattle were consuming low-quality forage.

## MATERIALS AND METHODS

The experiments were conducted at the University of Florida Range Cattle Research and Education Center, Ona, FL (27°26' N, 82°55' W) from June to September 2012 (yr 1) and 2013 (yr 2; Exp. 1) and from June to September 2013 (Exp. 2). The experimental procedures were approved by the University of Florida Institute of Food and Agriculture Sciences Animal Research Committee.

### Experiment 1

The soil at the research site was classified as Pomona fine sand (siliceous, hyperthermic, Ultic Alaquod). Before the initiation of the study, mean soil pH (in water) was 5.6 and Mehlich-I (0.05 M HCl + 0.0125 M H<sub>2</sub>SO<sub>4</sub>)

extractable P, K, Mg, and Ca concentrations in the Ap1 horizon (0 to 15 cm depth) were 25, 67, 190, and 1,230 mg/kg, respectively. 'Pensacola' bahiagrass (*Paspalum notatum*) pastures were established in 1998 and fertilized with 56 kg N/ha in May 2012 and 2013. Before the initiation of the study, the pastures were grazed with a stocking rate of 1 animal unit [AU]/ha.

Thirty Angus × Brahman crossbred heifers (*Bos taurus* × *Bos indicus*) with initial BW of 345 ± 7 kg in yr 1 and 341 ± 9 kg in yr 2 were randomly distributed in 12 bahiagrass pastures (*n* = 4 pastures/treatment and 1.2 ha/pasture). Pasture was used as the experimental unit, and 2 or 3 heifers were allocated per pasture to achieve the proposed stocking rate treatments. Pastures were stocked continuously using a fixed stocking rate.

Treatments were the factorial combination of 2 stocking rates (1.2 or 1.7 AU [500 kg BW]/ha) and supplementation with monensin (200 mg/d) or control (no monensin) distributed in a complete randomized design with 3 replicates. The stocking rates were selected according to Obour et al. (2011), who observed that 1.2 AU/ha resulted in adequate HM for heifers grazing bahiagrass in the same location. Conversely, the higher stocking rate treatment was selected with the objective to create a condition of decreased HM with limited forage quantity. Rumensin 90 (Elanco Animal Health, Greenfield, IN) was the commercial product used as a source of monensin and the proposed level of monensin was added daily to the concentrate at the time of feeding (0800 h). The heifers received 0.4 kg DM of a concentrate supplement daily for 86 d. The composition of the concentrate is presented in Table 1. Heifers had ad libitum access to a complete salt-based trace mineral mix (guaranteed analysis: 14% Ca, 9% P, 24% NaCl, 0.20% K, 0.30% Mg, 0.20% S, 0.005% Co, 0.15% Cu, 0.02% I, 0.05% Mn, 0.004% Se, 0.3% Zn, 0.08% F, and 82 IU/g of vitamin A; Lakeland Animal Nutrition, Lakeland, FL).

**Pasture Sampling.** Pastures were sampled for HM and nutritive value (CP and in vitro digestible organic matter [IVDOM]) just before initiation of grazing and every 14 d during the experimental periods, from June 18 to September 12, 2012, and from June 30 to September 24, 2013 (86 d). The double sampling technique was used to determine HM according to Gonzalez et al. (1990). The indirect measure was the settling height of a 0.25-m<sup>2</sup> aluminum disk, and the direct measure involved hand clipping all herbage to 2.5 cm above soil level using an electric clipper. Every 28 d, 2 double samples were taken from each of the 12 experimental units for a total of 24 double samples per date. Sites for double sampling were chosen to represent the range of HM present on the pastures. At each site, the disk settling height was determined

**Table 1.** Nutrient composition of the concentrate used in yr 1 and yr 2 (DM basis)<sup>1</sup>

Item	Concentrate
CP, %	14.3
ADF, %	11.2
NDF, %	22.8
TDN, <sup>2</sup> %	78.1
Ca, %	0.31
P, %	0.69
Mg, %	0.34
K, %	1.47
Fe, mg/kg	249
Zn, mg/kg	67
Cu, mg/kg	34
Mn, mg/kg	61

<sup>1</sup>Concentrate consisted of soybean hulls (40%), wheat middlings (30%), cottonseed meal (12%), cottonseed hulls (10%), molasses (6.25%), calcium carbonate (1.25%), and vitamin/mineral premix (0.50%). Rumensin 90 (Elanco Animal Health, Greenfield, IN) was the commercial product used as a source of monensin and 500 mg/kg was added daily to the concentrate at the time of feeding (0800 h).

<sup>2</sup>TDN was calculated as described by Weiss et al. (1992).

and the forage under the disk was clipped at ground level. Clipped forage was dried at 60°C for 72 h in a forced-air oven and weighed. Indirect measures (disk heights) were taken every 14 d at 20 sites per pasture. Sites were selected by walking a fixed number of steps between each drop of the disk to ensure that all sections of the pasture were represented. The average disk height of the 20 indirect measures was entered into the regression equation developed from double sampling to predict HM. The average  $r^2$  values for the equations were 0.83 and 0.92 for yr 1 and 2, respectively. Herbage allowance was calculated for each pasture as the average HM (mean across 2 sampling dates within each 28-d period) divided by the average total heifer BW during that period (Sollenberger et al., 2005).

Twenty hand-plucked samples were randomly taken at the average forage height from each pasture. Herbage was composited across sites, dried at 60°C for 48 h in a forced-air oven to constant weight, and ground to pass a 1-mm stainless steel screen in a Wiley mill (model 4 Thomas-Wiley Laboratory Mill; Thomas Scientific, Swedesboro, NJ). Samples were analyzed for IVDOM using the 2-stage technique described by Tilley and Terry (1963) and modified by Moore and Mott (1974). Nitrogen concentration was determined using a micro-Kjeldahl method, a modification of the aluminum block digestion technique described by Gallaher et al. (1975). Crude protein was determined by multiplying N concentration by 6.25.

**Animal Measurements.** Heifers were weighed on d 0 and then every 28 d. Individual BW was taken at 0800 h, following a 16-h period of feed and water

deprivation. The ADG was calculated as the difference between the initial and final BW divided by 28 d. Gain per hectare was calculated as the BW gain of the animals within the pastures adjusted to a hectare basis during the entire experimental period (86 d).

Blood samples were collected from the jugular vein at the termination of the experimental period. Samples were collected into 9-mL, Na-heparinized syringes (Luer Monovette; Sarstedt, Inc., Newton, NC) and immediately placed on ice. Blood was centrifuged ( $2,000 \times g$  for 30 min at 4°C) and plasma was harvested and kept frozen at -80°C until further analysis. Plasma concentrations of insulin were determined using Coat-A-Count solid phase 125I RIA kits (Siemens Healthcare Diagnostics, Los Angeles, CA) previously validated for bovine samples (Moriel et al., 2008). Plasma concentrations of glucose and plasma urea nitrogen (PUN) were determined using quantitative colorimetric kits (numbers G7521 and B7551, respectively; Pointe Scientific Inc., Canton, MI). Concentrations of IGF-1 were determined using a human-specific commercial ELISA kit (SG100; R&D Systems Inc., Minneapolis, MN) with 100% cross-reactivity with bovine IGF-1 and previously validated for bovine samples (Cooke et al., 2012). The intra- and interassay CV were 2.1 and 5.5% for insulin, 1.7 and 1.9% for glucose, 4.4 and 5.9% for PUN, and 7.2 and 8.5% for IGF-1. The minimum detectable concentration was 0.01  $\mu$ IU/mL for insulin and 0.056 ng/mL for IGF-1.

**Drylot Measurements.** On d 86 of yr 1, heifers ( $n = 16$ ) were allocated into 1 of 8 drylot pens to evaluate the effects of the monensin supplementation on voluntary forage and total DMI for 17 d. The heifers were randomly selected from the low stocking rate treatment from the grazing study and maintained in their respective monensin treatment. Heifers received stargrass (*Cynodon nlemfuensis*) hay (11% CP and 52% IVDOM) at amounts to ensure 10% DM refusals. The hay was processed through a hay chopper (Balebuster 2100; Haybuster, Jamestown, ND) to an approximately 5-cm particle size and offered 4 times daily. The heifers received 0.4 kg DM of the same concentrate described for the grazing portion of the experiment daily (Table 1) and had libitum access to a salt-based trace mineral mix (described previously) for 17 d.

Initial BW of heifers was recorded on d 0 after 16 h of feed restriction. The adaptation period was from d 1 to 10, and samples of the forage offered and refused were collected daily from each pen from d 11 to 17 and then dried at 60°C for 96 h for DM calculations. Daily DMI was determined for each pen.

**Statistical Analysis.** Data were analyzed using PROC MIXED of SAS (SAS Inst. Inc., Cary, NC) with pasture as experimental unit and the Satterthwaite approximation used to determine the denominator degrees

of freedom for the tests of fixed effects. Monensin, stocking rate, month, and their interactions were considered fixed effects. Months were analyzed as repeated measures using the unstructured covariance structure. The model statement used for gain per hectare and plasma variables contained the fixed effects of the monensin in the diet and the stocking rate. Replicates, year, and their interactions were considered random effects. For the drylot period (yr 1 only), the model statement contained only the effect of monensin. Treatments were considered different when  $P < 0.05$  and tendencies when  $0.05 \geq P < 0.10$ . The means reported were least squares means and was separated using PDIFF (SAS Inst. Inc.).

### Experiment 2

Four ruminally fistulated steers of approximately 24 mo of age (approximately  $392 \pm 12$  kg BW) were used in a  $4 \times 4$  Latin square design. Treatments were 4 levels of monensin (0, 125, 250, or 375 mg monensin/d) added to a daily concentrate supplement fed at 0.2% BW (Table 1). Considering a voluntary DMI of 2.2% BW, these monensin levels were designed to supply the equivalent of 0, 10, 20, and 30 mg/kg per animal per day and create a wide range of doses, including the minimum and maximum doses recommended for grazing beef cattle, 50 and 200 mg/d, respectively (Elanco Animal Health, 2014).

Steers received stargrass hay daily with a target 10% refusal. The hayfield was fertilized with 80 kg N/ha in April 2013 and harvested with a 5-wk regrowth interval. The nutritive value of the hay was 11% CP and 52% TDN. The hay was processed through a hay chopper (Balebuster 2100; Haybuster) to an approximately 5-cm particle size. Four consecutive 15-d periods were used, each consisting of 10 d for adaptation followed by a 5-d collection period for forage DMI evaluation and sampling of blood and rumen fluid.

**Animal Measurements.** Rumen fluid and blood samples were collected every 8 h from d 10 to 15 of each period. Blood samples were collected and analyzed with the same procedures described in Exp. 1. Rumen fluid was collected (50 mL) and filtered through 4 layers of cheesecloth into a 200-mL plastic container and pH was measured (Orion pH meter [model 330] Perpfect LOGR; Orion Research, Boston, MA). Rumen fluid was then transferred into a plastic container and 0.5 mL of a 20% sulfuric acid solution was added. The container was placed in ice and then stored at  $-20^{\circ}\text{C}$  until further analysis. Before the analysis for VFA and ammonia, rumen fluid was transferred to a plastic container and centrifuged (Avanti JE centrifuge with JA-20 rotor; Beckman Coulter, Inc., Fullerton, CA) for 15 min

at  $10,000 \times g$  at  $10^{\circ}\text{C}$ , and 3 mL of the solution was transferred to a container. Rumen fluid was analyzed for VFA using an Agilent 7820A Gas Chromatograph (Agilent Technologies, Palo Alto, CA; 2.5 m by 0.32 mm by 0.45 mm glass column). Analysis for rumen ammonia was performed according to Broderick and Kang (1980) and quantified using a spectrophotometer (Beckman Coulter AD340 microplate reader; Beckman Coulter, Fullerton, CA) at 630 nm.

**Statistical Analysis.** Data were analyzed using PROC MIXED of SAS (SAS Inst. Inc.) using steers as the experimental unit and the Satterthwaite approximation to determine the denominator degrees of freedom for the tests of fixed effects. Treatment, time of collections, and the interaction were considered fixed effects. Time was analyzed as a repeated measure using the unstructured covariance structure. Steer was considered the random variable. Single degree of freedom orthogonal polynomial contrasts were used to test the effects of equally spaced increasing levels of monensin. Treatments were considered different when  $P < 0.05$  and tendencies when  $0.05 \leq P < 0.10$ . The means reported were least squares means and was compared using PDIFF (SAS Inst. Inc.).

## RESULTS AND DISCUSSION

### Experiment 1

There was no effect of monensin, monensin  $\times$  stocking rate, monensin  $\times$  month, or monensin  $\times$  stocking rate  $\times$  month interactions on ADG (mean =  $0.4 \pm 0.1$  kg/d;  $P \geq 0.23$ ) and HA (mean =  $1.6 \pm 0.1$  kg DM/kg BW;  $P \geq 0.38$ ). However, there was a stocking rate  $\times$  month interaction ( $P \leq 0.04$ ) for ADG and HA (Table 2). Average daily gain was not different among treatments in July and September; however, the higher stocking rate decreased ( $P \leq 0.01$ ) ADG in August due to limited HA. Herbage allowance was not different ( $P = 0.26$ ) between stocking rate treatments in June, but the higher stocking rates had reduced ( $P \leq 0.02$ ) HA in July, August, and September. Gain per hectare was not different among monensin treatments (mean =  $75 \pm 5$  kg;  $P = 0.29$ ) and tended to be greater on the higher stocking rate treatment ( $88$  vs.  $60 \pm 5$  kg;  $P \leq 0.06$ ). The higher stocking rate offset the effect of decreased ( $P \leq 0.01$ ) ADG in August and resulted in greater total BW gain during the experimental period. There was no monensin  $\times$  stocking rate effects ( $P = 0.87$ ) on gain per hectare.

Heifers receiving monensin treatment had greater PUN concentrations than controls ( $P = 0.03$ ; Table 3); however, there was no effect of monensin  $\times$  stocking rate, monensin  $\times$  month, or monensin  $\times$  stocking rate  $\times$  month interactions ( $P \geq 0.32$ ) on plasma concentrations

**Table 2.** Herbage mass and allowance, and average daily gain of heifers grazed on bahiagrass pastures at different stocking rates (1.2 vs. 1.7 animal unit [AU]/ha; Exp. 1)

Response variable/ stocking rate	Months				SE
	June	July	August	September	
Herbage mass, kg/ha					
1.2 AU/ha	1,600 <sup>b</sup>	1,700 <sup>b</sup>	2,600 <sup>a</sup>	2,700 <sup>a</sup>	300
1.7 AU/ha	1,490 <sup>b</sup>	1,530 <sup>b</sup>	2,080 <sup>a</sup>	2,090 <sup>a</sup>	
<i>P</i> -value <sup>1</sup>	0.20	0.18	<0.01	<0.01	
SE	450				
Herbage allowance, kg DM/kg BW					
1.2 AU/ha	1.1 <sup>b</sup>	1.3 <sup>b</sup>	2.3 <sup>a</sup>	2.3 <sup>a</sup>	0.1
1.7 AU/ha	0.9 <sup>b</sup>	1.0 <sup>b</sup>	1.3 <sup>a</sup>	1.4 <sup>a</sup>	
<i>P</i> -value <sup>1</sup>	0.26	0.02	<0.01	<0.01	
SE	0.1				
ADG, kg/d					
1.2 AU/ha		0.3 <sup>b</sup>	0.6 <sup>a</sup>	0.6 <sup>a</sup>	0.1
1.7 AU/ha		0.1 <sup>b</sup>	0.3 <sup>b</sup>	0.6 <sup>a</sup>	
<i>P</i> -value <sup>1</sup>		0.15	<0.01	0.85	
SE		0.1			

<sup>a,b</sup>Within a row, means without a common superscript differ ( $P \leq 0.05$ ).

<sup>1</sup>*P*-value for the comparison of means between stocking rate treatments within month.

of glucose, insulin, and IGF-1. Plasma concentrations of PUN (mean = 18.3 ± 1.2 mg/dL), glucose (mean = 74.5 ± 6.3 mg/dL), insulin (mean = 7.3 ± 0.6 μIU/mL), and IGF-1 (mean = 68.2 ± 11.3 ng/mL) were not affected by stocking rate nor was there a stocking rate × month interaction ( $P \geq 0.36$ ). The blood samples were collected at the termination of the experimental period (d 86) and may not have captured the potential differences in forage intake and blood metabolites, which resulted in greater ADG for the 1.2 AU/ha treatment in August.

According to Bergen and Bates (1984), monensin acts on the flux of ions through the membranes dissipating cation and proton gradients and interfering with the update of solutes and the primary transport system in the cells. This process selects gram-negative bacteria in the rumen and potentially increases the proportion of propionic acid. Rouquette et al. (1980) compared the effects of monensin on beef calves grazing bermudagrass (*Cynodon dactylon*) and receiving 0.9 kg/d of concentrate. Calves receiving 200 mg/d of monensin had greater ADG (0.54 vs. 0.40 kg/d) than calves receiving only concentrate. It seems that monensin can improve the efficiency of use of greater levels of concentrate supplements to cattle grazing warm-season grasses; however, the predominant low-quality forage diet and limited supply of concentrate in the current study (0.4 kg/d of DM) may have limited the substrate for propionate formation in the rumen and decreased the potential benefit of monensin on ADG. In addition, monensin may also have the potential to decrease methanogenesis and increase fermentation efficiency in the

**Table 3.** Plasma concentrations of plasma urea nitrogen (PUN), glucose, insulin, and IGF-1 in heifers grazing bahiagrass pastures and receiving monensin supplementation (200 mg/d) or not (Control) for 86 d (Exp. 1)

Response variable	Treatment		SE	<i>P</i> -value
	Control	Monensin		
PUN, mg/dL	16.5	20.2	1.2	0.03
Glucose, mg/dL	76.0	73.8	6.9	0.56
Insulin, μIU/mL	7.3	7.9	0.6	0.45
IGF-1, ng/mL	65.1	71.8	11.1	0.49

rumen (Russell, 1987); however, this potential benefit did not translate into greater ADG in this study. Rowe et al. (1991) suggested that there is less likelihood of a positive response to ionophore in animals grazing low-quality pastures. According to the authors, low-quality forages have decreased concentration of nonstructural carbohydrates and limited intake, which may decrease the precursors of propionate in the rumen. Therefore, the limited nutritive value of the forage may have also decreased the potential benefit of monensin. Due to the lack of stocking rate × monensin interaction effects on ADG, it is expected that forage quantity may not alter the effect of monensin treatment on performance of beef heifers grazing low-quality warm-season grass pastures with limited concentrate supplementation.

Inyang et al. (2010) and Hernández Garay (2004) observed a similar decrease in animal performance with increasing stocking rates on beef cattle grazing warm-season grasses. According to Inyang et al. (2010), HA below 1.4 kg DM/kg of BW may decrease growth performance of heifers grazing bahiagrass pastures. In this study, HA was above or equal to 1.4 kg DM/kg BW in July and September for both stocking rate treatments.

The greater PUN concentration in heifers receiving monensin may be due to improved utilization of N (Muntiferi et al., 1980; Poos et al., 1979) associated with decreased proteolysis of dietary protein and altered site of protein digestion (Poos et al., 1979). According to Hammond (1997), the optimal levels of PUN concentration in growing heifers range from 15 to 19 mg/dL and the PUN concentrations of the heifers in all treatments were within the adequate range in this trial. The increase in plasma concentrations of glucose and IGF-1 would be expected with greater DMI and energy density in the diet (Cappelozza et al., 2014a,b) or greater efficiency in propionate production in the rumen (Harmon et al., 1993); however, the monensin treatment in the predominantly forage-based diet used in this trial may not have resulted in differences in propionate production or energy intake. Similar to this study, Harmon et al. (1993) observed that 240 mg/d of monensin did not increase propionate and blood glucose concentration of steers receiving alfalfa

**Table 4.** Crude protein and in vitro digestible organic matter (IVDOM) concentrations in hand-plucked forage from bahiagrass pastures grazed by beef heifers for 86 d during the summers of 2012 and 2013. The trial began on June 18 and 30 and ended on September 12 and 24 for 2012 and 2013, respectively

Response variable	Months				SE
	June	July	August	September	
CP, %	11.6 <sup>a</sup>	7.4 <sup>b</sup>	7.6 <sup>b</sup>	6.8 <sup>c</sup>	0.3
IVDOM, %	56 <sup>a</sup>	49 <sup>b</sup>	47 <sup>b</sup>	43 <sup>c</sup>	1

<sup>a-c</sup>Within a row, means without a common superscript differ ( $P \leq 0.05$ ).

(*Medicago sativa*) hay. In a meta-analysis to evaluate the effects of monensin on blood metabolites of dairy cows, Duffield et al. (2008) observed that monensin had a positive effect on glucose and PUN and had no effect on insulin concentrations; however, the data were collected from cows receiving levels of concentrate greater than the one used in the present study.

Monensin, monensin  $\times$  stocking rate, monensin  $\times$  month, or monensin  $\times$  stocking rate  $\times$  month interactions did not affect HM (mean = 2,000  $\pm$  300 kg/ha;  $P = 0.37$ ), herbage CP (mean = 8.3  $\pm$  0.2%;  $P = 0.27$ ), and IVDOM (mean = 49  $\pm$  0.7%;  $P = 0.20$ ); however, CP and IVDOM decreased ( $P = 0.001$ ) from June to September (Table 4). There was a stocking rate  $\times$  month interaction for HM ( $P \leq 0.01$ ; Table 2). The interaction occurred because HM was not different ( $P \geq 0.18$ ) during June and July, but pastures with the higher stocking rate had reduced ( $P \leq 0.01$ ) HM in August and September.

The reductions in CP and IVDOM concentrations during the summer months are likely due to higher temperatures resulting in rapid growth and increased deposition of lignin, which reduces digestibility (Mislevy et al., 2001; Ezenwa et al., 2006). In June and July, treatments were recently imposed on the pastures and the forage growth was likely decreased due to climatic conditions. Conversely, the pastures had greater forage growth in August and September, which likely increased the differences between treatments and the overall HM from June to September in both treatments. The increase in HM throughout the summer in Florida occurs because of the increasing rainfall and temperature as previously reported (Obour et al., 2011; Vendramini et al., 2013).

In the drylot period, forage DMI (mean = 2.0  $\pm$  0.05% BW;  $P = 0.61$ ) or total DMI (mean = 2.1  $\pm$  0.06%;  $P = 0.65$ ) did not differ between monensin treatments. It has been observed that monensin may have decreased total DMI in diets with greater proportion of concentrate (Poos et al., 1979) due to changes in rumen fermentation (Muntiferer et al., 1980) and slower liquid and solid turnover rates (Lemenager et al., 1978);

however, limited and inconsistent information is available on ruminants receiving diets with predominantly warm-season forages. Vagnoni et al. (1995) observed no difference in DMI on steers receiving bermudagrass hay. The changes in rumen fermentation and VFA proportion may have been minimized due to the lack of substrate for propionate production in these animals receiving predominantly warm-season forages with limited concentrate supplementation. The lack of response in forage intake appears to support the similar HM and HA found in the grazing portion of the trial.

## Experiment 2

There was no difference in forage DMI (mean = 1.9  $\pm$  0.5% BW;  $P = 0.93$ ) among treatments, which substantiates the DMI intake data from Exp. 1. Based on the forage DMI, the monensin treatments were the equivalent of 0, 14.5, 29, and 43.5 mg/kg DMI. There was a linear ( $P \leq 0.01$ ) increase in propionate and a tendency ( $P \leq 0.09$ ) for a decrease in acetate in the rumen with increasing levels of monensin (Table 5); however, the magnitude of the change in the propionate:acetate acid ratio was not sufficient to alter rumen pH (Table 5) and impact DMI. The concentrations of butyrate were not different ( $P \geq 0.72$ ) among treatments (Table 5).

In spite of the increase in ruminal propionate concentrations with greater monensin levels, there was no effect of levels of monensin on blood glucose (mean = 62.0  $\pm$  3.8 mg/dL;  $P = 0.72$ ), insulin (mean = 3.3  $\pm$  1.2  $\mu$ IU/mL;  $P = 0.84$ ), or IGF-1 (mean = 80.7  $\pm$  12.8;  $P = 0.65$ ). Levels of monensin did not affect ruminal ammonia-N concentrations ( $P \geq 0.17$ ) or blood PUN concentrations (mean = 25.0  $\pm$  2.4 mg/dL;  $P = 0.86$ ).

The increasing levels of monensin may have caused a change in microbial populations and the fermentation profile in the rumen (Bergen and Bates, 1984), thus optimizing propionate formation.

Harmon et al. (1993) observed that steers receiving alfalfa hay had increased propionic acid concentration in the rumen but similar arterial concentrations of glucose. The increased propionate in the rumen may have been metabolized by gut tissues, thereby sparing substrate for glucose synthesis. Increasing ruminal propionate may then result in greater metabolism of propionate by rumen epithelial and a decrease in blood glucose (Harmon et al., 1993). A concomitant increase in insulin and IGF-I would be expected with greater energy intake (Cappelozza et al., 2014b); however, it was not observed in this study likely due to the similar DMI observed at different levels of monensin supplementation.

The ammonia-N concentrations found in this study are similar to the 6 mM threshold proposed by Kang-Meznarich and Broderick (1981) as adequate to maxi-

**Table 5.** Effects of supplemental levels of monensin on ruminal fermentation parameters of steers receiving star-grass (*Cynodon nlemfuensis*) hay and supplemented with 0.4 kg/d of concentrate (Exp. 2)

Response variable	Levels of monensin, mg/kg				Orthogonal contrast			SE
	0	10	20	30	Linear	Quadratic	Cubic	
Ruminal pH	6.6	6.6	6.7	6.5	0.41	0.19	0.33	0.07
Propionate, mol/100 mol	16.9	17.9	19.1	19.4	0.004	0.56	0.64	0.5
Acetate, mol/100mol	74.0	73.1	71.3	71.1	0.09	0.91	0.82	1
Butyrate, mol/100 mol	8.7	8.4	8.3	8.5	0.82	0.72	0.98	0.7
Acetate:propionate	4.3	4.0	3.7	3.6	0.001	0.19	0.65	0.2
Ammonia-N, mg/100 ml	7.3	6.1	6.4	7.3	0.86	0.17	0.79	0.7

mize ruminal microbial growth. The lack of differences in PUN concentrations is in agreement with the similar ruminal ammonia-N concentrations among treatments. However, it differs from the findings in Exp. 1, in which heifers receiving monensin had greater PUN than in the control. The discrepancy between Exp. 1 and 2 may be related to differences in forage CP concentrations and animal category (growing heifers vs. mature steers).

### Overall Conclusions

Under the conditions of this study, monensin had a limited effect on growing heifers grazing low-quality warm-season grass with low levels of supplementation. Increasing levels of monensin supplementation may increase rumen propionate and decrease acetate; however, the magnitude of the change in fermentation profile may not be sufficient to improve performance of animals receiving low-quality warm-season forages with limited supplementation.

Stocking rates that result in HA below 1.4 kg DM/kg BW should be avoided if the primary goal is to optimize ADG. Greater stocking rates may result in increasing gain per hectare; however, only 2 stocking rates were tested in this study and greater levels of stocking rate would be necessary to identify the stocking rate for optimum ADG and gain per hectare.

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