EFFECTS OF FEEDING DIETARY CATION-ANION DIFFERENCE ON ANIMAL PERFORMANCE

by

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(Under the Direction of John Bernard)

ABSTRACT

Effects of supplementing calcareous marine algae (CMA) and optimal length of feeding negative dietary cation-anion difference (DCAD) were evaluated. In trial one, no differences were observed in DMI, milk yield, or concentrations of milk components when cows were fed diets supplemented with CMA starting either prepartum or postpartum. Milk protein yield was higher for cows fed CMA prepartum compared with those without supplemental CMA. Milk fat yield and ECM were higher for cows fed CMA prepartum during week 2 and 6. Concentrations of serum Na were greater and urinary K was lower for cows fed sodium bicarbonate postpartum compared with CMA. In trial two, no differences were observed in DMI, milk yield and components due to the length of time cows were fed a negative DCAD diet prepartum. Increasing time feeding negative DCAD prepartum linearly decreased prepartum serum total protein, albumin, and Ca, and increased postpartum serum total protein and globulin concentrations. Feeding a negative DCAD diet starting from 4 wk prepartum resulted in the lowest urine pH on day of calving and highest milk protein percentage during wk 3, 5, and 6 compared with cows fed a negative DCAD diet for 3 or 6 wk prepartum. These trials indicate that feeding CMA prepartum does not affect DMI or serum metabolites prepartum, but supported

improvements in animal performance postpartum. Feeding CMA postpartum supported similar performance and serum metabolite concentrations compared with feeding sodium bicarbonate. Extending the length of time a negative DCAD is fed prepartum up to 42 days does affect select serum metabolites, but has no negative impact on health, milk production and components.

INDEX WORDS: DCAD, Milk yield, Milk composition

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DEDICATION

I would like to dedicate this dissertation to my parents, who have always given me support and encouragement. Thank you!

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CHAPTER 1

INTRODUCTION

The transition from the late gestation to that of lactation is characterized by enormous physiological adaptations on dairy cows that significantly impact the subsequent lactation performance and reproduction. During this period of transition from gestation to lactation, cows are most susceptible to metabolic disorders such as milk fever, ketosis, fatty liver, and displaced abomasum. Milk fever, or hypocalcaemia, is a metabolic disorder commonly occurring at parturition due to low blood calcium concentrations. Cows that have milk fever have also been observed to develop other health disorders such as retained placenta and displaced abomasum that are also associated with a reduced smooth muscle contractibility due to low blood calcium concentrations.

Anionic diets, containing higher concentrations of Cl and S compared with Na and K, are fed prepartum to alter the normal calcium homeostatic control processes and elevate blood calcium concentrations at calving, thus reducing the incidence of milk fever. Improved DMI and milk yield postpartum have been observed when cows were fed with anionic diets prepartum in the studies of Block (1984) and DeGroot et al. (2010). Increasing length of exposure to anionic diets prepartum increased ECM and milk protein yield (Degaris et al., 2008). However, no differences were observed on DMI or milk yield when cows were exposed to anionic diets starting either 21 d or 42 d prepartum (Weich et al., 2013). After calving, feeding cationic diets postpartum to achieve an alkalinizing effect is optimal to help prevent rumen acidosis and further improve DMI, milk yield and composition (West et al., 1991, 1992).

The objective of these projects were to investigate 1) the effect of supplemental calcareous marine algae fed pre- and postpartum and 2) the effect of length of feeding prepartum of an anionic diet prepartum on select blood and urine metabolites during late gestation and early lactation, and milk yield and composition. Results of these trials will provide new information on the potential of supplemental calcareous marine algae for improving the transition of cows from late gestation into lactation and to determine if there are any negative or positive effects of feeding an anionic diet for longer than 21 d prepartum which is the current recommendation.

CHAPTER 2

LITERATURE REVIEWED

Dietary Cation-anion Difference

The concept of dietary cation-anion difference (DCAD) was first used by Dishington (1975) and Mongin (1981). The DCAD is the balance of the dietary cations and anions. Formulating diets with different DCAD concentrations has been used as a means of manipulating Ca homeostasis prepartum and blood buffering postpartum and further improves animal performance. After calving, the demand of Ca is increased due to large amounts of Ca secreted into milk. Subclinical hypocalcaemia commonly occurs as a result of low circulating Ca concentrations and can lead to clinical hypocalcaemia. To maintain Ca homeostasis, parathyroid hormone is secreted and initiates bone calcium reabsorption and renal production of 1, 25-dihydroxyvitamin D. Bichara et al. (1990) reported that metabolic acidosis enhanced circulating parathyroid hormone. Cations such as Na, K, and Mg have a positive charge and create a more alkaline metabolic state, while anions such as Cl, S, and P with a negative charge promote a more acidic metabolic state. According to Oetzel (1991), feeding anionic diets prepartum, high in Cl^- and S^{2-}, is effective for preventing milk fever. Goff et al. (2014) reported that plasma Ca concentrations increased at a significantly lower rate in cows fed a DCAD of 18.8 mEq/100g of DM compared with -18.1 mEq/100g when receiving a synthetic PTH injection, with a lower production of 1, 25-dihydroxyvitamin D as well. Under heat stress, body temperature and respiration rate are elevated in lactating cows, resulting in additional loss of CO_2 and reducing blood carbonic acid content (Benjamin, 1978). To compensate for respiratory alkalosis, cows

excrete bicarbonate in the urine to maintain carbonic acid:bicarbonate ratio in the blood. Elevated pCO_2 and urinary bicarbonate excretion were observed for lacatating cows fed a DCAD (Na+K-Cl) of 59.3 compared with 29.7 mEq/100g, suggesting increased buffering capacity with high DCAD (Wildman et al., 2007a). West et al. (1991) reported increased DMI when lactating cows were fed DCAD (Na+K-Cl) at 19.14 compared with -16.66 mEq/100g under heat stress, along with blood base excess (the deviation of buffer base of blood from the normal value) nearest to zero, suggesting that manipulating DCAD is effective for regulating blood acid-base status and improve animal performance. Serum total AA and essential AA concentrations and ratio of essential AA:total AA increased when cows were fed at a DCAD (Na+K-Cl) of 50 compared with 25 mEq/100g (Wildman et al., 2007a). These increases suggested that elevating DCAD improves AA availability for protein synthesis by sparing AA used to maintain acid-base balance. These authors also observed lower blood urea N with a DCAD of 58 compared with 41 mEq/100g, suggesting the possibility of enhanced microbial ammonia utilization by altering protein metabolism at high DCAD (Wildman et al., 2007b).

Calculating DCAD

For dairy cattle, DCAD is most commonly defined as the milliequivalents of Na + K – Cl – S per kilogram of DM. However, various equations have been proposed for calculating DCAD. These have included different ions in the equation (Horst et al., 1997) and inclusion of absorption coefficients to further refine the equation (Tucker et al., 1991; Delaquis and Block, 1995). Tucker et al. (1991) suggested S should be included due to its blood buffering effect and the acidification potency is relatively lower than Cl. The absorption coefficient for S was further proposed by Horst et al. (1997) indicating 60% of bioavailability of S based on NRC (1989).

Horst et al. (1997) also recommended Ca, Mg and P be included in the equation due to their contribution on acid-base balance, even though their bioavailability is not as high as Na, K or Cl.

A meta-analysis of 137 trials indicated there was no additional gain in model predictions or fit from the inclusion of multivalent ions other than S (Lean et al., 2006). This analysis also indicated that increasing dietary concentrations of Mg and decreasing P concentrations reduce milk fever risk markedly, which is contrast to the predicted effect of the DCAD equations proposed by Horst et al. (1997) and Tucker et al. (1991). Thus Lean et al. (2006) concluded that the best DCAD equation supported by the meta-analysis for predicting the risk of milk fever was [(Na + K) - (Cl + S)].

Blood Acid-base Chemistry

Tucker et al. (1988b) reported linear increases in blood pH as DCAD increased from -10 to 20 meq/(Na+k)-Cl/100g of dietary DM via decreasing blood bicarbonate concentration. Similar findings were observed by West et al. (1991) where blood pH and bicarbonate were lowest for cows receiving DCAD (Na+K-Cl) of -11.66 mEq/100g compared with those fed 19.14, 18.0 and 31.24 mEq/100g. Apper-Bossard et al. (2006) reported increased blood pH and HCO_3 as DCAD increased from 0, 15.0 to 30.0 mEq/100g. Partial pressure of CO_2 (pCO_2) in blood increased linearly because of elevated respiratory rates observed with increasing DCAD from -11.66 to 31.24 mEq/100g (West et al., 1991). Similar increases in blood pH, pCO_2, HCO_3 of dairy calves were observed as DCAD increased from 0 to 52 mEq/100g (Jackson et al., 1992). For pasture dairy systems, cows fed diets with a DCAD of 21 mEq/100 g had lower blood pH compared with DCAD of 52, 102 or 127 mEq/100g (Roche et al., 2003).

Blood Minerals

Feeding negative DCAD prepartum increases plasma Ca concentrations at parturition. According to Block (1984), plasma Ca and P concentrations at parturition were lower for cows offered the cation diet of 33.05 mEq/100g compared with the anion diet of -12.85 mEq/100g. Siciliano-Jones et al. (2008) reported higher plasma Ca concentrations at parturition for multiparous cows fed negative DCAD prepartum achieved by feeding supplemental chloride compared with positive DCAD. Increased plasma ionized Ca concentrations at calving were observed when DCAD was reduced from 0 to -15 mEq/100g (Moore et al., 2000). According to Grünberg et al. (2011), protein-corrected plasma Ca concentration was higher for cows fed a DCAD of -9 mEq/100g compared with 11 mEq/100g at 1 d postpartum.

Tucker et al (1988b) concluded that the plasma mineral concentrations were less responsive to dietary mineral manipulation than urinary mineral excretion. Tucker et al. (1988b) observed decreased serum Cl as DCAD increased from -10 to +20 mEq/100g, but serum Na and K were not affected which is explained by the accompanying removal of Cl with excretion of excess Na and K. No effect of prepartum anionic diets on plasma P, Mg and K was observed at calving in normal multiparous cows (Siciliano-Jones et al., 2008; Vagnoni and Oetzel, 1998). For dairy calves, plasma Ca increased linearly with increasing DCAD while plasma Mg and Cl decreased without effect on plasma Na (Jackson et al., 1992). However, a quadratic response of serum K and Mg on lactating dairy cows was observed by increasing dietary DCAD (Na+K-Cl) from -11.66 to 31.24 mEq/100g (West et al., 1991).

Urine Metabolites

Moore et al. (2000) observed that prepartum urine pH was reduced from 7.3 to 6.0 when DCAD was reduced from 0 to -15 mEq/100 g. This is consistent with the study of Vagnoni and

Oetzel (1998) where urinary pH and bicarbonate excretion were reduced when cows were fed anionic diets prepartum. These authors also reported a reduction in ion difference in urine by feeding anionic salts due to the relatively greater excretions of Cl^- and S^{2-} than Na^+ and K^+. Tucker et al. (1988a) reported that urine pH increased quadratically with increasing DCAD, which can be explained by the compensatory renal excretion of bicarbonate. Also urine cation-anion balance increased as DCAD increased. Increased urine pH was also observed in dairy calves as DCAD (Na+K-Cl) increased from 0 to 52 mEq/100 g of dietary DM (Jackson et al., 1992). The authors also observed a linear decrease of urinary Ca, Mg and Cl excretion, while P excretion increased as DCAD increased from 0 to 52 mEq/100 g.

Rumen Parameters

According to Tucker et al. (1988a), rumen pH increased linearly as DCAD increased from -10 to 20 mEq/100g (Na+K-Cl) without affecting rumen fermentation patterns. However, Apper-Bossard et al. (2010) reported that increasing DCAD from 0 to 30 mEq/100g (Na+K-Cl) did not affect mean rumen pH, molar proportion of VFA or fiber digestibility.

Feed Intake

Some studies have reported decreased DMI when feeding anionic diets due to poor palatability (Oetzel, 1993; Joyce et al., 1997; Moore et al. 2000; Charbonneau et al., 2006). However, other studies have reported improved DMI postpartum of multiparous cows fed anionic diets prepartum (DeGroot et al., 2010), which was consistent with the study of Joyce et al., 1997; and Goff and Horst, 1998). This positive postpartum response to an anionic diet fed prepartum is related to improved Ca homeostasis which prevented milk fever and facilitated increased DMI.

Hu and Murphy (2004) conducted a meta-analysis using the results from 17 published trials examining the effects of postpartum DCAD ranging from -19.1 to 63.57 mEq/100 g DM on DMI and performance. Maximum DMI was achieved at a DCAD of 40 mEq/100 g of DM, but DMI was depressed when DCAD was negative, which may be explained by the unfavorable palatability or metabolic acidosis induced by the anionic salt source (Hu and Murphy, 2004). The optimal postpartum DCAD to maximize DMI was 18.0 and 19.14 mEq/100g (Na+K-Cl), during cool and hot environment respectively (West et al., 1991). Also West et al. (1992) reported a linear increase in DMI as DCAD (Na+K-Cl) increased from 12 to 46 mEq/100g. According to Chan et al. (2005), DMI (kg/100 kg of BW) was greater for diets with DCAD of [(Na+K)-(Cl-S)] of 20 and 35 compared with 50 mEq/100g postpartum. Also, Roche et al. (2003) demonstrated that DCAD greater than 52 mEq/100 g may have deleterious effects on DMI. Optimum DMI was observed for dairy calves (56 to 70 d after birth) fed a diet formulated for 37 mEq/100g of DM compared with 0, 21 and 52 mEq/100 g (Jackson et al., 1992).

Lactating Performance

According to the study of Block (1984), feeding negative DCAD prepartum increased milk yield, which is also observed by Siciliano-Jones et al. (2008) that supplementing negative DCAD prepartum increased milk yield of cows with 3 or more lactations. This positive postpartum effect of feeding negative DCAD prepartum is potentially achieved by improved blood Ca circulation at parturition. DeGroot (2004) reported feeding anionic prepartum increased milk yield resulting from increased DMI postpartum for multiparous cows. No difference was observed in percentage and yield of fat, protein or 3.5% FCM for primiparous cows fed anionic diets versus control.

Various studies showed that increasing DCAD postpartum elevated milk yield. Tucker et al. (1988a) reported that increasing DCAD from -10 to +20 mEq/100g supported increased milk yield without any change in fat yield. These authors also reported highest milk protein percentage and yield with a DCAD of 0 mEq/100g. According to West et al. (1991), increasing dietary DCAD linearly increased milk yield and 4% FCM in both the cool and hot environment. However, Roche et al. (2003) reported decreased milk protein yield and numerical decreases in yield of milk, fat, and lactose as DCAD from 21 to 127 mEq/100g

Health

Milk fever (parturient paresis) is a metabolic disorder typically associated with parturition and beginning lactation due to the excessive excretion of Ca with the onset of lactation. Supplementing anions can be an effective way to elevate total or ionized blood Ca concentrations, thus preventing milk fever (Ender and Disghington, 1967; Dishington, 1975; Goff and Horst, 1998; Oetzel et al., 1988). Block (1984) did not observe any cases of milk fever when cows were fed an anionic diet, whereas cows fed a cationic diet had a milk fever incidence rate of 47.4%. According to Goff et al. (2014), milk fever affects approximately 5% of dairy cows each year, and subclinical hypocalcaemia may affect half of all multiparous cows.

Curtis et al. (1983) reported significant associations between parturient hypocalcaemia and dystocia, retained fetal placenta, ketosis, mastitis and coliform mastitis. A path analysis conducted by Curtis et al. (1985) showed that a cow that develops one disorder is at greater risk for developing other disorders. Siciliano-Jones et al. (2008) reported that the prevalent of clinical ketosis was numerically higher in cows fed positive DCAD diet compared with negative DCAD prepartum diet, and 10.5 and 7.3, respectively. Hypocalcemia at parturition further exacerbated the decline in the peripheral blood mononuclear cells releasable Ca^{2+} stores (Kimura et al.,

2006). Decreased rumination and smooth muscle contraction was observed when hypocalcaemia was induced through infusion of Na_2EDTA (Jorgenson et al., 1998). According to Martinez et al. (2014), subclinical hypocalcaemia induced by infusion of a 5% EGTA solution decreased rumen contractions, percentage of neutrophils undergoing phagocytosis and oxidative burst response after incubation of pathogenic bacteria.

Length of Feeding Negative DCAD

Most studies have focused on the effect of varying levels of DCAD, while limited research has been conducted on the length of adjusting a DCAD to cows in late gestation. Frequently, effects of negative DCAD were studied 21 d and 14 d prepartum (Chan et al., 2006; DeGroot et al., 2010). Degaris et al. (2008) reported that increasing length of exposure to anionic diets prepartum increased ECM and milk protein yield with a maximum response at 25 d and 22 d prepartum, respectively. No difference in DMI and milk yield was observed when cows were exposed anionic diets either 21 days or 42 days prepartum (Weich et al., 2013). Lean et al. (2006) speculated that longer exposure to a low DCAD prepartum may exacerbate the hypercalciuric effect, which is in contrast with study of Weich et al. (2013) where feeding negative DCAD 42 d prepartum tended to increase total blood Ca concentrations postpartum compared with 21 d. However, predictive models of DeGaris et al. (2010) reported increasing days exposed to negative DCAD prepartum would decrease prepartum blood calcium but would have no effect on postpartum blood calcium concentrations.

Buffering Effect of Calcareous Marine Algae

Calcareous marine algae (CMA) is the skeletal remains of the seaweed *lithothamnium calcareum,* harvested off the southwest coast of Ireland and contains Ca, Mg and other trace minerals. The minerals in CMA have been reported to support improved bone health profile and

mineralization (O'Gorman et al., 2012; Aslam et al., 2013; Slevin et al., 2014). Also the rumen buffering effect of CMA was widely investigated. Inclusion of multiple buffers on reducing the rate of pH decline was compared according to Cruywagen et al. (2007). They reported that the rumen pH resulting from CMA remained higher than the pH from bicarbonate treatment, and took shorter time to buffer the rumen acidity as 4 h for CMA compared with 7.7 for sodium bicarbonate and 13 h for limestone. The minimum rumen pH was 5.19 for limestone, 5.37 for sodium bicarbonate and 5.42 for CMA. They also concluded that supplementing 90 g/d of CMA may have a greater impact on rumen acidity than 180 g/d of sodium bicarbonate. The study of Calitz (2009) also confirmed that CMA had a higher buffering capacity compared to sodium bicarbonate, and there's no gain in overall buffering capacities when additional sodium bicarbonate were included compared with only contained CMA. According to Cruywagen et al. (2004), increasing the dose of CMA from 0.125 to 1.2% of dietary DM increased rumen pH throughout the period of measurement which was monitored 2 h before the morning feeding, 2, 6 and 10 h after this feed allocation. However the optimum dose of CMA to optimize milk yield and feed efficiency was 0.3% of dietary DM or 80 g/day (Cruywagen et al., 2004). Calitz et al., (2009) reported that daily intake of 80 g of CMA had lower acetate: propionate ratio and numerically higher milk yield compared with adding additional sodium bicarbonate in the diet. According to in vitro study of Mesgaran et al. (2013), the diet containing CMA and sodium bicarbonate had a relatively low acidogenic value and acidogenic value: in vitro dry matter disappearance, and could maintain a relatively high rumen fluid pH compared with bentonite or magnesium oxide.

Based on the above literature review, the following experiments were carried out to investigate the effect of supplementing negative DCAD prepartum and positive DCAD

postpartum on animal production and metabolic parameters. The first trial was conducted to determine the effect of supplementing CMA either starting prepartum or postpartum on DMI, milk yield and composition, blood and urine metabolites. The second trial will determine the optimal length of time feeding a negative DCAD, beginning either 42, 28 or 21 days prior to predicated calving date to maximize animal production.

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CHAPTER 3

EFFECTS OF FEEDING CALCAREOUS MARINE ALGAE TO HOLSTEIN COWS BEGINNING PREPARTUM ON POSTPARTUM PERFORMANCE AND SERUM METABOLITES[1]

[1] Wu, Z., J. K. Bernard, and S. J. Taylor. To be submitted to *J. Dairy Sci.*

Abstract

Thirty-six multiparous Holstein cows and 12 springing heifers were used in a 9 wk randomized design trial to determine the effect of feeding calcareous marine algae (CMA) from 3 wk prepartum through 6 wk postpartum on dry matter intake (DMI) , blood and urine metabolites, milk yield and composition. Within parity, animals were assigned randomly to one of four treatments with a 2 × 2 factorial arrangement. Prepartum diets were supplemented with 0 (CON) or 50 g/d CMA with a resulting DCAD of -5.17 and -3.99 meq/100g, respectively. Postpartum diets were supplemented with either 317 g/d sodium bicarbonate (NBC) or 100 g/d CMA providing a DCAD of 35.58 and 15.64 meq/100g, respectively. No differences were observed in prepartum DMI among treatments. Postpartum DMI, milk yield, percentage of milk fat, protein, lactose, and SNF were not different among treatments. Milk protein yield was higher for cows fed CMA prepartum compared with CON. Interactions of prepartum treatment and wk were observed for yield of milk fat and energy-corrected milk (ECM) because of higher yields for cows fed CMA during wk 2 and 6 compared with CON. An interaction of prepartum treatment and wk was also observed for milk efficiency (ECM/DMI) because of lower efficiencies for cows fed CMA during wk 1 and 4 compared with CON; however, efficiencies tended to reverse and were numerically higher for CMA compared with CON during wk 6. Serum Na concentrations were greater for cows fed CON prepartum or NBC postpartum compared with CMA. Postpartum urinary concentrations of Na exhibited an interaction among treatments and were higher for CON-NBC and CMA-NBC compared with CON-CMA and CMA-CMA. Similar interactions of treatments were also observed for serum urea N and creatinine postpartum. Postpartum urinary K concentrations were higher for cows fed CMA postpartum compared with NBC. Results of this trial indicate that cows fed CMA prepartum

does not affect DMI or serum metabolites prepartum, but supported higher milk protein yields and increased ECM with time postpartum. Performance and serum metabolite concentrations of cows fed CMA postpartum were comparable with that of cows fed NBC except for changes in serum and urinary concentration of Na that was related to dietary intake of Na.
KEY WORDS: DCAD, milk yield, milk composition

Introduction

Transition cows are highly susceptible to periparturient diseases due to tremendous physiological adaptation during that period. Milk fever, hypocalcaemia, is a metabolic disease occurring at parturition as a result of low blood Ca concentration and is economically important due to lower milk yield. Curtis et al. (1983, 1985) reported that cows diagnosed with clinical milk fever are at greater risk for developing other disorders such as dystocia, retained fetal placenta, ketosis or coliform mastitis. Feeding anionic diets prepartum has been shown to reduce clinical and subclinical hypocalcaemia (Oetzel et al., 1988; Goff et al., 1991). Incidence rate of milk fever was significantly reduced by feeding anionic diets compared with feeding cationic diets prepartum (Block, 1984). However, feeding cationic diets is recommended postpartum to prevent rumen acidosis and further improve DMI and animal performance postpartum (West et al., 1991, 1992; Hu and Murphy, 2004).

Calcareous marine algae (CMA, Calmin, Celtic Sea Minerals, Ireland.) is produced from *Lithothamnium calcareum* and contains Ca, Mg and other trace minerals. The minerals in CMA have been reported to support improved bone strength and mineralization (Aslam et al., 2010) and function as an effective rumen buffer based on in vitro (Calitz, 2009; Mesgaran et al., 2013) and in vivo (Cruywagen et al., 2007) research. However, limited data are available on effects of feeding CMA to dairy cattle in prepartum and postpartum diets. The objective of this study was

to evaluate the effects of feeding calcareous marine algae from 3 wk prepartum through 6 wk postpartum on DMI, milk yield and composition, blood and urine metabolites.

Materials and Methods

Thirty-six Holstein cows and 12 springing heifers were used in a randomized block trial with a 2 × 2 factorial arrangement of treatments beginning 3 wk prepartum through 6 wk postpartum. Cows were blocked by parity and expected calving date. All protocols were approved by the University of Georgia Institutional Animal Care and Use Committee.

Animals were trained to eat behind Calan doors (American Calan, Inc., Northwood, NH) before beginning the trial. Cows were housed in a 4-row free stall barn with supplemental evaporative cooling provided by fans and misters and were allowed free access to a grassed exercise lot. At calving, cows were moved to grassed lot or a box stall and returned to the free stall area after calving. Cows were fed once daily in amounts to provide a minimum of 5% refusal and had free access to water. The amount of feed offered and refused was recorded daily. Experimental diets were formulated to meet NRC (2001) requirements for late gestation and early lactation, respectively (Table 3.1). Supplemental CMA was included in prepartum diets at the rate of 50 g/d and in the postpartum diets at the rate of 100 g/d according to the manufactures recommendations. The CMA contains (DM basis) 30.00% Ca; 0.05% P; 6.00% Mg, and 0.70% K. A basal diet was mixed daily for prepartum or postpartum treatments (Model 3120 Reel Mixer, Kuhn North America, Brodhead, WI). Treatment premixes (Table 3.1) were prepared and included with the other ingredients prior to final mixing using a Data Ranger (American Calan, Inc.). The amount of feed offered and refused was recorded daily. Samples of dietary ingredients and TMR were collected three times each week and DM determined by drying in a forced air oven 50 °C for 48 h. Samples were ground to pass through a 6-mm screen, composited by week,

and a subsample ground to pass through a 1-mm screen. Samples were analyzed for concentrations of ash (AOAC, 2000), N (Leco FP-528 Nitrogen Analyzer, St. Joseph, MO), ADF, (AOAC, 2000), NDF (Van Soest et al., 1991), and minerals (AOAC, 2000).

After calving, cows were milked three times daily at 0000, 0800 and 1600. Milk weights were recorded electronically at each milking (Alpro, DeLaval, Kansas City, MO), totaled each day, and a weekly average calculated. Milk samples were collected from three consecutive milkings each week. Samples were shipped for next day delivery to Dairy One Cooperative (Ithaca, NY) for analysis of fat, protein, lactose, SNF, and MUN concentrations by mid-infrared spectrophotometric analysis using a Foss 4000 instrument (Foss North America, Eden Prairie, MN) as described by AOAC (2000).

Body weight of cows was recorded on three consecutive days at the beginning of the trial and during wk 3 and 6 postpartum and once immediately after parturition. Access to water and feed was restricted until measurements were recorded immediately after the 0800 milking. Body condition scores were assigned by two individuals when BW was recorded as described by Wildman et al. (1982).

Two whole blood samples were collected from the coccygeal vein at 0900 once during wk -3, -2, -1 prepartum, at calving, and wk 1, 2, 3 and 6 postpartum. One sample was used for determination of serum glucose, urea N, total protein, albumin, creatinine, total bilirubin, aspartate aminotransferase (AST), creatine kinase, gamma-glutamyltransferase (GGT), Ca, P, Mg, Na, K, Cl, bicarbonate, and anion gap using a Bodhringer Mannheim/Hitachi 912 automated chemistry analyzer (Roche Laboratory Systems, Indianapolis, IN). Serum was separated from the second sample and analyzed for non-esterified fatty acids (NEFA) concentrations using an enzymatic procedure (Waco Chemicals USA, Inc., Richmond, VA). Blood ketone

concentrations were determined using a Nova Max Ketone Strips and a Nova Max Plus reader (Nova Biomedical, Waltham, MA). Urine samples were collected at the same times as blood samples for analyses for urine pH and electrolyte concentrations as described above.

Data were analyzed using PROC MIXED procedures of SAS (SAS Institute, Cary, NC). The model included block, prepartum and postpartum treatment, week, and the appropriate interactions. Genetic merit (PTA of multiparous cows and ETA of springing heifers) was included as a covariate for production variables. Cow within treatment and block was included as a random effect and week as a repeated measure.

Results and Discussions

The chemical composition of the experimental diets is presented in Table 3.2. Nutrient concentrations were similar for prepartum diets and postpartum diets except that Na concentrations were greater for NBC than CMA postpartum as expected. The DCAD [(Na + K) - (Cl + S)] of the diets was -5.17, -3.99, 35.58, 15.64 meq/100g for CON, CMA prepartum, NBC, CMA postpartum, respectively.

Prepartum DMI was not different among treatments for CON and CMA and averaged 11.9 and 12.2 kg/d, respectively. Postpartum DMI, milk yield, and composition are presented in Table 3.3. No differences among treatments were observed in DMI, milk yield, percentage of milk fat, protein and lactose. Milk protein yield ($P = 0.0453$) was higher for the cows fed CMA prepartum compared with CON, 1.09 and 1.02 kg/d respectively. Interactions of prepartum treatment and wk were observed for milk fat yield ($P = 0.0222$, Figure 3.1) and ECM ($P = 0.0395$, Figure 3.2) because of higher yields for cows fed CMA during wk 2 and 6 compared with CON. An interaction of prepartum treatment and wk ($P = 0.0337$) was observed for milk efficiency (ECM/DMI, Figure 3.3) because of lower efficiencies for cows fed CMA during wk 1

and 4 compared with CON; however efficiencies tended to reverse and tended to be slightly higher for CMA compared with CON during wk 6. Daily milk yield exhibited an interaction of postpartum treatment and DIM (P = 0.0093) as milk yield was higher for NBC compared with CMA during 28-31 DIM but was not different during 1-27 or 32-42 DIM (Figure 3.4). No differences were observed among treatments in BW (Figure 3.5) or BW change throughout the trial.

Concentrations of select metabolites and minerals in serum and urine prepartum are presented in Table 3.4. Prepartum serum Na was higher (P = 0.0461) for CON compared with CMA; 143.32 to 142.55 meq/L, respectively. Prepartum urine pH tended to be higher (P = 0.0910) for CON compared with CMA, 7.35 and 7.00, respectively. No differences were observed in the other metabolites and minerals.

Concentrations of select metabolites and minerals in serum and urine of cows postpartum are presented in Table 3.5. Interactions of prepartum and postpartum treatments were observed for serum urea N (P = 0.0148) and creatinine (P = 0.0005). Concentrations of serum urea N were highest for CON-NBC, CMA-CMA, intermediate for CON-CMA, and lowest for CMA-NBC, 12.18, 11.70, 11.20 and 10.31 mg/dl, respectively. Concentrations of creatinine were highest for CON-NBC and CMA-CMA, intermediate for CMA-NBC, and lowest for CON-CMA, 0.77, 0.77, 0.70 and 0.65 mg/dl, respectively. Postpartum concentrations of serum Na were higher (P = 0.0154) for NBC compared with CMA, 140.60 to 139.81 meq/L, respectively. An interaction of prepartum and postpartum treatments (P = 0.0353) was observed for urine Na concentration which was higher for CON-NBC and CMA-NBC, and lower for CON-CMA, CMA-CMA, 121.21, 105.43, 41.24 and 59.55 mmol/L, respectively. Postpartum urine K (P =

0.0030) was higher for the cows fed CMA postpartum compared with NBC, 155.60 and 118.40 mmol/L, respectively.

Cows fed diets supplemented with CMA prepartum had higher milk protein yield and yields of milk fat and ECM during wk 2 and 6 compared with CON. The DMI ($P > 0.1$) of cows fed supplemental CMA prepartum was 2.5% greater compared with CON. While not different, this small improvement would have provided additional nutrients which would have maintained a more positive energy balance prepartum which is consistent with the observed increases in yield of milk fat and ECM during wk 2 and 6 compared with CON. Cruywagen et al. (2004) reported increased milk yield for 80 g/d of CMA compared with 33 g/d of CMA. In second trial, Cruywagen et al (2007) reported higher milk yield for cows fed diets supplemented with CMA compared with sodium bicarbonate or the control diet without supplemental buffers, and 31.6, 29.1 and 27.6 liters/cow, respectively. Additional research is needed to verify the effects of CMA on milk yield and composition.

Hu and Murphy (2004) conducted a meta-analysis of trials examining the response of lactating cows to increasing dietary DCAD and concluded that maximum DMI and milk yield occurred when dietary DCAD (Na+K-Cl) increased to 34 and 40 meq/100 g DMI, respectively. Similar results were also reported by Wildman et al. (2007) when postpartum DCAD was increased from 14.2 to 38.7 meq/100g. In current trial, the inclusion of CMA postpartum was as effective as NBC for maintaining milk yield and composition even though diets containing CMA had lower DCAD compared with NBC, 16 and 36 meq/100g, respectively. The diets in the current trial was based primarily on corn silage and supplemented with finely groud corn and would be expected to benefit from increasing dietary DCAD. Cruywagen et al. (2004, 2007) reported greater milk yield for cows fed CMA compared with sodium bicarbonate. These authors

attributed the improvements to the slow-release buffer effect and higher bio-availability of the Ca, which may explain the similar performance from CMA at lower DCAD level compared with NBC in our study.

Serum bicarbonate and mineral concentrations were not different among treatments except that Na was higher for cows fed CON prepartum and for cows fed NBC postpartum compared with those fed CMA. The reason for higher serum Na concentrations prepartum is not clear since dietary concentrations of Na were similar for both CON and CMA. The increased concentration of serum Na postpartum is in response to the higher dietary concentrations of Na for cows fed NBC postpartum compared with CMA, 0.77 and 0.34 % of DM, respectively. Urinary concentrations of Na were also higher; however postpartum urinary concentrations of K were lower for cows fed NBC compared with CMA. Dietary concentrations of K were slightly higher for NBC compared with CMA, 1.48 and 1.39 % of DM, respectively. The increased concentration of urine K observed with CMA is not related to dietary concentrations or intake. As the kidneys work to maintain homeostatic conditions, it would appear that greater amounts of K were excreted than Na for CMA to maintain electrostatic balance. Urinary pH tended to be higher prepartum for cows fed CON compared with CMA, but no differences were observed postpartum. Hu and Murphy (2004) reported a linear increase in blood bicarbonate concentrations as DCAD increased although concentrations of Na and K were not altered. These authors also reported increased urinary excretion of Na and K as DCAD increased and proposed that the increased excretion prevented any increase in blood concentrations.

Conclusions

Feeding CMA prepartum did not alter prepartum DMI, but supported higher milk protein yield postpartum and supported higher yields of milk fat and ECM during wk 2 and 6. Overall

efficiency (ECM/DMI) was not different among treatments, but was greater for CON during wk 1 and 4 but tended to reverse trends after wk 4. Feeding supplemental CMA postpartum maintained similar DMI, milk yield and composition as NBC. Changes in serum and urine metabolite concentrations primarily reflected differences in dietary mineral supplementation. The results of this trial suggest that feeding CMA prepartum potentially has positive effects on postpartum performance. The results of this trial suggest that supplemental CMA supports performance similar to that of NBC in corn silage based diets.

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Table 3.1. Ingredient composition of experimental diets (% of DM)

	Prepartum		Postpartum	
Ingredient	CON[1]	CMA	NBC	CMA
Corn silage	39.32	39.32	41.25	41.25
Alfalfa hay	6.21	6.21		
Ryegrass baleage			5.38	5.38
Bermudagrass baleage	5.17	5.17	1.79	1.79
Ground corn	10.35	10.35	14.89	14.89
Cottonseed hulls[2]	0.46	0.41		1.26
Citrus pulp	7.24	7.24	5.38	5.38
Soybean hulls	7.24	7.24	4.48	4.48
Brewers grains, wet	8.28	8.28	11.66	11.66
Megalac[3]			1.35	1.35
Biochlor[4]	5.59	5.59		
Soybean meal, 47.5% CP	6.21	6.21	4.93	4.93
AminoPlus[5]			1.79	1.79
Prolak[6]			3.14	3.14
Urea			0.18	0.18
Sodium bicarbonate[1]			1.26	
Magnesium oxide[1]	0.29	0.21	0.18	0.18
CMA[1,7]		0.46		0.39
Calcium carbonate[2]	1.32	0.99	0.32	
Calcium monophosphate	0.21	0.21	0.18	0.18
Salt			0.63	0.63
Potassium carbonate[2]			0.22	0.18
Potassium magnesium sulfate	0.21	0.21	0.09	0.09
Zinpro Availa-4[8]	0.08	0.08	0.04	0.04
Yeast culture	0.52	0.52	0.22	0.22
OmniGen-AF[9]	0.52	0.52	0.22	0.22
Vitamin E, 20,000 IU/454 g	0.04	0.04	0.02	0.02
Monensin, 3 g/454g[10]	0.54	0.54	0.23	0.23
Trace mineral – vitamin premix[11]	0.20	0.20	0.14	0.14

[1]CON = control diet, CMA = supplemental calcareous marine algae, and NBC = supplemental sodium bicarbonate

[2]Ingredients were blended into a premix and added to mixer prior to feeding.

[3]Calcium salts of long chain fatty acids, Arm Hammer Animal Nutrition, Church & Dwight Co., Inc. Princeton, NY

[4]Feed supplement providing a source of dietary anions, Arm Hammer Animal Nutrition, Church & Dwight Co., Inc. Princeton, NY

[5]Ruminally protected soybean meal, Ag Processing, Inc. Omaha, NE

[6]Marine and animal rumen undegradable protein supplement, H. J. Baker & Bros., Inc. Westport, CT

[7]Calcareous marine algae, Calmin, Celtic Sea Minerals, Cork, Ireland

[8]Organic zinc, manganese, copper, and cobalt, Zinpro Corporation, Eden Prairie, MN.

[9]Immune stimulant, Prince Agri Products, Inc., Quincy, IL

[10]Rumensin, Elanco Animal Health, Indianapolis, IN.

[11]Mineral-vitamin premix contained (DM basis):): 26.1% Ca; 0.38% Mg; 1.76% S; 144 ppm Co; 9,523 ppm Cu; 1,465 ppm Fe; 842 ppm I; 28,617 ppm Mn; 220 ppm Se; 25,343 ppm Zn; 4,210,830 IU/kg Vitamin A; 1,684,330 IU/kg Vitamin D; 21,045 IU/kg Vitamin E.

Table 3.2. Chemical composition of experimental diets fed prepartum and postpartum

	Prepartum		Postpartum	
Ingredient	CON[1]	CMA	NBC	CMA
DM, %	43.8 ±3.3	44.4 ± 3.3	39.5 ± 3.4	39.7 ± 3.3
	---------- % of DM ----------			
CP	16.6 ± 0.5	16.4 ± 0.5	17.0 ± 0.9	16.9 ± 0.9
ADF	24.9 ± 1.3	24.3 ± 1.9	24.1 ± 1.7	24.1 ± 1.7
NDF	41.6 ± 1.4	41.1 ± 1.7	41.8 ± 1.1	40.8 ± 2.4
Ash	7.59 ± 0.17	7.65 ± 0.19	8.90 ± 0.53	8.06 ± 0.40
Ca	1.08 ± 0.05	1.16 ± 0.09	0.88 ± 0.05	0.86 ± 0.08
P	0.41 ± 0.02	0.41 ± 0.01	0.48 ± 0.02	0.47 ± 0.02
Mg	0.40 ± 0.02	0.40 ± 0.02	0.34 ± 0.02	0.34 ± 0.03
K	1.26 ± 0.04	1.30 ± 0.03	1.48 ± 0.11	1.39 ± 0.11
S	0.35 ± 0.03	0.35 ± 0.03	0.24 ± 0.02	0.25 ± 0.04
Na	0.14 ± 0.02	0.15 ± 0.03	0.77 ± 0.06	0.34 ± 0.07
Cl	0.77 ± 0.02	0.78 ± 0.06	0.74 ± 0.06	0.68 ± 0.04
	---------- meq/100g ----------			
DCAD[2]	-5.17	-3.99	35.58	15.64
DCAD[3]	16.70	17.88	50.58	31.27

[1]CON = control diet, CMA = supplemental calcareous marine algae, and NBC = supplemental sodium bicarbonate.

[2]Calculated as: (Na + K) - (Cl + S)

[3]Calculated as: Na + K - Cl

Table 3.3. Dry matter intake, performance of cows fed diets CON or CMA prepartum with NBC or CMA postpartum

Prepartum	CON[1]	CMA	CON	CMA		*P*		
Postpartum	NBC	NBC	CMA	CMA	SE	Prepartum	Postpartum	Interaction
DMI, kg/d	18.0	19.8	19.4	20.0	0.9	0.1743	0.3526	0.4633
Milk, kg/d[d]	38.1	39.0	37.3	38.0	2.2	0.3585	0.3526	0.9864
Fat, %	4.02	3.96	4.01	4.24	0.25	0.7276	0.5884	0.5436
Fat, kg/d[c]	1.53	1.54	1.49	1.61	0.08	0.2918	0.3820	0.6223
Protein, %	2.71	2.85	2.71	2.81	0.08	0.1204	0.7763	0.7766
Protein, kg/d	1.03	1.11	1.00	1.07	0.04	0.0453	0.3754	0.5307
Lactose, %	4.75	4.74	4.75	4.72	0.04	0.6103	0.8849	0.7974
Lactose, kg/d	1.81	1.85	1.77	1.80	0.05	0.6625	0.8628	0.9323
SNF, %	8.37	8.38	8.41	8.37	0.08	0.8634	0.8755	0.7209
SNF, kg/d	3.19	3.27	3.12	3.18	0.09	0.6107	0.9795	0.9137
3.5% FCM,	41.3	41.9	40.2	42.6	1.6	0.2733	0.6618	0.7540
ECM, kg/d[c]	40.2	41.3	39.1	41.5	1.4	0.1428	0.7925	0.8876
ECM/DMI[c]	2.23	2.08	2.01	2.08	0.11	0.3262	0.4739	0.5176
MUN, mg/dl	11.20	10.07	9.95	10.49	0.55	0.5720	0.4342	0.1179

[1]CON = control diet, CMA = supplemental calcareous marine alge, and NBC = supplemental sodium bicarbonate.

[c]Interaction of prepartum treatment and week ($P < 0.05$).

[d]Interaction of postpartum treatment and day ($P < 0.05$).

Table 3.4. Concentrations of select metabolites and minerals in serum and urine of cows prepartum

	Treatment			*P*	
	CON[1]	CMA	SE	Treatment	TRT x WK
Serum					
NEFA, meq/L	0.82	0.83	0.16	0.9723	0.4483
Ketone, mmol/ml	0.38	0.37	0.04	0.6943	0.4194
Total protein, g/dl	6.51	6.44	0.12	0.6648	0.4161
Albumin, g/dl	3.50	3.48	0.06	0.7730	0.7050
Globulin, g/dl	3.02	3.00	0.14	0.9112	0.4732
Albumin/Globulin	1.18	1.14	0.09	0.8640	0.7512
Urea N, mg/dl	13.92	13.51	0.45	0.4863	0.6603
Creatinine, mg/dl	0.90	0.91	0.03	0.8756	0.3939
BUN/Creatinine	15.94	15.43	0.72	0.5954	0.4170
Total billrubin, mg/dl	0.17	0.17	0.05	0.9374	0.4823
Glucose, mg/dl	73.09	74.60	3.29	0.7290	0.7612
AST, U/L	75.46	74.93	4.68	0.9308	0.3786
Creatinine kinase, U/L	210.79	189.92	33.57	0.7889	0.5909
GGT, IU/L	13.80	13.02	1.30	0.6444	0.4172
Ca, mg/dl	9.21	9.08	0.11	0.3109	0.6691
P, mg/dl	6.09	6.28	0.15	0.3083	0.2002
Mg, mg/dl	2.42	2.36	0.05	0.3612	0.3208
Na, meq/L	143.32	142.55	0.29	0.0461	0.8850
K, meq/L	4.64	4.70	0.06	0.4879	0.7388
Cl, meq/L	104.55	104.86	0.43	0.5838	0.4934
Bicarbonate, mmol/L	28.15	27.73	0.43	0.4535	0.7686
Anion gap, mmol/L	14.75	15.52	1.04	0.5017	0.7030
Urine					
pH	7.35	7.00	0.16	0.0910	0.9116
Na, mmol/L	38.25	25.30	5.97	0.1039	0.0732
K, mmol/L	144.91	137.86	11.49	0.6387	0.4605
Cl, mmol/L	125.40	127.00	10.58	0.9076	0.4766

[1]CON = control diet and CMA = supplemental calcareous marine algae.

Table 3.5. Concentrations of select metabolites and minerals in serum and urine of cows postpartum

Prepartum	CON[1]	CMA	CON	CMA		*P*		
Postpartum	NBC	NBC	CMA	CMA	SE	Prepartum	Postpartum	Interaction
Serum								
NEFA, meq/L	1.92	1.92	2.31	2.29	0.33	0.9755	0.2306	0.9826
Ketone, mmol/ml	0.60	0.63	0.75	0.74	0.18	0.9463	0.4423	0.8810
Total protein, g/dl	7.01	6.92	7.15	7.01	0.16	0.4601	0.4330	0.8530
Albumin, g/dl	3.71	3.55	3.61	3.56	0.06	0.1029	0.4515	0.3366
Globulin, g/dl	3.30	3.36	3.55	3.45	0.18	0.9201	0.3371	0.6342
Albumin/Globulin	1.16	1.09	1.03	1.05	0.06	0.6564	0.1586	0.4384
Urea N, mg/dl	12.18[a]	10.31[b]	11.20[ab]	11.70[a]	0.51	0.1923	0.6026	0.0148
Creatinine, mg/dl	0.77[a]	0.70[ab]	0.65[b]	0.77[a]	0.03	0.3168	0.2696	0.0005
BUN/Creatinine	16.93	16.23	22.79	16.29	2.62	0.1516	0.2502	0.2431
Total billrubin,	0.26	0.28	0.25	0.24	0.04	0.9345	0.6075	0.7688
Glucose, mg/dl	52.47	53.45	49.45	52.14	2.66	0.4727	0.3755	0.7510
AST, U/L	97.93	99.61	90.81	96.48	6.70	0.5676	0.4252	0.7546
Creatinine kinase,	188.47	184.74	179.18	130.79	31.28	0.3875	0.2943	0.4574
GGT, IU/L	18.57	22.95	22.24	24.48	4.12	0.4030	0.5110	0.7848
Ca, mg/dl	9.53	9.38	9.41	9.37	0.10	0.3039	0.4873	0.6113
P, mg/dl	6.03	6.00	5.72	5.68	0.22	0.8953	0.1434	0.9815
Mg, mg/dl	2.37	2.36	2.43	2.36	0.04	0.3291	0.5566	0.5042
Na, meq/L	140.80	140.40	139.61	140.00	0.33	0.9981	0.0154	0.2120
K, meq/L	4.64	4.55	4.52	4.52	0.06	0.3626	0.1902	0.4343

Cl, meq/L	98.68	98.51	97.77	98.15	0.51	0.8297	0.1974	0.5681
Bicarbonate,	29.18	29.13	29.10	29.18	0.57	0.9818	0.9803	0.9109
Anion gap, mmol/L	17.59	17.34	17.39	17.39	0.49	0.7868	0.8640	0.7867
Urine								
pH	8.04	8.01	7.97	8.03	0.07	0.8105	0.6097	0.4791
Na, mmol/L	121.21[a]	105.43[a]	41.24[b]	59.55[b]	8.27	0.8723	<0.0001	0.0353
K, mmol/L	122.66	114.13	159.51	151.68	12.39	0.4890	0.0030	0.9762
Cl, mmol/L	61.76	60.60	71.22	66.89	5.67	0.6128	0.1525	0.7699

[1]CON = control diet, CMA = supplemental calcareous marine algae, and NBC = supplemental sodium bicarbonate.

[abc]Means in the same row with different superscripts differ ($P < 0.05$)

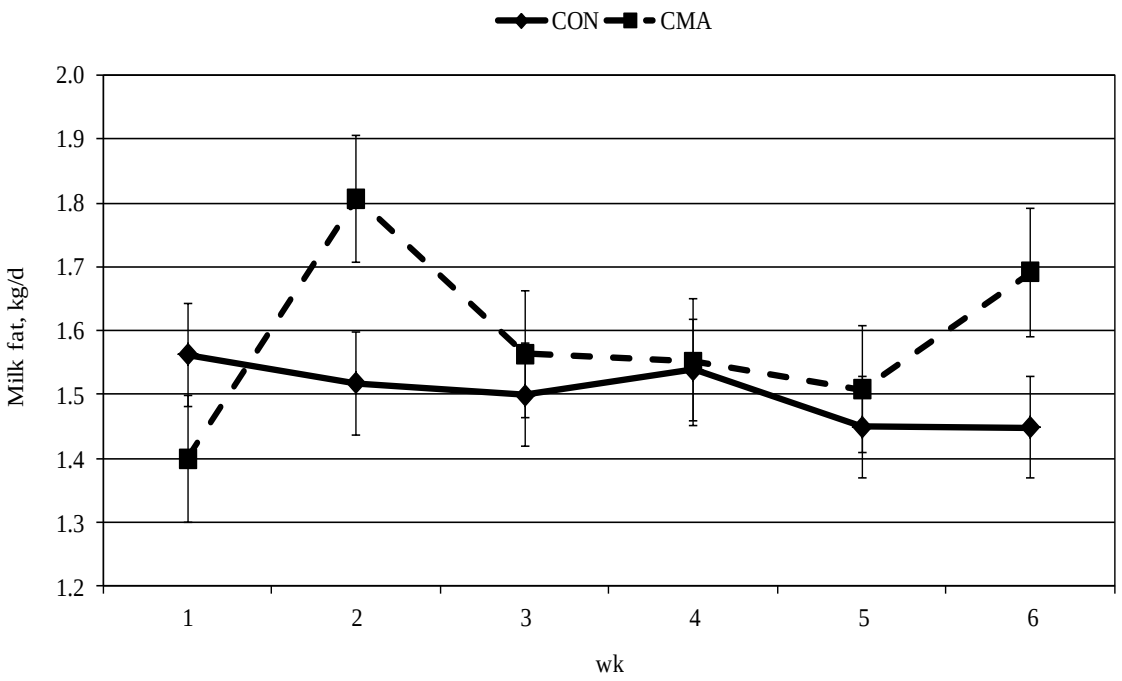

Figure 3.1. Interaction of prepartum treatment and week on milk fat yield (P = 0.0222, CON = control and CMA = cows fed supplemental calcareous marine algae)

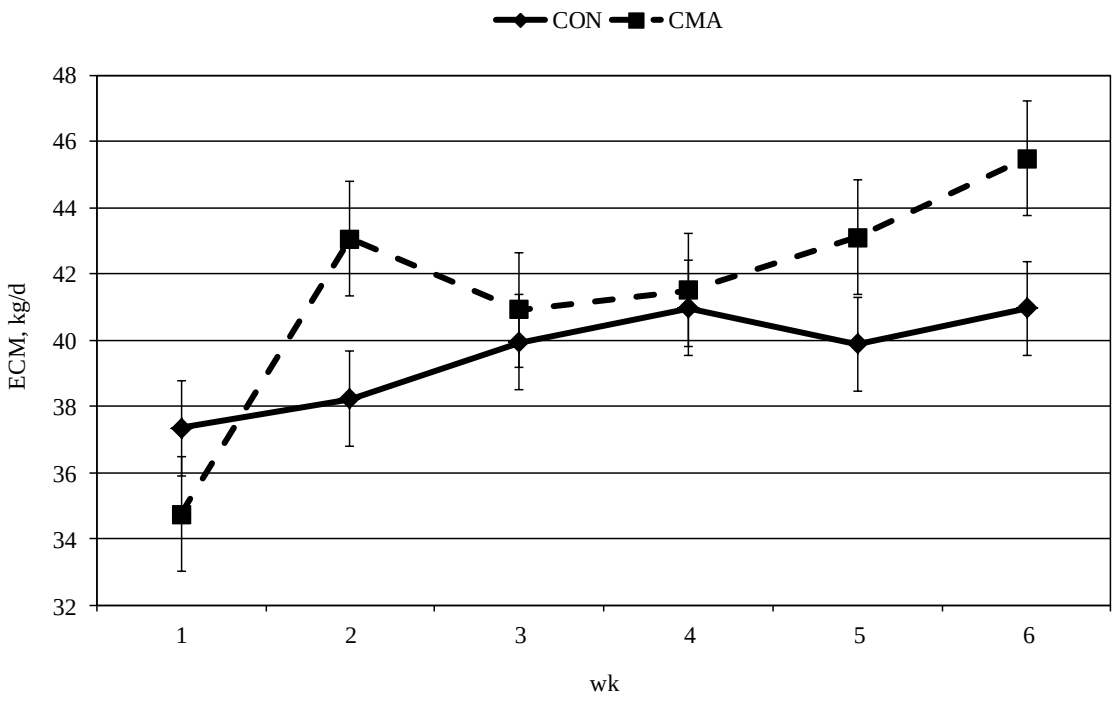

Figure 3.2. Interaction of prepartum treatment and week on ECM (*P* = 0.0395, CON = control and CMA = cows fed supplemental calcareous marine algae)

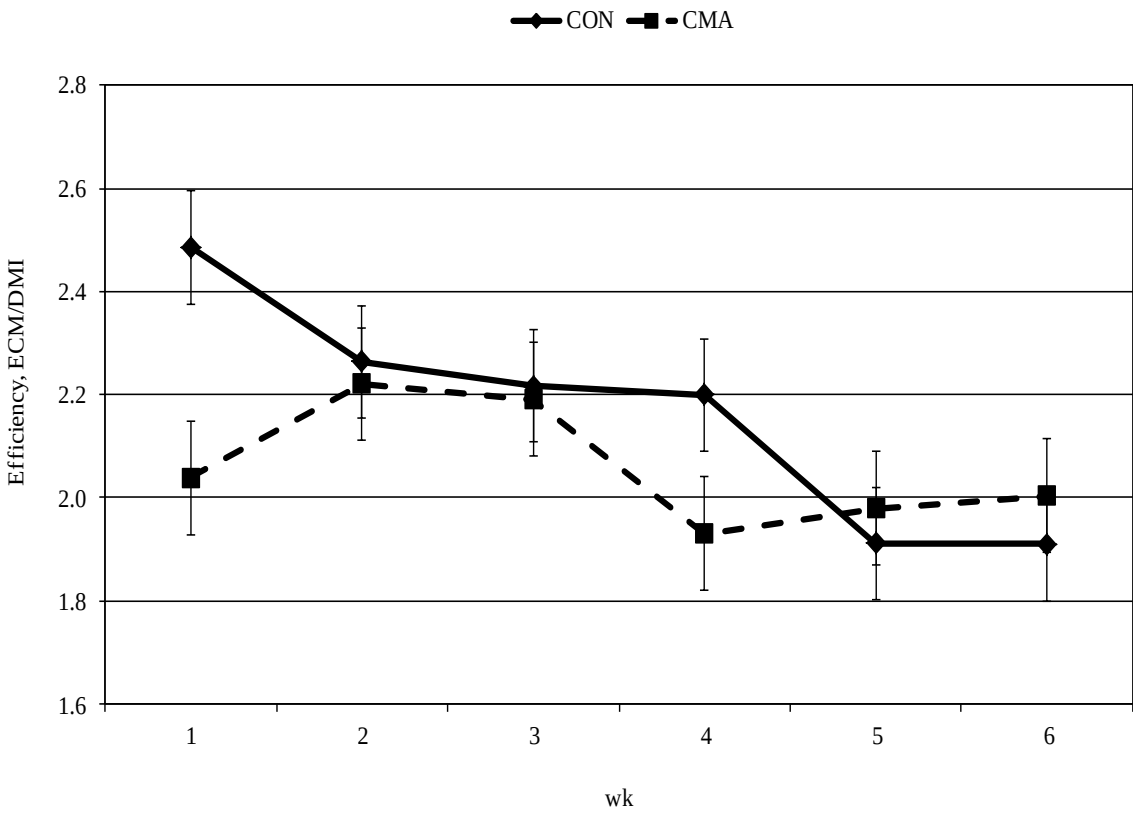

Figure 3.3. Interaction of prepartum treatment and week on milk efficiency, ECM/DMI (*P* = 0.0337, CON = control and CMA = cows fed supplemental calcareous marine algae)

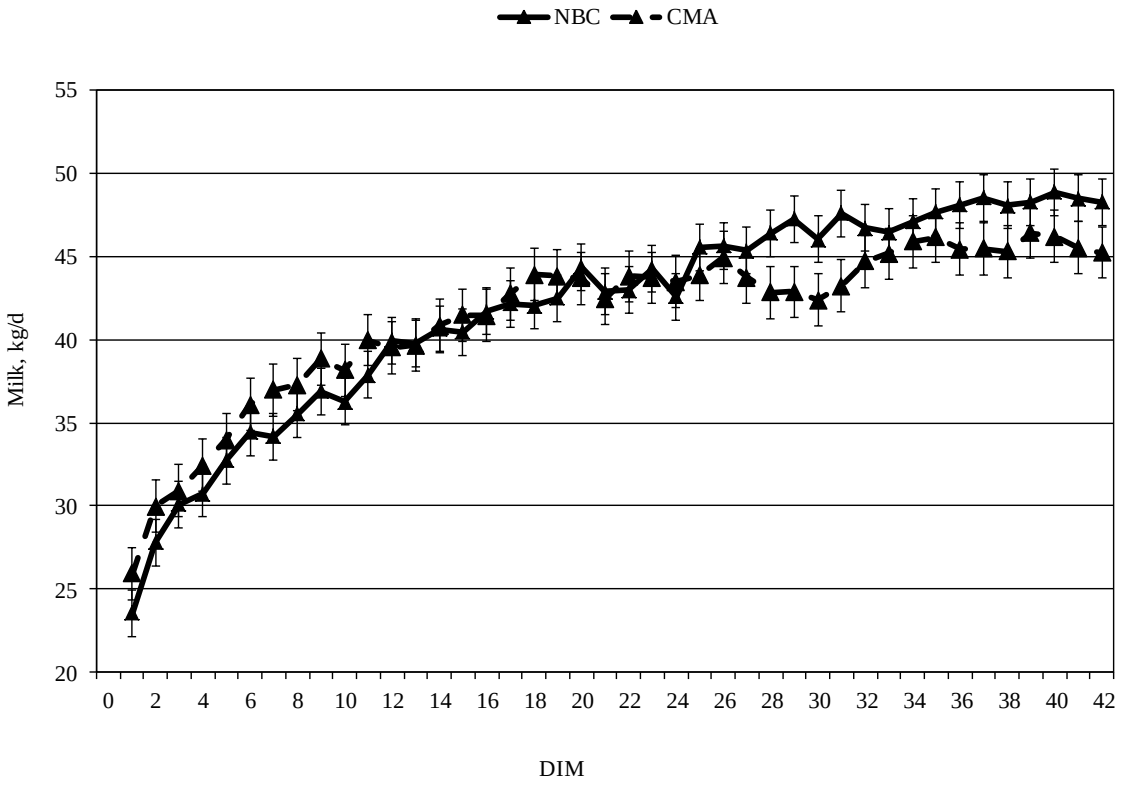

Figure 3.4. Interaction of postpartum treatment and day in milk yield (P = 0.0093, CMA = cows fed supplemental calcareous marine algae and NBC = supplemental sodium bicarbonate)

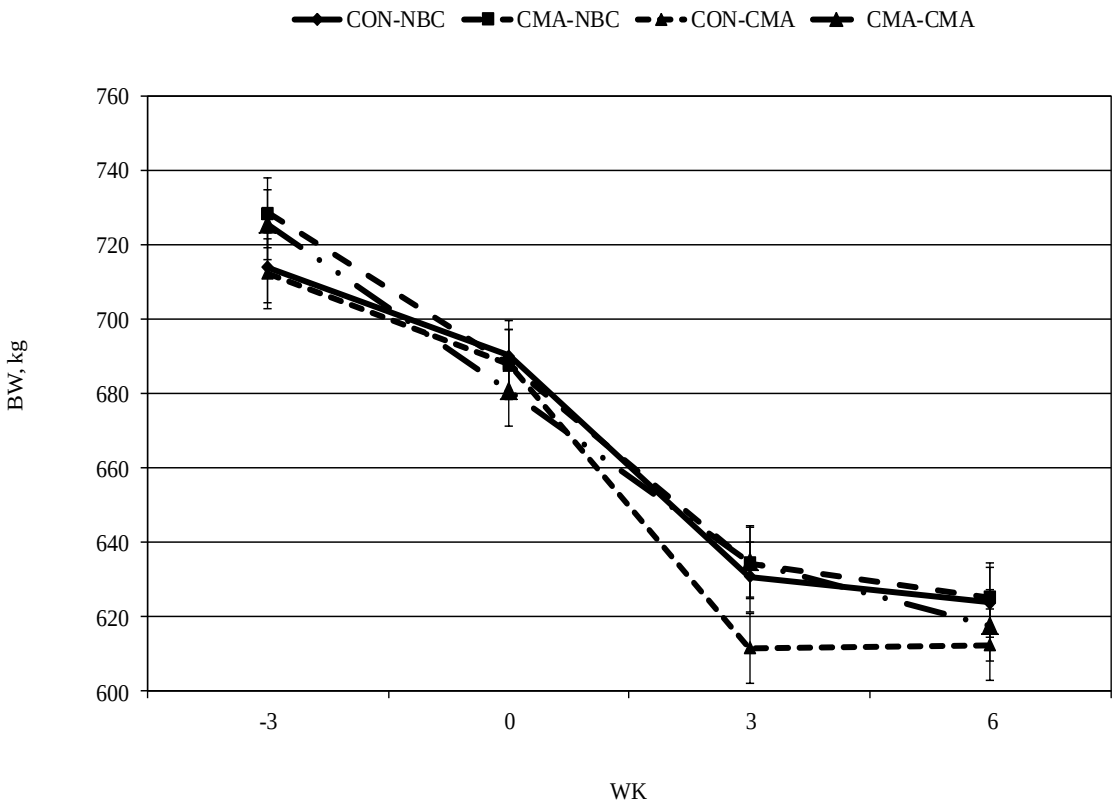

Figure 3.5. Body weight of cows throughout the trial (CON = no supplemental calcareous marine algae prepartum; CMA = supplemental calcareous marine algae prepartum or postpartum; or NBC = supplemental sodium bicarbonate postpartum). Interaction of pre- and postpartum treatment and wk (P = 0.6574)

CHAPTER 4

THE EFFECTS OF FEEDING A NEGATIVE DCAD DIET FOR VARIED LENGTHS OF TIME PREPARTUM ON DRY MATTER INTAKE, MINERAL BALANCE AND MILK YIELD[1]

[1] Wu, Z., J. K. Bernard, K. Zanzalari, and J. Chapman. To be submitted to *J. Dairy Sci.*

Abstract

Forty-five multiparous Holstein cows and 15 primiparous Holstein heifers were used in a randomized block design trial to determine the effect of length of feeding a negative dietary cation anion difference (DCAD) diet prepartum on serum and urine metabolites, dry matter intake (DMI), milk yield and composition. Within parity, cows were assigned randomly to one of 3 treatments beginning 21 ± 3, 28 ± 3, or 42 ± 3 d prepartum based on expected calving date. Diets were formulated for late gestation and early lactation cows, with a DCAD of -21.02 and 20.55 mEq/100g, respectively. Final treatment assignment was based on actual calving date and defined as cows that were fed negative DCAD diets prepartum for less than 24 d (19.2 ± 4.1 d, 3WPC), 25 to 34 d (27.9 ± 3.1 d, 4WPC) or longer than 36 d (41.5 ± 4.1d, 6WPC) providing 23, 18, and 18 animals for each treatment, respectively. No differences ($P > 0.10$) were observed in prepartum DMI which averaged 11.4, 11.5 and 11.7 kg/d for 3WPC, 4WPC and 6WCP, respectively. A linear decrease was observed prepartum for serum concentrations of total protein ($P = 0.0270$), albumin ($P = 0.0110$), Ca ($P = 0.0203$), K ($P = 0.0657$), and anion gap ($P = 0.0063$) with increased time feeding negative DCAD diets. A quadratic response was observed for Na ($P = 0.0329$) and bicarbonate ($P = 0.0544$) because of lower concentrations for 4WPC compared with 3WPC and 6WPC. No differences ($P > 0.10$) were observed in serum metabolites on day of calving except for a quadratic response for bicarbonate ($P = 0.0154$) which was lower for 4WPC than 3WPC or 6WPC. Urine pH ($P = 0.0327$) and K ($P = 0.0225$) exhibited a quadratic response because of lower concentrations for 4WPC compared with 3WPC and 6WPC. A linear increase was observed for postpartum serum total protein ($P = 0.0382$), globulin ($P = 0.0223$), and Na ($P = 0.0228$) as time of feeding negative DCAD diets increased. An interaction of treatment x wk was observed for postpartum serum gamma-glutamyl transferase ($P = 0.0096$),

and tended to be higher for 4WPC during wk 2 and 3. Postpartum DMI, milk yield and components were not different ($P > 0.10$) among treatments. An interaction of treatment x wk was observed for milk protein percentage ($P = 0.0001$) because of higher concentrations during wk 1 for 3WPC compared with 4WPC and 6WPC and higher concentrations for 4WPC during wk 3, 5 and 6 compared with 3WPC and 6WPC. An interaction of treatment x wk was also observed for milk SNF percentage ($P = 0.0122$) which was highest for 4WPC during wk 5. Results of this trial indicate that extending length of time on negative DCAD diets prepartum does affect select serum metabolites prepartum, on day of calving or postpartum other than serum total protein which increased linearly with increasing length of feeding negative DCAD diets postpartum. Pre- and postpartum DMI and milk yield postpartum was not affected by length of time fed a negative DCAD diet, but milk protein percentage tended to be higher for 4WPC. No other differences were observed in composition or yield of components. This trial demonstrates that feeding a negative DCAD diet for 42 days has no negative impact on health, milk yield and components and can be fed in one-group dry cow feeding programs for periods beyond the traditional 21 to 28 day prepartum period.

Key words: DCAD, milk yield, milk composition.

Introduction

The transition from late-gestation to lactation requires enormous physiological adaptations by the dairy cow which can significantly impact the following lactation and subsequent reproduction. Nutrition management during the transition period is challenged by reduced DMI during the late gestation period coupled with a drastic increase in nutrient requirements following calving. One of the most significant challenges involves Ca homeostasis

and can result in clinical or subclinical hypocalcaemia. Block (1984) reported that cows experiencing clinical hypocalcaemia during the immediate periparturient period produced 14% less milk than cows with normal serum Ca concentrations. In addition to decreased milk yield, cows that experienced clinical or subclinical hypocalcaemia are at greater risk for developing other metabolic disorders (Curtis et al., 1985). Feeding negative DCAD diets prepartum results in a more acidic metabolic state stimulating Ca mobilization, thus preventing hypocalcaemia which maintains DMI and improves milk yield postpartum (Block, 1984; DeGroot et al., 2010).

Animate ® (Prince Agri. Products, Inc., Quincy, IL.) is an anionic mineral supplement containing (% of DM), 13.9% Cl, 5.4% S, 4.8% Mg and 39.0% CP that is designed for use in close-up dry cow diets to acidify the diet, reducing the incidence of clinical and subclinical hypocalcaemia, resulting in greater DMI and milk yield postpartum (Puntenney, 2006). Feeding a negative DCAD diet starting 21 d prepartum was shown to be effective in preventing hypocalcaemia (Chan et al., 2006; DeGroot et al., 2010). Degaris et al. (2008) reported increased ECM and milk protein yield postpartum when cows were fed prepartum transition diets with a DCAD of -15 mEg/100g for 25 and 22 d prepartum, respectively. Most studies have focused on the effect of feeding variable levels of DCAD, while limited research has been conducted on the length of feeding a DCAD diet to transition cows. The objective of this study was to evaluate the effects of length of time feeding a negative DCAD diet prepartum on serum metabolites and performance postpartum.

Materials and Methods

Forty-five multiparous dairy cows and 15 primiparous Holstein heifers were used in a randomized block design starting 21 ± 3, 28 ± 3, or 42 ± 3 d prepartum. Cows were blocked by

expected calving date and parity. Final treatment assignments were based on days fed the negative DCAD diet based on actual calving date and was defined as: less than 24 d (3WPC), 25 to 34 d (4WPC) or longer than 36 d (6WPC) providing 23, 18, and 18 animals for each treatment, respectively. All protocols were approved by the University of Georgia Institutional Animal Care and Use Committee.

Prior to beginning the trial, all cows were trained to eat through Calan doors (American Calan, Inc., Northwood, NH). Cows were housed in a free stall barn equipped with fans and misters and were allowed unlimited access to an exercise lot. Cows were moved to a box stall at calving and returned to the free stall area after calving.

All dry cows and heifers were fed on same far-off dry diets before starting on experimental prepartum anionic diets. Experimental diets were formulated to meet requirements (NRC, 2001) for late gestation and early lactation (Table 4.1). Animate was included in the late gestation diet as an acidifying agent and the amount used was adjusted after measuring urinary pH to maintain a pH within the range of 6.0 to 6.5. Experimental diets were mixed and fed once daily using a DataRanger (American Calan, Inc.). Cows had free access to water throughout the day. The amount of feed provided was adjusted to maintain a minimum of 5% refusal. The amount of feed offered and refused was recorded daily.

Samples of dietary ingredients, TMR, and orts were collected 3 d each week and analyzed for DM content by drying samples at 50 °C for 48 h in a forced air oven. Individual samples were ground to pass through a 6-mm screen using a Wiley mill (Thomas Scientific, Swedesboro, NJ), and composited by week. A subsample was ground to pass through a 1-mm screen before analysis of ash (AOAC, 2000), N (Leco FP-528 Nitrogen Analyzer, St. Joseph,

MO), NDF (Van Soest et al., 1991), ADF (AOAC, 2000), starch (Hall, 2009), starch (Dubois, 1956), ether extract, and minerals (AOAC, 2000).

After calving, cows were milked three times daily beginning at 0000, 0800 and 1600. Milk weights were recorded electronically at each milking (Alpro, Delaval, Kansas City, MO), totaled each day, and a weekly average calculated. Milk samples were collected from three consecutive milkings each week for analysis of fat, protein, lactose, SNF, and MUN concentrations by mid-infrared spectrophotometric analysis with a Foss 4000 instrument (Foss North America, Eden Prairie, MN; Dairy One Cooperative, Ithaca, NY).

Body weight of cows were recorded on three consecutive days during -3 wk prepartum and wk 3 and 6 postpartum and once immediately after parturition. Access to water and feed was restricted until measurements were recorded. Body condition scores were assigned at the same time by two individuals on a 1 to 5 scale as described by Wildman et al. (1982).

Two whole blood samples were collected from the coccygeal vein at 0900 once during wk -6, -5, -4, -3, -2, -1 prepartum, at calving, and during wk 1, 2, 3 and 6 postpartum. One sample was used for determination of serum glucose, urea N, total protein, albumin, creatinine, total bilirubin, aspartate aminotransferase (AST), creatine kinase, gamma-glutamyl transferase (GGT), Ca, P, Mg, Na, K, Cl, bicarbonate, and anion gap using a Bohringer Mannheim/Hitachi 912 automated chemistry analyzer (Roche Laboratory Systems, Indianapolis, IN). Serum was separated from the second sample and analyzed for non-esterified fatty acids (NEFA) concentration using an enzymatic procedure (Waco Chemicals USA, Inc., Richmond, VA). Serum ketone concentrations were determined using a Nova Max Ketone Strips and a Nova Max Plus reader (Nova Biomedical, Waltham, MA). Urine samples were collected at the same times for analyses for pH and electrolyte concentrations as described above.

Data was analyzed using PROC MIXED procedure of SAS (SAS Enterprise 4.2, 2008. SAS Institute, Cary, NC). The model included block, treatment, week and the appropriate interactions. Genetic merit (PTA of multiparous cows and ETA of springing heifers) was included as a covariate for production variables. Cow within treatment and block was included as a random effect and week as a repeated measure. Significance was declared when $P < 0.05$ and trends when $P > 0.05$ and < 0.10.

Results and Discussions

The chemical composition of experimental diets is presented in Table 4.2. Nutrient concentrations in each of the diets were consistent with formulated values. The DCAD [(Na+K) - (Cl+S)] of the prepartum and postpartum diets were -21.02 and 20.55 mEq/100g, respectively.

Prepartum Responses

Urine pH was maintained within the desired range prepartum (between 6.0 and 6.5) and averaged 6.44, 6.22 and 6.43 for 3WPC, 4WPC and 6WPC, respectively (Figure 4.1). Prepartum urine pH is an important indicator of systemic acidification. One inconsistency in prepartum acidification research has been the large variation between and among studies with respect to level of systemic acidification. This difference may account for the different responses observed in serum metabolite concentrations and postpartum health and production responses among studies. Although not significant, urine pH for 6WPC cows tended to rise more during the final wk of gestation than for cows fed either of the other two treatments. Weich et al. (2013) reported a rise in pH for cows fed a negative DCAD diet for 42 d compared with 21 d. However, the increase in urine pH began at wk -3.

Prepartum DMI, either kg/d or % of BW was not different ($P > 0.10$) among treatments and averaged 11.4, 11.5, and 11.7 kg/d for 3WPC, 4WPC, and 6WPC, respectively (Table 4.3).

However, daily DMI was slightly higher for 3WPC compared with 4WPC on d -21 and lower for 4WPC compared with 3WPC and 6WPC on d -1 resulting in a tendency (P = 0.0617) for an interaction of treatment by d prepartum (Figure 4.2).

Concentrations of select metabolites and minerals in serum and urine during wk -3 through wk -1 are presented in Table 4.4. Linear decreases were observed in concentrations of serum total protein (P = 0.0270), albumin (P = 0.0110), Ca (P = 0.0203), K (P = 0.0657), and anion gap (P = 0.0063) as time of feeding negative DCAD diets increased. A quadratic response was observed prepartum for lower concentrations of serum Na (P = 0.0329) for 4WPC compared with 3WPC and 6WPC which most likely reflects differences in urinary secretion of Na which was numerically higher for 4WPC (P = 0.1135). A similar tendency for a quadratic response was observed for serum bicarbonate (P = 0.0544) which reflects changes in acid-base balance to maintain homeostasis. No differences were observed in the remaining serum metabolites.

The linear increase observed in serum concentrations of total Ca and anion gap prepartum for 3WPC compared with 4WPC and 6WPC are consistent with the linear increase in serum albumin concentrations, which is a Ca-binding protein and a negatively charged protein associated with other negatively charged ions. Although serum Ca concentrations were lower for 4WPC and 6WPC compared with 3WPC, concentrations were within the normal range. Ionized Ca (iCa) is a more reliable indicator than total serum Ca to indicate biological effect of Ca (Ballantine and Herbein, 1991; Dauth et al., 1984; Saccon et al., 1995) and should be examined in future trials to monitor actual changes in serum Ca.

Day of Calving Responses

At parturition, a quadratic response was observed for urine pH which was lower for 4WPC compared with 3WPC and 6WPC (Table 4.5). Acidification of the blood stimulates PTH

secretion and initiates bone Ca mobilization and renal production of 1, 25-dihydroxyvitamin D, thus increasing Ca level after parturition (Bichara et al., 1990) which would presumably reduce the rise of milk fever or subclinical hypocalcemia. No differences ($P > 0.10$) were observed in serum Ca concentrations on day of calving for 3WPC, 4WPC and 6WPC which averaged 7.82, 7.32 and 7.87 mg/dl, respectively. Although cows fed negative DCAD diets longer than 24 d prepartum had lower concentrations of serum Ca prepartum (Table 4.4, Figure 4.3), it does not appear to have negatively affected Ca homeostasis at calving. This is consistent with the report by Weich et al. (2013) who reported increased total serum Ca concentration after parturition for cows fed negative DCAD diets for 42 d compared with 21 d.

Based on serum Ca concentrations on day of parturition, 7.0% of all cows would be classified as clinical hypocalcaemia (< 5.5 mg/dl), 66.7% as subclinical hypocalcaemia (5.5 to 8.0 mg/d), and 26.3% would have been classified as normal (>8.0 mg/dl). The number of cows classified as clinical and subclinical hypocalcaemia was higher than expected given the level of dietary acidification. One possible explanation for this may have been that Ca intake was less than desired. Chan et al. (2006) did not observe any difference in serum Ca concentration for cows fed diets containing either 0.99 or 1.50% Ca and suggested that an intake of approximately 109 g/d was adequate based on 11 kg/d DMI. In their trial, average milk yield for the first 21 DIM was 21.7 kg/d. In our trial, prepartum Ca intake averaged 112 g/d, but average milk yield during the first 6 wk postpartum was 41.0 kg/d. Moore et al. (2000) reported that cows fed a fully acidified diet containing 1.5% dietary Ca prepartum had higher serum iCa concentrations (4.35 vs 3.85 mg/dl) resulting in fewer cows classified as clinically hypocalcemic hypocalcemic on the day of calving (0 vs 50%) compared with cows fed a fully acidified diet containing only 1.0% dietary Ca prepartum. Oetzel (1988) reported that cows fed a negative

DCAD diet (-7.5 mEq/100g) with 1.17% dietary Ca prepartum had higher iCa (4.05 vs 3.56 mg/dl) and total serum Ca concentrations (8.40 vs 7.40 mg/dl) on the day of calving than cows fed a negative DCAD diet with 0.60% dietary Ca. In addition, Oba et al. (2011) observed that cows fed a negative DCAD diet (- 6.4 mEq/100g) with 0.9% dietary Ca had faster rates of serum Ca recovery following an EDTA challenge compared to cows fed the negative DCAD diet containing 0.3% Ca. These data suggest cows fed negative DCAD diets have improved response to a Ca challenge when higher concentrations of Ca are fed.

No differences were observed among treatments in concentrations of other serum metabolites or minerals on the day of calving except for a quadratic response ($P = 0.0154$) for bicarbonate, which was lower for 4WPC compared with 3WPC and 6WPC (Table 4.5). However, urine pH ($P = 0.0327$) and K ($P = 0.0225$) exhibited a quadratic response and was lowest for 4WPC compared with 3WPC and 6WPC.

Postpartum Responses

Linear increases were observed for postpartum concentrations of serum total protein ($P = 0.0382$), globulin ($P = 0.0223$) and Na ($P = 0.0228$) with increased time feeding negative DCAD diets (Table 4.6). A quadratic response was observed for the ratio of albumin and globulin ($P = 0.0501$) because of lower concentrations for 4WPC compared with 3WPC and 6WPC. A treatment x wk interaction was observed for postpartum serum gamma glutamyl transferase (GGT, $P = 0.0096$, Figure 4.4), and tended to be higher for 4WPC during wk 2 and 3. No differences were observed in urine pH and concentrations of minerals among treatments. No differences were observed among treatments in animal health throughout the trial.

Concentrations of serum total protein and globulin were within normal ranges and the increase observed with increasing time cows were fed the negative DCAD diet most likely reflect small differences in protein absorption and metabolism. Serum aspartate transaminase (AST) and GGT are frequently used as markers of liver disease resulting from metabolic disease or stress (González et al., 2011; Kartaria and Kartaria, 2012). Concentrations of AST are used as a sensitive marker of liver damage. In our trial, AST concentrations were within normal ranges suggesting that the cows did not experience any abnormal hepatic lipidosis (González et al., 2011). The reason for the observed interaction of treatment x wk (Figure 4.4) is unclear. González et al. (2011) did not observe any difference in GGT concentrations of cows classified as high (primarily early lactation) or low (primarily mid-lactation) lipid mobilization based on serum NEFA and BHB concentrations.

No differences were observed between treatments in serum Ca concentrations postpartum (Figure 4.3). Recovery of serum Ca concentrations above 8.0 mg/dl occurred within the first wk postcalving. Martinez et al., (2013) demonstrated in a retrospective study that cows with serum Ca concentration below 8.59 mg/dl within the first 3 d postpartum had reduced concentrations of neutrophils in blood, impaired neutrophil function and increased incidence of both metritis and puerperal metritis. Serum Ca concentration of the periparturient dairy cow has two dynamics. The first involves the magnitude of drop in serum Ca concentration following parturition and the second involves the rate of recovery of serum Ca concentration following the initiation of lactation. In our study, the magnitude of decrease in serum Ca concentration comparing wk -1 vs day of calving averaged 1.5 mg/dl. The rate of recovery within 1 wk of calving averaged 1.1 mg/dl. Weich et al. (2013) observed no increase in total serum Ca concentration for any treatment groups (control, positive DCAD prepartum; 21 d, negative DCAD prepartum for 21 d;

and 42 d, negative DCAD prepartum for 42 d) between 12 h and 24 h postcalving. However, by 72 h increases in total serum Ca were observed but these values were still below the 8.0 mg/dl, a concentration that has been used for many years as the expected normal serum Ca concentration for periparturient dairy cows. In contrast to total serum Ca concentration, Weich et al. (2013) reported higher concentrations of ionized Ca (iCa) in serum postcalving. Oetzel et al. (1988) also reported different responses between total serum Ca and iCa in periparturient dairy cows fed ammonium chloride and ammonium sulfate. In that study, total serum Ca concentration was a better predictor of Ca status on the day before calving whereas iCa concentration was a better predictor of Ca status on the day of calving. These studies demonstrate that differences between total serum Ca concentration and ionized serum Ca concentration may exist and that interpretation of data by one of these measures may differ from and potentially contradict interpretation of the data using the other measure.

Daily DMI postpartum was not different among treatments ($P > 0.10$, Figure 4.5). No differences among treatments were observed in average postpartum DMI, yield and percentage of milk, milk fat, lactose, or SNF (Table 4.3). Milk protein percentage tended to be higher for 4WPC compared with 3WPC and 6WPC (quadratic effect, $P = 0.0951$). There was an interaction of treatment x wk for milk protein percentage ($P = 0.0001$, Figure 4.6) with higher percentage for 3WPC compared with 4WPC and 6WPC during wk1 and higher percentage for 4WPC compared with 3WPC and 6WPC during wk 5 and 6. Milk SNF percentage exhibited an interaction of treatment X wk and was higher ($P = 0.0122$) for 4WPC during wk 5 compared with 3WPC and 6WCP, resulting in a treatment x wk interaction ($P = 0.0122$, Figure 4.7). No differences were observed in ECM, efficiency (ECM/DMI) or MUN concentrations among treatments.

The higher milk protein percentage and SNF percentage observed for 4WPC is consistent with the higher serum total protein and globulin concentrations. The increased serum protein concentration observed postpartum suggested an up- regulation of metabolism following increased exposure to negative DCAD diets (DeGaris et al., 2010).

Conclusions

Results of this trial indicate feeding a negative DCAD diet for increasing lengths of time prior to calving may slightly decrease concentrations of total serum protein, albumin, Ca, K, and anion gap prepartum and support linear increases in serum total protein, globulin and Na postpartum. The changes in K, Na, and anion gap most likely reflect changes in response to dietary supplementation and homeostasis. The slight decrease in serum Ca prepartum observed with increased length of feeding the negative DCAD diet did not affect serum Ca on day of calving or postpartum. The changes in milk protein possibly reflect differences in protein balance which supported slightly higher milk protein percentage for cows fed negative DCAD diets for 4 wk prior to calving. Overall, the results of the trial indicate minor differences in serum metabolite concentrations and urinary excretion of minerals as a result of feeding a negative DCAD diet for 3, 4 or 6 wk prior to calving, but the effects do not appear to alter cow health or performance postpartum. These data suggest that feeding a negative DCAD diet for longer than the traditional feeding period of 21 days prepartum does not negatively impact cow health or performance. These results are consistent with previous research documenting the effects of extended feeding of negative DCAD diets (Block, 1984; Weich et. al., 2013). From an applied application standpoint these collective data support the use of negative DCAD diets in one-group dry cow programs.

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Table 4.1. Ingredient composition of experimental diets (% of DM)

Ingredient	Prepartum	Postpartum
Corn silage	42.86	35.59
Alfalfa hay	5.95	
Ryegrass baleage		9.79
Bermudagrass baleage	7.94	
Whole cottonseed		8.90
Ground corn	8.93	16.55
Brewers grains, wet	7.94	12.46
Citrus pulp	3.97	4.45
Soybean hulls, pelleted	7.94	
Animate[1]	4.56	
Soybean meal, 48% CP	5.95	3.56
AminoPlus[2]		1.78
Prolak[3]	0.79	3.20
Sodium bicarbonate		0.89
Magnesium oxide	0.16	0.36
Calcium carbonate	1.19	1.07
Salt		0.18
Potassium carbonate		0.27
Yeast culture	0.50	0.22
Trace mineral	0.14	0.14
Vitamin E	0.14	0.02
Rumensin, 3g/lb[4]	0.48	0.34
Zinpro Availa-4[5]	0.08	0.04
OmniGen-AF[6]	0.50	0.22

[1]Anionic mineral supplement, Prince Agri Products, Inc., Quincy, IL

[2]Ruminally protected soybean meal, Ag Processing, Inc. Omaha, NE

[3]Marine and animal rumen undegradable protein supplement, H. J. Baker & Bros., Inc. Westport, CT

[4]Rumensin, Elanco Animal Health, Indianapolis, IN

[5]Organic zinc, manganese, copper, and cobalt, Zinpro Corporation, Eden Prairie, MN

[6]Immune stimulant, Prince Agri Products, Inc., Quincy, IL

Table 4.2. Chemical composition of experimental diets

Ingredient	Prepartum	Postpartum
DM, %	52.6 ± 3.0	50.2 ± 3.3
	% of DM	
CP	14.6 ± 1.0	18.0 ± 0.9
ADF	26.2 ± 1.9	21.6 ± 1.4
NDF	42.3 ± 2.3	36.4 ± 1.9
Starch	20.5 ± 2.3	24.2 ± 2.3
Sugar	2.8 ± 0.5	3.2 ± 0.8
Ether extract	3.3 ± 0.4	5.0 ± 0.5
Ash	7.1 ± 0.8	8.1 ± 1.1
Ca	0.97 ± 0.23	0.94 ± 0.16
P	0.37 ± 0.04	0.51 ± 0.02
Mg	0.52 ± 0.10	0.47 ± 0.09
K	1.07 ± 0.15	1.30 ± 0.10
S	0.42 ± 0.09	0.26 ± 0.08
Na	0.08 ± 0.04	0.41 ± 0.12
Cl	0.97 ± 0.19	0.45 ± 0.17
	meq/100g	
DCAD[1]	-21.02	20.55
DCAD[2]	3.98	39.3

[1] Calculated as: (Na+K)-(Cl+S)

[2] Calculated as: Na+K-Cl

Table 4.3. Dry matter intake, milk yield and composition of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC) or 6 wk prepartum (6WPC)

	TRT				P			
	3WPC	4WPC	6WPC	SE	TRT	TRT*WK	LINEAR	QUADRATIC
Prepartum DMI								
kg/d	11.4	11.5	11.7	0.6	0.9190	0.6804	0.6849	0.9682
% of BW	1.70	1.68	1.73	0.12	0.9614	0.6987	0.8646	0.8266
Postpartum DMI								
kg/d	19.1	19.6	18.6	0.8	0.6867	0.4650	0.6383	0.4789
% of BW	3.12	3.15	3.01	0.19	0.8528	0.7402	0.6727	0.7190
Milk, kg/d	40.6	41.5	41.0	1.5	0.9181	0.3965	0.8263	0.7425
Fat, %	4.30	4.50	4.30	0.13	0.5322	0.4001	0.9842	0.2654
Fat, kg/d	1.74	1.70	1.73	0.08	0.9259	0.9091	0.8857	0.7138
Protein, %	2.80	2.90	2.73	0.06	0.1490	0.0001	0.3769	0.0951
Protein, kg/d	1.14	1.10	1.09	0.03	0.5752	0.1552	0.2968	0.8980
Lactose, %	4.69	4.75	4.78	0.06	0.5362	0.7075	0.2750	0.8451
Lactose, kg/d	1.96	1.83	1.92	0.06	0.3672	0.3466	0.6439	0.1793
SNF, %	8.46	8.58	8.45	0.10	0.6488	0.0122	0.9914	0.3561
SNF, kg/d	3.49	3.29	3.37	0.10	0.4342	0.4196	0.4162	0.3065
ECM, kg/d	44.8	42.9	43.4	1.6	0.7199	0.9379	0.5176	0.6158
ECM/DMI	2.46	2.30	2.42	0.13	0.6965	0.9378	0.8020	0.4156
MUN, mg/dl	14.08	12.90	13.60	1.00	0.7228	0.7545	0.7280	0.4681

Table 4.4. Concentrations of select metabolites and minerals in serum and urine prepartum (wk -1 through -3 prepartum) in cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC) or 6 wk prepartum (6WPC)

		TRT				P			
	UNIT	3	4	6	SE	TRT	TRT*WK	LINEAR	QUADRATIC
Serum chemistry									
NEFA	mEq/l	0.73	0.84	0.56	0.38	0.2392	0.4970	0.5353	0.1156
Ketone	mmol/ml	0.31	0.29	0.36	0.03	0.1897	0.8278	0.1651	0.2918
Total protein	g/dl	6.44	6.49	6.10	0.11	0.0301	0.8135	0.0270	0.1625
Albumin	g/dl	3.51	3.45	3.36	0.04	0.0345	0.3197	0.0110	0.8469
Globulin	g/dl	2.93	3.04	2.74	0.10	0.1527	0.9077	0.1695	0.1763
Albumin/Globulin	%	1.22	1.16	1.27	0.04	0.2285	0.1035	0.4322	0.1311
Urea nitrogen	mg/dl	10.25	9.79	11.23	0.46	0.0954	0.7499	0.1152	0.1528
Creatinine	mg/sl	0.78	0.74	0.73	0.03	0.6435	0.4584	0.3700	0.8162
Total bilirubin	mg/dl	0.12	0.12	0.14	0.01	0.5332	0.7767	0.4326	0.4418
Glucose	mg/dl	59.58	61.02	57.56	1.40	0.2362	0.2039	0.2949	0.2164
AST[1]	U/L	72.84	67.55	64.67	3.74	0.2661	0.3676	0.1080	0.8204
Creatine Kinase	U/L	181.17	228.27	148.41	40.88	0.4259	0.9435	0.5573	0.2710
GGT[2]	IU/L	18.16	18.95	17.46	1.29	0.7542	0.8979	0.6816	0.5408
Ca	mg/dl	9.46	9.09	9.12	0.10	0.0380	0.2143	0.0203	0.1813
P	mg/dl	5.78	5.73	5.85	0.16	0.8923	0.4752	0.7647	0.7302
Mg	mg/dl	2.39	2.43	2.33	0.05	0.3784	0.8133	0.3535	0.3163
Na	mEq/l	143.71	139.78	141.53	0.92	0.0362	0.3050	0.0885	0.0329

K	mEq/l	4.90	4.85	4.75	0.06	0.1626	0.9506	0.0657	0.7829
Cl	mEq/l	107.86	105.72	106.39	0.82	0.2387	0.5063	0.1951	0.2239
Bicarbonate	mmol/L	25.58	24.18	25.95	0.57	0.1264	0.8775	0.6291	0.0544
Anion gap	mmol/L	15.13	14.78	13.96	0.30	0.0168	0.0629	0.0063	0.5667
Urine Chemistry									
pH		6.44	6.22	6.43	0.14	0.5444	0.7795	0.9776	0.2740
Na	mmol/L	18.98	23.72	14.79	2.99	0.1476	0.0993	0.3035	0.1135
K	mmol/L	159.64	138.63	142.62	12.08	0.4493	0.3057	0.3020	0.4508
Cl	mmol/L	183.72	188.55	176.44	18.00	0.8956	0.2344	0.7685	0.7200

[1]AST = Aspartate transaminase

[2]GGT = gamma glutamyl transferase

Table 4.5. Concentrations of select metabolites and minerals in serum and urine on day of calving in cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC) or 6 wk prepartum (6WPC)

		TRT				P		
	UNIT	3WPC	4WPC	6WPC	SE	TRT	LINEAR	QUADRATIC
Serum chemistry								
NEFA	mEq/l	1.10	1.18	1.10	0.22	0.9453	0.9967	0.7387
Ketone	mmol/ml	0.43	0.50	0.44	0.04	0.4431	0.8970	0.2108
Total protein	g/dl	6.12	6.03	6.06	0.16	0.9193	0.7862	0.7757
Albumin	g/dl	3.54	3.37	3.45	0.08	0.3765	0.4855	0.2467
Globulin	g/dl	2.58	2.66	2.61	0.10	0.8684	0.8662	0.6257
Albumin/Globulin	%	1.42	1.29	1.37	0.07	0.3926	0.6293	0.2092
Urea nitrogen	mg/dl	12.54	13.55	13.71	1.02	0.6642	0.4178	0.7364
Creatinine	mg/sl	0.80	0.82	0.85	0.05	0.8317	0.5533	0.8727
Total bilirubin	mg/dl	0.33	0.35	0.38	0.10	0.9446	0.7368	0.9853
Glucose	mg/dl	95.27	74.17	86.76	7.54	0.1348	0.2811	0.1065
AST[1]	U/L	91.50	76.61	84.06	5.29	0.1334	0.3196	0.0941
Creatine Kinase	U/L	276.81	367.80	284.75	89.22	0.7277	0.9496	0.4326
GGT[2]	IU/L	18.78	19.98	20.48	1.52	0.5253	0.4289	0.3861
Ca	mg/dl	7.82	7.32	7.87	0.42	0.5627	0.9323	0.2859
P	mg/dl	3.91	4.16	4.51	0.39	0.5533	0.2789	0.9195
Mg	mg/dl	2.46	2.49	2.38	0.07	0.6096	0.4683	0.4665
Na	mEq/l	143.45	139.89	143.00	2.37	0.5154	0.8915	0.2604
K	mEq/l	5.29	5.42	5.54	0.19	0.6484	0.3566	0.9794
Cl	mEq/l	106.82	105.78	105.88	2.00	0.9158	0.7402	0.8186
Bicarbonate	mmol/L	27.73	25.27	27.86	0.81	0.0515	0.9142	0.0154
Anion gap	mmol/L	14.23	14.28	14.94	0.56	0.6181	0.3666	0.6605
Urine Chemistry								
pH		6.37	6.15	6.80	0.16	0.0257	0.0633	0.0327

Na	mmol/L	20.59	15.91	27.32	6.13	0.4494	0.4369	0.2944
K	mmol/L	98.99	62.95	86.14	10.17	0.0430	0.3713	0.0225
Cl	mmol/L	94.23	71.17	79.35	11.95	0.3659	0.3782	0.2962

[1]AST = Aspartate transaminase

[2]GGT = gamma glutamyl transferase

Table 4.6. Concentrations of select metabolites and minerals in serum and urine postpartum (wk 1 through 6 postpartum) in cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC) or 6 wk prepartum (6WPC)

		TRT				P			
	UNIT	3WPC	4WPC	6WPC	SE	TRT	TRT*WK	LINEAR	QUADRATIC
Serum chemistry									
NEFA	mEq/l	1.59	1.57	1.37	0.35	0.8745	0.3792	0.6589	0.7846
Ketone	mmol/ml	0.60	0.51	0.75	0.09	0.1902	0.2556	0.1860	0.1988
Total protein	g/dl	6.88	7.23	7.12	0.09	0.0255	0.8459	0.0382	0.0699
Albumin	g/dl	3.65	3.53	3.60	0.07	0.4927	0.1920	0.6673	0.2740
Globulin	g/dl	3.24	3.68	3.55	0.10	0.0131	0.9782	0.0223	0.0504
Albumin/Globulin	%	1.13	0.96	1.06	0.05	0.0782	0.9801	0.2558	0.0501
Urea nitrogen	mg/dl	15.05	14.58	14.84	1.15	0.9567	0.1999	0.8970	0.7940
Creatinine	mg/sl	0.67	0.67	0.63	0.02	0.4556	0.8067	0.2661	0.5182
Total bilirubin	mg/dl	0.30	0.31	0.30	0.03	0.9149	0.4142	0.8171	0.7272
Glucose	mg/dl	50.34	53.81	50.00	1.75	0.3113	0.0578	0.8869	0.1304
AST[1]	U/L	109.92	97.93	103.38	4.82	0.2671	0.4443	0.3073	0.2054
Creatine Kinase	U/L	143.67	179.48	152.87	22.96	0.5991	0.2038	0.7624	0.3372
GGT[2]	IU/L	26.67	26.73	24.45	1.57	0.4956	0.0096	0.2901	0.6009
Ca	mg/dl	9.28	9.23	9.27	0.09	0.9224	0.4790	0.9695	0.6912
P	mg/dl	5.52	5.62	5.56	0.15	0.9161	0.1662	0.8275	0.7246
Mg	mg/dl	2.27	2.21	2.24	0.04	0.6087	0.8520	0.5592	0.4222
Na	mEq/l	138.80	139.30	140.30	0.48	0.0680	0.5930	0.0228	0.7091

K	mEq/l	4.40	4.56	4.49	0.07	0.3373	0.4390	0.3795	0.2431
Cl	mEq/l	96.77	95.67	95.51	0.86	0.2260	0.5488	0.1331	0.3953
Bicarbonate	mmol/L	32.96	32.51	32.98	0.57	0.8448	0.5671	0.9765	0.5647
Anion gap	mmol/L	16.47	15.78	16.20	0.33	0.4005	0.4924	0.5451	0.2284
Urine Chemistry									
pH		8.21	8.18	8.22	0.05	0.8845	0.8913	0.9526	0.6245
Na	mmol/L	91.72	98.44	92.51	7.72	0.8379	0.5941	0.9383	0.5582
K	mmol/L	146.66	146.80	131.90	9.49	0.4432	0.7828	0.2556	0.5512
Cl	mmol/L	37.24	38.67	35.34	3.30	0.7862	0.1631	0.6745	0.5753

[1]AST = Aspartate transaminase

[2]GGT = gamma glutamyl transferase

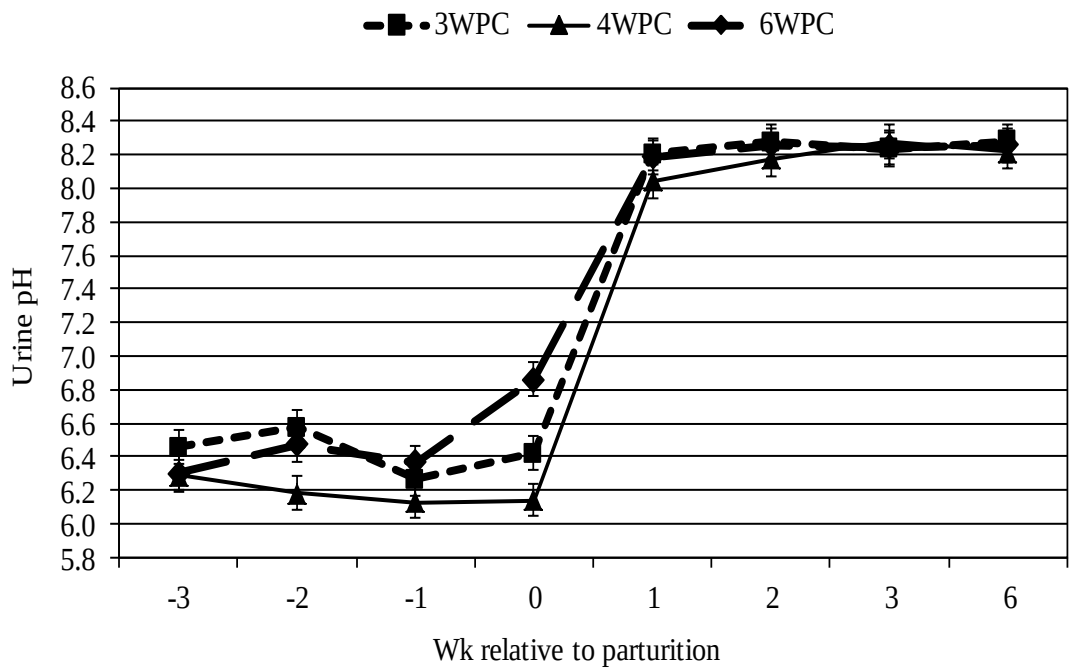

Figure 4.1. Urine pH of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC) or 6 wk (6WPC) prepartum and a positive DCAD diet postpartum (Treatment × wk, $P = 0.3847$)

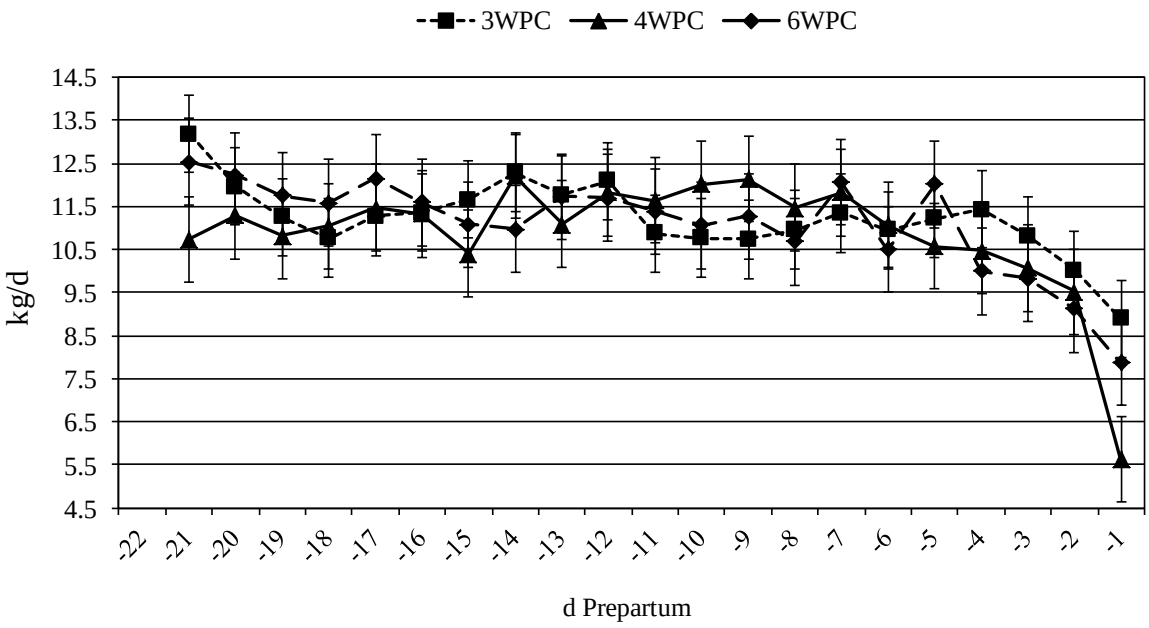

Figure 4.2. Prepartum DMI of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC) or 6 wk (6WPC) prepartum (Treatment × d, $P = 0.0617$)

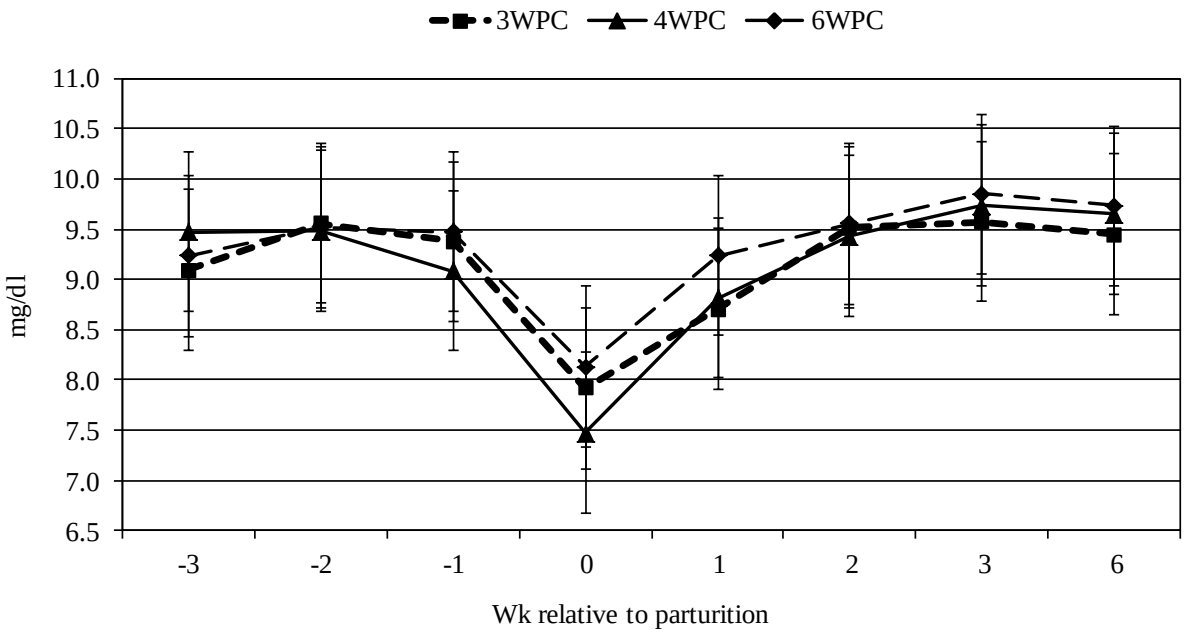

Figure 4.3. Blood Ca concentrations of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC), or 6 wk (6WPC) prepartum (Treatment × week, $P = 0.2384$)

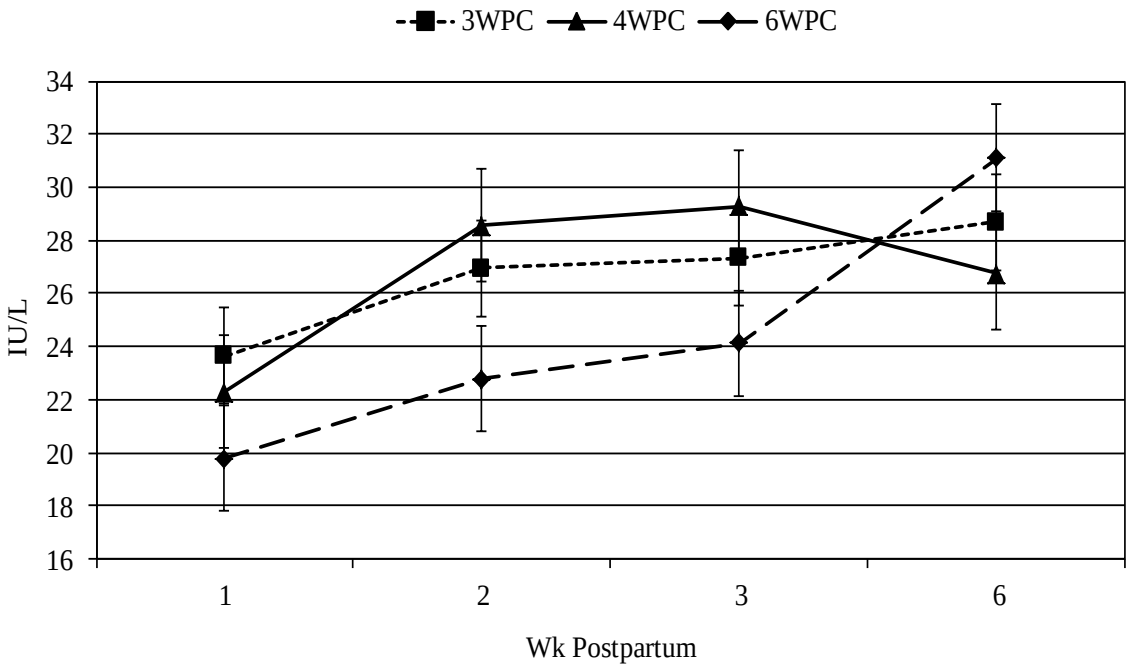

Figure 4.4. Postpartum serum GGT concentrations of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC), or 6 wk (6WPC) prepartum (Treatment × wk, $P = 0.0122$)

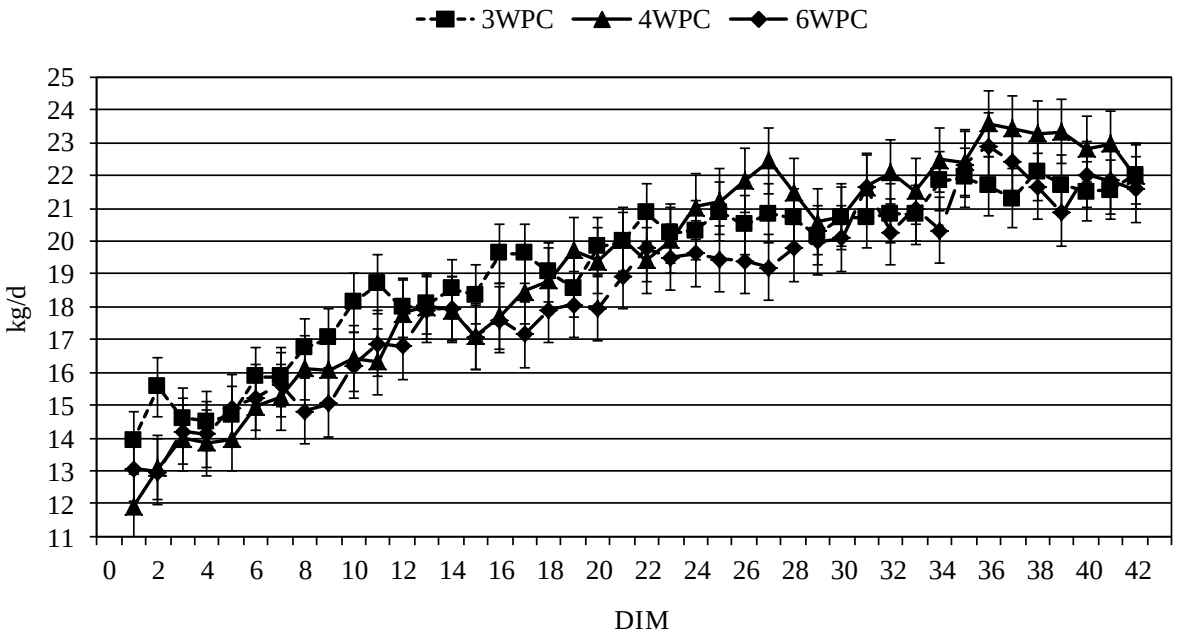

Figure 4.5. Postpartum DMI of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC), or 6 wk (6WPC) prepartum (Treatment × d, $P = 0.4650$)

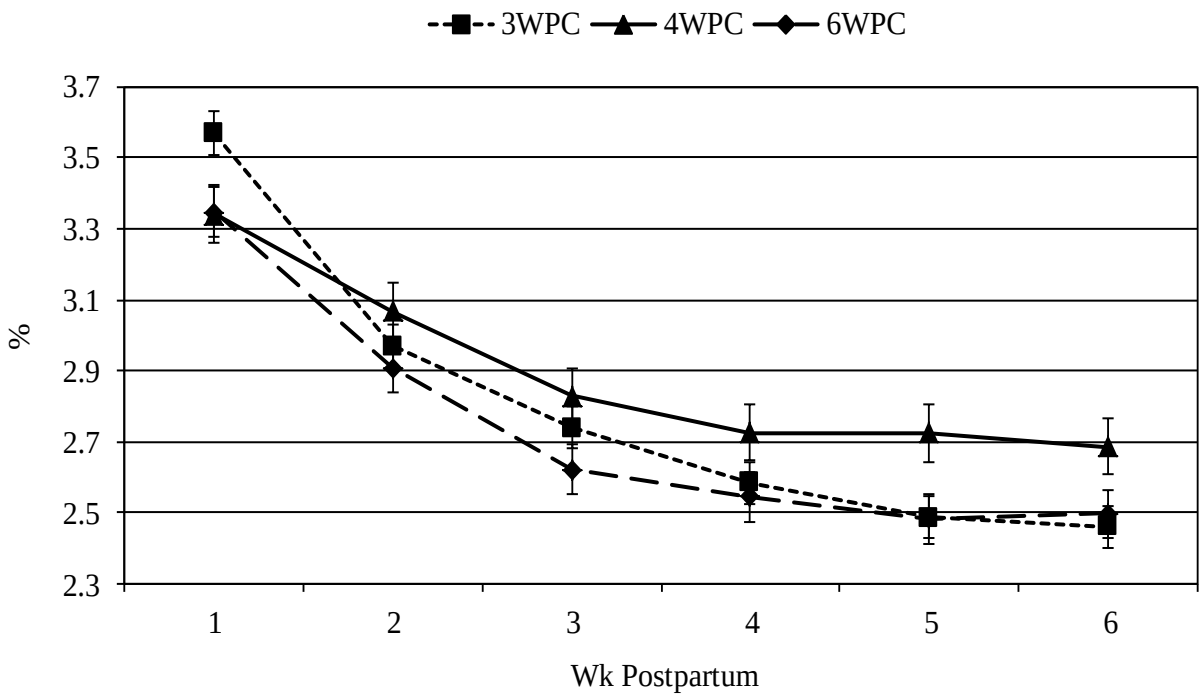

Figure 4.6. Milk protein percentage of cows fed a negative DCAD diet for 3 (3WPC), 4 (4WPC), or 6 wk (6WPC) prepartum (Treatment × wk, $P = 0.0001$)

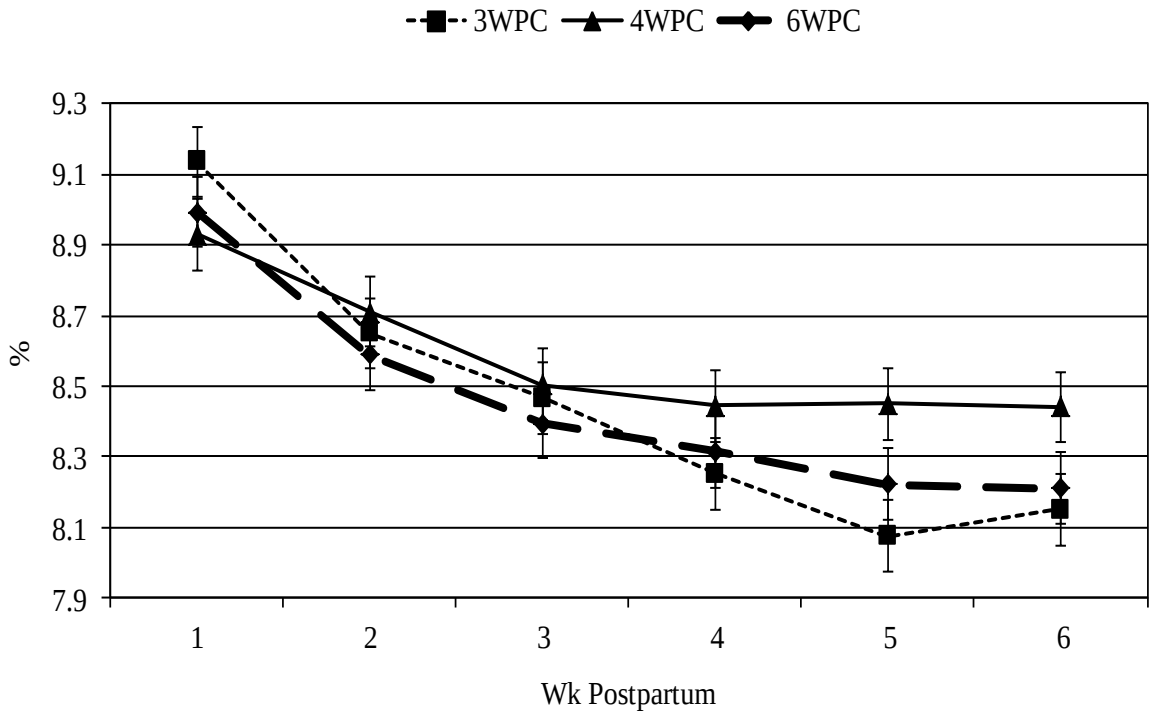

Figure 4.7. Milk SNF percentage of cows fed a negative DCAD diet for 3 (WPC), 4 (4WPC), or 6 wk (6WPC) prepartum (Treatment × wk, $P = 0.0122$)

CHAPTER 5

CONCLUSIONS

Results of these trials suggest that feeding calcareous marine algae prepartum supports higher milk protein yield postpartum. Dry matter intake, milk yield and composition were not different among pre- and postpartum treatments, but milk fat and ECM yields were higher at 2 and 4 wk postpartum for cows fed calcareous marine algae prepartum. Primarily, changes in serum and urine parameters reflected differences in dietary mineral supplementation. Overall, feeding calcareous marine algae prepartum potentially has positive effects on animal performance. Postpartum performance for cows fed calcareous marine algae was similar to those fed sodium bicarbonate. Extending lengths of time of feeding a negative DCAD diet did affect selected serum and urine metabolites, but the effects did not appear to affect cow health postpartum. Milk protein yield was higher for cows fed the negative DCAD diet for 4 wk prior to calving. Feeding a negative DCAD diet for 42 days can be fed in one-group dry cow feeding programs for periods beyond the traditional 21 to 28 day prepartum period. Additional research is need to evaluate the differences of ionized Ca concentrations to reflect actual biologic effect and the interaction between varied levels of DCAD and Ca sources.