

Study to Evaluate Inclusion of MegAnion in Diets for Non-lactating Dairy Cows

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Abstract

Low postpartum blood calcium remains one of the largest constraints to postpartum feed intake and performance in transitioning dairy cows. Decreasing dietary cation-anion difference (DCAD) by using anion sources during the dry period reduces the risk of hypocalcemia in cows at calving. MegAnion is a palatable anion supplement designed to lower the DCAD in diets for non-lactating cows as they transition into lactation. This product is unique compared to other anion sources on the market in that it is manufactured as an extruded organic complex to have significantly greater nutritional value and chloride content. The objective of this study was to assess if MegAnion, compared to a mix of typical anionic salts, had any major detrimental impacts on dry matter intake and mineral and electrolyte status in periparturient multiparous cows. Experimental cows were 12 pregnant multiparous cows fed 21 days before their expected calving date. Both treatments had a final DCAD value of -12 mEq/100g. Dry matter intake was evaluated. Fresh whole blood was collected throughout the duration of the study and analyzed for ionized calcium, pH and other acid-base indicators. Lactating cows were milked daily and milk weights recorded. Body weights and condition scores were recorded. Urine was collected by manual vulva stimulation and used to determine urine pH. Dry matter intake, acid base status, milk yield, body weight, body condition and overall performance did not differ between the MegAnion treatment and control. No detrimental effects were observed from the MegAnion treatment.

Introduction

Cows that successfully transition from pregnancy to lactation without health or metabolic disorders do not need to divert energy and nutrients toward addressing these disorders. As a result, these cows can have more productive lactations in terms of milk yield, reproductive performance, and profit. However, because this transition is the most stressful period of a cow's life, a smooth transition can be easily disrupted resulting in clinical or subclinical metabolic disorders. Most of these disorders are more prevalent in the multiparous than the primiparous cow.

Calcium is required for a number of important metabolic functions including muscle contractions. An insufficient supply of calcium (hypocalcemia) can reduce or alter activity of muscles including those responsible for abomasal contractions and mammary teat sphincter closing. This reduced activity can contribute to a displaced abomasum or mastitis, both of which can negatively impact the immune system and can contribute to the development of other health and metabolic disorders. The amount of calcium needed increases as gestation continues. The near-term fetus requires about 10 g/d and the cow needs about 2.3 g for every kilogram of colostrum she produces (NRC, 2001). Evidence indicates the high potassium content of forages fed to the late-gestation cow contributes to blood alkalosis which disrupts calcium mobilization and creates conditions that cause hypocalcaemia and, if severe enough, milk fever.

Diets with a negative dietary cation anion difference (DCAD) can produce blood acidosis, reduce the potential for development of hypocalcemia and increase the opportunity for a successful transition to lactation. These negative DCAD diets are achieved by including a mineral supplement that contains anionic salts that alter the relative amounts of dietary cations and anions in favor of the anions. Thus, the DCAD calculation (typically $DCAD = (Na^+ + K^+) - (Cl^- + S^-)$) becomes negative. Although all dietary cations and anions can contribute the DCAD value, Na, K, Cl, and S are usually the major contributors.

One challenge to this approach has been to find combinations of anionic salts that can reduce DCAD values without reducing feed intake. Anionic salts frequently are not very palatable. Anionic salts can also be difficult to handle due to their hygroscopic nature which contributes to processing and storage problems. Therefore, there is considerable interest in development of easy to handle, palatable alternatives. MegAnion is a low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, and hydrochloric acid. It has palatability and handling characteristics that make it a potentially valuable supplement for reducing DCAD values for diets of late-gestation multiparous cows.

The primary goal of this pilot study was to assess if MegAnion, compared to a mix of typical anionic salts, had any major detrimental impacts on dry matter intake and mineral and electrolyte status in periparturient multiparous cows. Thus, two supplements, one with MegAnion and one with typical anionic salts, were developed with identical DCAD values and fed to late-gestation, multiparous cows.

Materials and Methods

Animal Management and Sample Collection and Analyses

Twelve multiparous cows were housed in a tie-stall barn and randomly assigned to one of two close-up prepartum diets at 21 days before their expected calving date. The diets were fed ad libitum as total mixed rations (TMR) composed of corn silage, grass hay, and a grain mix that contained either a mixture of typical anionic salts (control) or MegAnion designed to provide a dietary cation anion difference (DCAD) of -12 meq/100g (Table 1, 2). Upon calving, all cows were fed ad libitum quantities of the same TMR (Table 2) designed to meet the nutritional needs of cows in early lactation (NRC, 2001). Dry cows were individually fed at 1000 h and lactating cows at 0830 and 1700 h daily. Amounts of TMR offered and refused were determined daily for each cow. Lactating cows were milked at 0300 and 1500 h daily and milk weights recorded. Body weights and condition scores from all cows were determined weekly at 0900 h on Mondays. Cows were observed daily and abnormalities treated when warranted. All animal procedures were approved by the University of Minnesota Institutional Animal Care and Use Committee.

Samples of each TMR were collected weekly, composited by TMR and analyzed (Dairy One Forage Lab, Ithaca, NY) using their wet chemistry package #315. Dry matter content of the TMRs was determined weekly. Urine (free-catch) and blood (coccygeal venipuncture) samples were collected between 0700 and 0900 h (before the morning feeding) every Monday, Wednesday and Friday from 21 days before expected calving until cows calved. Urine and blood was also collected on days 0 or 1, 7 ± 3 , 14 ± 3 , and 21 ± 3 after calving. Urine pH was determined (CMS Labcraft pH 101 with an Oakton pH probe) within 20 minutes of collection. Blood was collected in evacuated tubes (Vacutainer Beckton Dickinson and Co., Franklin Lakes, NJ), stored overnight at 4°C, centrifuged ($2,500 \times g$, 15 min) and harvested serum stored at -20°C until assayed for glucose (Wako Autokit Glucose 439-90901) content. Coccygeal blood was also collected in 1 mL syringes (Pro-Vent Plus) that contained lithium heparin (15 USP/mL

of blood) on -21 ± 3 , -7 ± 3 , 0 or 1, 7 ± 3 , and 21 ± 3 days of lactation and immediately analyzed on a hand-held iSTAT blood analyzer (Abbot Point of Care) with an EG7+ cartridge for ionized Ca (iCa), pH and other acid-base indicators.

Statistical Analyses

Statistical analyses were conducted with SAS programs (2001, SAS Inst. Inc., Cary, NC). Data were analyzed as a completely randomized design by the mixed model procedure for repeated measures. The first order autoregressive option was used as the covariance structure when measurements were evenly spaced and the spatial power law option when they were not. Time (day or week of sampling) was used as the repeated effect. The models contained the variable mean, treatment, time, all interactions, and error. Results are reported as least squares means and means were considered to differ when $P < 0.05$.

Results and Discussion

On a dry matter basis, MegAnion represented 9.17% of the grain mix and 2.88% of the prepartum TMR (Table 1). Both diets were formulated to have DCAD values of -12 mEq/100g DM but analyses indicated values of -3 and -7 mEq/100g for Control and MegAnion TMRs, respectively (Table 2). The reason(s) for this discrepancy are not known but an evaluation of TMR mixing logs did not reveal daily deviations sufficient to alter these values. Analytical variation could contribute to this discrepancy.

Feeding MegAnion and the control for 14 days had no impact on the whole blood components measured via iSTAT during the treatment period (Table 3). Ionized blood calcium (iCa) levels were maintained in both treatments well above critical calcium levels of 4.0 mg per dL during calving and the overall study, where the prevalence of subclinical hypocalcemia, is typically characterized by ionized calcium less than 4.0 mg/dL (or less than 1 mM). Prepartum and postpartum iCa was not different between treatments ($P=0.791$). Both treatments restored blood iCa at similar rates postpartum, increasing from 4.5 mg/dL at 1 d postpartum to 4.9 mg/dL at d 7. Cows fed the MegAnion diet for 21 d prepartum had similar postpartum iCa compared with the control. Concentrations of iCa decreased through 24 h postpartum, but increases in iCa were measured within 72 h postpartum, resulting in a significant time effect (WOS; $P=0.001$).

Urine pH was lower in cows fed MegAnion than the control during the treatment period, however there was no statistical difference between diets ($P=0.174$; Table 4). There was an interaction of treatment and week of study ($P = 0.046$) as urine pH was less in cows fed MegAnion from 9 to 14 days of treatment but greater in MegAnion cows during week 1 of lactation when cows were no longer fed MegAnion (Table 4, Figure 1).

The urine pH data indicate the amount of Control and MegAnion supplement fed to the cows was not sufficient to reduce pH to the desired range of 6.5 ± 0.3 . This is most likely due to the more positive dietary DCAD values than expected and to feeding an insufficient amount of each supplement.

It is recognized that coccygeal sampling collects venous or arterial blood. This difference in source has little impact on metabolites and endocrine components but can have large impacts on partial pressure measurement of gasses. Thus, these partial pressure measures should not be considered valid or be used unless source of the sample is considered. Because this was not

of primary interest, there was no attempt to identify source of coccygeal blood collected in this study.

The week of study effects on blood components (Table 5, Supplemental Figures) were expected as they typically occur when cows transition into lactation. There were no interactions of treatment and week of study (Table 5).

Prepartum DMI means were unaffected by treatment ($P=0.927$). Dry matter intake increased after calving in both treatments and followed similar increases in postpartum DMI from week 1 through 3. Body weight, body condition score, DMI and milk yield of cows fed Control or MegAnion diets did not differ (Table 6) but changes expected in these components as the cows transitioned into lactation were detected (Table 6, Supplemental Figures).

Summary

Although a sufficient reduction in urine pH was not achieved, the primary goal of this study was to assess if feeding MegAnion had a substantial impact on feed intake or any other detrimental impact. There were no indications that cows exhibited any refusal to consume the MegAnion supplement. These results indicate that inclusion of MegAnion in a TMR had no substantial negative impact on feed intake by prepartum dairy cows and indicate MegAnion can be fed to prepartum cows. No detrimental effects were observed from feeding MegAnion prepartum. Additionally feeding MegAnion did not have detrimental effects on postpartum performance in multiparous dairy cows and may provide similar or increased postpartum protection against hypocalcemia and metabolic disorders compared with traditional anionic salt feeding strategies.

Table 1. Ingredient contribution to grain and total mixed ration¹

Component	Anionic Salts	MegAnion ²	Early Lactation Protein Mix
Grain Mix, % of DM			
Soy hulls	39.00	44.77	----
Canola meal	20.00	13.90	14.07
Soybean meal (47% CP)	20.00	13.90	12.80
Soy Pass ³	----	----	11.31
Corn, ground shell	11.00	10.44	----
Corn, fine rolled	----	----	34.91
Distillers Dried Grains, dry	----	----	10.69
Calcium sulfate	2.36	----	----
Magnesium sulfate	1.64	----	----
Calcium chloride	1.45	----	----
MegAnion	----	9.17	----
Calcium carbonate	----	3.00	3.68
Potassium carbonate	----	----	1.08
Bio-Sel Dry Cow 1000 ⁴	1.64	1.63	----
Blood meal	1.09	1.09	4.28
Sodium Bicarb	----	----	3.90
Fat	0.91	0.91	----
UltraMet ⁵	----	----	1.90
Salt, white	----	----	0.92
DiCal, 21%	0.45	0.45	----
Magnesium oxide	0.45	0.73	0.43
Rumensin 90	0.01	0.01	0.03
TMR, % of DM			
Corn silage	40.63	37.14	34.91
Grass hay	25.00	31.43	4.55
Anionic salt grain mix	34.38	----	----
MegAnion grain mix	----	31.43	----
Alfalfa hay	----	----	8.18
Corn gluten feed	----	----	6.36
Corn, coarse ground	----	----	15.27
Cottonseed, fuzzy	----	----	3.64
Molasses	----	----	3.45
Early Lactation Protein mix	----	----	20.00
Energy Booster ⁶	----	----	3.64

¹TMR

²A palatable low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, processed grain by products and hydrochloric acid.

³A rumen bypass protein supplement

⁴A base mix formulated by VitaPlus to provide calcium and phosphorus and all essential minerals and vitamins

⁵A concentrated source of methionine for ruminants from VitaPlus

⁶Hydrolyzed animal fat from Milk Specialties

Table 2. Nutrient composition of total mixed rations (TMR)

Component ¹	Dry cow TMR		Early Lactation TMR
	Anionic Salts	MegAnion ²	
Dry matter, %	52.8	52.3	59.3
Crude protein, %	13.1	13.3	16.5
Available protein, %	12.2	12.4	15.6
ADICP, %	0.9	0.9	0.9
Soluble crude protein, %CP	40.0	47.0	39.0
ADF, %	28.8	33.2	24.2
Lignin, %	3.9	5.0	3.3
NFC, %	31.4	29.4	36.1
Starch, %	18.2	16.6	23.7
ESC (simple sugars), %	3.2	3.0	3.4
Crude fat, %	2.9	2.6	5.8
Ash, %	6.98	6.66	7.83
NEL, Mcal/kg	1.52	1.45	1.74
Ca, %	0.74	0.60	0.66
P, %	0.33	0.33	0.40
Mg, %	0.37	0.36	0.38
K, %	1.12	1.12	1.34
Na, %	0.09	0.70	0.48
Fe, ppm	302	301	291
Zn, ppm	59	67	95
Cu, ppm	15	16	25
Mn, ppm	63	70	77
Mo, ppm	1.3	1.2	1.2
S, %	0.37	0.26	0.29
Cl, %	0.46	0.80	0.56
DCAD, mEq/100g	-3	-7	21

¹Nutrient composition from analyses of composite samples collected during the study.

²A palatable low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, processed grain by products and hydrochloric acid.

Table 3. Effect of MegAnion¹ on blood components² in prepartum multiparous cows³ after 14 days of treatment⁴

Component	Treatment		SE ⁵	P-value	
	Control	MegAnion		COV ⁶	TRT ⁷
pH	7.42	7.47	0.02	0.918	0.115
pCO ₂ , mm Hg	39.65	35.42	1.80	0.976	0.153
pO ₂ , mm Hg	40.89	77.73	13.60	0.224	0.107
HCO ₃ , mmol/L	25.87	25.73	0.57	0.268	0.869
BEEcf ⁸ , mmol/L	1.71	1.95	0.58	0.266	0.783
sO ₂ , %	70.43	83.28	10.30	0.620	0.414
Na, mmol/L	142.49	143.61	0.58	0.052	0.247
K, mmol/L	3.98	4.19	0.11	0.420	0.235
tCO ₂ , mmol/L	26.97	26.84	0.61	0.304	0.887
iCA, mmol/L	5.05	5.16	0.07	0.677	0.290
HCT, % PCV	26.70	26.56	0.51	0.006	0.857
HGB, g/dL	9.06	9.04	0.18	0.008	0.933

¹MegAnion is a palatable low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, processed grain by products and hydrochloric acid. Treatments were fed from day 21 days before expected calving to calving.

²Measured via iSTAT technology (Abbot Labs) with an EG7+ cartridge

³Results from 6 Control and 5 MegAnion cows

⁴Components determined on day 0 and 14 of treatment (-21 and -7 days before expected calving) and the day 0 values were used as covariates.

⁵Standard error

⁶Covariate

⁷Treatment

⁸Calculated base excess in extracellular fluid

Table 4. Effect of MegAnion¹ on urine pH and serum glucose in multiparous cows

Component	Treatment			P-value			
	Control	MegAnion	SE ²	COV ³	TRT ⁴	Time ⁵	TRT*Time ⁶
Serum glucose, mg/dL							
Prepartum ⁷	78.6	81.2	1.64	0.051	0.279	0.396	0.811
Overall ⁸	67.1	69.5	3.40	----	0.639	0.397	0.701
Urine pH							
Prepartum ⁷	7.78	7.56	0.11	0.133	0.174	0.144	0.119
Overall ⁹	7.82	7.76	0.08	0.122	0.616	0.010	0.046

¹MegAnion is a palatable low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, processed grain by products and hydrochloric acid. Treatments were fed from day 21 days before expected calving to calving.

²Standard error

³Covariate

⁴Treatment

⁵Time as daily or weekly measures

⁶Interaction of treatment and time

⁷Measured every Monday, Wednesday, and Friday from day 2 to 16 of treatment with day 0 as a covariate

⁸Measured once during weeks -1, 0, 1 and 3 of lactation

⁹Measured every Monday, Wednesday, and Friday from day 2 to 16 of treatment and once during weeks -1, 0, 1 and 3 of lactation with values from day 0 of treatment as covariates

Table 5. Effect of MegAnion¹ on blood components² in multiparous cows³ during the study⁴

Component	Treatment		SE ⁵	P-value			
	Control	MegAnion		COV ⁶	TRT ⁷	WOS ⁸	TRT*WOS ⁹
pH	7.43	7.46	0.01	0.403	0.040	0.329	0.429
pCO ₂ , mm Hg	41.15	37.24	0.92	0.186	0.007	0.470	0.738
pO ₂ , mm Hg	47.35	56.90	5.69	0.024	0.258	0.079	0.173
HCO ₃ , mmol/L	26.96	26.85	0.49	0.224	0.876	0.021	0.818
BEEcf ¹⁰ , mmol/L	2.67	3.02	0.54	0.049	0.646	0.009	0.697
sO ₂ , %	70.76	76.13	5.15	0.211	0.474	0.302	0.347
Na, mmol/L	140.90	140.58	0.55	0.676	0.700	0.001	0.831
K, mmol/L	4.07	4.14	0.09	0.314	0.575	0.273	0.730
tCO ₂ , mmol/L	28.18	28.09	0.50	0.232	0.909	0.016	0.854
iCA, mmol/L	4.84	4.87	0.08	0.480	0.791	0.001	0.888
HCT, % PCV	26.60	27.56	0.69	0.060	0.359	0.001	0.522
HGB, g/dL	9.04	9.38	0.23	0.055	0.320	0.001	0.513

¹MegAnion is a palatable low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, processed grain by products and hydrochloric acid. Treatments were fed from day 21 days before expected calving to calving.

²Measured via iSTAT technology (Abbot Labs) with an EG7+ cartridge

³Results from 6 Control and 3 MegAnion cows

⁴Components were determined on day 0 and 14 of treatment, at calving (day 0 or 1 of lactation) and at 7 and 21 days of lactation with values from day 0 of treatment as covariates

⁵Standard error

⁶Covariate

⁷Treatment

⁸Week of study

⁹Interaction of treatment and week of study

¹⁰Calculated base excess in extracellular fluid

Table 6. Effect of MegAnion¹ on body weight (BW), body condition score (BCS)², dry matter intake (DMI) and milk yield of multiparous cows³

Component	Treatment		SE ⁴	COV ⁵	P-value		
	Control	MegAnion			TRT ⁶	Time ⁷	TRT*Time ⁸
BW, kg							
Prepartum ⁹	789	789	9.7	0.001	0.960	0.294	0.224
Postpartum ¹⁰	710	630	34	---	0.146	0.047	0.481
BCS							
Prepartum ⁹	3.38	3.27	0.08	0.028	0.396	0.788	0.855
Overall ¹⁰	3.17	3.00	0.11	----	0.323	0.184	0.999
DMI, kg/d							
Prepartum ¹¹	11.2	11.4	1.20	0.200	0.927	0.069	0.465
Overall ¹²	16.6	14.6	1.32	0.146	0.323	0.001	0.248
Milk yield, kg/d	40.9	35.0	3.00	----	0.212	0.013	0.370

¹A palatable low sulfur, high chlorine, anionic mineral supplement that contains plant protein products, ammonium chloride, magnesium sulfate, processed grain by products and hydrochloric acid.

²Five point scoring system of Wildman where 1 is thin and 5 is obese.

³Results are from 6 Control and 3 MegAnion cows.

⁴Standard error

⁵Covariate

⁶Treatment

⁷Time as daily or weekly measures for urine pH or as weekly means for DMI and milk yield

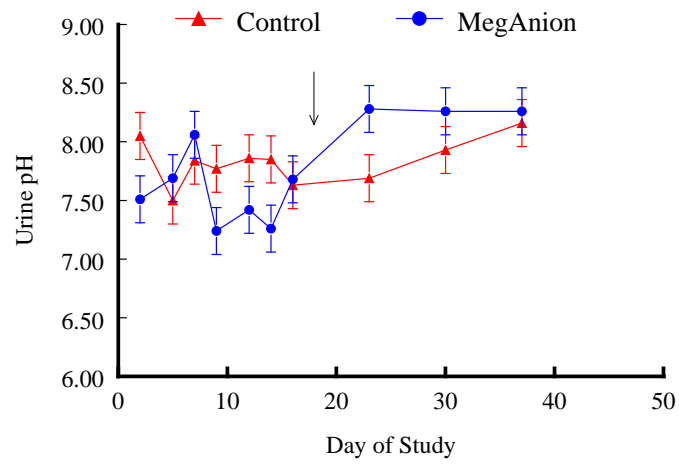
⁸Interaction of treatment and time

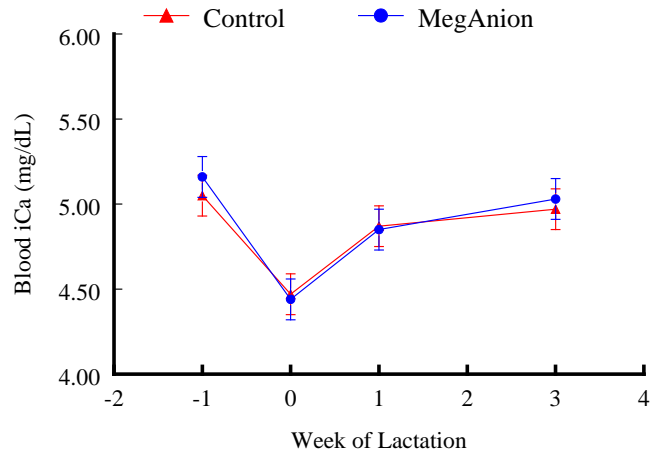
⁹Day 1 to 16 of treatment with day 0 as a covariate

¹⁰Measured once during weeks -1, 0, 1 and 3 of lactation

¹¹Weekly means from week 1 and 2 of treatment with the mean from the week before treatment used as a covariate

¹²Weekly means from week 1 and 2 of treatment and the first 3 weeks of lactation with the mean from the week before treatment used as a covariate





Supplemental figures (S1 to S14) are provided as a PowerPoint attachment.