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Mineral deficiencies in beef cattle from southern Brazil
Deficiências minerais em gado de corte no sul do Brasil

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Abstract

Minerals play an important role in the cattle metabolism with great impact in productive and reproductive performance of animals raised on extensive grasslands. At present, there is a lack of information concerning mineral deficiency throughout the analysis of biological fluids in Rio Grande do Sul State (southern Brazil). The Central Valley region of this State is characterized by extensive production on natural pastures with poor mineral supplementation. The low levels of minerals found by previous studies on pastures in this area motivated this research. The aim of the present work was to detect possible mineral deficiencies in beef cows by determining concentrations of minerals in biological fluids and in pasture. There were determined concentrations of phosphorus, calcium, copper, zinc, glutathion peroxidase (as indicator of selenium status) and thyroxine (as indicator of iodine status) in blood, sodium and potassium in saliva, and calcium, phosphorus, potassium, zinc, copper, sodium and iron in pastures, in four different periods of the reproductive cycle as follows: service period (artificial insemination), clean up bulls period, end of gestation and early lactation. Four herds were selected in Cachoeira do Sul County for this study which included 112 animals. The results showed marginal deficiencies of phosphorus, copper, iodine and selenium. Calcium concentration was below normal level and could be related to the low protein content of pasture. Most critical periods were the end of gestation and the early lactation.

Key-words:
Mineral profile.
Mineral deficiency.
Southern Brazil.
Beef cattle.

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Introduction

Brazil holds the second greatest livestock in the world (155 million heads). Rio Grande do Sul (southern Brazil) is the fourth greatest Brazilian State in beef cattle (13.7 million heads). This activity occupies 56% of the State area.[1] Almost all cattle in this State are raised on native pastures with poor mineral supplementation along the year. Grass native species are predominantly of low productivity and have low protein and mineral levels.[2] Consequently, productivity indicators of beef cattle in the State are poor.[3] Body weight losses of 30% have been reported during winter season (June to August) in cattle on native pasture without supplementation.[4]

Previous works have indicated mineral deficiencies in Rio Grande do Sul State, namely in phosphorus[5,6], copper[5,7,8,9], selenium[10], as well as molybdenum excess which predispose to copper deficiency.[8] Almost all those works have diagnosed mineral deficiencies on pathological or plant analysis basis. Identification of mineral deficiencies strategy must include, besides, animal tissues and fluid analysis of mineral

components, as well as performance response to supplementation.[6,11,12]

The aim of the present work was to detect possible mineral deficiencies in beef cows from a region of Rio Grande do Sul State by determining blood and saliva levels of mineral indicators along the reproductive cycle.

Materials and Methods

The Central Depression Region of Rio Grande do Sul State (southern Brazil) was selected for this work. This region holds 20% of beef cattle of the State (2.8 million heads). Its soils are classified as acid, moderately fertile and poor in phosphorus.[2] Native predominant pastures are *Paspalum notatum*, *Desmodium sp* and *Eryngium sp.*[13] Livestock is raised in extensive conditions, grazing continuously native pastures with poor mineral supplementation. Mineral deficiency in pasture reported by Cavalheiro and Trindade[2] and Senger et al.[14,15] and the economic relevance of this region were factors determining the choice of this geographic area of the State for this study.

Four similar herds were selected among representative farms in terms of breeds, husbandry, number of animals and type of native pasture (Table 1). The farms were not fertilized or used previously for agricultural purposes. All herds grazed native pastures and received weekly common salt (NaCl) *ad libitum*. Pasture, blood and saliva samples were collected in four periods during the 2001-2002 reproductive season, as follows: service period (artificial insemination: summer), clean up bulls period (autumn), end of pregnancy (winter) and early lactation (spring). Autumn and winter seasons are characterized by lesser forrage production and quality. In each period, seven cows of each herd were aleatory selected. A total of 112 samples were collected.

Blood samples were collected from coccygeal venipunction using vacutainer tubes (Becton Dickinson Co., Franklin Lakes, NJ, USA) with and without heparin. Samples were kept refrigerated during transportation to the laboratory. One aliquot of 50 mL of total blood was hemolized in a diluting agent (Randox, Antrim, United Kingdom) for determination of glutathion peroxydase (GSH-Px) activity in erythrocytes by UV method[16], as indicator of selenium status. Hemoglobin (Hb) concentration was also determined in total blood samples by cyanate colorimetric method (Labtest Diagnóstica, Lagoa Santa, MG, Brazil) in order to express activity of GSH-Px in U/g Hb. Remaining blood samples were centrifuged to obtain plasma (heparinized samples) or serum (non-heparinized samples), which were frozen (-20°C) until analysis of metabolites.

Calcium was determined by ftalein purple method and phosphorus by phosphomolibdenium colorimetric method (Labtest Diagnóstica, Lagoa Santa, MG, Brazil). Copper and zinc were analyzed by atomic absorption spectrophotometry using dilutions described by Fick et al.[17]. Serum thyroxine (T_4), as indicator of iodine status, was determined by radioimmunoassay (Diagnostic Products Co., Los Angeles, USA).

Saliva samples were collected from the lateral region of oral cavity with a 19 cm length steel probe connected to a plastic cannula and a 60 ml syringe. At the laboratory, saliva samples were centrifuged (2800 rpm, 10 min) and supernatant kept in eppendorf tubes at -20°C. Sodium and potassium were determining in saliva by in a Perkin Elmer instrument (Analyst 100) in acethylene/antioxidant flame emission mode.

Pasture samples were collected manually with stainless steel scissors, kept in plastic bags and dry at 75°C in an air circulating stove until constant weight. Samples were grounded in stainless steel mill to determine calcium, phosphorus, sodium, potassium, zinc, copper and iron by atomic absorption spectrophotometry.

All obtained data were organized in casualized blocks considering period sampling as treatment for analysis of variance using SAS program version 6.12 (SAS Institute, Cary, NC, USA). Tukey test was used for mean comparisons.

Results and Discussion

Values of blood and saliva components indicating mineral metabolism in the four periods of the reproductive cycle are shown in table 2. Mineral contents in pastures are shown in table 3. General mean of plasma phosphorus was 1.55 ± 0.32 mmol/L. Kaneko, Harvey and Bruss[18] reported reference range values for this mineral between 1.80 and 2.10 mmol/L. Plasma phosphorus values are good indicator of phosphorus intake in ruminants.[19] The results obtained in all periods, except in the service period (AI), showed that cows might be under phosphorus deficiency, which is worsened during early lactation period, when mean reached 1.32 mmol/L. This value is considered as critical by Timm[20] and worth mineral supplementation. González et al.[21] found a mean of 1.68 mmol/L for plasma phosphorus in beef heifers from a neighbor region, indicating phosphorus deficiency in native pastures along the year. Lisboa et al.[22] consider that cows with plasma phosphorus level bellow 1.16 mmol/L may show deficiency symptoms.

Phosphorus content in pasture was 0.15% ± 0.05, which is not adequate to fulfill recommended requirements of 0.16% for beef cattle[23]. Cavalheiro and Trindade[2] and Senger et al.[14] have found mean phosphorus of 0.13% in native pastures of southern Brazil. Correlation coefficient between phosphorus in plasma and in pastures was not significant (r= 0.04), suggesting that variations of plasma phosphorus levels were more related to mineral metabolic demand than to mineral intake. Lactation seems to be the period when phosphorus demand is higher and deficiency is more frequent. These finding is in according to Gióvine[24], who mentioned that plasma phosphorus levels are low in beef cows during lactation.

Mean plasma calcium was 1.94 ± 0.16 mmol/L. The level of this mineral was bellow minimum values (2.43 mmol/L) reported by Kaneko, Harvey and Bruss[18] and by Bauer, Santos and Mancuso[5] in southern Brazil (2.16 mmol/L). Plasma calcium concentration was significantly higher in cows during service period (2.43 mmol/L) and lower value was seen in lactation cows (1.86 mmol/L, possibly related to higher calcium demand in milk synthesis (Table 2).

Mean calcium content in pastures was 0.59% ± 0.11 (dry matter basis), which is similar to the value (0.60%) found by Senger et al.[15] but higher than that of Cavalheiro and Trindade[2] in the same region (0.25%). The requirements of Ca in feed recommended by NRC[23] for beef cows are 0.19-0.25%. Calcemia values bellow reference range found in this work in spite of Ca content in pastures, might reveal poor availability of the mineral in native pastures. Also, this may be due to low protein intake, which may reduce calcium intestinal absorption and calcium circulating levels bound to albumin.[25] Cavalheiro and Trindade[2] reported great fluctuations of protein quality and contents in native pastures of southern Brazil.

Mean plasma zinc was 15.93 ± 2.75 mmol/L. Mc Dowell[6] reported reference range of plasma zinc between 9.19 and 12.24 mmol/L. Cows in service period had the highest values of zinc (18.96 mmol/L). Although zinc contents in pastures (mean 21.5 ppm) were bellow NRC[23] requirements of 30 ppm (Table 3), plasma concentrations of this mineral do not indicate deficiency. Senger et al.[14] have found 16 ppm of zinc in pastures of the same region. This result may indicate that zinc requirements for beef cattle of the observed region could be lower than values reported by NRC[23]. This finding is in agreement with Morais et al.[26] who observed normal serum values of zinc in cows feeding pastures with low levels of this mineral in Brazil. In another work[27] was evidenced an increase in zinc plasma concentration of cows receiving zinc supplementation. Lower levels of plasma zinc in pregnant and lactating cows (Table 2) could suggest a higher demand of the mineral in those periods. Wittwer et al.[28] in Chile mentioned that lactating cows had lower plasma zinc than cows in another period.

Mean plasma copper was 10.93 ± 2.26 mmol/L. McDowell[6] cited reference range for plasma copper of 9.42 to 23.55 mmol/L. There was a significant lower copper value in pregnant cows (9.09 mmol/L), which may be considered in a deficient level as McDowell[6] describes (deficiency less than 9.42 mmol/L). Riet-Correa[29] mentioned that deficiency situations are observed when copper content in pasture is under 3 ppm. In the present work mean copper content in pastures was 7.31 ± 1.49 ppm and Cavalheiro and Trindade[2] have found 6 ppm in the same region. Wittwer et al.[28] affirmed that pregnant cows showed significant decrease of blood copper as gestation progress. The results evidence that, although copper contents in pasture is apparently adequate, pregnant cows may suffer marginal deficiency of this element.

Molybdenum is an important antagonist of copper, especially if feed level is above 10 ppm. In this work, mean Mo in pastures was 0.15 ppm, which is bellow levels reported by Cavalheiro and Trindade[2] for the same region (0.32 ppm). Nevertheless, mean iron content was 722 ppm, value that is beyond the maximum value (500 ppm) recommended by NRC[23]. Cavalheiro and Trindade[2] found similar iron levels in pastures of this area. It is possible that a marginal copper deficiency condition may be configured due to the high level of iron in pastures of the studied region, since iron interferes with copper metabolism.[30]

Glutation peroxidase (GSH-Px) activity has been used as indicator of selenium balance.[31] This enzyme is abundant in red blood cells and may be measured by photometric techniques.[16] Mean GSH-Px activity obtained in this work was 33.1 ± 21.7 U/g Hb. According to Wittwer[31] this value may be considered as compatible with selenium deficiency (<60U/g Hb). There are no works evaluating selenium content in pastures in southern Brazil, but the results obtained here may be suggesting a marginal deficiency of selenium in the studied region. Barros et al.[10] described a case of white muscle disease in cattle, possibly linked to selenium or vitamin E deficiency. Cows in service period had significant higher values of GSH-Px, revealing less selenium demand in that period. It is recommended more research to evaluate biological response to selenium supplementation in cattle from southern Brazil.

Serum thyroxine (T_4) levels are used as indicator of iodine balance because this mineral is almost exclusively used in thyroid hormone synthesis.[18] Mean value of serum thyroxine was 44.2 ± 15.4 nmol/L. Kaneko, Harvey and Bruss[18] cited a reference range for this hormone of 54 to 110 nmol/L in bovines. This finding strongly suggests iodine marginal deficiency in cattle of this region of southern Brazil, especially in gestation and lactation cows, which had significant lesser values of T_4 (Table 2). This result is in agreement with the values of T_4 reported by González, Dias and Riccó[32] for beef heifers in Rio Grande do Sul State (mean 30.24 nmol/L). Contreras et al.[33] found lower values of T_4 in lactating cows compared to other groups of cows in the same feed regime, and Dayrell[34] mentioned that lactating cows have higher iodine requirements as a consequence of iodine excretion by milk.

Sodium:potassium (Na:K) ratio in saliva has been used as indicator of sodium balance by several workers[19,35], who mentioned that in sodium deficiency, Na:K ratio in saliva may diminish bellow 10. Underwood and Suttle[30] observed a Na:K ratio of 0.45 in saliva from bovines with sodium deficiency. In the present work, mean saliva sodium (88.19 ± 31 mEq/L) and potassium (22.3 ± 17.1 mEq/L) made a mean Na:K ratio of 9.8. This suggests a marginal sodium deficiency, mainly in pregnant cows (Table 2).

Sodium content in pastures had a mean of 147 ± 85 ppm, which is bellow the value reported by Cavalheiro and Trindade[2] for the same region (300 ppm). In any case, sodium content of native pastures did not fulfill recommended requirements of 600-1000 ppm[23]. Potassium mean content in pastures was 0.51% ± 0.1.

Cavalheiro and Trindade[2] obtained 1% and Senger et al.[28] found 0.9% of K in pastures in the same area. It is necessary to perform more studies using supplementation response on saliva Na:K ratio to describe precisely the degree of sodium deficiency in beef cattle from this region of southern Brazil.

In abstract, using analysis of mineral indicators in blood and pastures, the present work detected deficiency of phosphorus in beef cows, more severely in early lactation period. Plasma calcium level was bellow normal reference range, possibly associated to low protein intake. It is necessary to perform more research to test this hypothesis. Although zinc content in pasture was bellow recommended level, plasma concentration was within normal reference values. There was detected a marginal (without evident symptoms) copper deficiency in pregnant

Table 1
Characteristics of studied beef herds in Rio Grande do Sul State, southern Brazil, 2002

Herd characteristic	Number of herd			
	A	B	C	D
Total area (Ha)	1200	270	700	1500
Number of animals	600	315	450	665
Grazing load (animal/Ha)	0.8	0.6	0.7	0.8
Pregnancy rate (%)	55	45	40	56
Predominant breeds	Zebu x Charolais	Zebu crossbred	Zebu x Angus	Zebu x Shorthorn

Table 2
Mean values of metabolites indicating mineral status of beef cows during four different periods of the reproductive cycle in southern Brazil, Rio Grande do Sul, 2002

Metabolite	Sampling period			
	Service (AI)	Clean up bulls	End pregnancy	Early lactation
N	28	28	28	28
Plasma calcium (mmol/L)	$2.43^{a} \pm 0.20$	$1.90^{b} \pm 0.12$	$1.94^{b} \pm 0.14$	$1.86^{b} \pm 0.12$
Plasma phosphorus (mmol/L)	$1.87^{a} \pm 0.14$	$1.47^{b} \pm 0.08$	$1.56^{b} \pm 0.10$	$1.32^{c} \pm 0.19$
Plasma Ca/P ratio	$1.11^{a} \pm 0.19$	$1.32^{b,c} \pm 0.20$	$1.27^{a,b} \pm 0.21$	$1.48^{c} \pm 0.36$
Plasma zinc (mmol/L)	$18.96^{a} \pm 2.09$	$15.47^{b} \pm 1.64$	$14.21^{c} \pm 2.08$	$14.86^{b,c} \pm 2.42$
Plasma copper (mmol/L)	$11.66^{a} \pm 0.71$	$12.06^{a} \pm 2.10$	$9.09^{b} \pm 1.17$	$10.94^{a} \pm 1.36$
Erythrocyte GSH-Px (U/g Hb)	$52.27^{a} \pm 25.52$	$30.03^{b} \pm 20.52$	$20.26^{b} \pm 7.25$	$22.94^{b} \pm 9.44$
Serum thyroxine (nmol/L)	$52.53^{a} \pm 13.72$	$51.24^{a} \pm 16.44$	$40.28^{b} \pm 12.07$	$32.83^{b} \pm 9.75$
Saliva sodium (mEq/L)	*	$89.16^{a} \pm 26.24$	$78.02^{b} \pm 29.19$	$97.38^{a} \pm 35.02$
Saliva potassium (mEq/L)	*	21.15 ± 17.03	22.89 ± 17.22	22.84 ± 17.12
Saliva Na/K ratio		8.18	9.83	11.44

Values with different letters significantly differ ($p < 0.05$) among sampling periods. * Unavailable samples

Table 3
Mean values of mineral components of native pastures during four different periods of the reproductive cycle in southern Brazil, Rio Grande do Sul, 2002

Mineral (dry matter basis)	Sampling period			
	Service (AI)	Clean up bulls	End pregnancy	Early lactation
Calcium (%)	0.55 ± 0.13	0.66 ± 0.18	0.60 ± 0.12	0.61 ± 0.04
Phosphorus (%)	0.16 ± 0.07	0.13 ± 0.04	0.15 ± 0.04	0.15 ± 0.05
Potássio (%)	*	$0,65 \pm 0,36$	$0,46 \pm 0,12$	$0,44 \pm 0,07$
Zinc (ppm)	21.50 ± 6.70	22.75 ± 6.08	20.00 ± 4.26	21.75 ± 14.85
Copper (ppm)	9.00 ± 1.15	6.75 ± 1.26	6.50 ± 0.58	7.00 ± 1.83
Sódio (ppm)	*	$246,5 \pm 158,5$	$96,3 \pm 9,0$	$101,0 \pm 26,2$
Ferro (ppm)	810 ± 647	726 ± 536	637 ± 278	714 ± 254

* Unavailable samples

cows, in spite of adequate copper levels in pasture. High levels of iron in pastures may be involved in copper availability.

This work reports for the first time GSH-Px activity in red blood cells and thyroxine plasma levels in beef cows from southern Brazil and the results suggest marginal deficiency of selenium and iodine, which must be experimented by supplemental response works.

Resumo

Os minerais são de grande importância no metabolismo dos bovinos, tendo impacto no desempenho produtivo e reprodutivo dos animais. Atualmente há poucos relatos no Rio Grande do Sul com relação ao diagnóstico de deficiências minerais mediante a análise de fluidos biológicos em animais a campo. A região da Depressão Central do Rio Grande do Sul é caracterizada pela produção extensiva de gado de corte em campo nativo com manejo precário da suplementação mineral, apresentando baixos níveis de alguns minerais nas pastagens. O objetivo do presente trabalho foi diagnosticar possíveis deficiências minerais em vacas de corte na região da Depressão Central mediante a dosagem de indicadores minerais em fluidos biológicos e na pastagem. Foram determinadas concentrações de fósforo, cálcio, cobre, zinco, glutation peroxidase (como indicador de selênio) e tiroxina (como indicador de iodo) no sangue, sódio e potássio na saliva e sódio, e cálcio, fósforo, potássio, zinco, cobre, sódio e ferro na pastagem. Foram estudadas quatro propriedades em Cachoeira do Sul, em quatro momentos do ciclo reprodutivo: período de serviço (inseminação artificial), período de repasse de touros, final da gestação e início da lactação, num total de 112 animais. O perfil mineral indicou deficiência marginal de fósforo, cobre, iodo e selênio. O cálcio sérico apresentou-se diminuído, possivelmente relacionado com o baixo conteúdo de proteína na pastagem. Os períodos mais afetados foram o final da gestação e o início da lactação indicando que essas categorias de produção têm maior exigências metabólicas.

Palavras-chave:
Perfil mineral.
Deficiência mineral.
Gado de corte.
Sul do Brasil.

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