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Cattle Temperament Alters the Metabolic Response to a Feed Restriction Challenge in Beef Steers

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Abstract

Recent studies have demonstrated metabolic differences between calm and temperamental cattle. Specifically, temperamental cattle exhibit greater concentrations of NEFAs, decreased blood urea nitrogen (BUN), and decreased insulin sensitivity compared to Calm cattle. It is hypothesized that these differences may influence the manner in which Temperamental cattle respond to feed restriction (FR). Therefore, the objective of this study was to determine whether cattle temperament would influence the metabolic responses to a FR challenge in beef cattle. Angus-cross steers (16 Calm and 15 Temperamental; 216 ± 6 kg BW) were selected based on Temperament Score measured at weaning. On d 1 of the study, steers were moved indoors into individual stanchions to allow measurement of individual feed intake. Feed and water was provided *ad libitum* from d 1- 4 in order to determine *ad libitum* feed intake. On d 6, steers were fitted with indwelling rectal temperature probes and jugular catheters, and were returned to individual stalls. Beginning at 0800 h on d 8, feed was removed for a 72-h period (d 8 - 10). Feed was then provided at 25%, 50%, 75%, and 100% of *ad libitum* on d 11, 12, 13, and 14, respectively. Blood samples were collected every 6 h from 0 to 156 h during the FR challenge. Serum was isolated and analyzed for cortisol, glucose, insulin, NEFA, and BUN concentrations. All variables changed over time ($P < 0.01$). For the duration of the study, Temperamental steers maintained greater ($P < 0.01$) serum NEFA and less ($P < 0.01$) serum BUN and insulin sensitivity (calculated using RQUICKI) compared to Calm steers. Additionally, Temperamental steers maintained greater ($P = 0.001$) serum glucose and less serum ($P = 0.001$) insulin than Calm steers. These data suggest that Temperamental and Calm cattle have metabolically different responses to FR, and further implicate metabolic differences as the primary factor associated with differences observed in immune function and performance traits between temperamental and calm cattle. These differences accentuate the need for different management strategies for feeding temperamental versus calm cattle.

Keywords: Cattle; Feed Restriction; Glucose; Insulin; Insulin Sensitivity; Temperament

Introduction

Variability in metabolism are common within mammals, including humans, rodents, and cattle and can result from innate or natural (e.g., genetic or epigenetic origin) and (or) imposed (e.g., diet) factors. For example, obesity in humans and rodents typically results in insulin resistance and intolerance to glucose, among other symptoms [1,2]. Additionally, the ob/ob mutant mouse, which lacks the leptin receptor, leads to obesity, insulin resistance and infertility [3]. Differences in metabolism are also observed between different breeds of cattle [4].

While inherent differences exist within cattle for a plethora of reasons, it has been well-established that temperamental cattle are behaviorally, physiologically, and immunologically different than calm cattle. Recently, the dramatic differences in basal metabolism in cattle of differing temperaments

were reported [5]. Specifically, temperamental cattle exhibit greater basal circulating concentrations of non-esterified fatty acids (NEFA) compared to Calm cattle [5]. Furthermore, in response to lipopolysaccharide (LPS), temperamental cattle exhibited reduced glucose and insulin responses and reduced blood urea nitrogen (BUN) response [5]. Further study of the effect of temperament on metabolic responses has included glucose tolerance and insulin sensitivity tests, as described in a separate manuscript [6]. These studies have indicated altered glucose and insulin dynamics in temperamental steers compared to calm steers, including reduced insulin sensitivity.

Inherent metabolic differences exhibited between calm and temperamental cattle may be a foundation on which to elucidate other physiological and performance differences associated with temperament. For example, temperamental cattle have been associated with reduced average daily gain, decreased body condition scores, and decreased hot carcass weights [7-10]. Also, a decrease in overall 12th rib fat thickness and a decrease in tenderness have been reported in temperamental cattle [10-12]. Based on these data, understanding the mechanisms behind the altered metabolism and energy utilization observed in temperamental cattle will allow for development or modification of protocols aimed at mediating the negative effects of temperament on performance. Therefore, the current study was designed to determine the effect of temperament on the metabolic responses to a feed restriction challenge and gradual feed re-introduction in steers.

Materials and Methods

All experimental procedures were in compliance with the *Guide for the Care and Use of Agricultural Animals in Research and Teaching* and approved by the Institutional Animal Care and Use Committee of the Livestock Issues Research Unit.

Experimental Design

In March 2014, Angus-cross steers (216 ± 6 kg BW), 16 Calm and 15 Temperamental, were selected from a herd of 250 cattle at the Mississippi Agriculture and Forestry Experiment Station - Brown Loam in Raymond, MS. Steers were selected based on Temperament Score (TS) measured at weaning (see description below). Steers were weaned in November 2013. Based on TS, 16 steers with the lowest TS and classified as Calm (TS: 2.00 ± 0.15; EV: 1.92 ± 0.16 m/s; PS: 2.19 ± 0.19), and 15 steers with the greatest TS and classified as Temperamental (TS: 3.94 ± 0.09; EV: 4.21 ± 0.21 m/s; PS: 3.67 ± 0.16) were selected. On d 0, steers were transported from Raymond, MS to Lubbock, TX (approximately 1184 km, 12 h). Steers were group housed in outdoor pens (7.6 x 18.3 m) overnight with access to feed and water. The steers were supplied a feed in Lubbock that was similar to the feed provided in Mississippi. The following day steers were moved into individual stanchions (2.28 m in length, 0.76 m in width, 1.67 m in height) to allow for measurement of individual *ad libitum* feed intake for 4 d. Steers diet was as follows: 60% certified timothy grass pellets (8% CP, 1.5% crude fat, and 35% crude fiber) and 40% concentrate (12% CP, 3% crude fat, 18% crude fiber). Steers had *ad libitum* access to water throughout the duration of the trial. Based upon the ort, daily allotment of feed delivered was formulated to meet *ad libitum* intake. Thus, steers were provided with an average of 5.23 kg of feed daily on d 1, 2, 3 and 4, respectively. On d 5, all orts from d 1 – 4 were weighed, and this was used to determine amount of feed consumed over d 1 – 4, and thus daily feed intake for each steer. Feed was provided daily at 0800 h. On the afternoon of d 5, steers were moved from the individual stanchions to the outdoor pens with *ad libitum* access to the same feed and water. On d 6, steers were weighed, fitted with indwelling jugular catheters and rectal temperature recording devices [13] that measured rectal temperature continuously throughout the study at 5-min intervals. The steers were then returned to the individual stanchions, where they remained for the remainder of the study, and fed. On d 7 at 0600 h, orts were removed from feed bunks and steers were administered a glucose tolerance test at 0900 h and an insulin sensitivity test at 1400 h [6]. Following the collection of the last blood sample from this challenge at 1630 h, steers were given their daily allotment of feed.

At 0800h on d 8, feed was removed for a 72-h period (d 8 - 10). Feed was then supplied at 25%, 50%, 75%, and 100% of *ad libitum* feed intake (based upon intake from d 1 to 5) on d 11, 12, 13, and 14, respectively. On d 11 - 14, the daily allotment of feed was provided following blood sample collection at 0800 h. Whole blood samples were collected into Sarstedt tubes containing no additive (Sarstedt, Inc., Newton, NC) at 6-h intervals from 0 to 156 h relative to feed removal (0 h). Specifically, for blood sample collection, a 3-mL waste sample was collected from the catheter extension (approximately 1.5 m in length, approximately 2 mL in volume), followed by collection of the blood sample. Next, 10 mL of sterile saline was administered to replace fluid loss, and 2.5 mL of heparinized saline (10 USP/mL) was administered to maintain catheter patency. The entire sample collection procedure was completed within 2-5 min. Whole blood samples were allowed to clot at room temperature for 30 min and were then centrifuged at 1500 g for 20 min at 4°C. Isolated serum was stored at -80°C until analyzed for cortisol, glucose, insulin, NEFA and BUN. After collection of the 156 h sample on d 14, steers were weighed, catheters and temperature monitoring devices were removed, and steers were allowed to rest overnight prior to transport back to Mississippi.

Temperament Measurements

Temperament score was calculated as an average of exit velocity and pen score measured at weaning. Exit velocity is an objective measurement that represents the rate of speed

of a calf traversing a distance of 1.83 m after its exit from a working chute [14,15]. This was determined using two infrared sensors (FarmTek Inc., North Wylie, TX, USA) and was done by calculating velocity [velocity = distance/time; expressed as m/s]. Pen score [16] is a subjective measurement obtained by separating cattle into small groups of three to five and scoring their reactivity to a human observer. In brief, a human observer approached each calf and assigned a score between 1 and 5 based on the calf's reaction to the observer. A score of 1 was given to calves that were docile, did not react to the observer, and allowed the observer to approach. A calf given a score of 2 was slightly flighty, was aware of the observer and likely stood in a corner away from the observer. A score of 3 was given to calves that moved away from the observer. These calves would run with a raised head alongside the fence, fully aware of the position of the observer. A calf given a score of 4 was considered flighty. They were aware of the observer and may have run along the fence or into gates or fences. A score of 5 was given to those considered very flighty. Those calves are often called 'crazy' and would often run at gates, fences, and humans in an attempt to exit the pen. Calves given a score of 5 were allowed to exit the pen in order to more accurately determine the scores of the remaining calves (i.e., to avoid the temperamental animals agitating the other calves, especially if they are of a calmer temperament). Pen score was assigned by the same observer, skilled in assigning pen scores on this herd for over 15 years; therefore an inter-observer reliability was not established.

Serum Analysis

All serum analyses were performed in duplicate. Serum cortisol concentrations were determined using a commercially available enzyme immunoassay kit according to the manufacturer's directions (Arbor Assays, Ann Arbor, MI) by comparison of unknowns to standard curves generated with known concentrations of cortisol. The minimum detectable cortisol concentration was 0.0454 ng/mL, and the intra- and inter-assay coefficients of variation were 17.0% and 14.6%, respectively.

Glucose concentrations were determined by modification of the enzymatic Autokit Glucose (Wako Diagnostics, Richmond, VA USA) to fit a 96-well format as previously described [5]. Briefly, 300 µL of prepared working solution was added to 2 µL of serum or prepared standards in a 96-well plate. Plates were incubated at 37°C for 5 min and absorption was recorded at 505 nm. The plate reader used for this assay (BioTek Powerwave 340; BioTek Instruments, Winooski, VT, USA) has an incubating and timing feature and therefore ensured that the sample absorbances were read immediately following the 5-min incubation. Concentrations of glucose were determined by comparing unknown samples to a standard curve of known glucose concentrations. The minimum detectable concentration was 3.8 mg/dL and the intra- and inter-assay coefficients of variation were 6.1% and 8.1%, respectively.

Insulin concentrations were determined by a bovine-specific insulin ELISA according to the manufacturer's instructions (Cat # 80-INSBO-E01; Alpco Diagnostics, Salem, NH). The minimum detectable concentration was 0.100 ng/mL and the intra- and inter-assay coefficients of variation were 11.0% and 12.8%, respectively.

Concentrations of NEFAs were determined by modification of the enzymatic HR Series NEFA-HR (2) assay (Wako Diagnostics, Richmond, VA USA) to fit a 96-well format as previously described [5]. Briefly, 200 µL of the prepared Color Reagent A were added to 5 µL of serum or prepared standards in a 96-well plate. Plates were incubated at 37°C for 5 min and then the absorbance was read using a spectrophotometer at 550 nm. Next, 100 µL of prepared Color Reagent B was added to all wells on the 96-well plate. Plates were incubated for an additional 5 min and read for a second time using a plate reader at 550 nm. The plate reader used for this assay has an incubating timing feature and therefore ensured that the sample absorbances were read immediately following the 5-min incubation. A final absorbance was obtained by subtracting the first reading, which was multiplied by a factor of 0.67 to account for changes in volume, from the second reading. The final absorbance values were used for all calculations (i.e., standard curve, sample concentrations). Concentrations of NEFAs were determined by comparing unknown samples to at standard curve of known NEFA concentrations. The minimum detectable concentration was 0.0014 mmol/L and the intra- and inter-assay coefficients of variation were 11.9% and 18.6%, respectively.

Serum concentrations of BUN were determined by a colorimetric assay according to the manufacturer's directions (K024-H1; Arbor Assays, Ann Arbor, MI) by comparison of unknowns to standard curves generated with known concentrations of urea nitrogen. The minimum detectable BUN concentration was 0.065 mg/dL and the intra- and inter-assay coefficients of variation were 3.0% and 11.1%, respectively.

Statistical Analysis

Prior to analysis, rectal temperature data were averaged into 1-h intervals, and insulin sensitivity was calculated based on the Revised Quantitative Insulin Sensitivity Check Index (RQUICKI) method as previously described [17]. Briefly, RQUICKI is calculated as 1/[log (Gb) + log (Ib) + log (FFAb)] where Gb is serum glucose concentration in mg/dL, Ib is serum insulin in µU/mL, and FFAb is free fatty acids (i.e, NEFA values) in mmol/L. Based on this index, less insulin sensitivity is demonstrated by lower values. Data were analyzed by the MIXED procedure of SAS specific for repeated measures (SAS Inst. Inc., Cary, NC USA). Temperament, time, and the temperament x time interaction were included as fixed effects

with steer within temperament group as the experimental unit. When the temperament x time interaction was significant, means were separated using the PDIFF option in SAS, with $P < 0.05$ considered significant and $P < 0.10$ considered a tendency. All data are presented as the LSM ± SEM.

Results

Basal feed intake, measured from d 1 - 4, did not differ ($P = 0.15$; 5.1 ± 0.2 and 4.6 ± 0.2 kg/d for Calm and Temperamental, respectively) between Calm and Temperamental steers. There were differences in body weight between the temperament groups. Specifically, Temperamental cattle weighed more on average throughout the study (226 ± 4 kg; $P < 0.001$) compared to Calm steers (203 ± 4 kg). Additionally, Temperamental steers lost more weight on average (-7.1 ± 1.1 kg; $P = 0.004$) than Calm steers (-2.4 ± 1.1 kg) throughout the study.

Rectal temperature in response to the feed restriction (FR) challenge was affected by temperament ($P < 0.001$) and time ($P < 0.001$), but was not affected by a temperament x time interaction ($P = 0.98$; Table 1). Temperamental steers maintained greater rectal temperature (38.75 ± 0.02°C) compared to Calm steers (38.65 ± 0.02°C). In regards to changes over time, rectal temperature fluctuated over time.

There was a temperament x time interaction ($P = 0.01$) for serum cortisol concentrations (Table 1). Specifically, Calm steers had greater ($P \leq 0.02$) serum cortisol than Temperamental steers at 0, 30, and 66 h relative to FR, while Temperamental steers had greater ($P \leq 0.05$) serum cortisol than Calm steers at 12 and 18 h relative to FR. Although there was an effect of time ($P < 0.001$), there was no temperament effect for serum cortisol concentrations ($P = 0.41$).

Serum glucose concentrations were affected by temperament ($P = 0.01$) and time ($P < 0.001$), but there was no temperament x time interaction ($P = 0.13$; Figure 1). During the FR challenge, Temperamental steers had greater serum glucose concentrations (66.90 mg/dL) compared to Calm steers (61.70 mg/dL), with concentrations of serum glucose fluctuating daily. There was a temperament x time interaction ($P < 0.001$) for serum insulin concentrations (Figure 2). Calm steers had greater ($P \leq 0.01$) serum insulin concentrations than Temperamental steers at 0, 6, and 12 h relative to feed removal. Calm steers had greater serum insulin concentrations (1.30 ± 0.28 ng/mL) than Temperamental steers (0.28 ± 0.25 ng/mL).

Serum concentrations of NEFA were affected by temperament ($P < 0.001$) and time ($P < 0.001$), but there was no temperament x time interaction ($P = 0.75$; Figure 3). Temperamental steers had greater serum NEFA concentrations (0.52 ± 0.01 mmol/L) compared to Calm steers (0.41 ± 0.01 mmol/L). Concentrations of serum NEFA initially increased during FR, and decreased once feed was reintroduced.

Serum BUN concentrations were affected by temperament ($P < 0.001$) and time ($P < 0.001$), but there was no temperament x time interaction ($P = 0.99$; Figure 4). Specifically, serum BUN concentrations were reduced in Temperamental steers (8.6 ± 0.2 mg/dL) compared to Calm steers (9.4± 0.2 mg/dL).

	Temperament			P-value		
Variable	Calm	Temperamental	SEM	Temperament	Time	Temperament x Time
RT, °C	38.65	38.75	0.02	<0.001	<0.001	0.98
Cortisol, ng/mL	7.03	6.70	0.29	0.41	<0.001	0.01
Glucose, mg/dL	61.69	66.90	1.26	0.01	<0.001	0.13
Insulin, ng/mL	1.30	0.26	0.28	0.01	<0.001	<0.001
NEFA, mmol/L	0.41	0.51	0.01	<0.001	<0.001	0.75
BUN, mg/dL	9.45	8.60	0.16	<0.001	<0.001	0.99
Insulin Sensitivity	0.53	0.48	0.01	<0.001	<0.001	<0.001

Abbreviations: RT: Rectal Temperature; NEFA: Non-Esterified Fatty Acids; BUN: Blood Urea Nitrogen

Table 1. Effect of temperament on rectal temperature and serum cortisol, glucose, insulin, non-esterified fatty acids and blood urea nitrogen and insulin sensitivity after a feed restriction challenge in beef steers.

Additionally, serum BUN concentrations displayed a similar temporal pattern as NEFA concentrations, with an initial increase during FR, and a decline following feed reintroduction.

There was a temperament x time interaction ($P < 0.001$) for insulin sensitivity during the FR challenge (Figure 5). Specifically, Temperamental steers had greater insulin sensitivity at 0 h ($P = 0.04$); however, Temperamental steers had reduced insulin sensitivity at 102 h and from 120 to 156 h relative to FR ($P \leq 0.04$). There were effects of time ($P < 0.001$) and temperament ($P < 0.001$) such that Temperamental

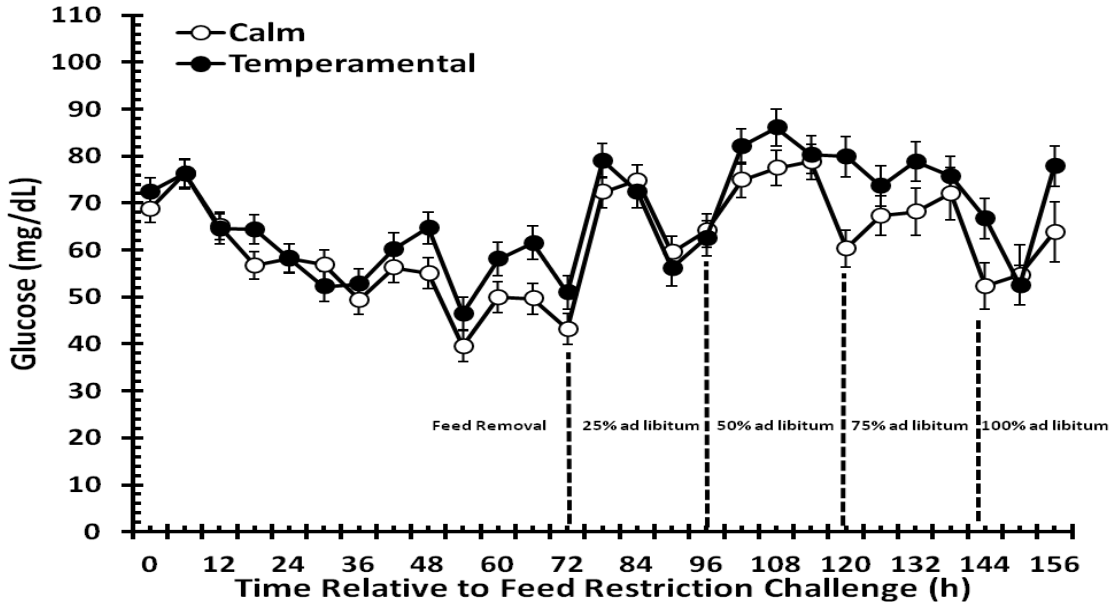

Figure 1. Effect of temperament on the glucose response in Angus-cross steers in response to a feed restriction challenge. Angus-cross steers (n = 16 Calm and 15 Temperamental) were selected based on Temperament Score measured at weaning. Blood samples were collected at 6-h intervals from 0 to 156 h relative to feed restriction (0 h). Data presented as LSM ± SEM. Serum glucose concentrations were affected by temperament ($P = 0.01$) and time ($P < 0.001$), but there was no temperament x time interaction ($P = 0.13$).

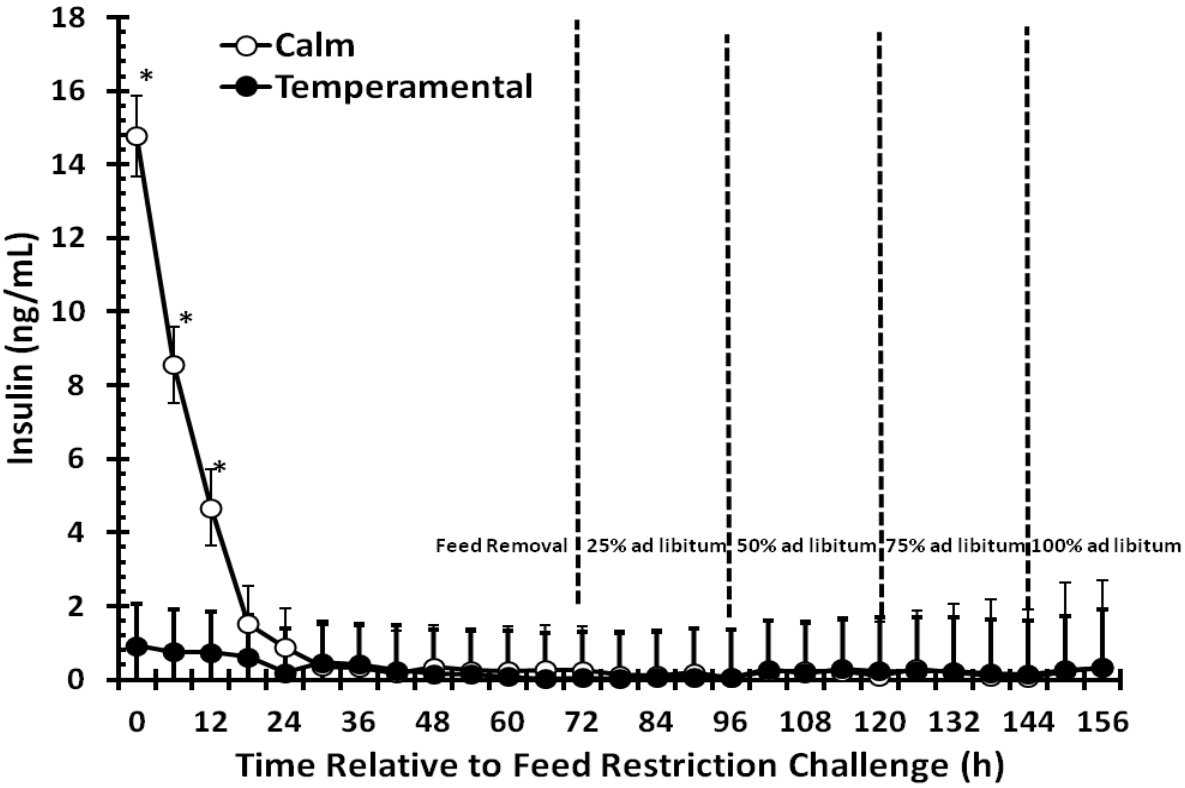

Figure 2. Effect of temperament on the insulin response in Angus-cross steers in response to a feed restriction challenge. Angus-cross steers (n = 16 Calm and 15 Temperamental) were selected based on Temperament Score measured at weaning. Blood samples were collected at 6-h intervals from 0 to 156 h relative to feed restriction (0 h). Data presented as LSM ± SEM. There was a temperament x time interaction ($P < 0.001$) for serum insulin concentrations.

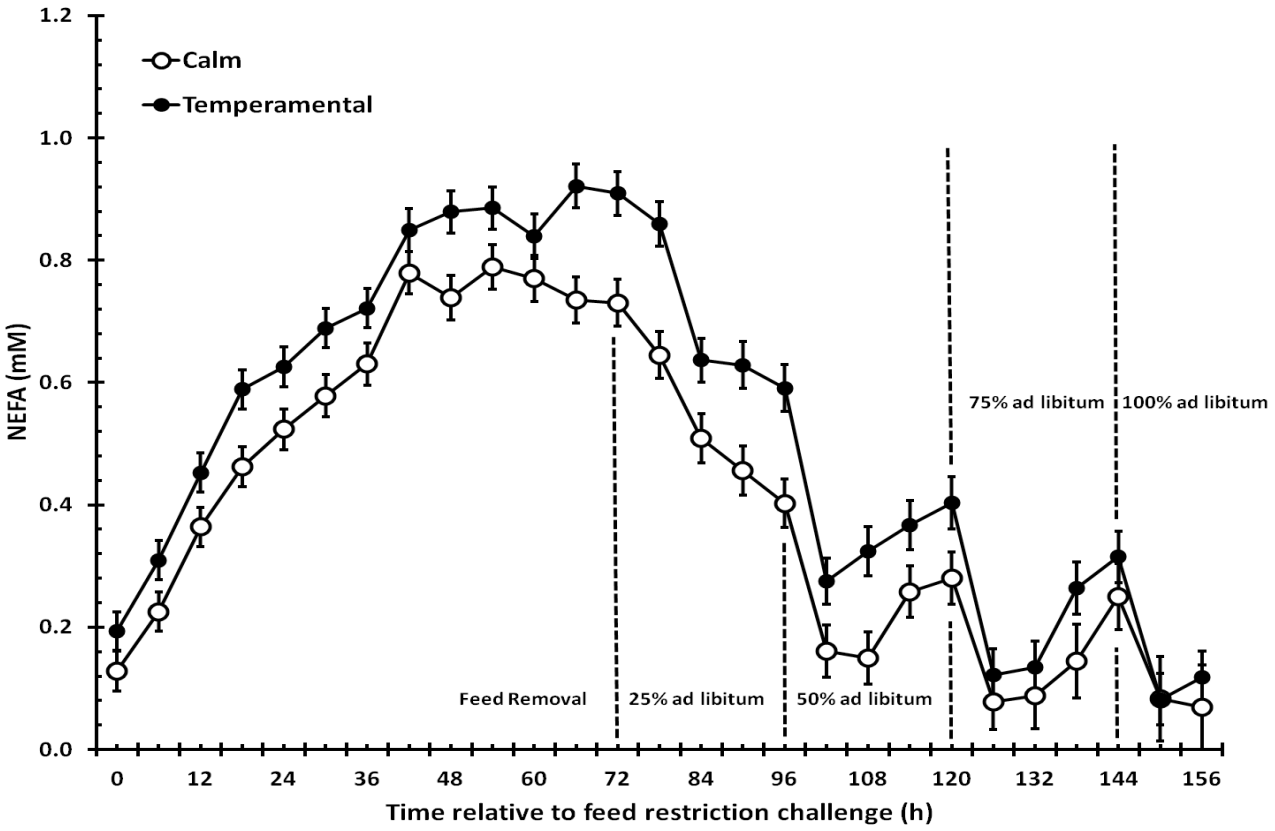

Figure 3. Effect of temperament on the NEFA response in Angus-cross steers in response to a feed restriction challenge. Angus-cross steers (n = 16 Calm and 15 Temperamental) were selected based on Temperament Score measured at weaning. Blood samples were collected at 6-h intervals from 0 to 156 h relative to feed restriction (0 h). Data presented as LSM ± SEM. Serum concentrations of NEFA were affected by temperament ($P < 0.001$) and time ($P < 0.001$), but there was no temperament x time interaction ($P = 0.75$).

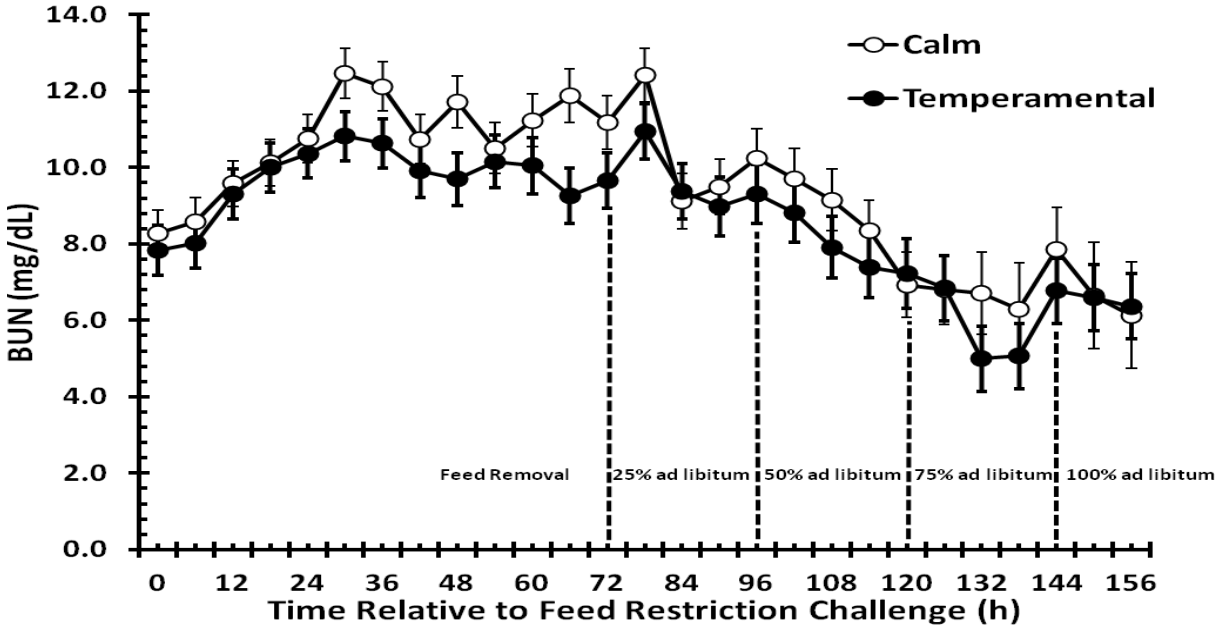

Figure 4. Effect of temperament on the blood urea nitrogen (BUN) response in Angus-cross steers in response to a feed restriction challenge. Angus-cross steers (n = 16 Calm and 15 Temperamental) were selected based on Temperament Score measured at weaning. Blood samples were collected at 6-h intervals from 0 to 156 h relative to feed restriction (0 h). Data presented as LSM ± SEM. Serum BUN concentrations were affected by temperament ($P < 0.001$) and time ($P < 0.001$), but there was no temperament x time interaction ($P = 0.99$).

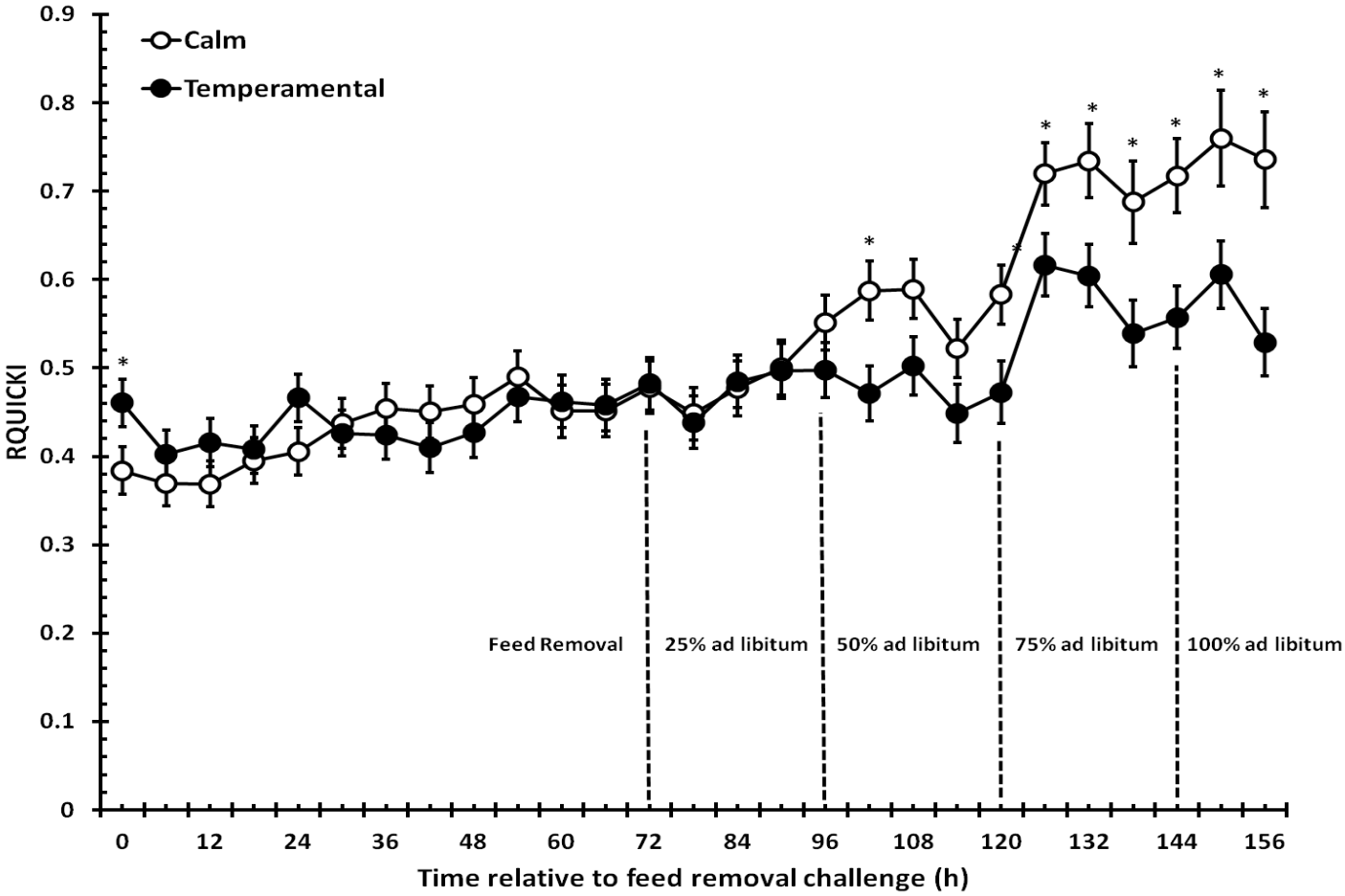

Figure 5. Effect of temperament on the insulin sensitivity response in Angus-cross steers in response to a feed restriction challenge. Angus-cross steers (n = 16 Calm and 15 Temperamental) were selected based on Temperament Score measured at weaning. Insulin sensitivity was calculated based on the Revised Quantitative Insulin Sensitivity Check Index (RUICKI) as previously described [17]. Data presented as LSM ± SEM. There was a temperament x time interaction ($P < 0.001$) for insulin sensitivity.

steers on average had reduced insulin sensitivity (0.48 ± 0.01) compared to Calm steers (0.53 ± 0.01).

Discussion

In the livestock industry it is imperative to maintain and enhance growth in order to improve production and overall profit. Therefore, identifying naturally-occurring deviations that can influence overall health and performance of livestock is essential in developing and modifying management practices to enhance performance. Cattle temperament has been documented to negatively influence performance [7,9,10]. Thus, it is essential to elucidate the mechanisms driving the altered metabolism in order to potentially alleviate the negative effects of temperament on performance.

Rectal temperature is typically one of the first signs that an animal is ill. However, elevations in rectal temperature also occur upon activation of the stress axis, and further, can be elevated by temperament, both in absence of an infection [18]. The observed greater rectal temperature in Temperamental steers in the current study is in concert with previous reports in temperamental cattle [18-19]. The elevated rectal temperature observed in Temperamental steers may be associated with the differences in metabolism. While only a 0.1°C difference was observed between Calm and Temperamental cattle in the current study and thus may not be biologically significant, previous reports have observed a greater difference (greater than 0.5°C) between calm and temperamental bulls [18,19]. Increasing body temperature 1°C has been associated with a 10-13% increase in metabolic rate [20]. An altered state of homeostasis in temperamental cattle may result in an increased metabolic rate, and thus elevated body temperature. However, further research is necessary to support this hypothesis.

Previous studies utilizing temperamental and calm cattle have explored differences in the hypothalamic-pituitary-adrenal (HPA; stress) axis. For example, Curley et al. [21] reported a correlation between serum concentrations of the stress hormone, cortisol, and exit velocity, an objective

method used to evaluate temperament. Decreased serum cortisol and adrenocorticotropic hormone (ACTH) responses were observed when temperamental cattle were challenged with corticotropin releasing hormone (CRH), ACTH, or lipopolysaccharide (LPS), all of which stimulate the HPA axis[19,21]. Thus, temperament is noted to alter physiologic stress responsiveness. While there was a temperament x time interaction for cortisol in the current study, the variation observed in serum cortisol concentrations, in addition to the relatively low serum concentrations of cortisol observed, lend to the difficulty in drawing conclusions relative to temperament. However, it appears that stress was alleviated once feed was reintroduced following FR, as serum cortisol concentrations decreased over time.

Serum glucose concentrations were greater, but serum insulin was less in Temperamental steers throughout the study. This data is similar to the response of these same steers to a glucose tolerance test, in which serum glucose concentrations were elevated yet serum insulin concentrations were decreased in temperamental steers compared to calm steers [6]. Thus, it appears that there is some degree of insulin insensitivity in temperamental cattle. Studies in rodents have determined that insulin insensitivity can decrease the transport of glucose into cells via decreased translocation of glucose transporters to the cell surface [28,22]. This may be one mechanism by which serum glucose remains elevated in temperamental compared to calm cattle, even in response to feed restriction.

It is unclear why serum insulin concentrations were elevated during the first 12 h of feed restriction in the Calm steers. There was no difference in *ad libitum* feed intake between temperaments in the 4 days prior to the start of the study. However, it is possible that more Calm calves were consuming feed immediately prior to feed removal at 0 h. Additionally, the increased insulin concentrations may be a residual response from the glucose and insulin challenges the day prior [6]; however, that does not explain why insulin was elevated in Calm while the Temperamental cattle were observed to have greater insulin in response to the insulin challenge. Yet, that does not rule out whether the elevated insulin concentrations in the Calm calves are an artifact from the prior day's challenges.

Maintenance of glucose homeostasis is vital to all animals, including ruminants. However, while monogastrics absorb glucose directly from the gastrointestinal tract, more than 70% of the energy requirements in ruminants are provided by volatile fatty acids, produced by fermentation of starches by the microbiome in the rumen [23,24]. Mcdowell [23] has reported that while propionate and amino acids are the primary contributors to glucose production in the fed ruminant, there is a shift in the fasted animal to glycerol and amino acids due to the reduction in propionate concentrations. In our voyage to gain a better understanding of the metabolic differences between calm and temperamental cattle, the measurement of volatile fatty acids appears to be the next logical step. Further, as the liver is the major source of glucose production (85-90%), analyses that detect differences in gene and protein expression within this organ are of great interest [23].

Recent studies have demonstrated that temperamental cattle have elevated serum concentrations of NEFAs [5], which is supported by the results from the current study. The concentrations of NEFA exhibited by the Temperamental cattle are similar to what have been observed in transition dairy cows [25,26]. It has been reported that ruminants utilize free fatty acids for energy when energy requirements are high [23,27]. Furthermore, decreases in the absorption of glucose when serum NEFAs are elevated and during times of insulin insensitivity have been reported [22,28-30]. Therefore, it is possible that the limited ability of cattle to absorb serum glucose, due to their inherent decreased insulin sensitivity, has made the utilization of NEFAs a requirement rather than an optional pathway for energy utilization in ruminants.

It should be noted that less insulin sensitivity in Temperamental steers relative to Calm steers was not observed during the 72 h feed removal period, or when cattle were feed 25% *ad libitum*. Rather, this phenomenon was only apparent when cattle were fed 50% or more of *ad libitum*. Why a decrease in insulin sensitivity is not observed when cattle are fed less feed is not clear. It is possible that temperamental cattle maintain a similar level of insulin sensitivity regardless of amount fed, and thus the increase in insulin sensitivity in the Calm steers is due to an increased need for insulin following reintroduction of feed and increasing serum glucose concentrations. Studies aimed at monitoring insulin receptors within various tissues are necessary in order to fully understand the mechanisms behind the changes in insulin sensitivity in calm and temperamental cattle.

Alternative management strategies may allow mitigation of some of the negative effects of temperament on production parameters. Separating temperamental cattle at a feedlot and managing these cattle differently could reduce costs. As temperamental cattle less efficiently deposit adequate adipose tissue [11] it may be more advantageous to market these cattle to a programmed finishing weight rather than to level of finish. Studies aimed at determining if changing the composition of the diet, based on the basal metabolic responses of temperamental cattle, would increase performance and enhance carcass composition are needed. Separating temperamental cattle from the rest of the herd may be advantageous to the producer, considering that temperamental cattle do not exhibit typical sickness behaviors when ill [19], and thus these cattle could potentially serve as 'persistent infectors' of the other cattle in the pen because they remain untreated. Thus, this data, in conjunction with other reports regarding the effect of temperament on immunity and metabolic parameters, provides areas for future research aimed at modifying management strategies to reduce the negative impacts of cattle temperament on health and production.

Conclusion

Cattle temperament can influence various physiological systems including metabolism as well as performance. Data from this study demonstrate that Calm and Temperamental steers have different metabolic responses to feed restriction and gradual feed reintroduction. Specifically, Temperamental steers have greater serum NEFA and glucose, yet less serum BUN, insulin, and insulin sensitivity compared to Calm steers. Through elucidating the mechanisms associated with the differences in metabolism between Calm and Temperamental cattle, methodologies can be developed or modified in order to reduce the negative effects of temperament on production parameters.

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