

# Beef cows housed in mud during late gestation have greater net energy requirements compared with cows housed on wood chip bedding

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## ABSTRACT

Mud increases net energy requirements for cattle because mud and precipitation compromise the ability of the hair coat to insulate and maintain core body temperature of the cow. The increase in energy required for a gestating cow to compensate for a muddy environment is unknown. The objective of this study was to evaluate effects of muddy conditions on cow body weight (BW) and fetal growth during late gestation. Sixteen multiparous Angus cows ( $n = 8/\text{treatment}$ ) were paired based on initial BW and one cow from each pair was randomly allocated to either the mud (MUD) or control (CON) treatment on day 213 of gestation. Pens in the CON group were bedded with wood chips, while pens in the MUD group were designed to create a muddy lot (average depth of  $23.6 \pm 5.8$  cm). Cows were housed outdoors individually and fed the same diet that consisted of a limit-fed total mixed ration. Each pair was fed to meet energy and protein requirements for maintenance and gestation. From day 213 to 269 of gestation, cows were weighed and sampled for blood metabolites weekly. Data were analyzed as a randomized complete block design with repeated measurements (SAS 9.4). Though cows consumed the same amount of dry matter, cows in the MUD treatment weighed 37.4 kg less than cows in the CON treatment ( $P < 0.01$ ) by day 269 of gestation. Cows in the MUD treatment decreased approximately half a body condition score (BCS), while cows in the CON treatment gained approximately 1 BCS during the treatment period ( $P < 0.01$ ). There was no evidence of a treatment  $\times$  day of gestation effect for 12th rib back fat ( $P = 0.85$ ), rump fat ( $P = 0.48$ ), total plasma protein concentrations ( $P = 0.85$ ), or plasma 3-methylhistidine ( $P = 0.84$ ); however, there was a marginally significant treatment  $\times$  day of gestation effect for plasma non-esterified fatty acid concentration ( $P = 0.09$ ). Despite differences in cow BW at the end of the treatment period, calf birth weight ( $P = 0.66$ ) and calf total plasma protein ( $P = 0.27$ ) were not different; however, the divergence in cow BW remained marginally significant at parturition ( $P = 0.06$ ). These results indicate that mud increased net energy requirements for cows in the MUD treatment, as calf birth weight was not different but maternal BW was decreased compared with cows in the CON treatment.

## Lay Summary

Winter and spring precipitation are expected to increase 20% to 30% by the end of the century in the Midwest, United States. As a result of this environmental change, beef cattle in this region of the United States are increasingly becoming exposed to muddy conditions in pastures and lots. These muddy conditions cause a cow to increase her metabolic heat production to maintain her internal body temperature, thus increasing her net energy requirements. The increase in net energy required during late gestation for a beef cow is unknown; therefore, we completed a study to estimate the increase in net energy requirements. Based on the present results, cows housed in muddy conditions that were consuming the same amount of dry matter as cows housed in wood chips lost body weight and needed an additional 3.9 Mcal/d in order to maintain their body weight. While cows housed in muddy conditions lost body weight and body condition, we do not believe fetal growth was affected as calf birth weight was similar between the cows housed in pens with the muddy conditions and the cows housed in pens with wood chips.

**Key words:** fetal growth, late gestation, mud, net energy requirements

## INTRODUCTION

Late-gestation nutrient restriction can negatively affect the dam and fetus because most of the fetal growth occurs during the last 2 mo of gestation in beef cattle (NASEM, 2016). Decreased calf birth weights have been observed when cows were nutrient restricted during late gestation (Hight, 1968; Tudor, 1972; Spitzer et al., 1995; LeMaster et al., 2017). While maternal undernutrition during late gestation has been shown to have negative effects on the calf, late-gestation energy

restriction is common in spring-calving beef cow herds as forage quality and quantity are often limited during the winter months (Meyer et al., 2010). In addition to being subjected to poor forage quality and quantity during late gestation, beef cows are often housed outside where they are exposed to climatic changes and cold stress (Roland et al., 2015). When beef cows are unprotected from wind and rain, the insulative properties of the hair coat are compromised, and the cow must increase her rate of metabolic heat production to maintain

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homeothermy (Webster et al., 1970; Webster, 1974; Young, 1983). Climate predictions indicate that annual precipitation and intense rainfall events will continue to increase and come to characterize the U.S. Midwest, impacting livestock agriculture (Walthall et al., 2012; Hatfield et al., 2014). Thus, it is reasonable to infer that these future weather patterns will intensify the late-gestation energy restriction in beef cows by creating a severe environmental stressor during late winter months.

In general, it is accepted that mud and precipitation increase net energy requirements of cattle (NASEM, 2016). There is, however, a need to quantify the net energy requirement and the physiological response of beef cows in late gestation when challenged with muddy conditions in winter. There is limited information evaluating the effects of mud on cow body weight (BW) and body condition score (BCS); however, it is recognized by the NASEM (2016) that mud and precipitation increase nutrient requirements in addition to cold stress. We hypothesize that the NASEM (2016) net energy recommendations for maintenance and gestation are not sufficient to maintain cow BW and body condition during late gestation when cows are housed in muddy conditions. We further hypothesize that calves from cows exposed to mud will have a lesser birth weight compared with cows that were housed in bedding but exposed to the same weather. This leads to the objectives of this study which were to evaluate the effects of muddy environmental conditions on cow BW, BCS, body store mobilization, and calf birth weight to determine if the NASEM's (2016) net energy recommendations for maintenance and gestation are sufficient for beef cows exposed to muddy conditions.

## MATERIALS AND METHODS

All procedures were approved by The Ohio State University Institutional Animal Care and Use Committee (Animal Use Protocol # 2019A00000142).

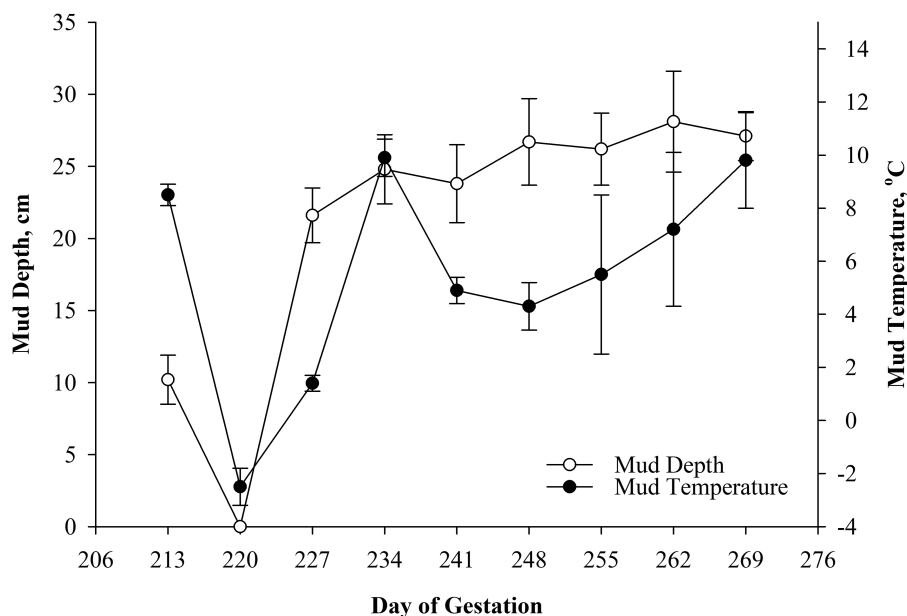
### Animals, Experimental Design, and Treatments

Mature Angus cows ( $n = 8/\text{treatment}$ ; initial mean age  $\pm$  SEM =  $7.3 \pm 0.5$  yr; range = 5 to 10 yr; initial mean BW  $\pm$  SEM =  $545.4 \pm 6.6$  kg; initial mean BCS  $\pm$  SEM =  $4.1 \pm 0.1$ ) were used in a randomized complete block design experiment at the Eastern Agricultural Research Station (Caldwell, OH). The treatment period lasted from day 213 to day 269 of gestation. All cows were removed from their pens 14 d before their expected calving date on day 269 of gestation to prevent any calves from being born in the individual pens that cows were housed in during the treatment period. After being removed from their pens on day 269 of gestation, all cows were housed together in a single pasture.

Cows were maintained as one herd and treated similarly before initiation of the study. All cows entered an estrous synchronization protocol to allow for fixed-time artificial insemination in June 2019. Pregnancy status was diagnosed using transrectal ultrasonography approximately 31 d after artificial insemination at the initiation of the study. All cows were confirmed to be bred to the first artificial insemination date and all cows had an expected calving date of March 22nd, 2020. Only cows that had conceived to the first artificial insemination were used to ensure that all cows were at the same days of gestation throughout the treatment period.

On January 14th, 2020 (day 213 of gestation), the group of cows at the research station that had conceived to first artificial insemination were weighed one time. The 16 cows that had the most similar BWs were selected, and cows were paired based on initial BW to create similar BWs in each pair. Pairs were created based on cow BW rather than BCS, as when nutrient requirements are calculated using NASEM (2016) for maintenance and gestation, cow BW is a variable in the estimated model. One cow from each pair was then randomly allocated to either the mud (MUD;  $n = 8$ ) or control treatment (CON;  $n = 8$ ), and the other cow in the BW pair was assigned to the opposite treatment. Cows in the CON treatment were individually housed in pens ( $4.9 \text{ m} \times 4.9 \text{ m}$ ) bedded with wood chips and not exposed to mud, while cows in the MUD treatment were housed in pens filled with mud to a depth of  $23.6 \pm 5.8$  cm. The 16 individual pens were created in the same outdoor lot and were uncovered and housed away from all buildings to avoid a windbreak effect, so that all cows were exposed to the same environmental conditions except for their allocated pen treatment. The outdoor lot that the individual pens were created on was scraped and graded using a skid loader before the treatment period began to provide a flat surface. The area occupied by the pens was previously used for holding pens outside of the chute area and had a geotextile fabric and stone base. Before the treatment period began and after the outdoor lot was scraped and graded, soil from the same area of the research station was added to the pens at a depth of approximately 30 cm as a target depth in all eight of the mud treatment pens. The target depth of 30 cm was based on the depth of mud that cows were typically subjected to at the research station in previous years. The soil that was used to create the mud in the mud treatment pens was previously analyzed by the National Resources Conservation Service as part of the National Cooperative Soil Survey and is classified as Vandalia-Guernsey silty clay loam. On the first day of the treatment period only, water was added to the mud treatment pens to create the muddy environment until the cows' hooves were sinking into the soil when walking rather than walking on top of the soil. No drainage system was created for the pens; however, to maintain the integrity of each treatment pen, each treatment pen was created in a line. As there were 16 pens created, this provided four rows of four pens. The treatment was randomly applied to row, rather than individual pen. This allowed for each row to house only one treatment and prevented the mud from entering the control pens. Furthermore, cows were randomly assigned to pen within their treatment. The CON pens were bedded weekly with saw dust and wood chips as needed such that no mud formed in those pens and to the same target depth as the mud pens of 30 cm. While it was not recorded, there were no observations of any consumption of the bedding by the cows in the CON treatment.

Pen mud depth and mud temperature were recorded weekly in only the MUD treatment pens. Mud depth was recorded following a procedure outlined by Castillo et al. (2012) in which mud depth was recorded using a steel rod of 1 m in length with a density of  $1.1 \text{ g cm}^3$ . To determine the depth of mud, the rod was dropped from a height of 1 m above the ground. To ensure that the rod was dropped vertically, a 1-inch (2.54 cm) diameter PVC pipe was used as a guide. The portion of the steel rod immersed in the mud was considered as the depth of mud. Mud temperature was recorded by inserting a thermometer 75 mm into the mud in the middle of



**Figure 1.** Mean mud depth (cm) and mud temperature (°C)  $\pm$  SD of the eight MUD treatment pens recorded at weekly intervals from the beginning of the treatment period on day 213 of gestation until the end of the treatment period on day 269 of gestation.

each individual pen. In addition to manually recording mud temperature in the experimental pens, weather data including mean ambient air temperature (°C), daily precipitation (mm), daily solar radiation (Cal/cm<sup>2</sup>), mean soil temperature (°C), mean wind speed (km/h), and mean relative humidity (%) were recorded using the EARS weather station that is 500 m from the experimental site (<https://weather.cfaes.osu.edu/stationinfo.asp?id=3>). These pen data and weather data are presented in Fig. 1 and Fig. 2A–F as descriptive results, respectively.

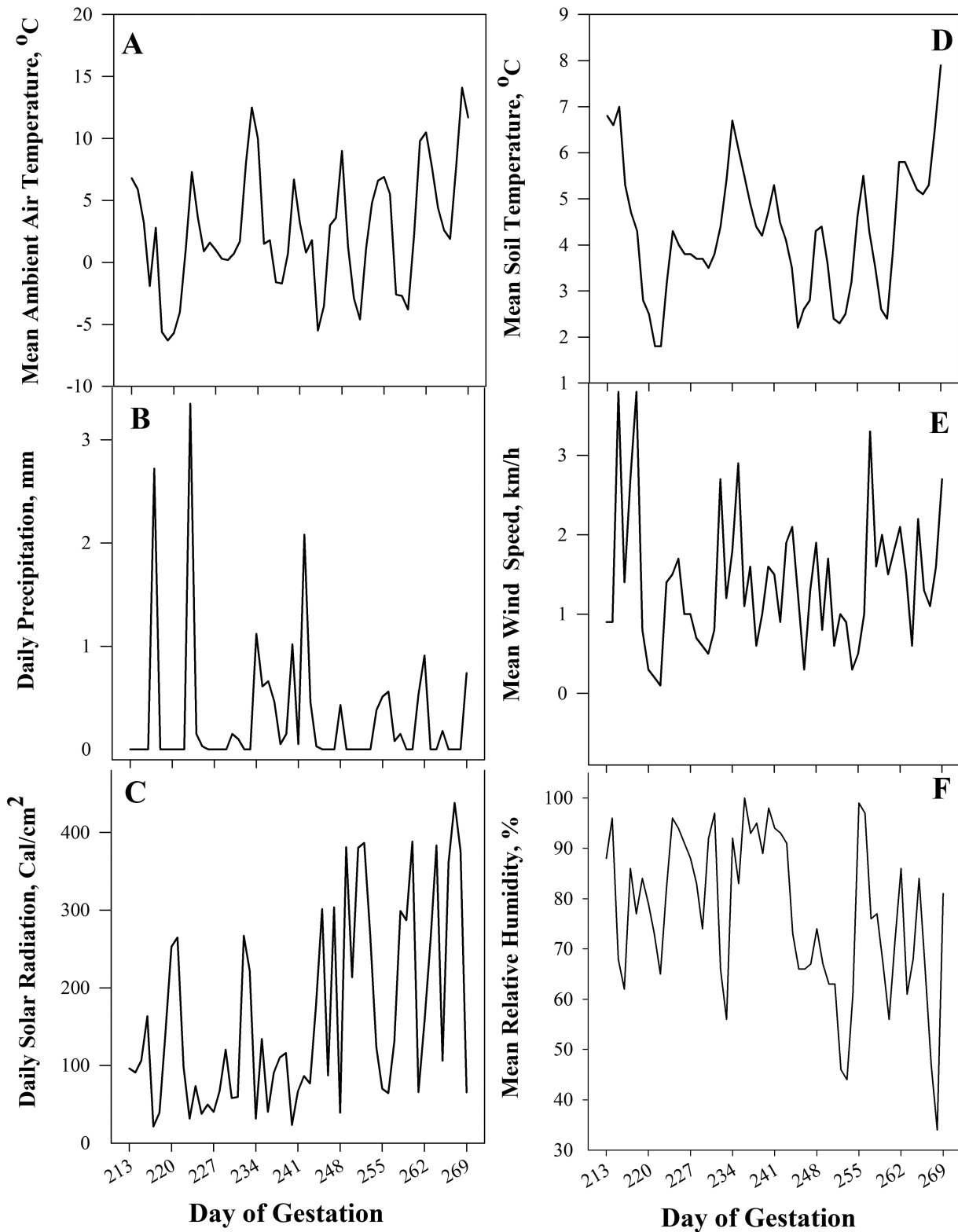
All cows were fed the same diet once daily (Table 1) at approximately 0830 h and dry matter intakes were recorded. The diet given to the cows was a total mixed ration of chopped hay and a supplement with vitamins and minerals included that was limit-fed. Each cow had her own feed bunk (1.5 m  $\times$  0.7 m) and water trough that was filled daily to provide ad libitum access to water. The diet was formulated to meet cow maintenance and gestation requirements (NASEM, 2016). Cow maintenance requirements were calculated using equation 11-1 of the NASEM (2016), cow gestation requirements for net energy and crude protein were calculated using equations 13-34 through 13-44 assuming a 35 kg calf birth weight, and cow mineral requirements were calculated using table 7-1. Each week, dry matter allowances for the pair were adjusted based on the CON cow BW and week of gestation so that each pair received the same dietary allowance each day throughout the study (Table 2). Dry matter allowances were adjusted on a weekly basis to keep cows as close as possible to meeting their net energy and crude protein requirements for both maintenance and gestation. However, cows were never limited and were always meeting or slightly exceeding their nutrient requirements. All 16 cows were pair fed, with one cow in each pair being in the CON and MUD treatment, respectively. Therefore, by design, dry matter intake was the same for each pair throughout the treatment period. Since each cow had her own bunk, there was no wastage of the diet that was fed, and there were no dry matter refusals at any point of the treatment period for

any of the BW pairs. During the first week of the treatment period (starting on day 213 of gestation), cows were fed to meet the requirements of 220 d of gestation and this dry matter allowance was kept the same for the second week of the treatment period (starting on day 220 of gestation) to allow for a transition to the experimental diet, since cows were previously on an ad libitum hay diet.

### Prepartum Measurements

Beginning on day 213 of gestation, cows were assigned a mud score at weekly intervals as follows: 1 = no tag, clean hide; 2 = small lumps of manure/mud attached to the hide in limited areas of the legs and underbelly; 3 = small and large lumps of manure/mud attached to the hide covering larger areas of the legs, side, and underbelly; 4 = small and large lumps of manure/mud attached to the hide in even larger areas along the hind quarter, stomach, and front shoulder; and 5 = lumps of manure/mud attached to the hide continuously on the underbelly and side of the animal from brisket to rear quarter (Busby and Strohbehn, 2008).

As previously mentioned, on day 213 of gestation, cows were weighed and assigned a BCS using a scale described by Wagner et al. (1988) (BCS; 1 = severely emaciated; 9 = very obese). Cows continued to be weighed and assigned a BCS once weekly until parturition. Body condition scores were excluded from analysis on day 262 of gestation, as there was a different individual recording the measurement on this day of the treatment period. Cows were weighed once at the beginning of each week at 0830 h. Cows were allowed ad libitum access to water before being weighed; however, cows were not fed until after they were weighed on these days. Using prediction equations from O'Rourke et al. (1991), an estimated conceptus-free live weight (CFLW) was calculated from the weekly recorded BW of the cows to calculate the weight of the cows without the gravid uterus. The estimated CFLW was calculated using the regression equation:  $y = \exp[a + bt + 0.001ct^2]$  and the coefficients for *Bos taurus* cows calculated by O'Rourke et al. (1991).



**Figure 2.** Climate variables throughout the experimental period (A–F). Mean air temperature (A; °C), daily precipitation (B; mm), daily solar radiation (C; Cal/cm<sup>2</sup>), mean soil temperature (D; °C), mean wind speed (E; km/h), and mean relative humidity (F; %) from the start of the treatment period on day 213 of gestation (January 14, 2020) to the end of the treatment period on day 269 of gestation (March 10, 2020) at the Eastern Agricultural Research Station (Caldwell, OH).

Each week, ultrasound measurements of back fat between the 12th and 13th ribs over the longissimus muscle were recorded, as well as rump fat measurements (Brethour, 1992).

Back fat and rump fat measurements on day 262 of gestation were excluded from the analysis because a different technician measured and recorded the variables on this day.



**Table 1.** Composition and nutritional profile of the prepartum diet offered to pair-fed cows housed in 23.6 ± 5.8 cm of mud (Mud) or wood chips (Control) from day 213 to day 269 of gestation

Item	
Composition, as-fed basis	
Whole shelled corn, %	35.95
Corn gluten meal, %	10.00
Soybean hulls, %	6.50
Cane molasses, %	7.00
Dicalcium phosphate, %	0.50
Availa <sup>4</sup> , %	0.05
Chopped first-cutting grass hay, %	40.00
Nutritional profile <sup>2</sup> , dry matter basis	
Net energy for maintenance <sup>3</sup> , Mcal/kg	1.67
Total digestible nutrients, %	69.62
Neutral detergent fiber, %	36.81
Acid detergent fiber, %	24.54
Crude protein, %	14.55

<sup>1</sup>Availa4 – Zinpro Performance Minerals (5.15% Zn, 2.86% Mn, 1.80% Cu, 0.18% Co).

<sup>2</sup>Based on wet chemistry procedures by a commercial laboratory (Rock River Laboratory, Wooster, OH).

<sup>3</sup>Calculations for net energy for maintenance and gestation of the diet using the feed composition estimates provided by [NASEM \(2016\)](#) for feed ingredients and the nutritional profile of the hay provided by Rock River Laboratory (Wooster, OH).

Blood samples were collected weekly via jugular venipuncture into a 10 mL vacutainer collection tube containing K2 ethylenediaminetetraacetic acid (EDTA). Blood samples were collected as cows were weighed each week; therefore, blood samples were collected at approximately 0830 h and before cows were fed that day. Blood samples were placed on ice until they were centrifuged at 2,500 × g for 25 min at 4 °C. Plasma from the collection tubes was frozen at –20 °C for quantification of plasma non-esterified fatty acid (NEFA), total plasma protein, plasma 3-methylhistidine, and plasma insulin concentration. A colorimetric assay was used to determine concentration of plasma NEFA (Wako Chemicals USA, Richmond, VA) according to a protocol by [Johnson and Peters \(1993\)](#). Intra-assay variation for plasma NEFA was 1.5%, and inter-assay variation was 1.7%. Total plasma protein was quantified using the bicinchoninic acid (BCA) method in a colorimetric assay using a 1:80 dilution (Pierce BCA Protein Assay Kit; ThermoFisher Scientific, Waltham, MA). Intra-assay variation for total plasma protein was 2.5%, and inter-assay variation was 3.3%. Plasma insulin was quantified using a commercial Porcine insulin radioimmunoassay kit previously validated for bovine by [Miqueo et al. \(2019\)](#) (100% bovine insulin specificity, MilliporeSigma, Burlington, MA). The intra-assay coefficient of variation was 4.4%. Plasma 3-methylhistidine was quantified using a commercial human competitive binding enzyme-linked immunoassay (MyBioSource Inc., San Diego, CA). Intra-assay variation for plasma 3-methylhistidine was 9.9%, and inter-assay variation was 12.9%.

### Postpartum Measurements

To avoid calves being born in muddy conditions and in the individual pens, all cows were removed from their pens 2 wk

before the expected calving date on day 269 of gestation. After cows were removed from their individual pens, they were placed on pasture as a single group and were allowed ad libitum access to hay. The university COVID-19 restrictions on personnel at day 269 to calving resulted in no BW sampling of the cows until parturition. Within the first 24 h after parturition, cow weight and calf birth weight were recorded. In addition, a single blood sample was collected from each calf within the first 12 to 24 h after parturition when calves were weighed as an indicator of passive transfer after birth. Each blood sample was collected via jugular venipuncture into a 5 mL vacutainer collection tube containing K2 EDTA. Samples were placed on ice until they were centrifuged at 2,500 × g for 25 min at 4 °C. Plasma from the collection tubes was frozen at –20 °C for quantification of total plasma protein. Similar to the cow total plasma protein, calf total plasma protein was quantified using the BCA method in a colorimetric assay using a 1:80 dilution (Pierce BCA Protein Assay Kit; ThermoFisher Scientific) and were run on the same plates as the cow total plasma protein samples (intra-assay variation = 2.5%, and inter-assay variation = 3.3%).

### Energetic Cost of Mud and NASEM (2016) Modeling Equations

Using the data produced in this experiment, we then aimed to compare our estimation of the energetic cost of mud to the mature beef cow to the [NASEM \(2016\)](#) calculations. To our knowledge, the [NASEM \(2016\)](#) only incorporates mud into model equations for net energy in equations 11-7 to calculate external insulation in the “Maintenance Considerations” chapter. In this equation, the reader has the option to use a value of 1 (no mud), 0.8 (some mud on lower body), or 0.2 (heavily covered with mud) when calculating the external insulation value. While the [NASEM \(2016\)](#) acknowledges that external insulation is related to hair depth and is affected by wind, precipitation, mud, and hide thickness, we propose that the energetic cost of mud is much greater than that predicted by the equations in the cold stress section of the [NASEM \(2016\)](#). Using calculations 11-3 through 11-13, we calculated the additional net energy that the [NASEM \(2016\)](#) predicts for cattle that are heavily covered with mud (MUD = 0.2). We also made the assumptions based on the average weather data during the treatment period of this study that wind speed was 1.4 km/h and effective ambient temperature was 2.6 °C. Additionally, we assumed that hair depth of our cows was 7.5 cm, the hide thickness of our cows was average (HIDE = 1), and that tissue insulation was equal to 9 (average of recommended 6.0 to 12.0 for mature cattle). Using these assumptions, we modeled the additional net energy required for day 269 of gestation (end of the treatment period) and for the average BW of the cows in the MUD treatment which was 462.6 kg.

### Statistical Analyses

Cow was considered the experimental unit with eight replications per treatment. Body weight pair was considered the blocking criteria. Cow BW, CFLW, BCS, back-fat thickness, rump fat thickness, mud score, NEFA concentration, plasma insulin concentration, total plasma protein concentration, and plasma 3-methylhistidine concentration were analyzed using the MIXED procedure of SAS (9.4, SAS Inst. Inc., Cary, NC). The model included treatment, day of gestation, and their interaction as fixed effects. The model also included block

**Table 2.** The body weight (BW; kg) of the control cow in each BW pair used to calculate maintenance and gestation requirements (NASEM, 2016), and the net energy for maintenance (NEm; Mcal/d) and crude protein (CP; g/d) provided to pair-fed cows housed in 23.6 ± 5.8 cm of mud (Mud) or wood chips (Control) from day 213 to day 269 of gestation

	Day of gestation								
	213 <sup>1</sup>	220	227	234	241	248	255	262	269
Pair 1									
Control BW, kg <sup>2</sup>	519.3	462.6	448.1	444.4	441.7	452.6	458.0	459.9	465.3
NEm provided, Mcal/d <sup>3</sup>	10.02	10.02	9.55	9.71	10.05	10.55	10.89	11.22	—
CP provided, g/d <sup>4</sup>	862.52	862.52	799.77	813.80	841.86	883.95	912.02	940.08	—
Pair 2									
Control BW, kg	530.6	482.30	476.2	451.7	459.0	475.3	468.9	478.0	477.1
NE provided, Mcal/d	10.33	10.33	9.88	9.88	10.38	10.72	11.05	11.56	—
CP provided, g/d	889.47	889.47	827.83	827.83	869.92	897.98	926.05	968.14	—
Pair 3									
Control BW, kg	535.1	485.3	487.5	467.1	477.1	493.4	497.1	503.4	497.1
NE provided, Mcal/d	10.33	10.33	10.05	10.05	10.38	10.89	11.39	11.89	—
CP provided, g/d	889.47	889.47	841.86	841.86	869.92	912.02	954.12	996.20	—
Pair 4									
Control BW, kg	542.0	507.9	505.7	478.5	488.9	491.6	492.5	492.5	496.1
NE provided, Mcal/d	10.49	10.49	10.22	10.22	10.55	10.89	11.22	11.72	—
CP provided, g/d	902.95	902.95	855.89	855.89	883.95	912.02	940.08	982.17	—
Pair 5									
Control BW, kg	546.5	473.9	462.6	455.8	468.9	478.0	475.3	488.9	492.5
NE provided, Mcal/d	10.49	10.49	9.71	9.88	10.38	10.72	11.05	11.56	—
CP provided, g/d	902.95	902.95	813.80	827.83	869.92	897.98	926.05	968.14	—
Pair 6									
Control BW, kg	557.8	517.0	498.9	496.6	509.8	531.5	529.7	528.8	530.6
NE provided, Mcal/d	10.49	10.49	10.05	10.38	10.89	11.39	11.72	12.06	—
CP provided, g/d	902.95	902.95	841.86	869.92	912.02	954.12	982.17	1010.23	—
Pair 7									
Control BW, kg	573.7	528.3	498.9	494.3	488.9	494.3	508.8	515.2	517.9
NE provided, Mcal/d	10.80	10.80	10.05	10.38	10.89	11.05	11.56	11.89	—
CP provided, g/d	929.90	929.90	841.86	869.92	883.95	926.05	968.14	996.20	—
Pair 8									
Control BW, kg	580.5	510.2	505.7	496.6	502.5	522.4	528.8	537.0	523.4
NE provided, Mcal/d	10.80	10.80	10.22	10.38	10.72	11.22	11.72	12.23	—
CP provided, g/d	929.90	929.90	855.89	869.92	897.98	940.08	982.17	1024.26	—

<sup>1</sup>213 = Diets were formulated to meet the net energy and crude protein requirements of 220 d of gestation.

<sup>2</sup>Control BW, kg = the body weight (kg) of the control cow in each body weight pair used to calculate maintenance and gestation requirements (NASEM, 2016).

<sup>3</sup>NEm provided, Mcal/d = net energy for maintenance provided to each pair that was adjusted weekly (Mcal/d).

<sup>4</sup>CP provided, g/d = crude protein provided to each pair that was adjusted weekly (g/d).

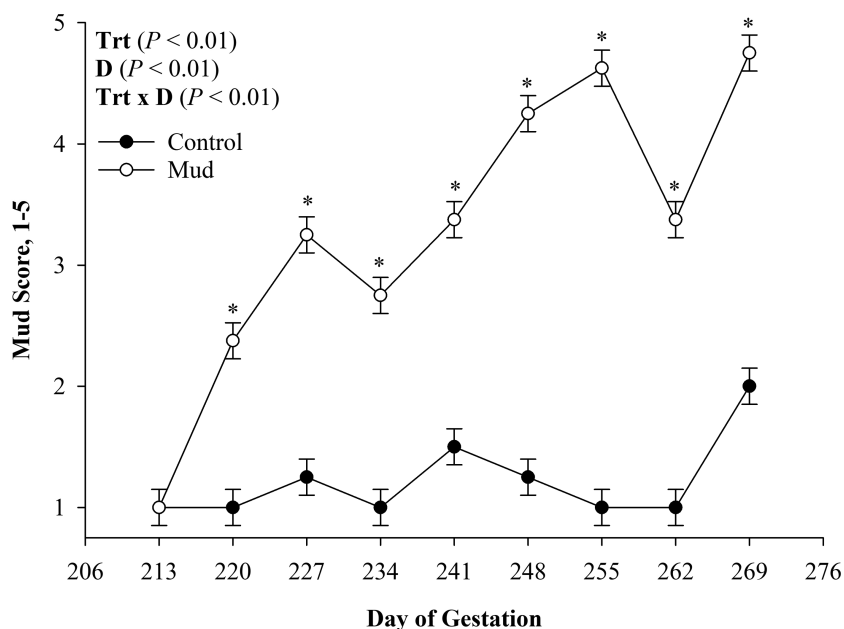
and cow within block by treatment as the random effects. A covariance structure was used to account for the error's correlation due to the repeated measures over time. For all repeated measures data, a first-order autoregressive structure with heterogeneous variances was used, as it produced the lowest Akaike information criterion for each model. Linear regression was performed using day of gestation as the independent variable and CFLW as the dependent variable to evaluate the effect of treatment on CFLW change over the treatment period. Cow BW at parturition, gestation length, calf birth weight, and 24-h calf total plasma protein concentration were analyzed using the MIXED procedure of SAS. The model included cow BW pair as the random effect. The assumptions of normality and homogeneity of variance were

evaluated using the residuals plots in SAS for all variables. No variables violated these assumptions; therefore, there were no transformations. Differences were considered significant if  $P \leq 0.05$  and marginally significant if  $0.05 < P \leq 0.10$ . If the day of gestation × treatment interaction was significant, the PDIF option of SAS was used for mean separation. Data are presented as least squares means ± SEM.

## RESULTS

### Mud Measurements, Weather Observations, and Mud Scores

Mud temperature and mud depth of the experimental pens were recorded weekly and are presented in Fig. 1 as



**Figure 3.** Mean prepartum cow mud score  $\pm$  SEM measured weekly from day 213 of gestation (start of treatment period) to day 269 of gestation (end of treatment period) of pair-fed cows housed in 23.6  $\pm$  5.8 cm of mud (Mud) or wood chips (Control) presented with Treatment (Trt), Day of Gestation (D), and Treatment  $\times$  Day of Gestation (Trt  $\times$  D) effects. \* $P \leq 0.05$ .

descriptive statistics. On day 220 of gestation, the average daily ambient temperature was  $-5.7$   $^{\circ}\text{C}$ , and the mud was frozen and could not be measured. Throughout the treatment period, the manually recorded temperatures of the MUD pens were comparable to the research station's measurements for soil temperature. Weather data obtained from the EARS weather station is presented in Fig. 2A–F as descriptive statistics. Of the 56 d cows were housed in the individual mud pens, there were 3 d where the mud was frozen (5% of the treatment period). Of the 56 d during the treatment period, cows experienced precipitation (rain or snow) for 27 d (48% of the treatment period). This accumulated to a total of approximately 176.5 mm of precipitation throughout the 56-d treatment period.

Prepartum cow mud score is presented in Fig. 3. There was evidence of a treatment  $\times$  day of gestation ( $P < 0.01$ ) effect for mud score. By design, all cows in both treatments started the study at day 213 of gestation with mud scores of 1 (no tag; clean hide). As expected, cows in the MUD treatment continued to increase their mud scores as the treatment period progressed, having significantly greater scores compared with CON cows every week for the rest of the experiment ( $P < 0.01$ ). In weeks where mud scores decreased in the MUD treatment, it had rained and cleaned some of the mud and tag that was previously on the cows. By the end of the treatment period, the MUD treatment's average mud score was 4.8, while the CON treatment had an average score of 2.0 ( $P < 0.01$ ).

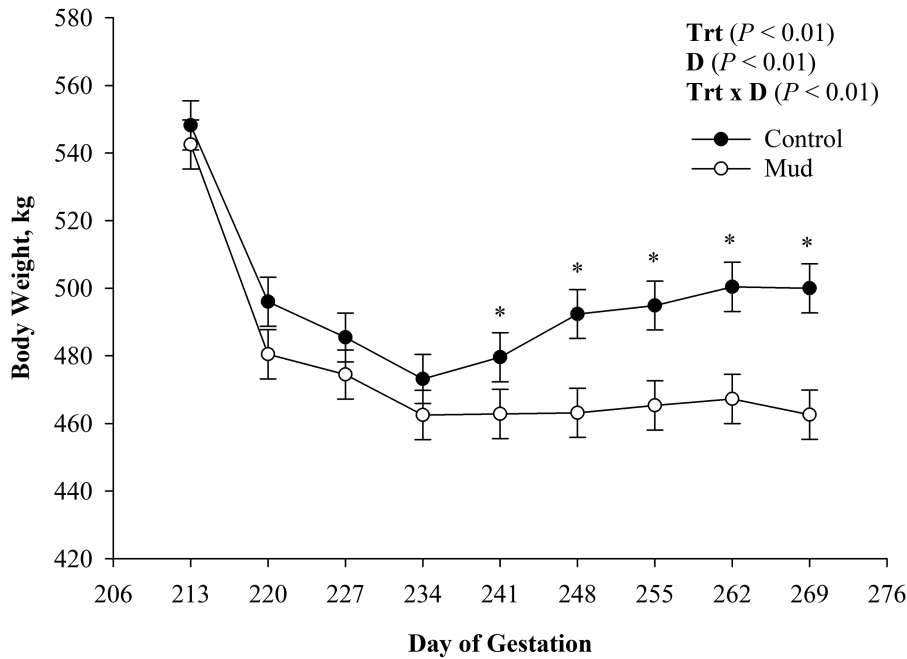
### Cow Prepartum Measurements

There were no dry matter refusals recorded for any cow throughout the treatment period. Prepartum cow BW is presented in Fig. 4. There was evidence of a treatment  $\times$  day of gestation effect for prepartum cow BW ( $P < 0.01$ ). Both treatment groups began the treatment period with similar BW ( $P = 0.53$ ). Beginning on day 241 of gestation, the CON cows

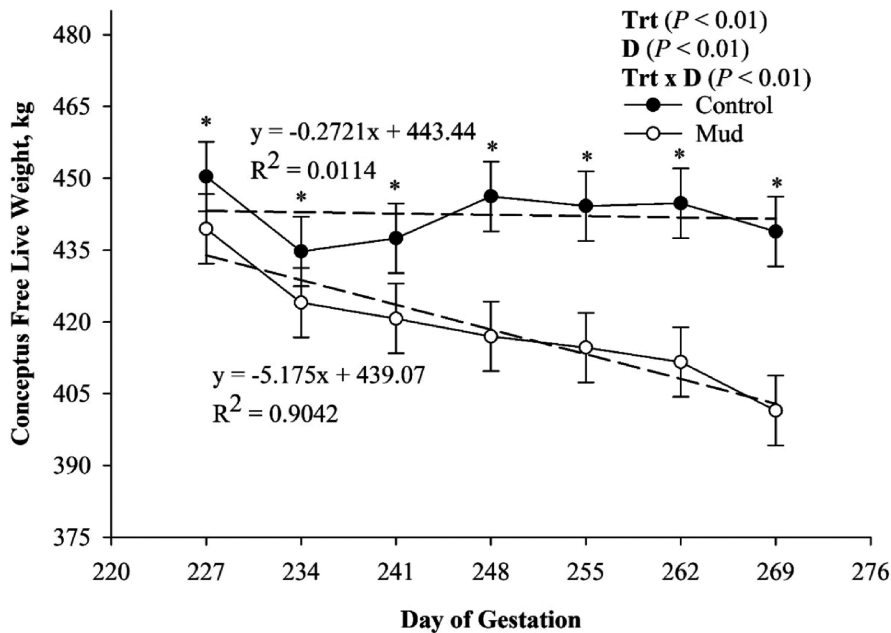
had greater BW compared with cows in the MUD treatment ( $P < 0.01$ ). Though the CON and MUD treatments started at similar BW at day 213 of gestation and consumed the same amount of dry matter throughout the treatment period, the CON cows were 37.4 kg heavier at day 269 of gestation. A similar treatment  $\times$  day of gestation effect was observed when the estimated CFLW was calculated ( $P < 0.01$ ; Fig. 5). Excluding the first 2 wk of the treatment period to allow for adaptation to the treatment diet, a linear regression was performed from day 227 to day 269 of gestation for CFLW (Fig. 5). The slopes of the regressions indicate that cows in the CON treatment lost 0.3 kg/wk, while cows in the MUD treatment lost 5.2 kg/wk. This indicates that cows in the CON treatment were able to maintain their estimated CFLW and cows in the MUD treatment were not able to maintain their estimated CFLW throughout the treatment period, even though they consumed the same amount of net energy.

Prepartum cow BCS is presented in Fig. 6. There was evidence of a treatment  $\times$  day of gestation effect for prepartum cow BCS ( $P < 0.01$ ). Corresponding to prepartum cow BW, cows in both treatments started on day 213 of gestation at similar BCS ( $P = 0.62$ ). The MUD treatment lost body condition throughout the study while the CON treatment gained body condition. Body condition scores of the two treatments were not different on day 213 or day 220 of gestation; however, from day 227 to day 269 of gestation, the CON treatment had greater BCS compared with the MUD treatment ( $P < 0.05$ ). At the end of the treatment period, the CON treatment had gained approximately 1 BCS unit (BCS of  $4.1 \pm 0.2$  to  $5.1 \pm 0.2$ ;  $P < 0.01$ ) and the MUD treatment had lost approximately half a BCS unit (BCS of  $4.0 \pm 0.2$  to  $3.6 \pm 0.2$ ;  $P = 0.04$ ).

Prepartum cow back-fat thickness is presented in Supplementary Fig. S1. There was evidence of a treatment ( $P = 0.03$ ) and day of gestation ( $P < 0.01$ ) effect, but no evidence of a treatment  $\times$  day ( $P = 0.85$ ) effect for back-fat



**Figure 4.** Mean prepartum cow body weight  $\pm$  SEM measured weekly from day 213 of gestation (start of treatment period) to day 269 of gestation (end of treatment period) of pair-fed cows housed in  $23.6 \pm 5.8$  cm of mud (Mud) or wood chips (Control) presented with Treatment (Trt), Day of Gestation (D), and Treatment  $\times$  Day of Gestation (Trt  $\times$  D) effects. \* $P \leq 0.05$ .



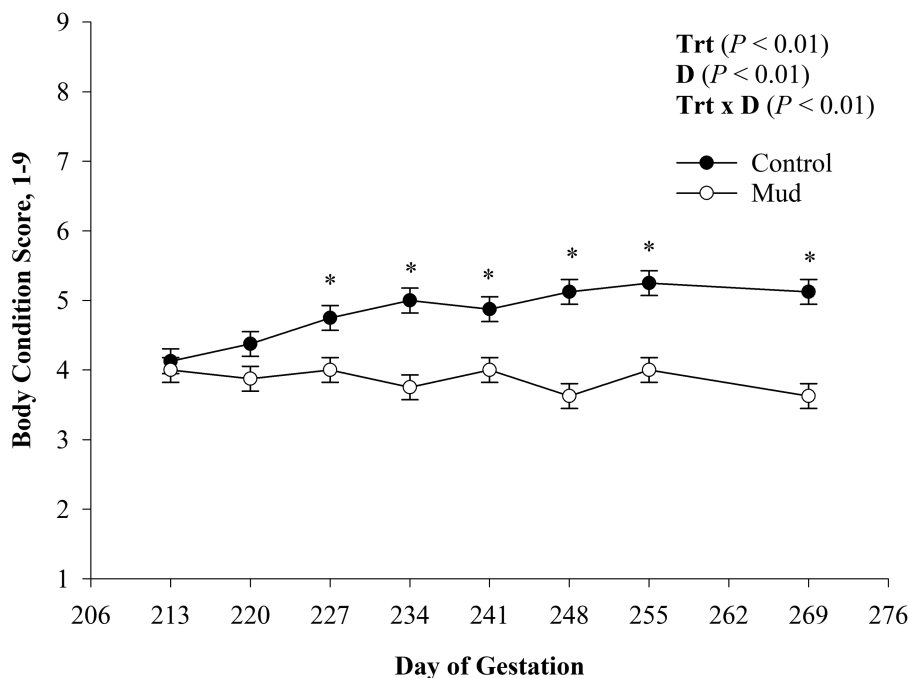
**Figure 5.** Mean prepartum conceptus-free live weight (CFLW)  $\pm$  SEM estimated using the weekly cow body weight from day 213 of gestation (start of treatment period) to day 269 of gestation (end of treatment period) of pair-fed cows housed in  $23.6 \pm 5.8$  cm of mud (Mud) or wood chips (Control) presented with Treatment (Trt), Day of Gestation (D), and Treatment  $\times$  Day of Gestation (Trt  $\times$  D) effects. \* $P \leq 0.05$ . CFLW was estimated using equations by O'Rourke et al. (1991).

thickness. Cows in the CON treatment had greater back-fat thickness throughout the entire study compared with the MUD cows ( $P = 0.03$ ). By the conclusion of the treatment period on day 269 of gestation, the CON treatment had a marginally significant increase in back-fat thickness compared with their day 213 back-fat thickness ( $P = 0.08$ ). Additionally, the MUD treatment had a significant increase

in back-fat thickness compared with their day 213 back-fat thickness ( $P < 0.01$ ).

Prepartum cow rump fat thickness is presented in [Supplementary Fig. S2](#). There was evidence of a treatment ( $P = 0.05$ ) effect for rump fat thickness; however, there was no evidence of a day of gestation ( $P = 0.97$ ) or treatment  $\times$  day of gestation ( $P = 0.48$ ) effect. The CON treatment had





**Figure 6.** Mean prepartum cow body condition score  $\pm$  SEM measured weekly from day 213 of gestation (start of treatment period) to day 269 of gestation (end of treatment period) of pair-fed cows housed in 23.6  $\pm$  5.8 cm of mud (Mud) or wood chips (Control) presented with Treatment (Trt), Day of Gestation (D), and Treatment  $\times$  Day of Gestation (Trt  $\times$  D) effects. \* $P \leq 0.05$ .

greater rump fat thickness throughout the treatment period compared with the MUD treatment ( $P = 0.05$ ).

### Cow Prepartum Plasma Metabolites

There was a marginally significant treatment  $\times$  day of gestation (Fig. 7;  $P = 0.09$ ) effect for plasma NEFA concentration. While both treatments increased their plasma NEFA concentration from day 213 of gestation to day 234 of gestation, cows in the MUD treatment had a marginally significant greater plasma NEFA concentration on day 220 of gestation compared with the CON treatment. Prepartum plasma insulin concentration is presented in Supplementary Fig. S3. There was no evidence of a treatment ( $P = 0.73$ ) or treatment  $\times$  day of gestation ( $P = 0.45$ ) effect; however, there was evidence of a day of gestation ( $P < 0.01$ ) effect for plasma insulin. Both treatments experienced a decrease in plasma insulin concentration as the treatment period progressed. Prepartum total plasma protein concentration is presented in Supplementary Fig. S4. There was no evidence of a treatment ( $P = 0.26$ ), day of gestation ( $P = 0.20$ ), nor treatment  $\times$  day of gestation ( $P = 0.85$ ) effect observed for total plasma protein. Prepartum plasma 3-methylhistidine concentration is presented in Supplementary Fig. S5. There was evidence of a day of gestation effect ( $P < 0.01$ ) such that both treatments had their least plasma 3-methylhistidine concentration on day 241 of gestation; however, there was no evidence of a treatment ( $P = 0.90$ ) or treatment  $\times$  day of gestation ( $P = 0.84$ ) effect.

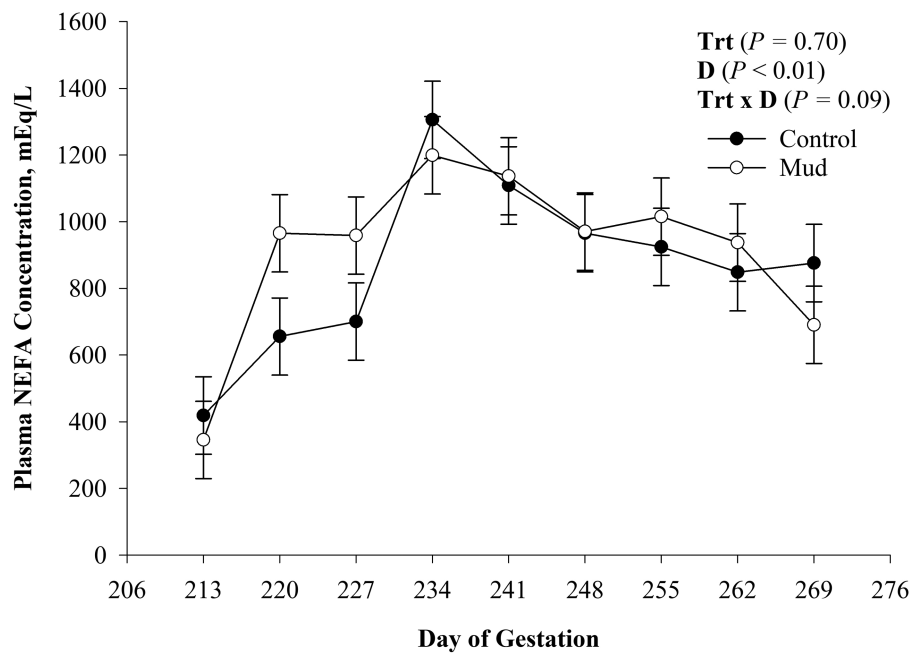
### Cow and Calf Data at Parturition

Cow BW at parturition, gestation length, calf birth weight, and calf total plasma protein are presented in Table 3. There was evidence for a marginally significant effect ( $P = 0.06$ ) for cow BW at parturition, such that cows in the MUD

treatment continued to weigh less at calving. Similarly, calf birth weight was not different between the two treatments ( $P = 0.66$ ). There was no evidence for a difference in gestation length (days) between treatments ( $P = 0.21$ ). Additionally, there was no evidence for differences in calf total plasma protein ( $P = 0.27$ ) within 24 h after birth.

## DISCUSSION

There is evidence that environmental stressors such as cold temperatures, wind, and mud will increase the net energy requirements for cattle (NASEM, 2016). There is, however, no data on the effects of a muddy environment on the energy requirements of gestating cows. Moreover, it is unknown if calf birth weight will be affected when cows are housed in muddy environments during late gestation. In the present study, cows housed in muddy conditions from day 213 to day 269 of gestation lost greater BW and body condition compared with cows housed in pens with wood chip bedding. We speculate that all cows decreased their BW in the beginning of this study because of dietary adjustments that caused a decrease in rumen fill, as they were allowed ad libitum access to hay before the initiation of the study and were then transitioned to an energy-dense diet at the start of the treatment period. Although the cows in the MUD treatment were pair fed to the CON cow's maintenance and gestation requirements based on the CON cow's BW and the week of gestation, cows in the MUD treatment weighed 37.4 kg less at the end of the treatment period, and this difference persisted and was marginally significant at parturition. This led us to accept our hypothesis that mud increases a cow's net energy requirements beyond the NASEM (2016) recommendations for maintenance plus gestation, because the cows in the MUD treatment were not able to maintain their BW or body condition throughout late gestation. However, both treatments



**Figure 7.** Mean prepartum cow plasma non-esterified fatty acid concentration  $\pm$  SEM measured weekly from day 213 of gestation (start of treatment period) to day 269 of gestation (end of treatment period) of pair-fed cows housed in  $23.6 \pm 5.8$  cm of mud (Mud) or wood chips (Control) presented with Treatment (Trt), Day of Gestation (D), and Treatment  $\times$  Day of Gestation (Trt  $\times$  D) effects.

**Table 3.** Least squares mean  $\pm$  SEM cow body weight (BW) at parturition (kg), gestation length (d), calf birth weight (kg), and calf total plasma protein (g/100 mL) for pair-fed cows housed in  $23.6 \pm 5.8$  cm of mud (Mud) or wood chips (Control) from day 213 to day 269 of gestation

	Control	Mud	P-value
Cow BW at parturition, kg	488.3 $\pm$ 9.8	461.8 $\pm$ 9.8	0.06
Gestation length, d	278.5 $\pm$ 1.0	276.5 $\pm$ 1.0	0.21
Calf birth weight, kg	35.7 $\pm$ 1.4	36.4 $\pm$ 1.4	0.66
Calf total plasma protein, g/100 mL	9.0 $\pm$ 0.8	7.7 $\pm$ 0.8	0.27

started the treatment period at a BCS of approximately 4. This is below the recommendation of a BCS of 5 to 6 for cows at calving to avoid negative consequences on reproduction after calving (Richards et al., 1986; Soca et al., 2013). It is possible that all the cows in this study were not meeting their nutrient requirements prior to the start of the treatment period. This is a limitation of this study, as we do not know the maternal nutritional status before the start of the study, and previous nutrition and nutrient stores of the dam can influence the fetal nutrient supply (Barker and Clark, 1997). In addition, while cows in the CON treatment increased their BCS as the treatment period progressed, this could have been an observation bias caused by the mud score of the cows.

Since calf birth weight was not different, it was assumed that fetal growth was not different between the two treatments and the gravid uterine weight was subtracted from both treatment's weekly mean BW. The gravid uterine weight was subtracted from the total maternal weight each week to determine if cows in both treatments were able to maintain their estimated CFLW, as the estimated CFLW is an indication

of the ability of the diet to meet the net energy requirements for the cow. The linear regression of estimated CFLW from this study demonstrated the CON cows decreased their estimated CFLW by 0.3 kg each week, while the MUD cows decreased their CFLW by 5.2 kg each week. The small decrease in estimated CFLW each week from the CON cows suggests to us that the NASEM (2016) recommendations for net energy requirements for maintenance and gestation were sufficient within the climatic conditions of this study for the CON cows. The cows in the MUD treatment, however, did not consume sufficient dietary energy to maintain their CFLW. Using table 13-4 of the NASEM (2016) and the BCS of the cows in the MUD treatment at the start of this study (BCS =  $4.0 \pm 0.2$ ), it is estimated that 1 kg of empty BW loss is equivalent to 5.3 Mcal of energy. The decrease in estimated CFLW for the mud cows each week suggests an energetic cost of 27.5 Mcal/wk. This 27.5 Mcal/wk multiplied by the 8-wk treatment period suggests a total energetic cost of 220 Mcal. The average energetic cost of mud on a per-day basis for the cows in this study is estimated by dividing the 220 Mcal by the 56-d treatment period. This equation results in an estimated energetic cost of 3.9 Mcal/d because of the MUD treatment in this study. Using the NASEM (2016), we calculated the estimated additional net energy required for cows under our experimental conditions and heavily covered in mud and compared this with our estimation based on the current results. The NASEM (2016) equations estimated an additional 1.1 Mcal of net energy required for cows that are heavily covered in mud and exposed to the average climatic conditions that occurred during the treatment period in this study. This calculated value from the NASEM (2016) is approximately 2.8 Mcal less than the 3.9 Mcal/d that we have estimated the energetic cost of mud to be under our experimental conditions. At the start of the treatment period, on day 213 of gestation, we estimated the average net energy requirement for maintenance to be 8.40 Mcal/d for the cows in the MUD treatment. We suggest that the net energy requirement

for maintenance in gestating beef cows exposed to mud and the climatic conditions of this study should be increased by 46% to 12.3 Mcal/d to avoid a decrease in CFLW. It is possible that as the treatment period progressed and cows in the MUD treatment increased the mud and tag on their hides, that the actual BW difference between the two treatments was greater than what is reflected in the data presented, and that this energetic cost may be slightly underestimated because of this. Additionally, [Dijkman and Lawrence \(1997\)](#) demonstrated that the energetic cost of walking was increased for cattle and buffaloes as the soils became more saturated with water. It is likely that muddy conditions increase a cow's net energy requirements from a temperature homeostasis standpoint, but also by making locomotion more difficult in muddy conditions. While we did not measure locomotion and cows were housed in relatively small pens that limited locomotion and exercise, it is possible that our estimate for the energetic cost of mud may increase in pasture settings where cows will have to travel greater distances to feed and water under muddy conditions.

To evaluate body fat mobilization in the cows, we measured 12th rib back-fat thickness, rump fat thickness, and plasma NEFA concentration. When nutrients are restricted, fat is mobilized first, followed by muscle, and lastly bone ([Chilliard et al., 1998](#)). Body tissues are generally mobilized in the inverse order that they were deposited, as the latest maturing tissues are more sensitive to mobilization because of their lower physiological priority. Additionally, adipose tissue mobilization occurs in the order of subcutaneous fat being mobilized first, followed by perirenal, omental/mesenteric, intermuscular, and finally intramuscular fat ([Chilliard et al., 1998](#)). Therefore, we hypothesized that cows in the MUD treatment would be in a negative energy balance and would first mobilize their subcutaneous fat stores to provide energy for maintenance and fetal growth. While the cows in the MUD treatment consistently lost BW each week and lost body condition over the treatment period, we reject our hypothesis, as we did not observe this same decrease in 12th rib back-fat thickness. [Long et al. \(2021\)](#), however, did observe a decrease in 12th rib back-fat and ribeye area when mature cows were nutrient restricted to 70% of the [NASEM \(2016\)](#) recommendations for total net energy requirements from day 158 of gestation until parturition. In this study of [Long et al. \(2021\)](#), however, it is unclear if net energy requirements were adjusted daily or weekly for each individual cow. Additionally, the authors state that requirements were estimated per individual based on BW. It is unclear if cows were provided 70% of [NASEM \(2016\)](#) recommendations for total net energy requirements based on their own BW, or if this was 70% of the control treatment receiving 100% of their total net energy requirements from day 158 of gestation to parturition, making it difficult to directly compare the present results. Furthermore, while we did not observe a decrease in rib fat, the cows in the MUD treatment had consistently less rump fat than the cows in the CON treatment throughout the study. We additionally measured plasma NEFA concentration as an indicator of fat catabolism. Previous studies have observed increases in plasma NEFA concentration when cattle were exposed to cold stress ([Thompson and Clough, 1972](#); [Young, 1975](#); [Broucek et al., 1987](#); [Andreoli et al., 1988](#); [Tucker et al., 2007](#); [Nonnecke et al., 2009](#)). We observed a marginally significant increase in plasma NEFA concentration in the cows allocated to the MUD treatment on day 220 of

gestation, and both treatments had their greatest concentration on day 234 of gestation. During the first 3 wk of the treatment period, the CON cows decreased their BW while simultaneously increasing their BCS. This indicates that we were meeting or exceeding the [NASEM \(2016\)](#) requirements for maintenance plus gestation and that this was likely a reduction in rumen fill and a reduction in total gastrointestinal tract size. We believe this peak in plasma NEFA concentration in both treatments could be because of cows adjusting from consuming ad libitum hay before the treatment period to the diet that was fed during the treatment period. During the dietary transition in the first 3 wk of the treatment period, it is likely that the cows regulated their basal metabolic rate in response to the new diet and became more efficient on a limit-fed, energy-dense diet. This adjustment to basal metabolism could have resulted in an abnormal metabolic profile in the blood and could be why we observed increases in plasma NEFA concentration in both treatment groups during the first 3 wk of the study. Not only were cows limit-fed, but blood sampling occurred before cows were fed on the weigh days. Therefore, it is possible that sampling time may have affected NEFA concentrations, as both treatment groups could have increased their plasma NEFA concentrations in response to having not eaten since the previous morning when they were fed their dry matter allowance of the limit-fed diet.

To further evaluate energy status in the cows, we measured plasma insulin concentration, total plasma protein, and plasma 3-methylhistidine concentration. We hypothesized that cows in the MUD treatment would be in a negative energy balance and would therefore have decreased plasma insulin concentration compared with cows in the CON treatment. Both treatment groups, however, decreased their plasma insulin as the treatment period progressed. This aligns with previous studies that have observed decreases in serum insulin as gestation progresses in cows ([Grigsby et al., 1974](#); [Rhoads et al., 2004](#); [Rico et al., 2015](#)). Total plasma protein and plasma 3-methylhistidine were quantified to evaluate protein catabolism, and more specifically skeletal muscle breakdown. The catabolism of actin and myosin in skeletal muscle leads to an increase in 3-methylhistidine in circulation, and plasma concentration of 3-methylhistidine is a good indicator for protein metabolism in cows because it is not further metabolized in the body ([Harris and Milne, 1981](#); [Houweling et al., 2012](#)). We hypothesized that cows in the MUD treatment would be in a negative energy balance and would therefore mobilize both fat and protein stores to provide for fetal growth. While we reject our hypothesis that cows in the MUD treatment would have increased total plasma protein and plasma 3-methylhistidine, it is possible that we are not accurately measuring protein accretion as this involves both degradation and synthesis ([Jones et al., 1990](#)). Protein accretion is greatly impacted by nutritional status, and nutritional restriction has been observed to decrease both rates of synthesis and degradation ([Haverberg et al., 1975](#)). While the cows in the MUD treatment were not necessarily restricted of all nutrients, it is possible that energy-restricted cows may use the supplied protein as an energy source and therefore could affect protein synthesis and degradation rates. [Jones et al. \(1990\)](#) found that when beef steers were restricted feed, myofibrillar protein degradation and synthesis both decreased during the restriction period and increased following the subsequent repletion period. While it is difficult to compare gestating cows to growing beef steers, it is possible that gestating cows will

similarly decrease their myofibrillar protein degradation and synthesis in response to being energy restricted.

In this study, our initial hypothesis was that cows in the MUD treatment would mobilize their two largest stores of maternal tissues, muscle and fat, to support vital maintenance functions and to support fetal growth. Our data, however, do not support this hypothesis as we did not observe treatment differences in plasma insulin, total plasma protein, or plasma 3-methylhistidine. Though the data do not support our hypothesis, there is still evidence that the cows in the MUD treatment were mobilizing their tissue stores, as the estimated CFLW continued to decrease but fetal growth was unaffected by the restriction imposed on net energy requirements. We believe that we were meeting the cows' requirements for crude protein, and that the MUD treatment was affecting their net energy requirements. Therefore, it is possible that the weight loss that was observed in the MUD treatment cows is partly because of a decrease in the mass of the visceral organs, as we were likely meeting all nutrient requirements except for their net energy requirements. It has been documented in ruminants that metabolically active tissues such as the gastrointestinal tract and the liver will decrease in size and activity under feed restriction (Murray et al., 1977; Ferrell et al., 1986; Aziz et al., 1993; Yambayamba et al., 1996; Hornick et al., 2000). The pancreas (Aziz et al., 1993) and spleen (Yambayamba et al., 1996) have also been reported to decrease in size during nutrient restriction. Though we did not measure any of these components, as this would have required slaughter of the cows, we speculate that the BW lost by the cows in the MUD treatment is partly due to shrinking of several of the visceral organs.

It has been speculated that malnourishment of the dam during different trimesters of gestation will have impacts on offspring birth weight and subsequent growth and health, depending on the severity of the nutrient restriction imposed (Hight, 1968). The dam acts as a buffer between environmental stressors and the fetus. Therefore, it seems likely that development of the fetus would only be affected if the environmental demands exceed the nutrient intake of the dam plus the maternal tissue stores that she is able to mobilize (Hight, 1968). While there has been evidence that nutrient restriction of the dam during late gestation decreases birth weight (Tudor, 1972; Corah et al., 1975; LeMaster et al., 2017), we did not observe a difference in birth weight between the CON and MUD treatments. LeMaster et al. (2017) observed a 39.4 kg decrease in BW when cows were nutrient restricted during the last 90 to 100 d of gestation, which is similar to Tudor (1972) that recorded a 36.8 kg decrease in BW when cows were provided a low plane of nutrition during the last 100 d of gestation. Alternatively, Corah et al. (1975) only observed a 5.8 kg decrease in BW when heifers were fed a low energy ration during the last 100 d of gestation. We observed similar decreases in cow BW during late gestation as LeMaster et al. (2017) and Tudor (1972); however, we did not observe similar differences in calf birth weight. In beef cattle, skeletal muscle development matures by approximately 210 d of gestation, and it has previously been suggested that maternal nutrient restriction after this period has no major impacts on the number of muscle fibers present (Du et al., 2010). It is likely that any observed decrease in birth weights of calves due to nutrient restriction during the last trimester of gestation is because of a reduction in muscle fiber size, as the fibers cannot undergo hypertrophy nearing

parturition (Du et al., 2010). It is possible that the calves in this study had already undergone primary muscle fiber development, and therefore the energy restriction of the cows in the MUD treatment did not affect the number of muscle fibers in the calves. Additionally, with no differences being observed in calf birth weight or calf total plasma protein concentration, we speculate that the mature cows in this study were able to mobilize their own body stores to adequately support fetal growth. Furthermore, while it seems that mature cows are capable of mobilizing their body stores to provide for fetal growth, this energy restriction imposed by the MUD treatment may be detrimental to first calf heifers, as they are expected to continue to grow to their mature weight while also growing a fetus. This agrees with several other studies that have demonstrated the ability of mature cows to reduce BW and BCS during late gestation with no effects on their progeny (Bond and Wiltbank, 1970; Mulliniks et al., 2015).

In conclusion, the present study demonstrates the negative effects that a muddy environment can have on cow BW and body condition. Cows housed in mud decreased their CFLW during the treatment period, although they were being pair fed to the cows that were housed in wood chip bedding. This decrease in CFLW equated to the cows in the muddy environment needing approximately 3.9 Mcal of extra energy per day because of the mud treatment. Though cows in the mud treatment significantly decreased their BW and body condition, there were no notable differences in calf birth weight; however, the difference in cow BW was still marginally significant at parturition. We believe that we were meeting all other nutrient requirements of the cows except for their net energy requirements. We conclude that the cows in this study when treated with a muddy pen utilized their body tissue for homeostatic processes such as thermogenesis. The effects of muddy pen conditions on cows at lesser BW and BCS than the cows reported in the present study are unknown. Additionally, these results indicate that if mature cows are in good-to-moderate body condition, they are capable of mobilizing their own body stores to adequately provide for fetal growth.

## Supplementary Data

Supplementary data are available at *Translational Animal Science* online.

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## Conflict of Interest Statement

None declared.

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