DIEGO ZANETTI

MINERAL RELEASE FROM DIFFERENT FEEDS, MINERAL BALANCE FOR NELLORE YOUNG BULLS, AND PREDICTION OF WATER INTAKE BY BEEF CATTLE

Thesis submitted to the Animal Science Graduate Program of the Universidade Federal de Viçosa as partial fulfillment of the requirements for the degree of *Doctor Scientiae.*

VIÇOSA
MINAS GERAIS – BRAZIL
2017

Ficha catalográfica preparada pela Biblioteca Central da Universidade Federal de Viçosa - Câmpus Viçosa

T
Z28m
2017

Zanetti, Diego, 1990-
Mineral release from different feeds, mineral balance for Nellore young bulls, and prediction of water intake by beef cattle / Diego Zanetti. – Viçosa, MG, 2017.
xi, 139f. : il. ; 29 cm.

Orientador: Sebastião de Campos Valadares Filho.
Tese (doutorado) - Universidade Federal de Viçosa.
Inclui bibliografia.

1. Minerais na nutrição animal. 2. Ruminantes - Alimentação e rações. 3. Cálcio na nutrição animal. 4. Fósforo na nutrição animal. 5. Cobre no organismo. 6. Água - Consumo. I. Universidade Federal de Viçosa. Departamento de Zootecnia. Programa de Pós-graduação em Zootecnia. II. Título.

CDD 22 ed. 636.208527

DIEGO ZANETTI

MINERAL RELEASE FROM DIFFERENT FEEDS, MINERAL BALANCE FOR NELLORE YOUNG BULLS, AND PREDICTION OF WATER INTAKE BY BEEF CATTLE

Thesis submitted to the Animal Science Graduate Program of the Universidade Federal de Viçosa as partial fulfillment of the requirements for the degree of *Doctor Scientiae.*

APPROVED: July 28th, 2017.

Ana Luiza Costa Cruz Borges

Stefanie Alvarenga Santos

Edenio Detmann
(Co-adviser)

Luciana Navajas Rennó
(Co-adviser)

Sebastião de Campos Valadares Filho
(Adviser)

To my niece, Cecília, who already bring happiness to who nears.

In memory of my grandfathers, Dico and Valentim, always with me!

ACKNOWLEDGMENTS

To God: source of strength, protection and faith, both necessary to move forward!

To my family, Paulo, Imaculada, and Paula: always my "Safe Harbor"! That in spite of the numerous challenges, they know as nobody, how create ties of affection and companionship.

To Katia: an indescribable companion. Person who knows how to take me along. You do not know how much you make me grow up.

To my relatives, especially to my grandparents: more than a vision of our tomorrow, an bridge with our past. Example of care among ourself.

To the Universidade Federal de Viçosa: for the teachings and opportunities offered since 2008, in particular to the Department of Animal Science.

To Professor Tiao: an example of a professional. For trust, patience, prompt service, dedication, guidance and teachings.

To Professors Edenio, Terry, Luciana, Mário Paulino and Mário Chizzotti, willing to contribute whenever necessary, for the excellent coorientation.

To the Ruminant Nutrition Group of Colorado State University. My year in Fort Collins was really memorable! An year to learn about science, languages, culture… life! Professors Terry, Archibeque, and John, Karen, Kessel, Jose, Maddie, Flor, Fernando, Sam, Octavio, Ashley, Hugo and Colton! Hope see you all soon! To my friends from FoCo, 203 will always be with me.

To the interns, undergraduate research fellows and graduate students (or better: to the friends who became interns and interns who became friends, not respectively): Ana Clara, Breno, Caio, Danillo, Douglas, Érica, Éverton, Flávão, Jarbas, Jéssiquinha, Laura, Leticia, Luiz Fernando, Magal, Markin, Meneguelli, Nathália, Pauliane, Mariana, and friends from LCC.

To the partners of experiment, Maurício e Letícia.

To Professors Stefanie and Ana Luiza and for the contribution to improve this work.

To the friends of SIMCORTE, Laura, Sinhá, Pedrão, Janaúba, Markin, Cajuru, Peão, Márcia, Thiago, Jéssiquinha, Ranyeri, and "calouros".

To the employees of DZO, Jojô, Pum, Zezé, Vanor, Marcelo Cardoso, Sérvulo, Monteiro, Seu Mário, Fernanda, Gabriel, Edson, Plínio and Seu Jorge.

To CNPq, Capes, INCT – Ciência Animal and Fapemig for the partial financing of this research, and CNPq, for the scholarship.

To all who have contributed in some way, Thank you for the moments of help, relaxing and personal growth.

BIOGRAPHY

DIEGO ZANETTI, son of Paulo Afonso Zanetti and Maria Imaculada Pinheiro Zanetti, was born in Viçosa/MG - Brazil on December 31st, 1990.

In 2008, joined the Universidade Federal de Viçosa, in an Animal Science course. In November of 2012, he obtained a Bachelor of Science.

In this date started the Master's degree course in the Department of Animal Science of Universidade Federal de Viçosa, concentrating his studies in Ruminant Nutrition and Production area, concluding this course in July of 2014.

In August of 2014 was started the PhD course in the same area and department. Between November of 2015 and October of 2016, he was Visiting Scholar at Colorado State University, USA, where part of his research was developed. The thesis has been submitted to the committee in July of 2017, to obtain the Doctor Scientiae degree in Animal Sciences.

CONTENTS

ABSTRACT

ZANETTI, Diego, D.Sc., Universidade Federal de Viçosa, July, 2017. **Mineral release from different feeds, mineral balance for Nellore bulls, and water intake prediction by beef cattle.** Adviser: Sebastião de Campos Valadares Filho. Co-advisers: Edenio Detmann and Luciana Navajas Rennó.

For the thesis composition were prepared four scientific manuscripts based on studies with mineral release, absorption, metabolism and balance, and water intake of beef cattle. In the first manuscript the objective was to quantify the dry matter (DM) digestibility and total ash (TA) and mineral release from 12 concentrate and 12 forage feedstuffs commonly fed to cattle using *in situ* and *in vitro* methods. The concentrate and forage feedstuffs were incubated in the rumen of ruminally cannulated beef bulls at 8 different time points. Two different trials were conducted for concentrates and forages, with maximum incubation time of 72 and 120 h, respectively. The residue from samples incubated for 24 h were treated with pepsin and HCl to simulate abomasum digestion. The initial and residual samples after *in situ* and *in vitro* incubations were measured. An asymptotic model was adopted for estimating solubility of minerals, degradation rate of DM, and TA. Correlations between feedstuff contents and mineral release were evaluated. Cluster analysis was performed to group feedstuffs, in relation to TA release. Large variability was observed between concentrate and forage feedstuffs for all analyzed constituents. Large variability was observed for the effective ruminal degradation of TA, and individual mineral release. When feedstuffs were clustered according with the α, β and kd estimates of TA ruminal release, 4 groups were identified. From group "1" to group "4", was observed an increase in the soluble fraction, and a reduction in both moderate releasable fraction and release rate. Neutral detergent fiber content has a negative correlation with mineral release in the rumen, while mineral content has positive correlation. These results demonstrate that mineral solubilization in digestive tract is not the limiting factor to mineral absorption from the feedstuffs tested. The objectives of the second and third manuscripts were to measure the effects of mineral supplementation on nutrient intake and digestibility, performance, mineral balance and requirements and mineral concentrations in the body of Nellore beef cattle fed with and without calcium (Ca), phosphorus (P) and micromineral (MM) supplemental sources during the growing and finishing phases. Nellore cattle (n = 51, initial body weight = 270.4 ± 36.6 kg, age = 8 months) were assigned to one of three

groups: reference (n = 5), maintenance (n = 4), and performance (n = 42). Reference group was slaughtered at the beginning of the experiment to measure initial mineral status. The maintenance group was used to collect values of animals at low gain and reduced mineral intake. Animals of performance group were assigned to one of six treatments: sugarcane as a roughage source and a soybean meal and soybean hull-based concentrate with (SH100) and without (SH0) Ca, P and MM supplementation; sugarcane as the roughage source and a soybean meal and ground corn-based concentrate with (SC100) and without (SC0) Ca, P and MM supplementation; and corn silage as the roughage source with a soybean meal and corn-based concentrate with (CS100) and without (CS0) Ca, P and MM supplementation. The experiment was conducted as a completely randomized design with a 3 × 2 factorial arrangement of treatments. Nutrient intake and digestibility, bone and serum parameters related to Ca and P metabolism, and liver mineral concentrations were measured. Orthogonal contrasts were adopted to compare mineral intake, fecal and urinary excretion and apparent retention among treatments. Maintenance requirements and true retention coefficients were generated with the aid of linear regression between mineral intake and mineral retention. Mineral composition of the body and gain requirements were assessed using non-linear regression between body mineral content and mineral intake. Nutrient intake, digestibility and performance were not affected by mineral factor ($P > 0.10$). Intakes of Ca, P, S, Cu, Zn, Mn, Co, and Fe were reduced in the absence of Ca, P, and MM supplementation ($P < 0.05$). Fecal excretion of Ca, Cu, Zn, Mn, and Co were also reduced in treatments without supplementation ($P < 0.01$). Overall, excretion and apparent absorption and retention coefficients were reduced when minerals were not supplied ($P < 0.05$). Rib bone breaking strength and densitometry were reduced ($P < 0.04$) in absence of supplementation. Metatarsus parameters were not affected ($P > 0.10$). Liver Cu content was reduced ($P < 0.01$) in diets without supplementation. Dietary mineral requirements were lower for P, Cu, and Zn and greater for Fe when compared to previously published recommendations.Therefore, absence of mineral supplementation does not influence intake and performance of Nellore beef cattle. However, this absence may influence serum, liver and bone parameters according to dietary type. This study provides useful information about mineral requirements and mineral supplementation to obtain adequate dietary mineral supply of Nellore cattle in tropical conditions. In the fourth manuscript, the objective was to validate six current water intake (WI) equations for beef cattle using water intake data from four experiments conducted in North America (n = 1 experiment; crossbred Angus beef steers) and Brazil (n = 3

experiments; Nellore beef cattle). Animal performance, diet composition, and environmental data were collected for all experiments. The prediction of WI using the current published WI equations was tested though the regression between predicted and measured WI values. All tested equations differed from the measured WI data from the four experiments. Several factors can help explain why the published equations did not predict the WI obtained from the three experiments, including that the tested equations were developed in temperate climates using predominantly *Bos taurus taurus*. From the current data, was generated new WI equations based on metabolic BW, DMI, humidity and temperature-humidity index for Nellore cattle in Brazil and metabolic BW, DMI, maximum daily temperature, and dietary concentrate level for Angus crossbred cattle in North America.

RESUMO

ZANETTI, Diego, D.Sc., Universidade Federal de Viçosa, julho de 2017. **Liberação de minerais em alimentos, balanço mineral em bovinos machos Nelore não castrados, e predição do consumo de água para gado de corte.** Orientador: Sebastião de Campos Valadares Filho. Coorientadores: Edenio Detmann e Luciana Navajas Rennó.

Para esta tese foram preparados quatro artigos científicos baseados em estudos de liberação de minerais de alimentos para ruminantes, absorção, metabolismo e equilíbrio de minerais para bovinos de corte, e ingestão de água de bovinos de corte. No primeiro artigo, o objetivo foi quantificar a digestibilidade da matéria seca (MS) e a disponibilidade de cinzas totais (CT) e liberação dos minerais de 12 alimentos concentrados e 12 volumosos comumente adotados em dietas para ruminantes, utilizando métodos *in situ* e *in vitro*. Os alimentos foram incubados no rúmen de machos não castrados por 8 deferentes tempos. Foram realizados dois ensaios, um para concentrados e outro para volumosos, com tempo máximo de incubação de 72 e 120 h, respectivamente. O resíduo das amostras incubadas durante 24 h foi tratado com pepsina e ácido clorídrico para simular a digestão no abomaso. As amostras iniciais e residuais após incubação *in situ* e *in vitro* foram avaliadas quanto aos teores de MS, CT e minerais. Um modelo assintótico foi adotado para estimar a liberação dos minerais, e as taxas de degradação da MS e CT. Foram avaliadas as correlações entre os teores nos alimentos e a liberação de cada mineral. "Clusteres" foram formados para agrupar os alimentos, em relação à liberação de CT. Observou-se grande variabilidade entre os alimentos concentrados e volumosos para todos os constituintes analisados. Grande variabilidade foi observada para a degradação ruminal efetiva da CT e para a liberação de cada mineral. Quando os alimentos foram agrupados de acordo com as estimativas α, β e kd da liberação ruminal de CT, foram identificados 4 grupos. Do grupo "1" para o grupo "4", observou-se um crescimento na fração solúvel e uma redução na fração de liberação lenta e na taxa de liberação. O conteúdo de fibra de detergente neutro apresentou uma correlação negativa com a liberação de mineral no rúmen, enquanto o conteúdo de minerais apresentou correlação positiva. Esses resultados demonstram que a solubilização mineral no trato digestivo não é o fator limitante para a absorção mineral dos alimentos testados. Os objetivos do segundo e terceiro artigos foram medir os efeitos da suplementação mineral sobre ingestão de nutrientes e digestibilidade,

desempenho, balanço e exigência de minerais, e concentrações de minerais no corpo de bovinos de corte Nelore alimentados com ou sem cálcio (Ca), fósforo (P) e fontes suplementares de microminerais (MM) durante as fases de crescimento e terminação. Os animais (n = 51, Nelore, peso corporal inicial = 270,4 ± 36,6 kg, idade = 8 meses) foram atribuídos a um dos três grupos: referência (n = 5), mantença (n = 4) e desempenho (n = 42) . O grupo referência foi abatido no início do experimento para estimar a composição corporal dos demais animais. O grupo mantença foi utilizado para coletar valores de animais com baixos ganho corporal e ingestão de minerais. Os animais do grupo de desempenho foram designados para um dos seis tratamentos: cana-de-açúcar como volumoso e concentrado a base de farelo de soja e casca de soja com (SH100) e sem (SH0) suplementação de Ca, P e MM; Cana-de-açúcar como volumoso e concentrado a base de farelo de soja e milho moído com (SC100) e sem (SC0) suplementação de Ca, P e MM; e silagem de milho como volumoso e concentrado a base de farelo de soja e milho moído com (CS100) e sem (CS0) suplementação de Ca, P e MM. O experimento foi conduzido como um delineamento inteiramente casualizado com um arranjo fatorial 3 × 2, sendo 3 tipos de dietas e presença ou ausência de suplementação inorgânica de minerais. O consumo e digestibilidade de nutrientes, os parâmetros ósseos e séricos relacionados ao metabolismo de Ca e P e as concentrações hepáticas de minerais foram mensurados. Para avaliar a ingestão de cada mineral, a excreção fecal e urinária e a retenção aparente entre os tratamentos, contrastes ortogonais foram adotados. As exigências de mantença e os coeficientes de retenção verdadeiros foram calculados com o auxílio da regressão linear entre o consumo e a retenção de cada mineral. A composição mineral do corpo e as exigências para ganho de peso foram avaliados, utilizando modelos não lineares entre o conteúdo corporal e o consumo de cada mineral. Os consumos, digestibilidade e desempenho de nutrientes não foram afetados pelo fator mineral ($P > 0,10$). Os consumos de Ca, P, S, Cu, Zn, Mn, Co e Fe foram reduzidas na ausência de suplementação de Ca, P e MM ($P < 0,05$). A excreção fecal de Ca, Cu, Zn, Mn e Co também foi reduzida em tratamentos sem suplementação ($P < 0,01$). Em geral, os coeficientes de absorção e retenção aparentes foram reduzidos quando os minerais não foram fornecidos ($P < 0,05$). A resistência à ruptura dos ossos da costela e a densitometria óssea foram reduzidas ($P < 0,04$) na ausência de suplementação. Os parâmetros do metatarso não foram afetados ($P > 0,10$). O conteúdo de Cu no fígado foi reduzido ($P < 0,01$) em dietas sem suplementação. As exigências totais dos minerais foram menores para P, Cu e Zn e maiores para o Fe quando comparados às recomendações previamente publicadas. Portanto, a ausência de suplementação mineral não

influencia o consumo e o desempenho de bovinos Nelore não castrados em confinamento. No entanto, esta ausência pode influenciar parâmetros séricos, hepáticos e ósseos de acordo com o tipo de dieta. Este estudo forneceu informações úteis sobre as exigências minerais e estratégias de fornecimento adequado de minerais para bovinos Nelore em condições tropicais. No quarto artigo, o objetivo foi validar seis equações já publicadas para predição da ingestão de água (IA) para bovinos de corte usando dados coletados em quatro experimentos realizados no Norte do Colorado (n = 1 experimento, novilhos de raça Angus) ou no Sudeste do Brasil (n = 3 experimentos, bovinos de corte Nelore). Características relacionadas ao desempenho animal, à composição da dieta e aos dados ambientais foram coletados em todos os experimentos. A predição de IA usando as equações publicadas atualmente foi testada através da regressão entre os valores previstos e mensurados. Todas as equações testadas diferiram dos dados observados. Vários fatores podem ajudar a explicar o porquê das equações publicadas não predizerem acuradamente a IA. Um dos fatores é que as equações testadas foram desenvolvidas em ambientes de clima temperado usando predominantemente animais *Bos taurus taurus*. A partir dos dados coletados no presente estudo, foram geradas novas equações baseadas no peso corporal metabólico, consumo de MS, umidade relativa e índice de temperatura e umidade para bovinos Nelore confinados no Sudeste brasileiro e baseadas no peso corporal metabólico, consumo de MS, temperatura máxima diária e nível de inclusão de concentrado na dieta para bovinos mestiços Angus no Norte do Colorado.

GENERAL INTRODUCTION

Among dietary constituents, mineral contents are lower than protein and carbohydrates. However, minerals are essential to keep animals' vital functions, and that is why mineral requirements are so important as other nutrients (Suttle, 2010). Dietary mineral absence may imply deficiencies, and consequently impair animal performance. Therefore, minerals are typically supplemented to cattle to prevent deficiencies and improve performance. So, most nutrient recommendations for cattle (USA: dairy cattle, NRC 2001; beef cattle, NRC 2016; United Kingdom: ARC 1980; AFRC 1993; and Brazil: BR-Corte, Valadares Filho et al*.,* 2010) recommends supply the nutritional requirements of macro and/or trace minerals as mineral premix. Therefore, minerals are normally supplemented to beef cattle diets without taking into account the inherent minerals within the feedstuffs (Spears, 1996). It should be noted that nutritional systems usually applied may overestimate minerals (safety margin).

Regarding mineral nutrition, it has been reported that, under standard feeding conditions, nutritionists supplement minerals in a way that exceeds the NRC (1996) recommendations (Vasconcelos and Galyean, 2007; Berret et al., 2015). It is important both from an economic perspective, due to financial inputs with supplementation, and an environmental perspective, due to excretion of potential pollutants, i.e. P (Jongbloed and Lenis, 1998; Sehested, 2004; Prados et al., 2017), to ensure adequate mineral supplementation.

Some researchers (Esser et al*.,* 2009; Prados et al*.,* 2016) have demonstrated that animals fed typical feedlot diets can meet their mineral requirements exclusively from basal feed ingredients. This is made possible because the mineral content in feedstuffs is, in general, high (Playne et al., 1978). Mineral bioavailability in feedstuffs is affected by the mineral distribution in plant cells, the form of the mineral, association with cell components, and interaction with other

minerals (Čerešňáková et al., 2005; Pogge et al,. 2014; Berrett et al., 2015). Mineral availability in the rumen depends on mineral content, fibre content and passage rate (Flachowsky and Grün 1992; Berrett et al. 2015). Furthermore, the solubility of minerals in general is increased under low pH conditions. Therefore, mineral content in feedstuffs can be divided into three fractions: very soluble, moderately soluble (minerals that are predominately associated with the fibre and/or protein fractions of the feedstuff), and un-releasable or insoluble minerals (Spears, 1994).

If mineral release from feedstuffs is not a limiting factor to absorption and use for cattle, it is an option for increase the sustainability of cattle production systems through the reduction of inputs. Despite this, two points may be considered in this context for reducing mineral input for cattle. First, in most tropical areas, cattle depend exclusively on forages to meet their mineral requirements (Perdomo, et al., 1977; Ibrahim et al., 1998; Costa e Silva et al., 2015; Costa et al., 2016). Second, when mineral supply in a production system is lowered, environmental excretion and costs are reduced (Khorasani et al., 1997; Humer and Zebeli, 2015).

Minerals that are not pottentiallyreleased from feedstuffs are unavailable for absorption. Minerals released from feedstuffs incubated in rumen have been reported to be highly correlated with mineral availability to the animal (Flachowsky et al., 1994). Therefore, this technique is an inexpensive, simple, rapid and reproducible technique that may allow absorption coefficients for minerals contained in feedstuffs to be estimated (Olubobokun et al., 1990).

Nellore beef cattle is a breed widely used in some parts of the world, but lacks substantial representation in the literature, mainly in relation to mineral nutrition. Furthermore, 83.1% of Brazilian feedlot nutritionists recommend providing supplemental minerals sources to beef cattle (Millen et al., 2009). Recently, Costa e Silva et al. (2015) reported no differences in feed intake or productive performance of Nellore bulls fed with or without Ca and P supplementation during the

growing and finishing phases of production. Likewise, research has demonstrated contradictory results for performance and mineral status based on micromineral (MM) supplementation for other cattle breeds (Engle and Spears, 2000; Mullis et al., 2003; Spears and Weiss, 2014). Besides this, different types of feedstuffs may propitiate different dietary minerals from the basal diet. For example, the sugarcane and corn silage are the most frequently used forage source in Brazilian feedlots (Millen et al., 2009; Pinto and Millen, 2016). However, these two forage sources are very different regarding Ca and P contents.

Recent research has demonstrated that suppling calcium (Ca) and phosphorus (P) to Nellore (Costa e Silva et al., 2015a) and Holstein × Zebu crossbreed (Prados et al., 2015) is not necessary in short terms feedlots. However, information about mineral requirements of *B. indicus* cattle is limited and the few existing studies did not include a negative control (Valadares Filho et al., 2010; Costa e Silva et al., 2015b). An accurate estimation of mineral requirements is therefore of economic and environmental importance (Jongbloed and Lenis, 1998; Humer and Zebeli, 2015; Spears and Weiss, 2014).

Similary to researches aiming measure mineral requirements of Zebu beef cattle, the water demands by cattle has been the aim of few studies, main under tropical conditions. This is worring when we know that beef production systems require a considerable amount of water. Mekonnen and Hoekstra (2012) reported that beef cattle production systems require more water than swine or poultry production systems. Moreover, Beckett and Oltjen (1993) reported that 3,682 L of water are required to produce one kg of boneless beef, considering drinking water, water in feedstuffs, water used to produce feedstuffs, and water used in carcass processing. However, in some parts of the world, water shortages have impacted livestock production (Ward and Michelsen, 2002; Pluske and Schlink, 2007).

Research aiming to increase the efficiency of water use for livestock production has been encouraged (Tilman et al., 2002; Rijsberman, 2006); however, recent research evaluating the amount of drinking water used by beef cattle remains limited (Araujo *et al.* 2010; Brew *et al.* 2011). Furthermore, several factors that affect water intake (WI) are known to exist in beef cattle, such as climatic variables, type of diet, breed, body weight, and physiological status. To our knowledge, the most recent report about WI by beef cattle is by Sexson et al. (2012), who studied this in temperate environmental conditions. Brazil has the largest commercial cattle herd (FAS/USDA, 2017), and the environmental conditions and beef cattle breeds are distinct from those described in the literature. Moreover, there is no equation to predict the water demand of beef cattle for Brazilian breeds. Based on constant changes in animal body, dietary type and environmental characteristics in beef cattle production systems, being able to accurately predict WI under different conditions is greatly important. It could help to sustainable usage of water in beef cattle production systems.

Thus, the aims of this study were:

- to evaluate the extent of mineral release from different feeds under ruminal and simulated abomasal conditions.
- to measure the effects of mineral supplementation on nutrient intake and digestibility, performance, and mineral status in Nellore beef cattle fed with and without calcium, phosphorus and microminerals supplemental sources during the growing and finishing phases of production.
- to quantify mineral balance in Nellore cattle fed with and without calcium, phosphorus and microminerals supplemental inorganic sources and to estimate net and dietary mineral requirements.

- to evaluate the accuracy of six current water intake equations for beef cattle, and suggest new equations if the current do not predict accurately water intake.

The chapters 1 and 3 presented in this thesis were accepted for publication and published as follow, respectively:

Zanetti, D., Menezes, A.C.B., Silva, F.A.S, Costa e Silva, L.F., Rotta, P.P., Detmann, E., Engle, T.E., Valadares Filho, S.C. 2017. In situ and in vitro estimation of mineral release from common feedstuffs fed to cattle. J. Agri. Sci. Doi: 10.1017/S002185961700034X

Zanetti, D., Godoi, L.A., Estrada, M.M., Engle, T.E., Silva, B.C., Alhadas, H.M., Chizzotti, M.L., Prados, L.F., Renno, L.N., Valadares Filho, S.C. 2017. Estimating mineral requirements of Nellore beef bulls fed with or without inorganic mineral supplementation and the influence on mineral balance. J. Anim. Sci. 95, 1696-1706. Doi: 10.2527/jas2016.1190.

References

AFRC. 1993. Energy and Protein Requirements of Ruminants. Wallingford, UK: CAB International.

ARC. 1980. The Nutrient Requirements of Ruminant Livestock. London, UK: CABI.

Beckett, J. L., Oltjen, J. W. 1993. Estimation of the water requirement for beef production in the United States. J. Anim. Sci. 71, 818-826.

Berrett, C.J., Wagner, J.J., Neuhold, K.L., Caldera, E., Sellins, K.S., Engle, T.E. 2015. Comparison of National Research Council standards and industry dietary trace mineral supplementation strategies for yearling feedlot steers. Profess. Anim. Sci. 31:237-247.

Brew, M. N., Myer, R. O., Hersom, M. J., Carter, J. N., Elzo, M. A., Hansen, G. R., Riley, D. G. 2011. Water intake and factors affecting water intake of growing beef cattle. Livestock Science 140, 297-300.

Čerešňáková, Z., Fľak, P., Poláčiková, M., Chrenková, M. 2005. In sacco NDF degradability and mineral release from selected forages in the rumen. Czech J. Anim. Sci. 50, 320-328.

Costa e Silva, L. F., Engle, T. E., Valadares Filho, S. C., Rotta, P. P., Valadares, R. F. D., Silva, B. C., Pacheco, M. V. C. 2015a. Intake, apparent digestibility, and nutrient requirements for growing Nellore heifers and steers fed two levels of calcium and phosphorus. Livest. Sci. 181, 17-24.

Costa e Silva, L. F., S. C.Valadares Filho, T. E. Engle, P. P. Rotta, M. I. Marcondes, F. A. S. Silva, E. C. Martins, and A. T. Tokunaga. 2015b. Macro minerals and trace element requirements for beef cattle. PloS ONE. 10:1-20.

Engle TE, Spears JW. 2000. Effects of dietary copper concentration and source on performance and copper status of growing and finishing steers. J. Anim. Sci. 78:2446-2451.

Esser, N. M., Hoffman, P. C., Coblentz, W. K., Orth, M. W., Weigel, K. A. 2009. The effect of dietary phosphorus on bone development in dairy heifers. J. Dairy Sci. 92, 1741-1749.

FAS/USDA – Livestock and Poultry: World Markets and Trade. 2017. Washington, USA: Foreign Agricultural Service/ United States Department of Agriculture.

Flachowsky, G., Grün, M. 1992. Influence of type of diet and incubation time on major elements release in sacco from Italian ryegrass, untreated and ammonia-treated wheat straw. Anim. Feed Sci. Tech. 36, 239-254.

Flachowsky, G., Grün, M., Polzin, S., Kronemann, H. 1994. In sacco dry matter degradability and Ca, Mg and P disappearance from Italian ryegrass, alfalfa hay and wheat straw in sheep and goats. J. Anim. Physiol. Anim. Nutr. 71, 57-64.

Humer, E., Q., Zebeli. 2015. Phytate in feed ingredients and potentials for improving the utilization of phosphorus in ruminant nutrition. Anim. Feed Sci. Tech. 209:1-15.

Ibrahim, M. N. M., Zemmelink, G., Tamminga, S. 1998. Release of mineral elements from tropical feeds during degradation in the rumen. Asian-Australasian J. Anim. Sci. 11, 530-537.

Jongbloed AW, Lenis NP. 1998. Environmental concerns about animal manure. J Anim Sci. 76:2641-2648.

Khorasani, G. R., Janzen, R. A., McGill, W. B., Kennelly, J. J. 1997. Site and extent of mineral absorption in lactating cows fed whole-crop cereal grain silage of alfalfa silage. J. Anim. Sci. 75, 239-248.

Mekonnen, M. M., Hoekstra, A. Y. 2012. A global assessment of the water footprint of farm animal products. Ecosystems 15, 401-415.

Millen, D.D., Pacheco, R.D.L., Arrigoni, M.D.B., Galyean, M.L., Vasconcelos, J.T. 2009. A snapshot of management practices and nutritional recommendations used by feedlot nutritionists in Brazil. J. Anim. Sci. 87:3427-3439.

Mullis, L.A., Spears, J.W., McCraw, R.L. 2003. Estimated copper requirements of Angus and Simmental heifers. J. Anim. Sci. 81:865-873.

NRC. 1996. Nutrient requirements of beef cattle. 7th rev. ed. Natl. Acad. Press, Washington, D.C.

NRC. 2001. Nutrient requirements of dairy cattle. 7th rev. ed. Natl. Acad. Press, Washington, D.C.

NRC. 2016. Nutrient requirements of beef cattle. 8th rev. ed. Natl. Acad. Press, Washington, D.C.

Olubobokun, J. A., Craig, W. M., Pond, K. R. 1990. Effects of mastication and microbial contamination on ruminal in situ forage disappearance. J. Anim. Sci. 68, 3371-3381.

Perdomo, J. T., Shirley, R. L., Chicco, C. F. 1977. Availability of nutrient minerals in four tropical forages fed freshly chopped to sheep. J. Anim. Sci. 45, 1114-1119.

Playne, M. J., Echevarría, M. G., Megarrity, R. G. 1978. Release of nitrogen, sulphur, phosphorus, calcium, magnesium, potassium and sodium from four tropical hays during their digestion in nylon bags in the rumen. J. Sci. Food Agric. 29, 520-526.

Pluske, J. M., Schlink, A. C. 2007. Managing water as a scarce resource in beef feedlots. Australasian Agribusiness Review 15, 1-13.

Pogge, D. J., Drewnoski, M. E., Hansen, S. L. 2014. High dietary sulfur decreases the retention of copper, manganese, and zinc in steers. J. Anim. Sci. 92, 2182-2191.

Prados L.F., Sathler, D.F.T., Silva, B.C., Zanetti, D., Valadares Filho, S.C., Alhadas, H.M., Detmann, E., Santos, S.A., Mariz, L.D.S., Chizzotti, M.L. 2017. Reducing mineral usage in feedlot diets for Nellore cattle: II. Impacts of calcium, phosphorus, copper, manganese, and zinc contents on intake, performance, and liver and bone status. J. Anim. Sci. 95:1-11.

Prados, L. F., S. C.Valadares Filho, S. A. Santos, D. Zanetti, A. N. Nunes, D. R. Costa, L. D. S. Mariz, E. Detmann, P. M. Amaral, F. C. Rodrigues, R. F. D. Valadares. 2015. Reducing calcium and phosphorus in crossbred beef cattle diets: impacts on productive performance during the growing and finishing phase. Anim. Prod. Sci. 55:1369-1374.

Rijsberman, F. R. 2006. Water scarcity: fact or fiction? Agricultural Water Management 80, 5-22.

Sehested J. 2004. Feeding phosphorus (P) to cattle: A new basis for Nordic recommendations. Acta Agriculturae Scandinavica, Section A-Animal Science, 54:169-180.

Sexson, J. L., Wagner, J. J., Engle, T. E., Eickhoff, J. 2012. Predicting water intake by yearling feedlot steers. Journal of Animal Science 90, 1920-1928.

Spears JW, Weiss WP. 2014. Invited Review: Mineral and vitamin nutrition in ruminants. Profess Anim Sci. 30:180-191.

Spears, J. W. 1994. Minerals in forages. In Forage Quality, Evaluation, and Utilization (Ed. G. C. Fahey Jr.), 281–317. Madison, WI, USA.

Spears, J. W. 1996. Organic trace minerals in ruminant nutrition. Anim. Feed Sci. Tech. 58, 151-163.

Suttle NF. 2010. Mineral nutrition of livestock. Cabi. 4th ed. London, UK.

Tilman, D., Cassman, K. G., Matson, P. A., Naylor, R., Polasky, S. 2002. Agricultural sustainability and intensive production practices. Nature 418, 671-677.

Valadares Filho S. C., M. I. Marcondes, M. L. Chizzotti, and P. V. R. Paulino. 2010. Nutrient requirements of Zebu beef cattle – BR-Corte. Suprema Gráfica e Editora, Visconde do Rio Branco, Brazil.

Vasconcelos JT, Galyean ML. 2007. Nutritional recommendations of feedlot consulting nutritionists: The 2007 Texas Tech University survey. J. Anim. Sci. 85:2772-2781.

Ward, F. A., Michelsen, A. 2002. The economic value of water in agriculture: concepts and policy applications. Water Policy 4, 423-446.

Chapter 1

In situ and *in vitro* estimation of mineral release from common feedstuffs fed to cattle

Short title: Mineral release from tropical feedstuffs

D. ZANETTI[1]*, A. C. B. MENEZES[1], F. A. S. SILVA[1], L. F. COSTA E SILVA[1], P. P. ROTTA[1], E. DETMANN[1], T. E. ENGLE[2], AND S. C. VALADARES FILHO[1]

[1] *Department of Animal Science, Universidade Federal de Viçosa, PH Rolfs St, 36570-900, Minas Gerais, Brazil*

[2] *Department of Animal Science, Colorado State University, 350 W Pitkin St, 80523, Colorado, United States*

(MS received 23 March 2017, revised 3 May 2017, accepted TBC May 2017)

Journal of Agricultural Science. DOI: https://doi.org/10.1017/S002185961700034X:

* To whom all correspondence should be addressed. Email: diego.zanetti@ufv.br ; diegozanetti_@hotmail.com

SUMMARY

The objective of the current study was to quantify the dry matter (DM) digestibility, and total ash (TA) and mineral release from 12 concentrate and 12 forage feedstuffs commonly fed to cattle using *in situ* and *in vitro* methods. Concentrate and forage feedstuffs were incubated in the rumen of ruminally cannulated beef bulls at eight different time points. Two different trials were conducted for concentrates and forages, with maximum incubation time of 72 and 120 h,

respectively. The residue from samples incubated for 24 h were treated with pepsin and hydrochloric acid (HCl) to simulate abomasum digestion *in vitro*. The initial and residual samples after *in situ* and *in vitro* incubations were measured. An asymptotic model was adopted for estimating solubility of minerals, disappearance rate of DM, and TA. Correlations between feedstuff contents and mineral release were evaluated. Residual samples from rumen fermentation after 24 h were incubated in simulated abomasal conditions and mineral release was measured. Cluster analysis was performed to group feedstuffs in relation to TA release. Large variability was observed between concentrate and forage feedstuffs for all constituents analysed. Large variability was observed for the effective ruminal degradation of TA and individual mineral release. When feedstuffs were clustered according to the immediately soluble fraction ('a'), the insoluble by potentially releasable fraction ('b') and the release rate of 'b' ('kd', /h) estimates of TA ruminal release, four groups were identified. From group '1' to group '4', an increase in the soluble fraction and a reduction in both moderate releasable fraction and release rate was observed. Neutral detergent fibre (NDF) content had a negative correlation with mineral release in the rumen, while mineral content had a positive correlation. These results demonstrate that mineral solubilization in the digestive tract is not the limiting factor for mineral absorption from the feedstuffs tested.

INTRODUCTION

The majority of nutrient recommendation publications and ration balancing programmes for cattle (USA: dairy cattle, NRC 2001; beef cattle, NRC 2016; United Kingdom: ARC 1980; AFRC 1993; and Brazil: BR-Corte, Valadares Filho *et al.* 2010) do not consider the inherent mineral contents from feedstuffs, or do not give a recommendation of the absorption coefficient for trace minerals within feedstuffs. Therefore, minerals are typically supplemented to beef cattle diets without taking into consideration the inherent minerals within the feedstuffs (Spears 1996).

Despite this, two points may be considered in this context for reducing mineral input for cattle. First, in most tropical areas, cattle depend exclusively on forages to meet their mineral requirements (Perdomo *et al.* 1977; Ibrahim *et al.* 1998; Costa e Silva *et al.* 2015; Costa *et al.* 2016). Second, when mineral supply in a production system is reduced, environmental excretion and costs are reduced (Khorasani *et al.* 1997; Humer & Zebeli 2015).

Some researchers (Esser *et al.* 2009; Prados *et al.* 2016; Zanetti *et al.* 2017) have demonstrated that animals fed typical feedlot diets can meet their mineral requirements exclusively from basal feed ingredients. This is made possible because the mineral content in feedstuffs is, in general, high (Playne *et al.* 1978). Mineral bioavailability in feedstuffs is affected by the mineral distribution in plant cells, the form of the mineral, association with cell components, and interaction with other minerals (Čerešňáková *et al.* 2005; Pogge *et al.* 2014; Berrett *et al.* 2015). Mineral availability in the rumen depends on mineral content, fibre content and passage rate (Flachowsky & Grün 1992; Berrett *et al.* 2015). Furthermore, the solubility of minerals in general is increased under low pH conditions. Therefore, mineral content in feedstuffs can be divided into three fractions: very soluble, moderately soluble (minerals that are predominately associated with the fibre and/or protein fractions of the feedstuff), and un-releasable or insoluble minerals (Spears 1994).

Minerals that are not released from feedstuffs are unavailable for absorption. Minerals released from feedstuffs incubated in rumen have been reported to be highly correlated with mineral availability to the animal (Flachowsky *et al.* 1994). Therefore, this technique is an inexpensive, simple, rapid and reproducible technique that may allow absorption coefficients for minerals contained in feedstuffs to be estimated (Olubobokun *et al.* 1990). Therefore, the current study hypothesized that a proportion of mineral content within feedstuffs is released under ruminal

and abomasal digestive conditions and these values should be considered in formulating diets for ruminants. The objectives were to evaluate the extent of mineral release under ruminal and simulated abomasal conditions.

MATERIAL AND METHODS

Two experiments were conducted in the Experimental Feedlot of the Animal Science Department of the Universidade Federal de Viçosa, Viçosa, MG, Brazil, following the recommendations of the Ethics Committee for Animal Use and Care. Twenty-four samples of feedstuffs were evaluated (12 concentrate and 12 forage typically fed to beef cattle) to determine mineral release.

Concentrate trial

Concentrate feedstuffs were divided into two groups where six ingredients were considered as energy concentrates (i.e. low protein content): ground maize (*Zea mays L.*), ground ear maize (*Zea mays L.*), ground sorghum (*Sorghum vulgare*), rice bran (*Oryza sativa*), soybean hulls (*Glycine max (L.) Merr*) and wheat bran (*Triticum aestivum*) while the other six ingredients were considered as protein concentrates (i.e. high protein content): maize gluten (*Zea mays L.*), cottonseed meal (*Gossypium hirsutum*), ground bean (*Phaseolus vulgaris L.*), peanut meal (*Arachis hypogaea L.*), soybean meal (*Glycine max (L.) Merr*) and sunflower meal (*Helianthus annuss*).

Concentrates were randomly divided into four groups and feedstuffs were incubated in the rumen of four cannulated crossbred bulls. The procedure of incubation was conducted four times, and within each of these periods, each concentrate group was incubated in the rumen of a different bull as part of a Latin square design. The bulls received *ad libitum* access to a diet composed of: 500 g maize silage/kg DM and a concentrate supplement composed of 800 g ground maize/kg, 86

g soybean meal/kg, 60 g wheat bran/kg, 14 g urea/kg, 1.0 g ammonium sulphate/kg, 9.0 g salt/kg, 9.0 g mineral premix/kg, 15 g sodium bicarbonate/kg and 5 g magnesium oxide/kg. The diet contained 120 g crude protein (CP)/kg DM.

Forage trial

The forages examined in the current experiment were maize silage (*Zea mays L*), brachiaria decumbens grass (*Urochloa decumbens*), elephant grass (*Pennisetum purpureum*), brachiaria humidicola grass (*Urochloa humidicola*), Marandu grass (*Urochloa brizantha* cv Marandu), brachiaria MG-4 grass (*Urochloa brizantha* cv MG-4), Mombaça grass (*Panicum maximum* cv Mombaça), mulato grass (hybrid *Urochloa decumbens* × *Urochloa ruziziensis*), peanut forage (*Arachis pintoi*), Tanzania grass *(Panicum maximum* cv Tanzânia), tifton 85 hay (*Cynodon* spp.) and brachiaria Xaraes grass (*Urochloa brizantha* cv Xaraés).

Samples of feedstuffs, except maize silage, were collected 45 d after regrowth, at the Agrostology Sector from Animal Sciences Department at Universidade Federal de Viçosa, Viçosa, MG, Brazil. The 45 d of regrowth, during the transition between rainy and dry seasons, was not irrigated, as usual for Brazilian pastures. The samples (leaves and stems) were harvested 10 cm above the soil. All samples were oven-dried at 55ºC for 72 h. Forages were randomly divided into three groups and incubated in the rumen of three cannulated crossbred bulls. The procedure of incubation was replicated three times and within each of these periods, each forage type was incubated in a different bull as part of a Latin square design. The bulls had *ad libitum* access to a diet that contained 1000 g/kg elephant grass containing 161 g CP/kg DM.

Ruminal incubation

All samples were ground through a 2-mm sieve using a knife mill (TECNAL, Piracicaba, São Paulo, Brazil) for *in situ* ruminal incubation. Nylon bags (Sefar Nitex, Switzerland; 50-µm porosity, 400 cm^2 surface area) were individually identified. Six g of each feedstuff were weighed into each bag and incubated in each animal. The bag surface area to mass ratio was 15 mg/cm^2. The number of bags varied as a function of incubation time to guarantee enough residual samples after incubation (i.e. more bags per sample were incubated for the longer incubation times relative to shorter incubations times). The incubation times were 0, 2, 4, 8, 16, 24, 48 and 72 h for concentrates and 0, 3, 6, 12, 24, 36, 48, 72 and 120 h for forages. The times were different for concentrate and forage feedstuffs due different degradation rates. Samples were incubated in the rumen by attaching the bags to a steel chain with a weight at the end to allow for continual immersion within the ruminal contents.

Bags were placed into the rumen in reverse order of incubation hours so that all bags were removed at the same time for washing. After the incubation period, bags were washed by hand with running cold tap water and the end-point for washing was when the rinsing water had a high clarity. The bags for time 0 were not incubated in the rumen, but as with incubated bags they were rinsed in running water. Samples were oven-dried at 55°C for 72 h. After drying, bags were placed in an oven at 105°C for 2 h and weighed. Residual samples in bags were used to estimate the parameters of ruminal DM degradation, and total ash (TA) and mineral release. Residues of each feedstuff were removed from nylon bags and placed in a labelled plastic bag, to obtain a sample of each feedstuff per animal/incubation time.

Abomasal digestion simulation

Residual samples of ruminal incubation at 24 h were submitted to a simulation of abomasal digestion as described by Berrett *et al.* (2015). Four hundred mg of sample was weighed into plastic tubes and 15 ml of a solution of 50 g/l pepsin and 1.2 N hydrochloric acid (HCl) was added to each tube to simulate abomasal digestion conditions. Tubes were then allowed to incubate in a water bath at 39°C with gentle swirling every 15 min. After 1 h, residual content was filtered through ashless filter paper (CAT nº 1541-090, Whatman, General Electric). The residual content was oven-dried at 105ºC for 12 h, and analysed for ash and mineral content.

Analytical procedures

All feedstuffs were analysed for dry matter (DM; method 934.01), TA (method 930.05), and total nitrogen (N, method 981.10), according to the AOAC protocols (AOAC 2012). Crude protein (CP) content was obtained by multiplying total nitrogen content by 6.25. Neutral detergent fibre (NDF) was obtained according to Mertens (2002) without addition of sodium sulphite and with addition of a thermostable alfa-amylase. Neutral detergent fibre was expressed in ash- and protein-free basis.

Mineral analysis was performed on all samples (initial and residual) by weighing the samples into pre-weighed acid-washed crucibles then drying the samples in a forced-air oven at 105°C for 12 h. After the drying period, samples were reweighed to measure DM content. Samples were then placed in muffle furnace and ashed at 600°C for 12 h. Samples were removed and placed in a desiccator for 30 min to cool. Samples were then weighed and re-suspended with 5 ml of HCl (1.2 N HCl). During re-suspension the crucibles were heated using a heat plate. Mineral concentrations (calcium [Ca], phosphorus [P,] sodium [Na], potassium [K], magnesium [Mg],

sulphur [S], copper [Cu], zinc [Zn], manganese [Mn], cobalt [Co] and iron [Fe]) of the samples were analysed through inductively coupled plasma optical emission spectroscopy methods (Optima 7300 DV, Perkin Elmer; Braselton *et al.* 1997).

Statistical analysis

A Latin square experimental design was used to assist and organize the information collected in the field, allowing for measurement of degradation of different feedstuffs without the confounding effect of animal. The objective was to control sources of variation and to avoid bias without estimating variability. The Latin square design, in this case, was not used as a way to analyse data for comparative differences among feedstuffs, animals, or collection periods, but to collect source data to generate the following equations.

The profiles for DM degradation, TA and mineral release in the rumen were estimated using the asymptotic model of Ørskov & McDonald (1979). The following model was used to estimate the parameters of degradability:

$$Y = a + b \times (1 - e^{(-kd \times t)}) \quad (1)$$

where Y is fraction degraded or released in the time t (g/100g), of DM, TA or individual mineral; 'a' is the immediately soluble fraction (g/100g); 'b' is the insoluble but potentially degradable/releasable fraction (g/100g); 'e' is the Euler's number (e = 2.71828183…) and *kd* is degradation/releasing rate of 'b' (/h). The effective degradation/release was estimated based on the model of Ørskov & McDonald (1979):

$$E = a + \frac{b \times kd}{kd + kp} \quad (2)$$

where E is effective degradation (or released) fraction, of DM, TA or each mineral; ‘a’, ‘b’ and *kd* are the parameters estimated from Eqn 1; and *kp* is passage rate, considered in the current study as 0.05.

The NLIN procedure of SAS (SAS Institute Inc., Cary, NC, USA) was used to estimate the parameters from the models. Pearson correlations between analysed contents in feedstuffs and ruminal TA and effective mineral release were performed (CORR procedure; SAS Institute Inc., Cary, NC, USA). All statistical procedures were conducted using 0.05 as the critical level for the probability for the type I error.

Additionally, to identify concentrate and forage sub-groups of mineral release, a cluster analysis was performed using ‘a’, ‘b’ and kd estimates for TA as response variable. Non-hierarchical k-means clustering method was performed (Johnson & Wichern 1998; Khattree & Naik 2000) by using the FASTCLUS procedure of SAS (SAS Institute Inc., Cary, NC, USA). Initially, a maximum of five clusters was established for each clustering procedure. If this procedure created at least one cluster with only one observation (feed), a new clustering was made with a maximum of four clusters. This procedure was repeated until each and every cluster had at least two observations (feeds). The efficiency of clustering was also evaluated through the over-all R^2 and the cubic clustering criterion (CCC).

RESULTS

Substantial variability was observed between concentrate and forage feedstuffs for all analysed constituents (Table 1). For certain feeds in the current study, the model proposed by Ørskov & McDonald (1979) did not converge due to different degradation responses. In this case, the model was not adopted because a negative slope was observed; indicative of a greater mineral amount in the residual than in the incubated sample. Calculated parameters of the model, ‘a’, ‘b’ and kd, for

concentrate and forage feedstuffs are presented in Tables 2 and 3, respectively. Substantial variability was observed for the effective ruminal degradation and total and individual mineral release (Table 4). When feedstuffs were clustered according with the 'a', 'b' and kd estimates of TA ruminal release, four groups were identified (Table 5). From group '1' to group '4', an increase in the fraction 'a', and a reduction in both fraction 'b' and release rate was observed. A negative correlation was observed between NDF content and mineral release (Table 6). In general, the solubility values for DM, TA and each mineral for majority of feedstuffs ranged between 0.6–0.8, 0.7–0.9 and 0.9–1.0, respectively except for Cu, Co and Fe (Table 7).

DISCUSSION

Mineral contents

Reports regarding the availability of minerals from feedstuffs are important to improve the precision of diet formulation and mineral supplementation for cattle. Moreover, mineral content of feedstuffs is an important factor for maintaining buffering capacity in ruminants (Jasaitis *et al.* 1987). Research in tropical countries has focused primarily on investigating the impact of protein and energy supplementation on cattle performance (Corah 1996; Sath *et al.* 2012). Furthermore, trace element content is extremely variable in feedstuffs when compared with CP or NDF content (Adams 1975; Playne *et al.* 1978; Genther & Hansen 2014). In the current study, while the coefficients of variation (CV) for CP and NDF in forage group were 35.4 and 15.2%, respectively, differences between mineral contents were even greater. The average coefficient of variation for minerals in forages was 82.6% and 99.7% when all groups were considered. The smallest CV between minerals was 54.1% for K and the greatest CV was 133.3% for Fe.

Feedstuff Ca, Mg, K, Cl, and Fe contents are in appropriate concentrations to meet the mineral requirements of beef cattle (Minson 1990; Chládek & Zapletal 2007). Smart *et al.* (1981), in a review about the interactions across Cu, Zn and Se in soil, plants and cattle, reported that basal feedstuffs can provide trace minerals to the animal, however, concentrations can be inadequate. Thus, Yoshihara *et al.* (2013) estimated mineral intake and nutritional quality for grazed cattle varying numbers and combinations of 17 temperate pasture species; they suggested that by improving the number of plant species or products in diet formulation or in grazed pastures, improvement in mineral balance can be obtained in cattle.

Rumen mineral release

Mineral requirements from ruminal fauna are not negligible (Bravo *et al.* 2000), consequently mineral release from ingested feeds is important. Although mineral solubilization does not guarantee mineral utilization, absorption, or tissue deposition, absorption is largely affected by mineral intake and mineral solubilization in the gastrointestinal tract prior to absorption (Field 1981; Flachowsky *et al.* 1994). Like this, Van Eys & Reid (1987) have reported that reduced mineral release from forages decreases performance in grazing cattle.

High variability in mineral content between feedstuffs, and different solubilities between minerals have been described (Čerešňáková *et al.* 2007). In the current study, high variability was observed for effective release among minerals and effective release in each mineral between feeds. Feeds with low DM degradation did not necessarily have low mineral release coefficients (Emanuele *et al.* 1991). Mineral release profiles in relationship to DM digestibility can be associated with concentration of minerals in the digestible part of the plant. Most minerals are associated with the organic constituents within feeds (Broadley *et al.* 2007; Maathuis & Diatloff

2013; He *et al.* 2014). This is most likely the reason for the lack of a relationship between DM digestibility and mineral solubility.

The average effective release of K was 0.98 for concentrates and forages, being the lowest variation measured among minerals. Potassium in the plant cell is not associated with organic matter and persists as the K^+ ion (Amtmann & Rubio 2012) and this helps to explain the high release of K. Phosphorus release was assumed to be similar to that of K. However, P within the concentrate feeds is primarily associated with phytate. Rumen microorganisms synthesize a phytase enzyme in significant amounts (Ray *et al.* 2013) that help to liberate P from the plant cell. Feeds with low P content were contaminated with P from rumen microorganisms and consequently P concentrations in the residual DM after rumen incubation were greater than the P content in the intact feeds, which caused P release to have negative values. This behaviour for P has been related to feeds with low P content and high NDF content (Bonhomme 1990; Bravo *et al.* 2000). However, P contamination was observed at times > 8 h, probably due to the lag time for bacterial growth and adherence to fibre. Phosphorus content in the residual DM of forages submitted to ruminal incubation (t = 8 h) indicates that 0.52 (maximum: maize silage, 0.77; minimum: Tanzania, 0.37) of initial P was released, and after this time, the P content of residue increased.

Group '1' was characterized by reduced fraction 'a' (3±4.9), but greater fraction 'b' and release rate (91±6.7 and 0.5±0.31, respectively). In contrast, group '4' was characterized by greater fraction 'a' (63±5.7), but reduced fraction 'b' and release rate (14±4.1 and 0.1±0.08, respectively). Only concentrates assigned to group '1' were characterized by 'highest release rate': wheat bran, maize gluten, ground bean, peanut meal and soybean meal. This group was composed only of concentrate feedstuffs considered as protein feeds, reaffirming a correlation of 0.41 between CP content and TA release. However, across minerals, a positive and significant relationship between

CP content and mineral release was observed only for Mg, Cu and Fe. Furthermore, from these five feeds, four are by-product feeds that were processed during manufacturing (i.e. heat, pressing, substances added). This may be the reason for a high mineral release rate from these feedstuffs. A second group (group '2') was similarly composed exclusively of concentrate feeds (ground sorghum, rice bran, soybean hulls, cottonseed meal, sunflower meal). These feeds presented a high release rate (0.5±0.29), like group '1', but greater fraction 'a' (22±5.2) and lower fraction 'b' (63±10.1). In contrast, group '4' was composed exclusively of forages (maize silage, Decumbens, Humidicola, Marandu, MG-4, Mulato, Tifton-85 hay and Xaraes). This could be related to high K content and solubility in these feeds. Group '3' was composed of ground ear maize, ground maize, elephant grass, Mombaça, peanut forage and Tanzania, with 'a' = 45±4.5, 'b' = 34±8.9, and kd = 0.09±0.032.

Relations between feed content and ruminal mineral release

Associations between nutrient contents and mineral release within feedstuffs do exist (Emanuele & Staples 1990). The correlation between NDF content and ruminal TA release was negative (r = –0.39), which may be related to the high capacity of NDF to exchange cations (McBurney *et al.* 1986; Jasaitis *et al.* 1987). Moreover, minerals associated with the plant cell wall have a lower bioavailability or require a longer fermentation time for maximal release (Emanuele & Staples 1990; Flachowsky & Grün 1992). A negative correlation between Ca release and NDF content, for example, can be explained by the function of Ca in the cell in that it helps to regulate the control of cell wall enzymes, assists with cell wall stabilization and binds with pectin in the cell wall to help support the plant (Demarty *et al.* 1984; Spears 1996). Furthermore, with the advancement of plant maturity, NDF and silica concentrations are increased (Smith *et al.* 1971; Jung & Allen 1995;

Spanghero *et al.* 2015). Silica content was not measured in the current study; however, this relationship may be confirmed when a correlation of 0.57 was observed between NDF and ruminal TA content, and the average correlation between each mineral and NDF contents was r = –0.31. Among minerals, only K and Mn were positively correlated with NDF content (0.56 and 0.34, respectively).

The correlation between TA content and ruminal TA release was negative (r = –0.51). Ma & Yamaji (2006) reported that insoluble silica is incorporated into the plant cell wall to improve cellular protection. However, ruminal release of each mineral was increased by the contents of other nutrients (average of r = 0.35). As minerals occur in feeds in soluble and insoluble forms, a greater concentration of each mineral is associated with the soluble fraction.

Abomasal mineral release

In general, minerals with nutritional importance have greater solubility as pH decreases. At a high pH, minerals typically become insoluble, thus decreasing the chances of absorption. Twenty-four h ruminal residues had negative mineral release values in some cases. This could be related to contamination from minerals of ruminal fluid origin (Moreira *et al.* 2013). In comparison with residual samples incubated in the rumen for 24 h, solubility of minerals with nutritional interest was numerically greater than DM and TA. Few reports have been published investigating mineral solubility of different feedstuffs in ruminant animals. It is difficult to explain the reason for the reduced availability of these minerals and the greater solubility of other minerals. In cattle, Berrett *et al.* (2015) reported solubilities ranging between 0.52 and 0.68 for Cu and 0.75 and 0.87 for Zn. These values are similar to those reported in the current experiment. Regarding Fe solubility,

Lestienne *et al.* (2005) reported that this reduced value could be related to interactions with phenolic compounds in the feed matrix.

In general, high solubility of minerals was observed after simulated abomasal digestion. As already mentioned, solubility is an essential step to mineral absorption. The absorption coefficients recommended by nutritionists are lower than solubility values reported in the current research, except for Na and K, which are considered highly soluble. A portion of absorption coefficients described in the literature is related to apparent absorption, which includes the endogenous fraction in the reported values. Costa e Silva *et al.* (2015) determined the true absorption coefficients for minerals. Among the minerals evaluated, only the absorption coefficient of Co was close to the average Co release under simulated abomasal conditions. The Mn and Fe absorption coefficients were close to the ruminal release of these minerals. Mineral homeostasis is controlled mainly by absorption mechanisms (Field 1981) and may help explain these differences. However, current understanding of the mechanisms for mineral absorption in beef cattle are limited (Spears 2003; Han *et al.* 2012).

In conclusion, mineral content and ruminal release are highly variable among plant species and between each mineral. In general, ruminal mineral release is high, with the greatest values for K, while Ca, S, Cu, Zn, Mn and Co have the lowest ruminal release, being proportional to the DM digested. Concentrated feeds have a greater mineral release rate than forages. Mineral release is affected by NDF content and mineral concentration. Under simulated abomasal conditions, most minerals are released, and consequently available for absorption. Based on this information, mineral content from basal feed ingredients should be considered as highly available and total mineral content may be used in diet formulation, thereby reducing the level of mineral supplement often routinely used and hence reducing costs and environmental excretion of minerals.

ACKNOWLEDGEMENT

The author gratefully acknowledges National Council of Scientific and Technological Development (CNPq), National Institute of Science and Technology in Animal Science (INCT – Ciência Animal), Coordination of Improvement of Personal Higher Education (CAPES), and Foundation for Research Support of the State of Minas Gerais (FAPEMIG) for providing a research grant for this research.

REFERENCES

ADAMS, R. S. (1975). Variability in mineral and trace element content of dairy cattle feeds. *Journal of Dairy Science* **58**, 1538-1548.

AFRC (1993). *Energy and Protein Requirements of Ruminants*. Wallingford, UK: CAB International.

AMTMANN, A. & RUBIO, F. (2012). Potassium in plants. In *eLS: Citeable Reviews in the Life Sciences* (Ed. A.M. Hetherington), doi: 10.1002/9780470015902.a0023737. Chichester, UK: John Wiley & Sons, Ltd. Available online from http://onlinelibrary.wiley.com/doi/10.1002/9780470015902.a0023737/abstract (accessed 8 May 2017).

AOAC (2012). *Official Methods of Analysis*. 19th edn. Arlington, VA, USA: Association of Official Analytical Chemists.

ARC (1980). *The Nutrient Requirements of Ruminant Livestock.* London, UK: CABI.

BERRETT, C. J., WAGNER, J. J., NEUHOLD, K. L., CALDERA, E., SELLINS, K. S. & ENGLE, T. E. (2015). Comparison of National Research Council standards and industry dietary trace mineral supplementation strategies for yearling feedlot steers. *The Professional Animal Scientist* **31**, 237-247.

BONHOMME, A. (1990). Rumen ciliates: their metabolism and relationships with bacteria and their hosts. *Animal Feed Science and Technology* **30**, 203-266.

BRASELTON, W. E., STUART, K. J., MULLANEY, T. P. & HERDT, T. H. (1997). Biopsy mineral analysis by inductively coupled plasma atomic emission spectroscopy with ultrasonic nebulization. *Journal of Veterinary Diagnostic Investigation* **9**, 395–400.

BRAVO, D., MESCHY, F., BOGAERT, C. & SAUVANT, D. (2000). Ruminal phosphorus availability from several feeds measured by the nylon bag technique. *Reproduction Nutrition Development* **40**, 149-162.

BROADLEY, M. R., WHITE, P. J., HAMMOND, J. P., ZELKO, I. & LUX, A. (2007). Zinc in plants. *New Phytologist* **173**, 677-702.

ČEREŠŇÁKOVÁ, Z., FĽAK, P., POLÁČIKOVÁ, M. & CHRENKOVÁ, M. (2005). *In sacco* NDF degradability and mineral release from selected forages in the rumen. *Czech Journal of Animal Science* **50**, 320-328.

ČEREŠŇÁKOVÁ, Z., FĽAK, P., POLÁČIKOVÁ, M. & CHRENKOVÁ, M. (2007). *In sacco* macromineral release from selected forages. *Czech Journal of Animal Science* **52**, 175-182.

CHLÁDEK, G. & ZAPLETAL, D. (2007). A free-choice intake of mineral blocks in beef cows during the grazing season and in winter. *Livestock Science* **106**, 41-46.

CORAH, L. (1996). Trace mineral requirements of grazing cattle. *Animal Feed Science and Technology* **59**, 61-70.

COSTA E SILVA, L. F., ENGLE, T. E., VALADARES FILHO, S. C., ROTTA, P. P., VALADARES, R. F. D., SILVA, B. C. & PACHECO, M. V. C. (2015). Intake, apparent digestibility, and nutrient requirements for growing Nellore heifers and steers fed two levels of calcium and phosphorus. *Livestock Science* **181**, 17-24.

COSTA, R. M., PONSANO, E. H. G., SOUZA, V. C. & MALAFAIA, P. (2016). Reduction of phosphorus concentration in mineral supplement on fertility rate, maternal ability and costs of beef cows reared in pastures of *Urochloa decumbens*. *Tropical Animal Health and Production* **48**, 417-422.

DEMARTY, M., MORVAN, C. & THELLIER, M. (1984). Calcium and the cell wall. *Plant, Cell and Environment* **7**, 441-448.

EMANUELE, S. M. & STAPLES, C. R. (1990). Ruminal release of minerals from six forage species. *Journal of Animal Science* **68**, 2052-2060.

EMANUELE, S. M., STAPLES, C. R. & WILCOX, C. J. (1991). Extent and site of mineral release from six forage species incubated in mobile Dacron bags. *Journal of Animal Science* **69**, 801-810.

ESSER, N. M., HOFFMAN, P. C., COBLENTZ, W. K., ORTH, M. W. & WEIGEL, K. A. (2009). The effect of dietary phosphorus on bone development in dairy heifers. *Journal of Dairy Science* **92**, 1741-1749.

FIELD, A. C. (1981). Some thoughts on dietary requirements of macro-elements for ruminants. *Proceedings of the Nutrition Society* **40**, 267-272.

FLACHOWSKY, G. & GRÜN, M. (1992). Influence of type of diet and incubation time on major elements release in sacco from Italian ryegrass, untreated and ammonia-treated wheat straw. *Animal Feed Science and Technology* **36**, 239-254.

FLACHOWSKY, G., GRÜN, M., POLZIN, S. & KRONEMANN, H. (1994). *In sacco* dry matter degradability and Ca, Mg and P disappearance from Italian ryegrass, alfalfa hay and wheat straw in sheep and goats. *Journal of Animal Physiology and Animal Nutrition* **71**, 57-64.

GENTHER, O. N. & HANSEN, S. L. (2014). Effect of dietary trace mineral supplementation and a multi-element trace mineral injection on shipping response and growth performance of beef cattle. *Journal of Animal Science* **92**, 2522-2530.

HAN, H., SO, H., DOMBY, E. & ENGLE, T. E. (2012). The relationship of pulmonary artery copper concentrations and genes involved in copper homeostasis in cattle, swine, and goats. *Asian-Australasian Journal of Animal Sciences* **25**, 194-199.

HE, H., VENEKLAAS, E. J., KUO, J. & LAMBERS, H. (2014). Physiological and ecological significance of biomineralization in plants. *Trends in Plant Science* **19**, 166-174.

HUMER, E. & ZEBELI, Q. (2015). Phytate in feed ingredients and potentials for improving the utilization of phosphorus in ruminant nutrition. *Animal Feed Science and Technology* **209**, 1-15.

IBRAHIM, M. N. M., ZEMMELINK, G. & TAMMINGA, S. (1998). Release of mineral elements from tropical feeds during degradation in the rumen. *Asian-Australasian Journal of Animal Sciences* **11**, 530-537.

JASAITIS, D. K., WOHLT, J. E. & EVANS, J. L. (1987). Influence of feed ion content on buffering capacity of ruminant feeds *in vitro*. *Journal of Dairy Science* **70**, 1391-1403.

JOHNSON, R.A. & WICHERN, D.W. (1998). *Applied Multivariate Statistical Analysis.* New Jersey, NY, USA: Prentice-Hall International.

JUNG, H. G. & ALLEN, M. S. (1995). Characteristics of plant cell walls affecting intake and digestibility of forages by ruminants. *Journal of Animal Science* **73**, 2774-2790.

KHATTREE, R. & NAIK, D.N. (2000). *Multivariate Data Reduction and Discrimination*. Cary, NC, USA: SAS Institute Inc.

KHORASANI, G. R., JANZEN, R. A., MCGILL, W. B. & KENNELLY, J. J. (1997). Site and extent of mineral absorption in lactating cows fed whole-crop cereal grain silage of alfalfa silage. *Journal of Animal Science* **75**, 239-248.

LESTIENNE, I., BESANÇON, P. CAPORICCIO, B., LULLIEN-PÉLLERIN, V. & TRÉCHE, S. (2005). Iron and zinc in vitro availability in pearl millet flours (*Pennisetum glaucum*) with varying phytate, tannin, and fiber contents. *Journal of Agricultural and Food Chemistry* **53**, 3240-3247.

MA, J. F. & YAMAJI, N. (2006). Silicon uptake and accumulation in higher plants. *Trends in Plant Science* **11**, 392-397.

MAATHUIS, F. J. & DIATLOFF, E. (2012). Roles and functions of plant mineral nutrients. In *Plant Mineral Nutrients: Methods and Protocols* (Ed. F. J. M. Maathuis), pp. 1-21. Methods in Molecular Biology 953. Dordrecht, the Netherlands: Springer.

MCBURNEY, M. I., ALLEN, M. S. & VAN SOEST, P. J. (1986). Praseodymium and copper cation-exchange capacities of neutral-detergent fibres relative to composition and fermentation kinetics. *Journal of the Science of Food and Agriculture* **37**, 666-672.

MERTENS, D. R. (2002). Gravimetric determination of amylase-treated neutral detergent fibre in feeds with refluxing beakers or crucibles: collaborative study. *Journal of AOAC International* **85**, 1217-1240.

MINSON, D. J. (1990). *Forage in Ruminant Nutrition*. San Diego, CA, USA: Academic Press.

MOREIRA, L. M., LEONEL, F. P., VIEIRA, R. A. M. & PEREIRA, J. C. (2013). A new approach about the digestion of fibers by ruminants. *Revista Brasileira de Saúde e Produção Animal* **14**, 382-395.

NRC (2001). *Nutrient Requirements of Dairy Cattle*. 7th revised. edn. Washington, DC, USA: The National Academy Press.

NRC (2016). *Nutrient Requirements of Beef Cattle.* 8th edn. Washington, DC, USA: The National Academy Press.

OLUBOBOKUN, J. A., CRAIG, W. M. & POND, K. R. (1990). Effects of mastication and microbial contamination on ruminal *in situ* forage disappearance. *Journal of Animal Science* **68**, 3371-3381.

ØRSKOV, E. R., & MCDONALD, I. (1979). The estimation of protein degradability in the rumen from incubation measurements weighted according to rate of passage. *The Journal of Agricultural Science, Cambridge* **92**, 499–503.

PERDOMO, J. T., SHIRLEY, R. L. & CHICCO, C. F. (1977). Availability of nutrient minerals in four tropical forages fed freshly chopped to sheep. *Journal of Animal Science* **45**, 1114-1119.

PLAYNE, M. J., ECHEVARRÍA, M. G. & MEGARRITY, R. G. (1978). Release of nitrogen, sulphur, phosphorus, calcium, magnesium, potassium and sodium from four tropical hays during their digestion in nylon bags in the rumen. *Journal of the Science of Food and Agriculture* **29**, 520-526.

POGGE, D. J., DREWNOSKI, M. E. & HANSEN, S. L. (2014). High dietary sulfur decreases the retention of copper, manganese, and zinc in steers. *Journal of Animal Science* **92**, 2182-2191.

PRADOS, L. F., VALADARES FILHO, S. C., SANTOS, S. A., ZANETTI, D., NUNES, A. N., COSTA, D. R., MARIZ, L. D. S., DETMANN, E., AMARAL, P. M., RODRIGUES, F. C. & VALADARES, R. F. D. (2016). Reducing calcium and phosphorus in crossbred beef cattle diets: impacts on productive performance during the growing and finishing phase. *Animal Production Science* **56**, 1643-1649.

RAY, P. P., JARRETT, J. & KNOWLTON, K. F. (2013). Effect of dietary phytate on phosphorus digestibility in dairy cows. *Journal of Dairy Science* **96**, 1156-1163.

SATH, K., PAULY, T. & HOLTENIUS, K. (2012). Mineral balance of Cambodian cattle based on their faecal and urinary excretion. *Journal of Animal and Veterinary Advances* **11**, 4221-4225.

SMART, M.E., GUDMUNDSON, J. & CHRISTENSEN, D. A. (1981). Trace mineral deficiencies in cattle: a review. *Canadian Veterinary Journal* **22**, 372-376.

SMITH, G. S., NELSON, A. B. & BOGGINO, E. J. (1971). Digestibility of forages *in vitro* as affected by content of 'silica'. *Journal of Animal Science* **33**, 466-471.

SPANGHERO, M., ZANFI, C., SIGNOR, M., DAVANZO, D., VOLPE, V. & VENERUS, S. (2015). Effects of plant vegetative stage and field drying time on chemical composition and in vitro ruminal degradation of forage soybean silage. *Animal Feed Science and Technology* **200**, 102-106.

SPEARS, J. W. (1994). Minerals in forages. In *Forage Quality, Evaluation, and Utilization* (Ed. G. C. Fahey Jr.), 281–317. Madison, WI, USA: ASA, CSSA, SSSA.

SPEARS, J. W. (1996). Organic trace minerals in ruminant nutrition. *Animal Feed Science and Technology* **58**, 151-163.

SPEARS, J. W. (2003). Trace mineral bioavailability in ruminants. *The Journal of Nutrition* **133**, 1506S-1509S.

VALADARES FILHO, S. C., MARCONDES, M. I., CHIZZOTTI, M. L. & PAULINO, P. V. R. (2010). *Nutrient Requirements of Zebu Beef Cattle – BR-Corte.* 2nd edn. Visconde do Rio Branco, Brazil: Suprema Gráfica e Editora.

VAN EYS, J. E. & REID, R. L. (1987). Ruminal solubility of nitrogen and minerals from fescue and fescue-red clover herbage. *Journal of Animal Science* **65**, 1101-1112.

YOSHIHARA, Y., MIZUNO, H., OGURA, S., SASAKI, T. & SATO, S. (2013). Increasing the number of plant species in a pasture improves the mineral balance of grazing beef cattle. *Animal Feed Science and Technology* **179**, 138-143.

ZANETTI, D., GODOI, L. A., ESTRADA, M.M., ENGLE, T.E., SILVA, B.C., ALHADAS, H.M., CHIZZOTTI, M.L., PRADOS, L.F., RENNO, L.N. & VALADARES FILHO, S.C. (2017). Estimating mineral requirements of Nellore beef bulls fed with or without inorganic mineral

supplementation and the influence on mineral balance. *Journal of Animal Science* **95**, 1696-1706. doi: 10.2527/jas2016.1190.

Table 1. *Chemical composition of concentrate and forage feedstuffs*

Feeds	DM*	Ash†	CP‡	NDF§	Ca	P	Na	K	Mg	S	Cu	Zn	Mn	Co	Fe
	g/kg	---------- g/kg DM ----------									---------- mg/kg ----------				
Energy concentrates															
Ground ear maize	875	45	146	588	0.9	2.0	0.3	2.5	0.9	0.2	6.7	40	11.9	0.37	167
Ground maize	865	13	96	56	0.3	2.0	0.3	3.4	1.3	0.3	2.2	12	3.6	0.02	18
Ground sorghum	863	13	104	66	3.6	2.8	1.2	10	1.8	0.3	2.4	17	10	0.04	42
Rice bran	865	96	164	179	1.1	7.7	0.2	11	9.1	1.8	5.5	56	192	0.25	131
Soybean hulls	875	45	146	588	3.8	1.2	1.2	13	3.2	0.8	7.9	31	9.0	0.07	307
Wheat bran	869	49	198	303	1.7	8.0	1.1	12	3.8	1.0	15	97	153	0.08	125
Protein concentrates															
Maize gluten	918	44	656	165	0.5	3.1	0.3	2.0	1.0	4.0	23	50	25	0.23	153
Cottonseed meal	891	66	413	192	2.6	9.3	0.3	12	6.1	1.0	26	63	17	0.21	123
Ground bean	879	63	525	186	3.0	5.9	0.7	17	5.5	0.9	14	43	26	0.33	263
Peanut meal	882	38	521	103	1.9	6.2	0.2	9.9	4.1	0.4	22	60	55	0.51	691
Soybean meal	935	49	465	150	4.3	7.6	0.7	20	3.9	1.5	18	63	42	0.33	247
Sunflower meal	851	63	322	451	3.4	8.4	1.0	11	6.2	0.5	31	90	36	0.30	176
*Forages***															
Maize silage	221	71	67	520	2.5	1.6	1.1	9.1	1.6	0.8	4.6	30	27	0.49	1244
Decumbens	194	87	115	607	5.0	0.9	0.1	9.4	0.9	0.4	3.8	28	85	0.28	470
Elephant grass	237	127	205	528	7.3	2.1	0.2	21	1.8	0.9	7.2	20	81	0.17	118
Humidicola	179	90	105	714	6.0	0.9	0.2	19	1.3	0.4	3.7	19	71	0.09	151
Marandu	179	81	107	666	4.2	0.9	0.2	6.8	0.9	0.3	15	10	56	0.46	157
MG- 4	185	72	117	685	4.0	0.9	0.1	5.4	0.9	0.3	2.9	19	68	0.09	70
Mombaça	242	108	146	659	14	1.8	0.1	27	1.5	1.2	11	34	192	0.41	137
Mulato	170	97	88	622	5.1	0.9	0.1	8.1	0.9	0.2	2.1	15	38	0.07	88
Peanut forage	269	77	207	412	24	2.1	0.2	7.2	2.8	1.2	14	29	162	0.48	83
Tanzania	236	91	142	607	7.8	1.1	0.1	15	1.2	0.9	6.3	22	51	0.23	54

Tifton 85 hay	868	50	87	753	3.3	1.6	0.1	12	0.9	2.0	21	79	91	0.13	92
Xaraes	209	79	106	644	6.0	0.9	0.1	6.9	1.1	0.3	4.2	22.	63	0.13	72

* Dry matter.
† Total mineral content.
‡ Crude protein.
§ Neutral detergent fibre.
** The forages: Decumbens grass (*Urochloa decumbens*), Elephant grass (*Pennisetum purpureum*), Humidicola grass (*Urochloa humidicola*), Marandu grass (*Urochloa brizantha* cv Marandu), MG-4 grass (*Urochloa brizantha* cv MG-4), Mombaça grass (*Panicum maximum* cv Mombaça), Mulato grass (hybrid *Urochloa decumbens* × *Urochloa ruziziensis*), Peanut forage (*Arachis pintoi*), Tanzania grass (*Panicum maximum* cv Tanzânia), Tifton 85 hay (*Cynodon* spp.) and Xaraes grass (*Urochloa brizantha* cv Xaraés) were collected after 45d of regrowth.

Table 2. *Dry matter and total and individual mineral parameters of ruminal release of concentrate feeds*

Feeds	Parameter*	DM†	Ash‡	Ca	P	Na	K	Mg	S	Cu	Zn	Mn	Co	Fe
Ground ear maize	a	10	46	9.0	6.1	8.4	5.7	8.1	6.9	43	12	4.6	7.1	8.1
	b	71	41	46	83	67	92	69	49	51	62	56	80	69
	Kd	5.0	14	7.0	73	43	171	36	7.0	2.0	25	7.0	65.0	36
Ground maize	a	18	52	9.0	12	11	65	73	11	32	18	8.4	11	13
	b	80	43	95	76	83	34	22	77	70	79	98	88	71
	Kd	6.0	7.0	1.0	63	137	169	47	232	3.0	5.0	2.0	218	47
Ground sorghum	a	18	18	7.3	69	3.7	9.2	1.0	10	7.7	29	10	-§	9.3
	b	78	68	85	24	97	90	99	82	54	55	75	-	82
	Kd	5.0	16	20	152	17	147	17	22	51	5.0	60	-	91
Rice bran	a	11	17	67	8.0	8.8	48	83	8.9	8.9	12	10	8.9	9.1
	b	55	45	30	64	90	49	10	64	64	64	72	65	84
	Kd	88	88	45	21	189	86	78	105	105	40	61	86	86
Soybean hulls	a	13	18	23	24	4.8	27	48	75	5.6	21	20	8.9	7.7
	b	80	71	70	62	94	72	57	33	66	52	68	84	97
	Kd	4.0	20	3.0	97	8.0	149	3.0	1.0	100	9.0	4.0	38	3.0
Wheat bran	a	11	0.3	17	75	88	10	16	-	12	8.1	11	10	10
	b	64	89	43	25	8	90	76	-	80	84	85	34	81
	Kd	36	51	17	24	14	156	38	-	29	20	18	21	38
Maize gluten	a	9.2	0.1	1.5	11	0.2	61	81	-	19	3.0	3.0	1.7	1.7
	b	54	90	34	74	85	37	12	-	58	56	56	91	91
	Kd	3.0	23	103	68	49	220	10	-	3.0	11	11	77	77
Cottonseed meal	a	19	28	25	16	-	16	17	61	17	14	-	15	16
	b	52	62	50	81	-	80	70	15	72	68	-	33	71
	Kd	14	46	8.0	40	-	100	31	191	35	14	-	100	31

Ground bean	a	15	11	43	9.0	8.1	8.9	64	9.1	12	13	9.4	16	14
	b	78	82	49	89	86	91	31	83	82	78	87	68	74
	Kd	14	77	5.0	83	13	162	24	93	24	29	2.0	26	24
Peanut meal	a	23	1.0	5.5	17	0.8	17	18	19	21	19	17	25	18
	b	64	96	80	79	86	82	76	63	69	61	76	59	76
	Kd	15	16	8.0	78	8.0	167	46	34	31	38	8.0	11	46
Soybean meal	a	15	0.5	8.6	11	9.1	14	12	55	16	6.7	13	17	11
	b	80	99	88	87	89	86	85	42	80	93	85	75	86
	Kd	12	87	10	40	21	178	27	59	9.0	6.0	7.0	16	27
Sunflower meal	a	10	27	8.9	10	55	6.4	19	8.0	10	7.0	1.0	6.4	8.2
	b	52	66	70	87	36	92	74	58	76	64	85	51	85
	Kd	21	57	11	49	22	122	36	22	31	36	8.0	125	36

* ‘a’ is the immediately soluble fraction; ‘b’ is the insoluble by potentially degradable/releasable fraction; and kd is degradation/release rate of ‘b’ (%/h).
† Dry matter.
‡ Total mineral content.
§ (-) means that the model proposed by Ørskov & McDonald (1979) did not converge due to different release response of minerals.

Table 3. *Dry matter and total and individual mineral parameters of ruminal release of forage feeds*

Feeds	Parameter*	DM†	Ash‡	Ca	P	Na	K	Mg	S	Cu	Zn	Mn	Co	Fe
Maize silage	a	39	56	34	79	-§	-	76	77	38	40	36	45	36
	b	42	20	35	9.1	-	-	12	18	37	41	32	52	31
	Kd	4.0	3.0	9.0	2.0	-	-	9.0	4.0	18	29	18	1.0	9.0
Decumbens	a	21	67	17	-	-	-	23	9.3	19	22	25	12	23
	b	63	14	67	-	-	-	58	78	41	46	66	60	58
	Kd	5.0	24	8.0	-	-	-	16	7.0	6.0	11	29	4	16
Elephant grass	a	16	38	10	57	65	99	50	69	11	16	18	17	17
	b	61	26	79	18	25	0.5	42	25	61	30	74	57	70
	Kd	5.0	9.0	7.0	62	5.0	32	10	4.0	4.0	15.0	12	0.1	10
Humidicola	a	18	54	18	-	22	99	34	22	8.3	18	20	12	21
	b	59	19	65	-	50	0.7	53	51	49	33	69	85	63
	Kd	5.0	11	8.0	-	20	29	26	2.0	10	14	33	6.0	26
Marandu	a	19	69	16	-	64	98	22	22	18	7.0	22	38	22
	b	64	11	66	-	23	1.9	57	63	42	28	64	40	57
	Kd	5.0	10	8.0	-	9.0	168	19	5.0	9.0	10	34.0	4.0	19
MG-4	a	21	68	16	-	-	98	24	18	6.4	18	24	25	24
	b	63	9.1	68	-	-	1.2	58	71	51	52	66	58	58
	Kd	5.0	23	8.0	-	-	56	18	0.1	8.0	13	27	8.0	18
Mombaça	a	17	44	29	-	25	99	22	41	31	-	20	18	22
	b	56	28	55	-	40	0.7	57	45	44	-	58	53	57
	Kd	4.0	5.0	10	-	13	60	30	0.0	11.0	-	42	9.0	30
Mulato	a	22	65	19	-	22	99	23	6.8	21	19	22	11	23
	b	60	12	63	-	55	0.8	60	71	56	38	62	80	60
	Kd	5.0	5.0	11	-	0.1	27	17	6.0	20	17	0.2	16	17

Peanut forage	a	28	42	34	41	18	97	16	34	21	26	28	17	16
	b	56	43	49	22	70	2.4	75	42	49	38	59	68	75
	Kd	8.0	12	14	32	11	32	8.0	20	7.0	12	17	10	8.0
Tanzania	a	20	46	16	-	16	95	47	27	23	20	25	8.5	23
	b	57	25	63	-	27	4.6	34	61	30	29	39	26	50
	Kd	4.0	8.0	8.0	-	10.0	166	6.0	4.0	5.0	7.0	12.0	7.0	6.0
Tifton 85 hay	a	12	62	13	-	10	98	22	87	2.3	5.3	12	3.0	13
	b	53	9.3	13	-	64	1.3	34	7.3	28	42	58	37	38
	Kd	4.0	4.0	191	-	6.0	39	18	10	7.0	6.0	21	28	18
Xaraes	a	18	62	12	-	20	98	22	8	4.8	11	21	23	22
	b	65	15	74	-	45	2.0	60	82	58	61	67	61	60
	Kd	4.0	9.0	8.0	-	19	34	20	4.0	8.0	13	21	22	20

* ‘a’ is the immediately soluble fraction; ‘b’ is the insoluble by potentially degradable/releasable fraction; and kd is degradation/releasing rate of ‘b’ (%/h).
† Dry matter.
‡ Total mineral content.
§ (-) means that the model proposed by Ørskov & McDonald (1979) did not converge due to different release response of minerals

Table 4. Coefficients of *dry matter rumen effective degradation, and total and individual mineral rumen release in concentrate and forage feeds considering passage rate = 0.05/h*

Feeds	DM*	Ash†	Ca	P	Na	K	Mg	S	Cu	Zn	Mn	Co	Fe
Concentrates													
Ground ear maize	0.45	0.76	0.36	0.84	0.68	0.95	0.69	0.35	0.57	0.64	0.37	0.81	0.69
Ground maize	0.61	0.77	0.30	0.82	0.91	0.98	0.93	0.86	0.59	0.58	0.36	0.97	0.76
Ground sorghum	0.57	0.70	0.75	0.92	0.79	0.96	0.78	0.76	0.57	0.57	0.79	-‡	0.87
Rice bran	0.63	0.60	0.94	0.59	0.97	0.93	0.93	0.70	0.70	0.69	0.76	0.70	0.89
Soybean hulls	0.48	0.74	0.48	0.83	0.62	0.96	0.68	0.81	0.68	0.54	0.50	0.83	0.41
Wheat bran	0.67	0.81	0.51	0.95	0.94	0.97	0.83	-	0.80	0.75	0.77	0.38	0.82
Maize gluten	0.29	0.74	0.34	0.80	0.77	0.98	0.89	-	0.38	0.42	0.42	0.87	0.87
Cottonseed meal	0.58	0.84	0.56	0.88	-	0.92	0.77	0.75	0.80	0.64	-	0.46	0.77
Ground bean	0.72	0.88	0.67	0.93	0.71	0.97	0.90	0.88	0.80	0.80	0.30	0.73	0.75
Peanut meal	0.71	0.74	0.54	0.92	0.54	0.97	0.87	0.73	0.80	0.73	0.63	0.65	0.87
Soybean meal	0.71	0.94	0.67	0.88	0.81	0.97	0.84	0.93	0.67	0.59	0.62	0.74	0.84
Sunflower meal	0.52	0.87	0.57	0.89	0.84	0.95	0.85	0.55	0.75	0.63	0.54	0.55	0.83
Forages													
Maize silage	0.57	0.63	0.56	0.82	-	-	0.83	0.84	0.67	0.75	0.61	0.52	0.56
Decumbens	0.52	0.78	0.58	-	-	-	0.67	0.56	0.41	0.54	0.80	0.40	0.67
Elephant grass	0.47	0.55	0.57	0.74	0.78	1.00	0.78	0.80	0.38	0.38	0.70	0.51	0.64
Humidicola	0.47	0.66	0.57	-	0.62	1.00	0.78	0.37	0.41	0.42	0.80	0.60	0.74
Marandu	0.51	0.76	0.57	-	0.79	0.99	0.67	0.53	0.45	0.26	0.77	0.54	0.67
MG- 4	0.52	0.76	0.56	-	-	0.99	0.69	0.63	0.37	0.55	0.80	0.59	0.69
Mombaça	0.41	0.58	0.66	-	0.54	1.00	0.71	0.57	0.60	-	0.71	0.51	0.71
Mulato	0.53	0.71	0.63	-	0.61	0.99	0.69	0.46	0.66	0.48	0.71	0.72	0.69

Peanut forage	0.61	0.73	0.70	0.60	0.66	0.99	0.62	0.67	0.50	0.53	0.73	0.63	0.62
Tanzania	0.46	0.61	0.54	-	0.34	0.99	0.67	0.55	0.38	0.37	0.52	0.24	0.51
Tifton 85 hay	0.35	0.66	0.26	-	0.44	0.99	0.49	0.92	0.18	0.27	0.59	0.34	0.43
Xaraes	0.48	0.72	0.57	-	0.55	0.99	0.70	0.46	0.40	0.56	0.88	0.73	0.70

* Dry matter.
† Total mineral content.
‡ (-) means that the model proposed by Ørskov & McDonald (1979) did not converge due to different release response of minerals.

Table 5. *Cluster classification based in soluble (*'a'*), insoluble but potentially degradable/releasable fraction ('b'); and release rate of 'b' (kd) fractions of total ash of feeds*

Cluster #	A	*b*	*Kd*
1	3±4.9	91±6.7	0.5±0.31
2	22±5.2	63±10.1	0.5±0.29
3	45±4.5	34±8.9	0.1±0.03
4	63±5.7	14±4.1	0.1±0.08

Cluster #1: Wheat bran, Maize gluten, Ground bean, Peanut meal, Soybean meal; **Cluster #2:** Ground sorghum, Rice bran, Soybean hulls, Cottonseed meal, Sunflower meal; **Cluster #3:** Ground ear maize, Ground maize, Elephant grass, Mombaça, Peanut forage, Tanzania; and **Cluster #4:** Maize silage, Decumbens, Humidicola, Marandu, MG-4, Mulato, Tifton-85 hay, Xaraes.

Table 6. *Pearson correlation coefficients between content and ruminal release of total ash, and individual minerals of different concentrate and forage feedstuffs (n = 24)*[*]

Item	Ash[†]	CP[‡]	NDF[§]	Ca	P	Na	K	Mg	S	Cu	Zn	Mn	Co	Fe
Ash	-0.51*	0.41*	-0.39*											
Ca	-0.27	-0.06	-0.20	0.49*										
P	0.52*	0.26	-0.23	0.29	0.66*									
Na	0.34*	0.11	-0.58*	0.10	0.49*	0.44*								
K	-0.22	-0.36*	0.56*	0.36*	0.14	-0.21	0.18							
Mg	0.28	0.49*	-0.75*	0.53*	-0.13	0.48*	-0.74*	0.54*						
S	-0.08	0.29	-0.54*	-0.16	-0.30	0.07	-0.07	0.15	0.53*					
Cu	0.48*	0.35*	-0.57*	0.00	0.28	0.19	-0.38*	0.43*	0.13	0.46*				
Zn	0.40*	0.29	-0.55*	-0.11	0.34*	0.22	-0.52*	0.19	0.30	0.39*	0.36*			
Mn	-0.40*	-0.46*	0.34*	0.45*	-0.58*	0.05	0.18	-0.15	-0.12	-0.05	0.02	0.53*		
Co	0.06	0.23	-0.42*	0.37*	-0.17	-0.10	-0.14	0.22	0.35*	0.42*	0.48*	-0.04	-0.09	
Fe	-0.09	0.47*	-0.69*	0.00	0.13	-0.15	-0.24	0.28	0.29	0.36*	0.46*	-0.15	0.01	-0.11

* $P < 0.05$.
† Total mineral content.
‡ Crude protein.
§ Neutral detergent fibre.

Table 7. *Coefficients of dry matter abomasal digestion and total ash, and individual mineral abomasal release of concentrate and forage feedstuffs*

Feed	DM*	Ash†	Ca	P	Na	K	Mg	S	Cu	Zn	Mn	Co	Fe
Average all feeds	0.68	0.82	1.00	1.00	1.00	1.00	1.00	1.00	0.74	0.93	0.92	0.75	0.79
Average concentrates	0.71	0.91	1.00	1.00	1.00	1.00	1.00	1.00	0.88	0.97	0.88	0.68	0.79
Ground ear maize	0.61	0.90	1.00	1.00	1.00	1.00	1.00	1.00	0.83	0.89	0.91	0.45	0.74
Ground maize	0.82	0.95	0.99	1.00	1.00	1.00	1.00	1.00	0.72	0.98	0.70	0.65	0.18
Ground sorghum	0.71	0.86	1.00	1.00	1.00	1.00	1.00	1.00	0.74	1.00	0.96	0.84	0.74
Rice bran	0.70	0.67	1.00	1.00	1.00	1.00	1.00	1.00	0.97	1.00	1.00	0.98	0.99
Soybean Hulls	0.63	0.97	1.00	0.99	1.00	1.00	1.00	1.00	0.86	0.92	0.43	0.49	0.77
Wheat bran	0.77	0.96	1.00	1.00	1.00	1.00	1.00	1.00	0.96	1.00	0.99	0.89	0.89
Maize gluten	0.38	0.97	1.00	1.00	1.00	1.00	1.00	1.00	0.88	0.94	0.79	0.44	0.85
Cottonseed meal	0.68	0.97	1.00	1.00	1.00	1.00	1.00	1.00	0.72	0.96	0.76	0.51	0.37
Ground bean	0.91	0.99	1.00	1.00	1.00	1.00	1.00	1.00	0.96	0.99	0.99	0.85	0.98
Peanut meal	0.87	0.73	1.00	1.00	1.00	1.00	1.00	1.00	0.96	0.99	1.00	0.79	0.99
Soybean meal	0.88	0.98	1.00	1.00	1.00	1.00	1.00	1.00	0.96	0.98	1.00	0.78	0.99
Sunflower meal	0.61	0.97	1.00	1.00	1.00	1.00	1.00	1.00	0.94	0.98	1.00	0.56	0.95
Average forages	0.65	0.74	1.00	1.00	0.99	1.00	1.00	1.00	0.60	0.90	0.97	0.81	0.79
Maize silage	0.65	0.64	1.00	1.00	1.00	1.00	1.00	1.00	0.72	0.95	0.94	0.91	0.89
Decumbens	0.67	0.77	1.00	1.00	0.99	1.00	1.00	1.00	0.58	0.93	0.97	0.82	0.71
Elephant grass	0.66	0.60	1.00	1.00	1.00	1.00	1.00	1.00	0.61	0.94	0.99	0.93	0.92
Humidicola	0.62	0.74	1.00	1.00	1.00	1.00	1.00	1.00	0.79	0.90	1.00	0.79	0.88
Marandu	0.67	0.77	1.00	0.98	0.98	1.00	1.00	0.99	0.38	0.87	0.89	0.89	0.71
MG- 4	0.68	0.74	1.00	1.00	0.99	1.00	1.00	1.00	0.72	0.91	0.96	0.98	0.80

Mombaça	0.56	0.72	1.00	1.00	0.99	1.00	1.00	1.00	0.43	0.83	1.00	0.61	0.79
Mulato	0.68	0.73	1.00	1.00	0.99	1.00	1.00	1.00	0.65	0.82	0.94	0.83	0.83
Peanut forage	0.81	0.96	1.00	1.00	1.00	1.00	1.00	1.00	0.72	0.92	1.00	0.90	0.59
Tanzania	0.60	0.67	1.00	1.00	0.98	1.00	1.00	1.00	0.82	0.88	0.99	0.44	0.79
Tifton 85 hay	0.51	0.72	1.00	1.00	0.99	1.00	1.00	1.00	0.12	0.90	0.99	0.79	0.68
Xaraes	0.68	0.76	1.00	1.00	0.99	1.00	1.00	1.00	0.63	0.95	0.99	0.79	0.82

* Dry matter.
† Total mineral content.

Chapter 2

Influence of inorganic mineral supplementation on nutrient intake and digestibility, productive performance, and mineral status in Nellore young bulls[1]

D. Zanetti[a*], L.A. Godoi[a], M.M. Estrada[a], T.E. Engle[b], M.V.C. Pacheco[a], J.M.V. Pereira[a], E. Detmann[a], M.F. Paulino[a], S.C. Valadares Filho[a]

[a] *Department of Animal Science, Universidade Federal de Viçosa, Viçosa, Brazil*

[b] *Department of Animal Science, Colorado State University, Fort Collins, United States of America*

*** Corresponding author:** Diego Zanetti, diegozanetti_@hotmail.com, diego.zanetti@ufv.br

[1] This work was supported by the National Council of Scientific and Technological Development (CNPq); National Institute of Science and Technology in Animal Science (INCT – Ciência Animal); Coordination of Improvement of Personal Higher Education (CAPES); and Foundation for Research Support of the State of Minas Gerais (FAPEMIG).

ABSTRACT

The objectives of this study were to measure the effects of mineral supplementation on nutrient intake and digestibility, performance, and mineral concentrations in the bodies of Nellore beef cattle fed with and without calcium (Ca), phosphorus (P) and micromineral (MM) supplemental sources during the growing and finishing phases. Five animals were slaughtered at the beginning of the experiment to measure initial mineral status. Forty-two Nellore beef cattle (initial body weight = 270.4 ± 36.6 kg, age = 8 months) were assigned to one of six treatments: sugarcane as a roughage source and a soybean meal and soybean hull-based concentrate with (SH100) and without (SH0) Ca, P and MM supplementation; sugarcane as the roughage source and a soybean meal and ground corn-based concentrate with (SC100) and without (SC0) Ca, P and MM supplementation; and corn silage as the roughage source with a soybean meal and corn-based concentrate with (CS100) and without (CS0) Ca, P and MM supplementation. The experiment was conducted as a completely randomized design with a 3 × 2 factorial arrangement of treatments. Nutrient intake and digestibility, bone and serum parameters related to Ca and P metabolism, and liver mineral concentrations were measured. Nutrient intake, digestibility and performance were not affected by mineral factor ($P > 0.10$). Water intake was greater in SH100 when compared to other diet types and to SH0 ($P < 0.05$). Rib bone breaking strength and densitometry were reduced ($P < 0.04$) in absence of supplementation. Metatarsus parameters were not affected ($P > 0.10$). Liver Cu content was reduced ($P < 0.01$) in diets without supplementation. Therefore, absence of mineral supplementation does not influence intake and performance of Nellore beef cattle. However, this absence may influence serum, liver and bone parameters according to dietary type.

Keywords – Calcium; Copper; Manganese; Phosphorus; Supplementation; Zinc

1. INTRODUCTION

Nellore beef cattle is a breed widely used in some parts of the world, but lacks substantial representation in the literature, mainly in relation to mineral nutrition. Regarding mineral nutrition, it has been reported that, under standard feeding conditions, nutritionists supplement minerals in a way that exceeds the NRC (1996) recommendations (Vasconcelos and Galyean, 2007; Berret et al., 2015; Zanetti et al., 2017a). It is important both from an economic perspective, due to financial inputs with supplementation, and an environmental perspective, due to excretion of potential pollutants, i.e. P (Jongbloed and Lenis, 1998; Sehested, 2004; Prados et al., 2017), to ensure adequate mineral supplementation.

Furthermore, 83.1% of Brazilian feedlot nutritionists recommend providing supplemental minerals sources to beef cattle (Millen et al., 2009). Recently, Costa e Silva et al. (2015) reported no differences in feed intake or productive performance of Nellore bulls fed with or without Ca and P supplementation during the growing and finishing phases of production. Likewise, research has demonstrated contradictory results for performance and mineral status based on micromineral (MM) supplementation for other breeds of cattle (Engle and Spears, 2000; Mullis et al., 2003; Spears and Weiss, 2014). Besides this, different types of feedstuffs may propitiate different dietary minerals from the basal diet. For example, the sugarcane and corn silage are the most frequently used forage source in Brazilian feedlots (Millen et al., 2009; Pinto and Millen, 2016). However, these two forage sources differ regarding Ca and P contents.

Therefore, we hypothesized that Nellore fed without Ca, P and MM supplemental sources in diets for growing and finishing cattle would have similar performances, but lower mineral concentrations in mineral reserve tissues than animals fed supplemental minerals. Our objectives were to measure the effects of mineral supplementation on nutrient intake and digestibility,

performance, and mineral status in Nellore beef cattle fed with and without Ca, P and MM supplemental sources during the growing and finishing phases of production.

2. MATERIAL AND METHODS

The experiment was carried out in the Experimental Feedlot of the Animal Science Department of the Universidade Federal de Viçosa, Viçosa, MG, Brazil, following the approval of the animal handling and procedures described herein by the Ethics Committee for Animal Use (protocol CEUAP/DZO/UFV 17/2015).

2.1. Animals, experimental design and treatments

Forty-seven Nellore young bulls with an average initial body weight of 270.4±6.6 kg were used. The animals were weaned at approximately eight months of age at the Beef Cattle Sector of the Animal Science Department of the Universidade Federal de Viçosa and immediately transported to the Experimental Feedlot at the same location. Five animals were slaughtered prior to the initiation of the experiment to measure mineral status. The 42 remaining animals were housed in a group pen (48.0 m^2) that contained electronic feeders (Model AF-1000 Master, Intergado Ltd., Contagem, Minas Gerais, Brazil) and waterers (Model WD-1000 Master, Intergado Ltd., Contagem, Minas Gerais, Brazil) for 125 d. Prior to initiation of the experiment, each animal was fitted with an ear tag containing a unique passive transponder (FDX – ISO 11784/11785; Allflex, Joinville, Santa Catarina, Brazil) in the left ear. Animals were weighed at 0800 h after a 16 h fasting (feed only) period at beggining and at the end of experiment. Average daily weight gain was calculated as the difference between the final and initial weights in relation to the number of days on feed.

Diets were formulated according to the BR-Corte (Valadares Filho et al., 2010) for a gain of 1.1 kg per day, and were isonitrogenous (13.5% CP). Sugarcane or corn silage were used as forage source. Inorganic mineral sources, soybean hulls, soybean meal, and/or grounded corn were used as concentrated ingredients. Feedstuffs were selected based on their adoption in Brazilian feedlots and to provide significant variation in the mineral content among treatments. For example, the corn presents a relationship between Ca and P of 1:8.3, while this relationship to soybean hulls is 3.4:1 (Valadares Filho et al., 2015).

The animals were randomly divided into six treatments. In the diets without supplementation, the same amount of mineral was replaced by inert sand, in order not to alter the concentrations of the other components of the diets. The proportions of the ingredients of the concentrates and diets, as well as their chemical compositions are shown in **Table 1**. Therefore, treatments were: 1) sugarcane as the forage source with a concentrate supplement composed of soybean meal and soybean hulls with (SH100) and without (SH0) Ca, P and MM supplementation; 2) sugarcane as the forage source with a concentrate supplement composed of soybean meal and ground corn with (SC100) and without (SC0) Ca, P and MM supplementation; and 3) corn silage as the forage source with a concentrate supplement composed of soybean meal and ground corn with (CS100) and without (CS0) Ca, P and MM supplementation.

[Table 1 near here]

A total mixed ration was provided twice a day, at 0800 and 1600 h. The Intergado monitoring systems were used to evaluate individual feed and water intake. For each feed bunk or waterer visit, the system recorded the animal number, feed bunk or waterer number, initial and final times of each feeding or drinking event, and the weight of the feeder or waterer. These data

were continuously recorded and transferred via a network cable, to the Intergado web software for data capture and storage (Chizzotti et al., 2015).

2.2. Digestibility trial

Water samples were collected on days 15, 65 and 115 from the waterers. Daily, all feeds were sampled. Feed samples were oven-dried (55°C), grounded in a knife mill using 2 and 1 mm screens sequentially, and were packed in plastic bags for further laboratory analyses. For the evaluation of the digestibility of the diets and bioavailability of minerals, fecal grab samples were obtained over a 5-day period from day 70 to day 75. The collections were conducted at 0600 h of the first day, at 0900 h of the second day, at 1200 h of the third day, at 1500 h of the fourth day and at 1800 h on the fifth day. Feces were packed in aluminum trays, dried in a forced-ventilation oven at 55°C, and then ground in a knife mill as described above.

Samples of feedstuffs and feces were analyzed for dry matter (DM; method 934.01), ash (method 930.05), and total nitrogen (N, method 981.10), according to the AOAC (2012) methods of analysis protocols. The organic matter (OM) content was obtained as the difference between DM content and ash content. The crude protein (CP) content was obtained by multiplying 6.25 by the total nitrogen content. Neutral detergent fiber (NDF) content was obtained according to Mertens et al. (2002) without addition of sodium sulfite and with the addition of a thermostable alpha-amylase. The indigestible neutral detergent fiber content (iNDF) was calculated after *in situ* incubation of the samples (ground to 2 mm using a knife mill), using F57 bags (Ankon®) for 288 h as described by Valente et al. (2011a) for tropical forages. The iNDF was adopted as a marker to calculate total fecal excretion. The daily fecal excretion was estimated by the relationship between iNDF intake and iNDF concentrations of fecal samples.

All feedstuffs were analyzed for calcium (Ca), phosphorus (P), sodium (Na), potassium (K), magnesium (Mg), sulfur (S), copper (Cu), zinc (Zn), manganese (Mn), cobalt (Co), iron (Fe) and molybdenum (Mo) contents. For mineral analysis, samples were weighed in pre-weighed acid-washed crucibles and placed in an ash oven and ashed at 600°C for 12 h. Samples were then removed and placed in a desiccator for an additional 30 min to cool. Finally, samples were weighed and suspended with 5 mL of HCl (1.2 N HCl). Water samples were analyzed without preparation, following the procedures described later. Mineral concentrations of the samples were determined through inductively coupled plasma optical emission spectroscopy methods (Optima 7300 DV, Perkin Elmer; Braselton et al., 1997).

2.3. Serum parameters

After 124 d in the feedlot, a blood sample was collected via jugular venipuncture by using tubes without coagulation accelerator (8 ml; LaborImport®, Osasco, Brazil). The samples were immediately centrifuged at 3,600 x g for 20 min at room temperature. The serum was analyzed for Ca and P (Automatic biochemical analyzer – Autoanalyzer. Mindray®, model BS200E, China), parathyroid hormone (PTH, Pth stat, Roche Cobas E601 Immunology Analyzer, USA) and bone alkaline phosphatase (BAP, MicroVue BAP Elisa kit, Germany).

2.4. Carcass characteristics

The animals were slaughtered after 125 d. Prior to slaughter, all animals were fasted (feed only) for 16 h. Animals were then slaughtered via captive bolt stunning followed by exsanguination. After slaughter, the carcass of each animal was divided into two halves, which were weighed, and were then cooled in a cold chamber at 4°C for 18 h. After this period, the half-carcasses were removed from the cold chamber for weighing and to perform the complete dissection of the left half of the carcass. Hot and cold carcass yields (HCY and CCY, respectively)

were calculated. Also, the subcutaneous fat thickness (SFT) was measured using a digital caliper in the region between 11^{th} and 12^{th} rib cut. For determination of the rib eye area, the *longissimus dorsi* muscle was sectioned between 11^{th} and 12^{th} ribs, the muscle outline was drawn on a transparent sheet, scanned, and the area was measured with the aid of AutoCAD software (Autodesk Inc.).

2.5. Liver, 10^{th} rib bone and metatarsus parameters

Samples of liver, 10^{th} rib bone and metatarsus were analyzed for mineral content, as described previously. Bone density was evaluated by dual energy X-ray absorptiometry (DEXA, GE Lunar, General Electric Company). Bone sections were scanned (Epson L355, Seiko Epson Corporation) and section areas were determined with the aid of AutoCAD software (Autodesk Inc.). Bone mechanical resistance was evaluated in bones with the aid of a mechanical universal testing machine. A flection test was performed on the 10^{th} rib bone, while a compression test was performed on the metatarsus. For both tests, breaking strengths were measured. Breaking strength was expressed in absolute values (kg_F) and scaled to bone sectional area (kg_F/cm^2).

2.6. Statistical analyses

The experiment was conducted as a completely randomized design with a 3 × 2 factorial arrangement of treatments (three types of diets and presence or absence of supplementary mineral). The data were submitted to variance analyses. All statistical analyses were performed using the MIXED procedure of SAS (SAS Institute Inc., Cary, NC). P=0.05 was utilized as the critical level of probability for the Type-I error.

3. RESULTS AND DISCUSSION

Due to the lower impact of minerals in animal performance and feedlot profitability, minerals have been less studied than other nutrients, such as protein or energy. Typically, minerals are included in diets to prevent deficiencies. However, data have suggested that minerals from feedstuffs can meet the mineral requirements of cattle (Geisert et al., 2010; Erickson et al., 1999). Thus, dicalcium phosphate, limestone, zinc sulfate, manganese sulfate, copper sulfate and cobalt sulfate were not supplied in treatments SH0, SC0 and CS0 (**Table 1**). By design, in diets without supplementation, the Ca, P, Zn, Mn, Cu and Co dietary concentrations were reduced. However, S and Fe concentrations were also reduced.

Analyzed water showed a small contribution from water to mineral intake in this experiment. Due to this, minerals consumed from water were not included in the total mineral intake. Among all treatments, the mean of the water intake was 18.29 ± 4.42 L/d. Considering the mineral content of water, the mineral intake was added, in mg/d, Ca = 111.33, Na = 97.06, K = 71.46, Mg = 46.97, S = 47.40, Fe = 0.05, Mn = 0.04, Mo = 0.14 and Zn = 0.05. Phosphorus content was less than 1 mg/L, Cu and Co were less than 10 ppb, and due to this, were not measured: these values were considered as a negligible contribution. In addition, mineral intake from water is not commonly measured, and considering it negligible tends to facilitate comparisons between this and other experiments.

Changes in nutritional aspects may affect animal feed and water intake (Mendes et al., 2015). However, limited studies have been published examining the influence of minerals on feed and water intake (Ganskopp, 2001; Porath et al., 2002). Of the studies that have been published, the work has focused on manipulating the salt content of the free-choice minerals, to influence mineral intake, animal distribution and grazing patterns. Considering water intake, an interaction (P<0.02) between diet and mineral factors was observed. Cattle given the SH100 diet consumed

more water than those given SC100 or CS100. Similarly, the SH100 group presented a greater water intake than the SH0 group. For dairy cattle, water intake is directly related with dietary sodium (Murphy et al., 1983), dietary potassium (Fraley et al., 2015) and dietary ash (Appuhamy et al., 2014). However, this relationship is most likely related to an increase in osmotic pressure of the rumen fluid, instead of to a specific mineral (Rogers et al., 1979). However, a negative relationship to dietary salt was reported by Hicks et al. (1988). This relationship is discussed in the NRC (1996), where, for each 1% increase in dietary salt daily, water intake decrease 4.44 L.

Feed intake is essential to profitability of feedlot systems, due to the direct relationship with weight gain (Steen and Kilpatrick, 2000; Zinn et al., 2008). Numerous factors may affect feed intake, such as gender (Zinn et al., 2008), dietary fiber (Detmann et al., 2014), salt content of the diet (Valente et al., 2011b), and dietary mineral concentration (Engle and Spears, 2000; Ward and Spears, 1997). The mineral aspect is complex and less-studied than other nutrients. This complexity is related to the impact that minerals may have on bacterial activity and interactions with other nutrients (Prados et al., 2015). Hosnedlova et al. (2007) reported a loss of appetite and inefficient use of nutrients when Zn deficiencies were detected in ruminants. This is in agreement with Ward and Spears (1997), who reported an increased DMI of growing steers fed with a Cu supplemented diet in contrast with steers fed a diet not supplemented with Cu (10.2 vs. 5 mg of Cu/kg DM).

In the present study, DM, OM and CP intakes were not affected ($P > 0.40$) by treatments (**Table 2)**. Intake of NDF was reduced ($P < 0.01$) in treatments SC100 and SC0. It is related to a greater forage content in corn silage-based treatments, and to a greater NDF content in soybean hulls in comparison to corn. Dry matter, OM, NDF and CP digestibility were not affected ($P > 0.33$) by mineral supplementation. Our results agree with the recently published studies of Costa

e Silva et al. (2015), Prados et al. (2015), and Zanetti et al. (2017b) who reported similar nutrient intakes and digestibilities among treatments with and without Ca and P supplementation, for Nellore heifers and steers, Holstein × Zebu bulls and Holstein × Zebu steers, respectively. Similarly, Flachowsky et al. (2009) reported no differences on apparent digestibility of crude nutrients of bulls fed 2, 3 or 4 mg P/kg DM and Mullis et al. (2003) reported similar DMI for heifers fed 0, 7 or 14 mg of supplemental Cu in two experiments (basal diets contained 6.4 and 4.4 mg of Cu/kg DM in experiments 1 and 2, respectively).

[Table 2 near here]

Dietary effects were observed ($P < 0.01$) for DM, OM, CP and NDF digestibilities. Sugarcane-based diets presented greater DM and OM digestibility coefficients, which could be related to greater amounts of concentrates in these diets. Neutral detergent fiber presented reduced digestibility in diets where sugarcane was the forage source and the concentrate was ground corn-soybean hulls based. This smaller coefficient is a consequence of the proportion of indigestible NDF in relationship to dietary NDF. While iNDF was 30.9 and 32.6% of NDF in SH and CS diets, respectively, this proportion was 38.5% in SC treatments. Regarding CP digestibility coefficients, corn silage-based diets presented smaller coefficients than sugarcane-based diets. This could be explained by a smaller amount of urea in the CS diets, and consequently smaller amounts of rumen degradable protein. Among sugarcane based treatments, SH diets presented greater CP digestibility coefficients than SC diets.

With regard to the productive performance and efficiency parameters measured, average daily gain, and empty body weight gain, hot and cold carcass yield, carcass length, rib eye area, and subcutaneous fat thickness were not affected ($P > 0.10$) by treatments (**Table 3**). Therefore, efficiencies of gain per DMI were not affected ($P > 0.10$). Rotta et al. (2014) reported similar

performance parameters between sugarcane and corn silage-based diets. However, the proportion of concentrates needs to be altered in order to retain performance and efficiency: forage in sugarcane based diets need to be 20% greater than in corn silage diets. Similarly, partial replacement of corn by soybean hulls have been used without compromising performance parameters.

[Table 3 near here]

Regarding mineral factors, these results agree with previously published studies reporting that removal of Ca and P supplementation to feedlot diets had no impact on cattle performance (Erickson et al., 1999; Prados et al., 2015). However, Malcolm-Callis et al. (2000) reported that Zn supplementation above NRC recommendations (at 20, 100 or 200 mg of Zn/kg DM) does not improve performance or carcass characteristics of finishing beef steers. Similarly, Rhoads et al. (2003) did not observe differences in ADG, gain: feed, SFT and KPH (kidney, pelvic, heart) fat between steers fed 1.5 or 3 times NRC (1996) requirements of Cu, Zn, Mn and Co supplied as inorganic sources. However, a reduction in rib eye area and dressing percentage of cattle supplied at three times the NRC requirements with inorganic sources, in comparison to cattle supplied 1.5 times NRC requirements have been reported (Rhoads et al., 2003). A reduction in subcutaneous fat thickness and a greater *longissimus muscle* area was reported by Ward and Spears (1997) in the growing and finishing phases for steers supplied with 5.2 mg of Cu/kg DM in a basal diet containing 5.0 mg of Cu/kg DM vs. animals fed without supplementation.

Serum Ca was greater ($P < 0.01$) in cattle receiving SH0 when compared to SH100 treatments (10.53 vs. 9.64 mg/dl, respectively; **Table 4**). Similarly, serum Ca was greater ($P < 0.01$) in SH0 than SC0 and CS0 (10.00 and 9.94 mg/dl, respectively). In all situations, serum Ca and P concentrations were within adequate concentrations (Goff, 2000). For all factors, bone

alkaline phosphatase concentrations were not altered ($P > 0.10$). Parathyroid hormone (PTH) was greater ($P < 0.05$) in cattle receiving CS0 than cattle receiving CS100. In this case, PTH action was measured in other parameters of treatment CS0 in comparison to CS100, with reduction of rib bone density, followed by Ca and P reduction in the bone. The possible reason for this was the Ca:P ratio. The NRC (2016) recommends a Ca:P ratio ranging from 1:1 to 7:1. In the CS0 treatment, this ratio was 0.75:1, showing an excessive P in relation to Ca amounts.

[Table 4 near here]

Among factors that can affect bone development, nutrition can affect chemical and physical bone properties (Loveridge, 1999). Williams et al. (1991) reported increases in metacarpal strength and calcification in Angus heifers fed with adequate dietary P (0.20% of DM) when compared to heifers fed with low dietary P (0.12% of DM). From baseline animals, animals of all treatments presented a numerically greater section area of bone after 125 d on feed. Initial and final section areas of rib bone were 2.46 and 3.68 cm^2, respectively; a growth of 1.22 cm^2 during the feedlot period. Similarly, metatarsus area growth from 8.09 to 12.03 cm^2; a growth of 3.94 cm^2 during the feedlot period. This growth was followed by an increased concentration of ash, Ca and P in both bones measured. Ash content was numerically increased from 37.35 to 55.24% and 38.85 to 57.21% in rib bone and metatarsus, respectively. Calcium content was numerically increased from 7.05 to 10.37% and 17.41 to 25.61% in rib bone and metatarsus, respectively. Similarly, the P content was numerically increased from 1.18 to 1.74% and 0.71 to 1.04% in rib bone and metatarsus, respectively. Besides this growth in area, and increase in ash, Ca and P contents during the feedlot period, rib bone breaking strength (in kg_F and kg_F/cm^2) and density were reduced ($P < 0.04$) by the absence of Ca, P and MM supplementation. Ca and P deposition in rib bone were

reduced ($P < 0.01$) in CS100 compared to CS0. Similar relationships were not observed for other treatments.

Metatarsus parameters were not affected ($P > 0.08$). These effects were found only on rib bone, and in detriment to effects on metatarsus, were related to different rates of mineral deposition and reabsorption according to the type and function of bones (Williams et al., 1990; Suttle, 2010). Rib bone has a primary function in thoracic protection, and the metatarsus has a primary function on locomotion and body sustentation, which makes the metatarsus more important with regard to physics and structural aspects.

Mineral liver concentrations in baseline animals were (mg/kg): Ca = 0.03, P = 0.92, Na = 0.40, K = 1.34, Mg = 0.09, S = 0.03, Cu = 914.43, Co = 0.51, Fe = 438.98, Mn = 11.31, Mo = 4.22 and Zn = 138.96. For a few minerals, e.g. Cu and Fe, a numerical reduction for liver content was observed from baseline animals to animals fed for 125 d. This reduction is reported during the growth phase, with high contents of few minerals in calves and smaller contents in young bulls (Gengelbach et al., 1994).

Dietary factors did not affect ($P > 0.10$) mineral liver concentrations (**Table 5**). The copper concentration in the liver was reduced ($P < 0.01$) in diets without Ca, P and MM supplementation. The copper status in the liver is dependent on several factors, including breed and dietary Cu content (Ward et al., 1995; Miranda et al., 2010). Correa et al. (2012, 2014) fed Nellore bulls a basal diet without Cu supplementation, or with 10 and 40 mg Cu/kg DM diets with copper sulfate or copper proteinate. This study showed that liver Cu concentrations were 237.3 mg Cu/kg DM for non-supplemented cattle, but were greater for cattle supplemented with Cu regardless of Cu source. The values reported in the present study are similar to those described by Correa et al. (2012, 2014): cattle that did not received supplemental Cu had lower liver mineral concentrations

than cattle that received supplementation of Cu (249.8 vs. 394.1 mg Cu/kg DM, respectively). In comparison to other breeds, liver Cu concentrations in Nellore cattle reported in the current experiment showed a lower reduction in the absence of Cu supplementation than previously reported in Angus and Simmental steers: Engle and Spears (2001) reported a reduction from 356.0 to 28.0 mg Cu/kg DM in the liver when Angus steers were fed with 40 and 0 mg Cu/kg DM, respectively. Similarly, Mullis et al. (2003) reported a reduction from 296.3 to 40.5 mg Cu/kg DM when the supplied Cu changed from 14 to 0 mg/kg DM for Simmental steers.

[Table 5 near here]

4. CONCLUSIONS

The absence of inorganic supplemental sources of Ca, P, Cu, Mn, Zn and Co in diets for Nellore beef cattle during both the growing and finishing phases does not affect intake, digestibility or performance. However, this absence may influence serum, liver and bone parameters according to dietary type.

Disclosure statement

No potential conflict of interest was reported by the authors.

References

AOAC. 2012. Official Methods of Analysis. 19th ed. AOAC International, Gaithersburg, MD.

Appuhamy JADRN, Wagner-Riddle C, Casper DP, France J, Kebreab E. 2014. Quantifying body water kinetics and fecal and urinary water output from lactating Holstein dairy cows. J Dairy Sci. 97:6177-6195.

Berrett CJ, Wagner JJ, Neuhold KL, Caldera E, Sellins KS, Engle TE. 2015. Comparison of National Research Council standards and industry dietary trace mineral supplementation strategies for yearling feedlot steers. Profess Anim Sci. 31:237-247.

Braselton WE, Stuart KJ, Mullaney TP, Herdt TH. 1997. Biopsy mineral analysis by inductively coupled plasma atomic emission spectroscopy with ultrasonic nebulization. J Vet Diagn Invest. 9:395–400.

Chizzotti ML, Machado FS, Valente EEL, Pereira LGR, Campos MM, Tomich TR, Coelho SG, Ribas MN. 2015. Technical note: Validation of a system for monitoring individual feeding behavior and individual feed intake in dairy cattle. J Dairy Sci. 98:3438-3442.

Coppock CE, Everett RW, Merrill WG. 1972. Effect of ration on free choice consumption of calcium-phosphorus supplements by dairy cattle. J Dairy Sci. 55:245-256.

Correa LB, Zanetti MA, Del Claro GR, Paiva FA, Silva SDL, Netto AS. 2014. Effects of supplementation with two sources and two levels of copper on meat lipid oxidation, meat colour and superoxide dismutase and glutathione peroxidase enzyme activities in Nellore beef cattle. Brit J Nutr. 112:1266-1273.

Correa LB, Zanetti MA, Del Claro GR, Melo MP, Rosa AF, Netto AS. 2012. Effect of supplementation of two sources and two levels of copper on lipid metabolism in Nellore beef cattle. Meat Sci. 91:466-471.

Costa e Silva LF, Engle TE, Valadares Filho SC, Rotta PP, Valadares RFD, Silva BC, Pacheco MVC. 2015. Intake, apparent digestibility, and nutrient requirements for growing Nellore heifers and steers fed two levels of calcium and phosphorus. Livest Sci. 181:17-24.

Detmann E, Valadares Filho SC. 2010. On the estimation of non-fibrous carbohydrates in feeds and diets. Braz J Vet Anim Sci. 62:980-984.

Detmann E, Gionbelli MP, Huhtanen P. 2014. A meta-analytical evaluation of the regulation of voluntary intake in cattle fed tropical forage-based diets. J Anim Sci. 92:4632-4641.

Engle TE, Spears JW. 2000. Effects of dietary copper concentration and source on performance and copper status of growing and finishing steers. J Anim Sci. 78:2446-2451.

Engle TE, Spears JW. 2001. Performance, carcass characteristics, and lipid metabolism in growing and finishing Simmental steers fed varying concentrations of copper. J Anim Sci. 79:2920-2925.

Erickson GE, Klopfenstein TJ, Milton CT, Hanson D, Calkins C. 1999. Effect of dietary phosphorus on finishing steer performance, bone status, and carcass maturity. J Anim Sci. 77:2832-2836.

Flachowsky G, Klosch M, Schneider A, Richter G.H, Löhnert HJ. 1998. Influence of various phosphorus supply on apparent digestibility of crude nutrients, growing and slaughtering performance as well as selected parameters of metabolism of fattening bulls. Arch Anim Nutr. 51:39-52.

Fraley SE, Hall MB, Nennich TD, 2015. Effect of variable water intake as mediated by dietary potassium carbonate supplementation on rumen dynamics in lactating dairy cows. J Dairy Sci. 98:3247-3256.

Ganskopp D. 2001. Manipulating cattle distribution with salt and water in large arid-land pastures: a GPS/GIS assessment. Appl Anim Behav Sci. 73:251-262.

Geisert BG, Erickson GE, Klopfenstein TJ, Macken CN, Luebbe MK, MacDonald JC. 2010. Phosphorus requirement and excretion of finishing beef cattle fed different concentrations of phosphorus. J Anim Sci. 88:2393-2402.

Gengelbach GP, Ward JD, Spears JW. 1994. Effect of dietary copper, iron, and molybdenum on growth and copper status of beef cows and calves. J Anim Sci. 72:2722-2727.

Goff JP. 2000. Pathophysiology of calcium and phosphorus disorders. Vet Clin North Am Food Anim Pract. 16:319-337.

Hicks RB, Owens FN, Gill DR, Martin JJ, Strasia CA. 1988. Water intake by feedlot steers. Miscellaneous publication - Agricultural Experiment Station, Oklahoma State University (USA).

Hosnedlová B, Trávníček J, Šoch M. 2007. Current view of the significance of zinc for ruminants: a review. Agricultura tropica et subtropica, 40:57-64.

Jongbloed AW, Lenis NP. 1998. Environmental concerns about animal manure. J Anim Sci. 76:2641-2648.

Lopez-Villalobos N, Garrick DJ, Holmes CW, Blair HT, Spelman RJ. 2000. Profitabilities of some mating systems for dairy herds in New Zealand. J Dairy Sci. 83:144-153.

Loveridge N. 1999. Bone: more than a stick. J Anim Sci. 77:190-196.

Malcolm-Callis KJ, Duff GC, Gunter SA, Kegley EB, Vermeire DA. 2000. Effects of supplemental zinc concentration and source on performance, carcass characteristics, and serum values in finishing beef steers. J Anim Sci. 78:2801-2808.

Mendes FBL, Silva RR, Carvalho GGP, Silva FF, Lins TOJD, Silva ALN, Macedo V, Abreu Filho G, Souza SO, Guimarães JO. 2015. Ingestive behavior of grazing steers fed increasing levels of concentrate supplementation with different crude protein contents. Trop Anim Health Prod. 47:423-428.

Mertens DR. 2002. Gravimetric determination of amylase-treated neutral detergent fibre in feeds with refluxing beakers or crucibles: collaborative study. J AOAC Int. 85:1217-1240.

Millen DD, Pacheco RDL, Arrigoni MDB, Galyean ML, Vasconcelos JT. 2009. A snapshot of management practices and nutritional recommendations used by feedlot nutritionists in Brazil J Anim Sci. 87:3427-3439.

Miranda M, Gutiérrez B, Benedito JL, Blanco-Penedo I, García-Vaquero M, López-Alonso M. 2010. Influence of breed on blood and tissue copper status in growing and finishing steers fed diets supplemented with copper. Arch Anim Nutr. 64:98-110.

Mullis LA, Spears JW, McCraw RL. 2003. Estimated copper requirements of Angus and Simmental heifers. J Anim Sci. 81:865-873.

Murphy MR, Davis CL, McCoy GC. 1983. Factors affecting water consumption by Holstein cows in early lactation. J Dairy Sci. 66:35-38.

NRC. 1996. Nutrient requirements of beef cattle. 7th rev. ed. Natl. Acad. Press, Washington, D.C.

NRC. 2001. Nutrient requirements of dairy cattle. 7th rev. ed. Natl. Acad. Press, Washington, D.C.

NRC. 2016. Nutrient requirements of beef cattle. 8th rev. ed. Natl. Acad. Press, Washington, D.C.

Porath ML, Momont PA, Del Curto T, Rimbey NR, Tanaka JA, McInnis M. 2002. Offstream water and trace mineral salt as management strategies for improved cattle distribution. J Anim Sci. 80:346-356.

Prados LF, Valadares Filho SC, Santos SA, Zanetti D, Nunes AN, Costa DR, Mariz LDS, Detmann E, Amaral PM, Rodrigues FC, Valadares RFD. 2015. Reducing calcium and phosphorus in crossbred beef cattle diets: impacts on productive performance during the growing and finishing phase. Anim Prod Sci. 55:1369-1374.

Prados LF, Sathler DFT, Silva BC, Zanetti D, Valadares Filho SC, Alhadas HM, Detmann E, Santos SA, Mariz LDS, Chizzotti ML. 2017. Reducing mineral usage in feedlot diets for

Nellore cattle: II. Impacts of calcium, phosphorus, copper, manganese, and zinc contents on intake, performance, and liver and bone status. J Anim Sci. 95:1-11.

Rhoads AR, Stanton TL, Engle TE, Kimberling CV. 2003. Effects of concentration and source of trace minerals on performance, immunity, mineral and lipid metabolism, and carcass characteristics of beef steers. Profess Anim Sci. 19:150-158.

Rogers JA, Marks BC, Davis CL, Clark JH. 1979. Alteration of rumen fermentation in steers by increasing rumen fluid dilution rate with mineral salts. J Dairy Sci. 62:1599-1605.

Rotta PP, Valadares Filho SC, Engle TE, Costa e Silva LF, Sathler DFT, Prado IN, Bonafe EG, Zawadzki F, Visentainer JV. 2014. The impact of dietary sugarcane addition to finishing diets on performance, apparent digestibility, and fatty acid composition of Holstein x Zebu bulls. J Anim Sci. 92:2641-2653.

Sehested J. 2004. Feeding phosphorus (P) to cattle: A new basis for Nordic recommendations. Acta Agriculturae Scandinavica, Section A-Animal Science, 54:169-180.

Spears JW, Weiss WP. 2014. Invited Review: Mineral and vitamin nutrition in ruminants. Profess Anim Sci. 30:180-191.

Steen RWJ, Kilpatrick DJ. 2000. The effects of the ratio of grass silage to concentrates in the diet and restricted dry matter intake on the performance and carcass composition of beef cattle. Livest Prod Sci. 62:181-192.

Suttle NF. 2010. Mineral nutrition of livestock. Cabi. 4th ed. London, UK.

Turner BL, Rhoades RD, Tedeschi LO, Hanagriff RD, McCuistion KC, Dunn BH. 2013. Analyzing ranch profitability from varying cow sales and heifer replacement rates for beef cow-calf production using system dynamics. Agr Syst. 114:6-14.

Valadares Filho SC, Marcondes MI, Chizzotti ML, Paulino PVR. 2010. Nutrient requirements of Zebu beef cattle – BR-Corte. Suprema Gráfica e Editora, Visconde do Rio Branco, Brazil.

Valente EEL, Paulino MF, Detmann E, Valadares Filho SC, Barros LV, Acedo TS, Couto VRM, Lopes SA. 2011b. Levels of multiple supplements or nitrogen salt for beef heifers in pasture during the dry season. Braz J Anim Sci. 40:2011-2019.

Valente TNP, Detmann E, Queiroz ACD, Valadares Filho SC, Gomes DI, Figueiras JF. 2011a. Evaluation of ruminal degradation profiles of forages using bags made from different textiles. Braz J Anim Sci. 40:2565-2573.

Vasconcelos JT, Galyean ML. 2007. Nutritional recommendations of feedlot consulting nutritionists: The 2007 Texas Tech University survey. J Anim Sci. 85:2772-2781.

Ward JD, Spears JW. 1997. Long-term effects of consumption of low-copper diets with or without supplemental molybdenum on copper status, performance, and carcass characteristics of cattle. J Anim Sci. 75:3057-3065.

Ward JD, Spears JW, Gengelbach GP. 1995. Differences in Copper Status and Copper Metabolism Among Angus, Simmental, and Charolais Cattle. J Anim Sci. 73:571-577.

Williams SN, Lawrence LA, McDowell LR, Warnick AC, Wilkinson NS. 1990. Dietary Phosphorus Concentrations Related to Breaking Load and Chemical Bone Properties in Heifers. J Dairy Sci. 73:1100-1106.

Williams SN, Lawrence LA, McDowell LR, Wilkinson NS, Ferguson PW, Warnick AC. 1991. Criteria to evaluate bone mineralization in cattle: I. Effect of dietary phosphorus on chemical, physical, and mechanical properties. J Anim Sci. 69:1232-1242.

Zanetti D, Godoi LA, Estrada MM, Engle TE, Silva BC, Alhadas HM, Chizzotti ML, Prados LF, Renno LN, Valadares Filho SC. 2017a. Estimating mineral requirements of Nellore beef

bulls fed with or without inorganic mineral supplementation and the influence on mineral balance. J Anim Sci., 95:1-12.

Zanetti D, Valadares Filho SC, Prados LF, Detmann E, Pacheco MVC, Godoi LA, Renno LN, Engle TE. 2017b. Impacts of reduction of phosphorus in finishing diets for Holstein× Zebu steers. Livest Sci., 198:45-51.

Zinn R A, Barreras A, Owens FN, Plascencia A. 2008. Performance by feedlot steers and heifers: Daily gain, mature body weight, dry matter intake, and dietary energetics. J Anim Sci. 86:2680-2689.

Table 1 – Experimental and chemical composition of diets.

Item	With supplementation			Without supplementation		
	SH100[1]	SC100[2]	CS100[3]	SH0[4]	SC0[5]	CS0[6]
Sugarcane	40.00	40.00	-	40.00	40.00	-
Corn Silage	-	-	58.00	-	-	58.00
Concentrate	60.00	60.00	42.00	60.00	60.00	42.00
Ground corn	49.74	80.37	63.49	49.74	80.37	63.49
Soybean meal	8.46	12.67	29.87	8.46	12.67	29.87
Soybean hulls	35.00	-	-	35.00	-	-
Urea	2.45	2.45	1.89	2.45	2.45	1.89
Sodium bicarbonate	1.50	1.50	1.50	1.50	1.50	1.50
Salt	1.00	1.00	1.00	1.00	1.00	1.00
Magnesium oxide	0.50	0.50	0.50	0.50	0.50	0.50
Ammonium sulfate	0.31	0.31	0.21	0.31	0.31	0.21
Dicalcium phosphate	0.61	0.27	0.63	-	-	-
Limestone	0.39	0.89	0.85	-	-	-
Micromineral premix[7]	0.04	0.04	0.06	-	-	-
Sand	-	-	-	1.04	1.20	1.54
Chemical composition						
Dry matter, %	50.64	50.64	41.50	50.64	50.64	41.50
Organic matter, % DM	95.72	96.17	93.69	95.40	96.03	93.47
NDF, % DM	37.60	28.91	37.37	37.60	28.91	37.37
Crude protein, % DM	13.95	13.67	14.05	13.95	13.67	14.05
Ether extract, % DM	2.95	3.23	3.55	2.95	3.23	3.55
NFC, % DM	42.68	51.93	39.72	42.68	51.93	39.72
iNDF, % DM	11.62	11.15	12.19	11.62	11.15	12.19
Calcium, g/kg	3.72	3.40	3.38	2.02	1.13	1.49
Phosphorus, g/kg	2.05	2.03	2.35	1.54	1.80	1.98
Sodium, g/kg	5.61	5.47	4.02	5.60	5.46	4.00
Potassium, g/kg	6.48	4.51	10.37	6.47	4.50	10.36
Magnesium, g/kg	2.92	2.67	2.48	2.90	2.66	2.47
Sulfur, g/kg	1.14	1.06	0.83	1.05	0.97	0.74
Copper, ppm	22.06	21.34	23.27	5.24	4.67	5.70
Zinc, ppm	72.05	66.99	69.22	22.88	18.02	17.73
Manganese, ppm	60.64	57.33	51.63	36.23	35.03	27.22
Cobalt, ppm	0.50	0.48	0.57	0.42	0.39	0.48
Iron, ppm	335.38	224.24	541.99	275.03	197.80	498.12
Molybdenum, ppm	0.46	0.47	0.70	0.46	0.47	0.70

[1] Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM. [2] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [3] Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [4] Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM. [5] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [6] Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [7] Micromineral premix composed of zinc sulfate (56.8%), manganese sulfate (26.2%), copper sulfate (16.8%), and cobalt sulfate (0.2%).

Table 2 – Nutrient intake and digestibility of Nellore bulls fed with or without calcium (Ca), phosphorus (P) and microminerals (MM)

Item	With supplementation			Without supplementation			SEM	P-Value		
	SH100[1]	SC100[2]	CS100[3]	SH0[4]	SC0[5]	CS0[6]		Diet	Min	Diet × Min
Intake, kg/d										
Water	23.23	15.7	19.31	16.29	17.01	18.19	1.48	0.060	0.103	0.019
Dry matter	7.68	7.55	7.47	7.56	7.67	7.32	0.30	0.784	0.914	0.915
Organic matter	7.27	7.19	6.94	7.15	7.30	6.80	0.29	0.401	0.857	0.906
Neutral detergent fiber	2.88a	2.18b	2.79a	2.83a	2.21b	2.74a	0.11	<0.001	0.877	0.935
Crude protein	1.08	1.04	1.06	1.06	1.06	1.04	0.04	0.890	0.903	0.916
Digestibility, g/100g										
Dry Matter	73.65a	71.85a	68.57b	73.70a	72.78a	68.11b	0.83	<0.001	0.941	0.565
Organic matter	74.21a	72.43a	69.81b	74.87a	73.96a	70.06b	0.85	<0.001	0.360	0.632
Neutral detergent fiber	59.46a	52.57b	58.62a	60.85a	53.29b	58.06a	1.00	<0.001	0.499	0.672
Crude protein	73.56a	70.84b	68.19c	73.8a	71.62b	67.08c	0.88	<0.001	0.727	0.330

[1] Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM. [2] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [3] Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [4] Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM. [5] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [6] Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.

Table 3 – Performance, carcass characteristics, and efficiency parameters of Nellore bulls fed with or without calcium (Ca), phosphorus (P) and microminerals (MM)

Item	With supplementation			Without supplementation			SEM	P-Value		
	SH100[1]	SC100[2]	CS100[3]	SH0[4]	SC0[5]	CS0[6]		Diet	Min	Diet × Min
Performance										
Initial body weight, kg	272.86	278.57	275.71	260.71	270.86	275.00	12.15	0.721	0.526	0.871
Average daily gain, kg/d	1.17	1.11	1.11	1.11	1.16	1.07	0.07	0.744	0.784	0.736
Empty body weight gain, kg/d	1.14	1.07	1.13	1.12	1.08	1.05	0.06	0.738	0.710	0.767
Carcass characteristics										
Hot carcass yield, %	58.99	58.84	59.08	58.90	57.53	58.75	0.56	0.321	0.219	0.516
Cold carcass yield, %	57.90	57.58	57.89	57.61	56.37	57.57	0.55	0.296	0.189	0.645
Carcass length, cm	125.33	129.17	126.67	129.17	127.75	126.42	2.00	0.629	0.661	0.397
Rib eye area, cm^2	65.94	68.49	63.10	61.66	60.81	64.29	3.71	0.961	0.244	0.490
SFT[7], mm	3.70	4.06	4.80	3.71	3.51	3.78	0.59	0.567	0.285	0.685
Efficiency parameters										
ADG/DMI[8], kg/kg	0.1522	0.1473	0.1483	0.1470	0.1506	0.1474	0.0066	0.950	0.801	0.824
EBWG/DMI[9], kg/kg	0.1480	0.1424	0.1506	0.1481	0.1410	0.1448	0.0062	0.684	0.711	0.779

[1] Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM. [2] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [3] Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [4] Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM. [5] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [6] Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [7] Subcutaneous fat thickness. [8] Average daily gain in relation to dry matter intake. [9] Empty body weight gain in relation to dry matter intake.

Table 4 – Serum, rib bone and metatarsus parameters of Nellore bulls fed with or without calcium (Ca), phosphorus (P) and microminerals (MM)

Item	With supplementation			Without supplementation			SEM	P-Value		
	SH100[1]	SC100[2]	CS100[3]	SH0[4]	SC0[5]	CS0[6]		Diet	Min	Diet × Min
Serum level										
Calcium, mg/dl	9.64	10.33	10.14	10.53	10.00	9.94	0.14	0.742	0.291	<0.001
Phosphorus, mg/dl	7.57b	8.88a	9.01a	6.86b	8.96a	9.19a	0.27	<0.001	0.477	0.223
Parathyroid hormone, pg/ml	40.66	68.76	55.53	59.87	52.56	103.43	18.16	0.215	0.237	0.048
Bone alkaline phosphatase, U/l	140.30	135.32	145.79	142.89	154.80	164.39	12.54	0.650	0.255	0.798
Rib bone										
Ash, % DM	56.40	54.54	57.60	53.43	55.27	54.21	1.26	0.664	0.079	0.216
Calcium, % DM	10.17	9.54	11.34	9.93	11.19	10.07	0.41	0.302	0.896	0.005
Phosphorus, % DM	1.65	1.51	1.99	1.68	1.92	1.67	0.11	0.325	0.670	0.009
Section area, cm^2	3.85	3.90	3.46	3.49	3.69	3.71	0.22	0.638	0.547	0.370
Densitometry, kg/cm^2	0.89	0.84	0.88	0.69	0.74	0.72	0.04	0.976	<0.001	0.584
Breaking strength, kg_F	263.50	258.00	229.33	201.33	224.00	227.00	18.66	0.783	0.039	0.290
Breaking strength, kg_F/cm^2	68.86	66.12	66.40	56.95	61.17	60.95	3.45	0.968	0.013	0.537
Metatarsus										
Ash, % DM	58.63	56.66	58.31	56.40	55.84	57.45	0.98	0.233	0.113	0.716
Calcium, % DM	25.62	25.15	25.00	26.18	24.97	26.73	0.74	0.457	0.257	0.448
Phosphorus, % DM	1.16	1.08	0.92	0.96	1.00	1.16	0.07	0.948	0.804	0.120
Section area, cm^2	12.09	12.12	11.81	11.94	12.44	11.79	0.46	0.584	0.899	0.873
Densitometry, kg/cm^2	2.06	2.11	2.20	1.97	2.03	2.13	0.07	0.083	0.158	0.990
Breaking strength, kg_F	6550.00	5591.67	5808.33	5833.33	6116.67	6225.00	357.67	0.658	0.802	0.194
Breaking strength, kg_F/cm^2	548.54	464.01	491.58	487.64	494.74	529.39	29.40	0.399	0.917	0.210

[1] Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM. [2] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [3] Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [4] Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM. [5] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [6] Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.

Table 5 – Liver mineral concentrations of Nellore bulls fed with or without calcium (Ca), phosphorus (P) and microminerals (MM)

Item	With supplementation			Without supplementation			SEM	P-Value		
	SH100[1]	SC100[2]	CS100[3]	SH0[4]	SC0[5]	CS0[6]		Diet	Min	Diet × Min
Macro minerals, %										
Calcium	0.025	0.017	0.016	0.019	0.017	0.019	0.003	0.266	0.671	0.366
Phosphorus	0.893	0.891	0.928	0.905	0.952	0.902	0.050	0.907	0.706	0.707
Sodium	0.400	0.477	0.378	0.383	0.356	0.351	0.060	0.715	0.299	0.671
Potassium	1.329	1.463	1.344	1.305	1.263	1.174	0.140	0.764	0.264	0.803
Magnesium	0.074	0.064	0.074	0.068	0.078	0.075	0.010	0.459	0.225	0.014
Sulfur	0.040	0.029	0.029	0.040	0.030	0.052	0.010	0.695	0.525	0.660
Microminerals, mg/kg DM										
Copper	371.86	397.37	413.11	264.44	268.11	216.72	54.59	0.941	0.002	0.700
Zinc	202.83	161.42	157.39	150.86	172.21	139.83	14.36	0.155	0.105	0.108
Manganese	11.15	11.64	12.34	12.12	11.81	11.32	1.23	0.987	0.966	0.720
Cobalt	0.35	0.38	0.32	0.46	0.23	0.27	0.08	0.369	0.710	0.318
Iron	252.77	284.14	169.29	354.57	263.39	274.38	51.74	0.292	0.152	0.394
Molybdenum	4.45	3.80	4.20	5.37	3.91	3.90	0.62	0.220	0.639	0.624

[1] Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM. [2] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [3] Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM. [4] Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM. [5] Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM. [6] Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.

Chapter 3

Running Head: Dietary mineral for Nellore beef cattle

Estimating mineral requirements of Nellore beef bulls fed with or without inorganic mineral supplementation and the influence on mineral balance[1]

Diego Zanetti* [2], Letícia Artuzo Godoi*, Maurício Miguel Estrada*, Terry Eugene Engle†, Breno de Castro Silva*, Herlon Meneguelli Alhadas*, Mário Luiz Chizzotti*, Laura Franco Prados*, Luciana Navajas Rennó*, Sebastião de Campos Valadares Filho*

***** Department of Animal Science, Universidade Federal de Viçosa, PH Rolfs St, 36570-900, Minas Gerais, Brazil

† Department of Animal Science, Colorado State University, 350 W Pitkin St, 80523, Colorado, United States

[1] Financial support for this research was provided by National Council of Scientific and Technological Development (CNPq), National Institute of Science and Technology in Animal Science (INCT – Ciência Animal), and Foundation for Research Support of the State of Minas Gerais (FAPEMIG).

[2] **Corresponding author:** diego.zanetti@ufv.br ; diego.zanetti@colostate.edu. Diego Zanetti is also affiliate as visiting scholar at Department of Animal Science, Colorado State University, 350 W Pitkin St, 80523, Colorado, United States.

ABSTRACT

The objectives of this study were to quantify mineral balance of Nellore cattle fed with and without calcium (Ca), phosphorus (P), and micromineral (MM) supplementation and to estimate the net and dietary mineral requirement for *B. indicus* cattle. Nellore cattle (n = 51, initial body weight = 270.4 ± 36.6 kg, age = 8 months) were assigned to one of three groups: reference (n = 5), maintenance (n = 4), and performance (n = 42). The reference group was slaughtered prior to the experiment to estimate initial body composition. The maintenance group was used to collect values of animals at low gain and reduced mineral intake. The performance group was assigned to one of six treatments: Sugarcane as roughage source with a soybean meal-soybean hull concentrate mix with (SH100) and without (SH0) Ca, P, and MM supplementation, sugarcane as roughage source with a soybean meal-ground corn based concentrate mix with (SC100) and without (SC0) Ca, P, and MM supplementation, and corn silage as roughage source with a soybean meal-ground corn based concentrate mix with (CS100) and without (CS0) Ca, P, and MM supplementation. Orthogonal contrasts were adopted to compare mineral intake, fecal and urinary excretion and apparent retention among treatments. Maintenance requirements and true retention coefficients were generated with the aid of linear regression between mineral intake and mineral retention. Mineral composition of the body and gain requirements were assessed using non-linear regression between body mineral content and mineral intake. Mineral intake and fecal and urinary excretion were measured. Intakes of Ca, P, S, Cu, Zn, Mn, Co, and Fe were reduced in the absence of Ca, P, and MM supplementation ($P < 0.05$). Fecal excretion of Ca, Cu, Zn, Mn, and Co were also reduced in treatments without supplementation ($P < 0.01$). Overall, excretion and apparent absorption and retention coefficients were reduced when minerals were not supplied ($P < 0.05$). The use of the true retention coefficient instead of the true absorption coefficient provided a better estimate of mineral requirements. Dietary mineral requirements were lower for P, Cu, and Zn and greater for

Fe when compared to previously published recommendations. This study provides useful information about mineral requirements and mineral supplementation to obtain adequate dietary mineral supply of Nellore cattle in tropical conditions.

Key Words – calcium; copper; manganese, phosphorus; supplementation; zinc.

INTRODUCTION

Minerals supplied to cattle in feedlots in the United States are approximately three times the recommended NRC (1996) concentrations (Vasconcelos and Galyean, 2007). An accurate estimation of mineral requirements is therefore of economic and environmental importance (Jongbloed and Lenis, 1998; Humer and Zebeli, 2015; Spears and Weiss, 2014). For Brazil, Millen et al. (2009) have demonstrated that 83.1% of feedlot nutritionists supplement minerals to beef cattle.

Recent research has demonstrated that suppling calcium (Ca) and phosphorus (P) to Nellore (Costa e Silva et al., 2015a) and Holstein × Zebu crossbreed (Prados et al., 2015) is not necessary in feedlots. However, information about mineral requirements of *B. indicus* cattle is limited and the few existing studies did not include a negative control (Valadares Filho et al., 2010; Costa e Silva et al., 2015b).

We therefore hypothesized that Nellore beef cattle fed diets not containing supplemental Ca, P, and microminerals (MM) have a reduced mineral excretion and that the actual recommendations for dietary minerals are overestimated for *B. indicus*. The objectives of this study were to quantify mineral balance in Nellore cattle fed with and without Ca, P, and MM supplemental inorganic sources and to estimate net and dietary mineral requirements.

MATERIAL AND METHODS

The experiment was conducted in the experimental feedlot of the Animal Science Department of the Universidade Federal de Viçosa, Viçosa, MG, Brazil, following the recommendations of the Ethics Committee for Animal Use (protocol CEUAP/DZO/UFV 17/2015).

Animals, experimental design, and treatments

Fifty-one Nellore bulls with an average initial body weight of 270.4 ± 36.6 kg were used in this study. The animals were weaned at approximately 8 mo of age at the Beef Cattle Sector of the Animal Science Department of Universidade Federal de Viçosa and immediately transported to the experimental feedlot at the same location. After a 21-d period of acclimation to the feeding system and recovery from the stress of weaning, the animals were randomly divided into three groups: reference (n = 5), maintenance (n = 4), and performance (n = 42).

The reference group was slaughtered to determine initial empty body weight (EBW) as well as mineral contents of the carcass and organs. Animals from the maintenance group were housed in individual pens equipped with concrete feeders and daily fed at 1.1% of their body weight on a DM basis. The maintenance group was included to generate data in reduced gain and mineral intake levels. Different intake levels are necessary since linear and nonlinear regressions are the main tools for determination of nutritional requirements. The maintenance group was therefore not considered in the statistical analyses regarding intake, fecal and urinary excretion, and apparent retention.

The performance group was housed in a group pen (48.0 m^2) with electronic feeders (Model AF-1000 Master, Intergado Ltd., Contagem, Minas Gerais, Brazil) and waterers (Model WD-1000 Master, Intergado Ltd., Contagem, Minas Gerais, Brazil) for 125 d. Prior to the

experiment, each animal was fitted with an ear tag (left ear) containing a unique passive transponder (FDX – ISO 11784/11785; Allflex, Joinville, Santa Catarina, Brazil). Animals were weighed at 0800 h after a 16 h fasting period (feed only) every 28 d and at the end of the experiment. Average daily gain was calculated as difference between final and initial weight in relation to number of days on feed.

Diets were formulated according to the BR-Corte (Valadares Filho et al., 2010) for a gain of 1.1 kg per day and were isonitrogenous (13.5 % of crude protein). Sugarcane or corn silage was used as roughage source. The performance group was randomly subdivided into six treatments. Treatments consisted of three diets formulated with feeds commonly used in Brazilian feedlots, with or without supplemental Ca, P, and MM (Co, Cu, Mn, Zn sources). In the diets without mineral supplementation, the same amount of minerals was replaced by inert sand to avoid changing the other constituents of the diets. The proportions of the ingredients of the concentrates and diets as well as their chemical composition are shown in **Table 1**. The treatments were as follows: 1) Sugarcane as roughage source with a concentrate supplement composed of soybean meal and soybean hulls with (SH100) and without (SH0) Ca, P, and MM supplementation; 2) sugarcane as roughage source with a concentrate supplement composed of soybean meal and ground corn with (SC100) and without (SC0) Ca, P, and MM supplementation; and 3) corn silage as roughage source with a concentrate supplement composed of soybean meal and ground corn with (CS100) and without (CS0) Ca, P, and MM supplementation.

Intake

A total mixed ration was provided twice a day, at 0800 and 1600 h. Feed delivery was adjusted daily to maintain minimum refusals the next day and *ad libitum* intake. The appropriate amount for each group was based on refusal weight every morning. According to the amount of

refusals, TMR offered was reduced or increased to reach *ad libitum* intake. Each treatment was delivered in a group of electronic feeders and consequently provided unique access to certain animals.

The Intergado monitoring system was used to determine individual feed intake for the performance group. For each feed bunk visit, the system recorded the animal number, feed bunk number, and initial and final weight of the feeder. These data were continuously recorded and transferred via a network cable to the Intergado web software for data capture and storage (Chizzotti et al., 2015). The maintenance group was housed in individual pens, doted of an individual feeder. For the maintenance group, TMR was provided once a day at 0800 h. The amount of feed for each animal from this group was based on individual body weight (1.1 % on dry matter basis).

Mineral bioavailability

Water samples were collected on d 15, 65, and 115 from the waterers. All feeds were sampled daily. The samples were dried in a forced-ventilation oven (55°C), ground in a knife mill using a 2-mm screen, and packed in plastic bags for further laboratory analyses. For evaluation of mineral bioavailability, fecal grab samples (per rectum) were obtained over a 5-d period, from d 70 to 75. The collections were conducted at 0600h of the first day, at 0900h of the second day, at 1200h of the third day, at 1500h of the fourth day, and at 1800h on the fifth day. Feces were packed in aluminum trays, dried in a forced-ventilation oven at 55°C for 72 h, and ground in a knife mill as described previously. For each animal, a composite sample was made that was proportional to the amount of dry matter collected during each collection period.

Subsamples of feedstuffs and feces were analyzed for dry matter (DM; method 934.01), according to the AOAC (2012) methods of analysis protocols. The indigestible neutral detergent

fiber (iNDF) content was calculated after *in situ* incubation of the samples (ground to 2 mm using a knife mill), using F57 bags (Ankon®) for 288 h as described by Valente et al. (2011) for tropical forages. The iNDF was adopted as a marker to calculate total fecal excretion. Daily fecal excretion was estimated based on the relationship between iNDF intake and iNDF concentration of fecal samples.

For mineral analysis, samples were weighed into pre-weighed acid-washed crucibles, placed in an ash oven and combusted at 600°C for 12 h. Samples were then removed and placed in a desiccator for an additional 30 min to cool. Finally, samples were weighed and suspended with 5 mL of HCl (1.2 N HCl). Water samples were analyzed without preparation, following the procedures described above. Mineral concentrations of the samples were determined through inductively-coupled plasma optical emission spectroscopy methods (Optima 7300 DV, Perkin Elmer; Braselton et al., 1997). The minerals analyzed were calcium (Ca), phosphorus (P), sodium (Na), potassium (K), magnesium (Mg), sulfur (S), copper (Cu), zinc (Zn), manganese (Mn), cobalt (Co), iron (Fe), and molybdenum (Mo).

Urinary excretion

During fecal collection periods, spot urine samples were collected by spontaneous urination four hours after feeding. Creatinine concentrations were determined for each urine sample via an automatic biochemical analyzer (Mindray, model BS200E, Shenzhen, China). The daily urinary volume was estimated by dividing the daily creatinine excretion (CE) by the creatinine concentration in the urine. The CE (g/d) was estimated using the following equation described by Costa e Silva et al. (2013):

$$CE = 0.0345 \times BW^{0.9491}, \quad (1)$$

where the BW was expressed in kg. Two hundred milliliter of each urine sample were lyophilized and the mineral concentration was quantified as described for the feedstuffs and feces.

Mineral retention

The difference between intake and excretion through urine and feces for each mineral was considered as the mineral content retained in the body of the animal. The relationship between retention and intake of each mineral was expressed through a linear equation using the linear model:

$$RMi = \beta_0 + \beta_1 \times CMi, \quad (2)$$

where RMi = retained mineral "i" expressed as mg/EBW/d, CMi = consumed mineral "i" expressed as mg/EBW/d, β0 was considered the net requirement for maintenance of the mineral "i" in mg/EBW, and β1 is the true retention coefficient for the mineral "i" in percent.

Body composition

The performance and maintenance groups were slaughtered after 125 d on feed. Prior to slaughter, all animals were fasted (feed only) for 16 h. Animals were then slaughtered via captive bolt stunning followed by exsanguination. Post slaughter, the gastrointestinal tract of each animal was emptied, washed, and weighed. The weight of the gastrointestinal tract was added to the weights of all other organs, blood, and carcass for the determination of the EBW. Organs and viscera were ground in an industrial cutter for 20 min to obtain a homogeneous sample. The hide, head, and limbs of the carcasses were removed and weighed. The hide was manually chopped and sampled and the head and legs were ground in a bone grinder to obtain a homogeneous sample. The blood was quantified and subsampled. All samples were packed in trays for lyophilization.

After slaughter, the carcass of each animal was divided into two halves which were weighed and subsequently cooled in a cold chamber at 4°C for 18 h. After this period, the half-

carcasses were removed from the cold chamber for weighing and complete dissection of the left half of the carcass. Bones, muscles, and subcutaneous adipose tissue were ground separately, sampled, and lyophilized. All samples were ground in a Willey knife mill through a 2-mm screen. Two samples per animal were reconstituted and termed either "carcass" or "non-carcass" samples. The carcass sample consisted of the lyophilized samples of bone, meat, and fat, which were grouped proportionally based on the dissection weights. The sample of non-carcass components consisted of lyophilized samples of blood, head and limbs, organs and viscera, and hide, which were also grouped proportionally based on the dissection weights. Dry matter and mineral content were analyzed similarly to feedstuffs and feces.

Calculation of the mineral requirements for weight gain

The mineral content in the body, as a function of EBW, was estimated by using equations relating the body content of each mineral to the performance and reference animals, according to the following exponential model (ARC, 1980):

$$M_i = \beta_0 \times EBW^{\beta_1}, \quad (3)$$

where M_i = mineral "i" of the animal's body, EBW = empty body weight, and 'β_0' and 'β_1' = regression parameters.

From the regression parameters presented above, the net requirements for each mineral per kilogram of empty body gain were obtained by the derivative of the equation above, according to the model below (ARC, 1980):

$$Y = \beta_0 \times \beta_1 \times EBW^{\beta_1 - 1}, \quad (4)$$

where Y = the net requirement for each mineral for weight gain (g/kg of EBG), EBW = empty body weight (kg), and 'β_0' and 'β_1' = regression parameters. The dietary requirements for each

mineral corresponded to the sum of the net requirements for maintenance and weight gain divided by the true retention coefficient.

Statistical analyses

Comparisons between means were made through orthogonal contrasts using the significance level of $P < 0.05$ for all procedures. The contrasts evaluated were: A – sugarcane as roughage source and concentrate based on soybean meal and soybean hulls with (SH100) versus without (SH0) Ca, P, and MM supplementation; B – sugarcane as roughage source and concentrate based on soybean meal and ground corn with (SC100) versus without (SC0) Ca, P, and MM supplementation; C – Corn silage as roughage source and concentrate based on soybean meal and ground corn with (CS100) versus without (CS0) Ca, P, and MM supplementation; and D – treatments with (SH100, SC100, and CS100) versus without (SH0, SC0, and CS0) Ca, P, and MM supplementation. The linear models were built using the REG procedure of SAS (SAS Institute Inc., Cary, NC); for the non-linear models, the NLIN procedure of SAS was used.

RESULTS AND DISCUSSION

Mineral intake from water

Water composition is extremely variable according to the area. In some areas, mineral uptake from water is a nutritional problem, e. g. excessive S intake. In the present study, water only slightly contributed to the overall mineral intake; we therefore did not add the minerals consumed through water to the total mineral intake. Among all treatments, mean water intake was 18.29 ± 4.42 l/d. Considering the mineral content of the water, mineral intake needs to be added as follows (in mg/d): Ca = 111.33, Na = 97.06, K = 71.46, Mg = 46.97, S = 47.40, Fe = 0.05, Mn = 0.04, Mo = 0.14, and Zn = 0.05. Phosphorus content was lower than 1 mg/l; Cu and Co

concentrations were below 10 ppb and therefore not measured. These values were considered as a negligible contribution and therefore disregarded in the overall mineral intake. In addition to that, mineral intake from water is not commonly measured, and considering it negligible enabled us to compare our results with those of other studies.

Mineral balance

Body mineral balance is strongly controlled, initially by intake (Huntington et al., 1981; Robertson et al., 1996) and then by intestinal absorption (Field, 1981) and kidney reabsorption (Pickering, 1965). Once consumed, minerals are generally absorbed by either transcellular or paracellular mechanisms (Bronner, 2003; Powell et al., 1999). While the transcellular process is saturable, the paracellular mechanism is highly dependent on the soluble mineral concentration in the intestine. Therefore, in diets with decreased mineral contents, a smaller proportion of the soluble minerals would be absorbed through the paracellular process. However, the amount of reabsorbed minerals is proportional to maintain homeostasis. In the present experiment, as expected, intakes of Ca, P, S, Cu, Zn, Mn, Co, and Fe were reduced when Ca, P, and MM were not supplemented ($P < 0.05$, **Tables 2 and 3**). However, the fraction of fecal minerals in relation to intake minerals was greater in treatments without Ca, P, and MM supplementation. These findings could be related to fecal endogenous losses, which are generated by microbial debris, sloughed gastrointestinal epithelium, and digestive secretions (i.e. biliary secretion) not reabsorbed by the intestines (Lucas, 1960). Furthermore, endogenous losses can be influenced by dietary characteristics such as fiber and fat contents (Miller and Cragle, 1965). Urinary excretion of Na was decreased in animals receiving no supplemental Ca, P, or MM ($P = 0.019$). With regard to mineral absorption in relation to intake, absorptions of P, Cu, Zn, Mn, and Fe were reduced ($P < 0.05$), while Co absorption was improved ($P < 0.01$) without Ca, P, and MM supplementation.

Macromineral retention as a percentage of intake was not affected ($P > 0.05$). However, micromineral retention was reduced ($P < 0.05$) in treatments without Ca, P, or MM supplementation.

Calcium and phosphorus

Calcium and P intakes were greater ($P < 0.01$) in SH100, SC100, and CS100 than in SH0, SC0, and CS0, due to Ca and P supplementation. Fecal excretion showed a similar trend. Although previous studies have observed a linear relationship between Ca and P intake and fecal excretion (Dou et al., 2001; Prados et al., 2015; Robertson et al., 1996), this was not the case in the present study. This finding can be attributed to different concentrations and resulting relationships of Ca and P in the basal diet (maximum relationship Ca: P = 1.82 in SH100; minimum relationship Ca: P = 0.63 in SC0). Furthermore, Ca and P mainly get excreted via feces, as described by Morse et al. (1992). In the present experiment, due decreased dietary Ca and P levels, urinary excretion may be more controlled by hormone and metabolite concentrations (Horst et al., 1994; Goff, 2014).

Sodium, potassium, and magnesium

Intakes of Na, K, and Mg were not reduced in treatments without supplementation ($P > 0.05$). However, urinary Na and Mg were reduced ($P < 0.05$) in the absence of Ca, P, and MM supplementation and urinary K was reduced ($P = 0.04$) in the CS0 treatment when compared to the CS100 treatment. This reduction may be related to the impact of these ions on urinary osmolarity. Kume et al. (2008) reported that urinary osmolarity is maintained easily through reabsorption or excretion of ions (i.e. K and Mg) compared to using other compounds such as urea. This also applies to the present study, as reduced ion intake results in reduced ion excretion. In addition, interaction between minerals has been reported in the literature and a reduction of Na and Mg excretions via urine may be a result of interactions with other minerals. Engle et al. (1997)

have reported that dietary Zn levels influence urinary Na and K excretion; in particular, they observed increased urinary Na and urine volumes and a reduction of urinary K in animals with a marginal Zn deficiency. Although signs of Zn deficiency were not observed, decreased dietary Zn levels could possibly be the reason for the reduced urinary excretion of Na and Mg in treatments without Zn supplementation.

Copper, zinc, and manganese

Intakes and fecal excretion of Cu and Zn were reduced ($P < 0.01$) in the absence of Ca, P, and MM supplementation. Sources (organic and inorganic) and supplementation level of Cu and Zn are the principal reason for variation in use and excretion of these minerals (Chapman and Bell, 1963; Jongbloed and Lenis, 1998; Mandal et al., 2006). Urinary excretion was similar ($P > 0.05$) between all treatments, indicating that Cu and Zn urinary excretion is altered only when dietary Cu and Zn are in excess (Kim et al., 2009; Miller et al., 1966). Consequently, Cu and Zn absorption and retention were reduced ($P < 0.01$). Similar to Cu and Zn, Mn intake, fecal excretion, absorption, and retention were reduced ($P < 0.01$), although urinary excretion was not affected ($P > 0.05$) by supplementation. This could be explained by the fact that excess Mn is excreted via biliary excretion and poorly reabsorbed in the intestines (Symonds and Hall, 1983). Hidiroglu (1979) have reported decreased apparent Mn absorption (approximately 1%) than the ones found in the present study.

Cobalt

Intakes and fecal excretion of Co were reduced ($P < 0.01$) in the absence of Ca, P, and MM supplementation. However, Co urinary concentrations are regulated by other factors since urinary excretion was not affected ($P > 0.05$) by treatments. The Co apparent absorption coefficient and, consequently, the apparent retention coefficients were increased ($P < 0.01$) in the absence of

supplementation. In terms of mineral balance, Co was most balanced in all treatments. In rats, Taylor (1962) has described an increase in the absorption coefficient in diets with reduced Co. In the present study, Co was absorbed similarly (2.03 mg/d) in all treatments, suggesting that ruminal synthesis of cobalamin among treatments was similar. In addition, symptoms of cobalamin deficiency, such as lack of appetite, dysfunction of energy metabolism, and anemia, were not detected. Bile and pancreatic juice are important excretion routes (Greenberg et al., 1943; Comae and Davis, 1947). To achieve greater apparent absorption, the excretion of Co in the bile must have been reduced.

Sulfur, iron, and molybdenum

Intakes of S and Fe were indirectly reduced ($P < 0.01$) in the absence of Ca, P, and MM supplementation. Molybdenum intakes were similar ($P > 0.05$) between all treatments. Excretion, absorption, and retention of these minerals presented irregular behavior in different types of diets. However, when all treatments were compared, Fe and Mo absorption and retention were reduced ($P < 0.05$) in the absence of supplementation, while S absorption and retention were not influenced ($P > 0.05$) by dietary treatments. It should be noted that these minerals have numerous interactions with other elements (Suttle, 1975).

Mineral requirements

In Brazil, the mineral requirements stated by the NRC (1996) have been adopted for ration formulation (Millen et al., 2009, Oliveira and Millen 2014). However, data used to determine the mineral requirements of beef cattle by the NRC (2000, 2016) is primarily based on data reported for *Bos taurus* breeds of cattle. However, Brazilian beef cattle are primarily of the Nellore (*Bos indicus*) breed and may therefore have different mineral requirements. The BR-Corte reports nutritional requirements for Nellore beef cattle, but does not state micromineral requirements in

tropical conditions for Zebu-based breeds. Recently, Costa e Silva et al. (2015b) have published the results of the first study on micromineral requirements for Nellore cattle. In this study, the authors used data from three experiments: one using young Nellore bulls (n = 37, average BW = 259 kg, 1.23 kg/d), one using young heifers (n = 32, average BW = 180 kg, ADG = 0.35 kg/d), and one using young steers (n = 18, average BW = 150 kg, ADG = 0.35 kg/d), following a methodology similar to that used in the present experiment. The authors have reported different estimates for the net requirements for maintenance and growth and true retention coefficients for microminerals compared to those reported for *Bos taurus* breeds. Different diet types (with feedstuffs commonly adopted in Brazil) were used in this study, due to the knowledge of dietary impacts on nutritional requirements. True retention coefficients, maintenance, and gain requirements were calculated, considering all treatments and the animals fed at maintenance. The values and equations are presented in **Table 4**. Daily net mineral requirements in relationship to body weight and weight gain are shown in **Table 5**.

When compared with other mineral requirement estimates (**Table 6**), different outcomes were observed for each mineral. These differences between these findings and the results of the recent study of Costa e Silva et al. (2015b) could be attributed to lower BW and ADG levels of steers and heifers adopted in that report in comparison with this study. However, the results of this study are in agreement with the findings of Costa e Silva et al. (2015b) that the true retention coefficient of each mineral should be used, and not the true absorption coefficient, to convert the net requirements to dietary requirements. Data from this experiment show the importance of considering urinary excretion of Na, K, Mg, S, and Mo, due to the great differences between apparent retention and absorption.

Calcium dietary requirements were similar to those reported by the BR-Corte (2010), albeit lower than those in the NRC (2016) and greater than those observed by Costa e Silva et al (2015b). In terms of P, the measured requirement was approximately two times lower than the requirements reported in the BR-Corte (2010) and the NRC (2016). However, the relationship between Ca and P (3:1) is similar in all publications. Magnesium requirements were similar in all cases, while sodium requirements predicted in this study were greater than in all other studies. Potassium and S requirements were similar to those predicted by Costa e Silva et al (2015b) and approximately half of those predicted by BR-Corte (2010) and the NRC (2016). Copper and Zn requirements were lower than those given in other reports. Manganese requirements were similar to those predicted by the NRC (2016), but three times higher than those reported by Costa e Silva et al. (2015b). Cobalt and Fe requirements were three times the NRC (2016) recommendations, but 11 times lower for Co and similar for Fe when compared to the recommendations of Costa e Silva et al. (2015b). Molybdenum requirement was not stated in Costa e Silva et al. (2015b), but the value calculated in the present study was about 34 times lower than the NRC (2016) recommendations.

This substantial variation in determination of mineral requirements has been described previously by Coelho da Silva (1995). However, due to recent research and standardization of methods, the differences between the studies could be considerably reduced. The reason why mineral requirement estimations still diverge between studies could be: 1) the difficulty in measuring microelement concentrations in feeds and animal tissues; 2) the role of feed sorting by animals; 3) the varying availability of each mineral from different feedstuffs; and (4) the metabolic status of the individual animal as determined by age, breed, nutritional status, and physiologic status (CSIRO, 2007).

CONCLUSIONS

Mineral excretion, and apparent absorption and retention coefficients, are generally reduced in the absence of Ca, P, and MM supplementation for Nellore cattle in growing and finishing phases of production. The use of the true retention coefficient instead of the true absorption coefficient provides better estimates of mineral requirements, since the urinary excretion is considered here. The present study provides useful information for dietary mineral supplementation in countries where *Bos indicus* cattle are used for meat production.

LITERATURE CITED

AOAC, 2012. 19th ed. Official methods of analysis. Assoc. Off. Anal. Chem., Arlington, VA.

ARC. 1980. The nutrient requirements of ruminant livestock. The Gresham Press, London, UK.

Braselton, W. E., K. J. Stuart, T. P. Mullaney, and T. H. Herdt. 1997. Biopsy mineral analysis by inductively coupled plasma atomic emission spectroscopy with ultrasonic nebulization. J. Vet. Diagn. Invest. 9:395–400. doi: 10.1177/104063879700900409

Bronner, F. 2003. Mechanisms of intestinal calcium absorption. J. Cell. Biochem. 88:387-393. doi: 10.1002/jcb.10330

Chapman, H. L., and M. C. Bell. 1963. Relative absorption and excretion by beef cattle of copper from various sources. J. Anim. Sci. 22:82-85. doi:10.2134/jas1963.22182x

Chizzotti, M. L., F. S. Machado, E. E. L. Valente, L. G. R. Pereira, M. M. Campos, T. R. Tomich, S. G. Coelho, and M. N. Ribas.2015. Technical note: Validation of a system for monitoring individual feeding behavior and individual feed intake in dairy cattle. J. Dairy Sci. 98:3438-3442. doi:10.3168/jds.2014-8925

Coelho da Silva, J. F. 1995. Exigências de macroelementos inorgânicos para bovinos: O sistema ARC/AFRC e a experiência no Brasil. In: J. C. Pereira, Editor, International Symposium of Nutritional Requirements of Ruminants. Viçosa, Brazil.

Comae, C., and G. K. Davis. 1947. Cobalt metabolism studies. 3. Excretion and tissue distribution of radioactive cobalt administered to cattle. Arch. Biochem. Bioph. 12:257-266.

Costa e Silva, L. F., S. C. Valadares Filho, E. Detmann, P. P. Rotta, D. Zanetti, F. A. C. Villadiego, and R. M. G. Pereira. 2013. Performance, growth, and maturity of Nellore bulls. Trop. Anim. Health Prod. 45:795-803. doi: 10.1007/s11250-012-0291-1

Costa e Silva, L. F., S. C.Valadares Filho, T. E. Engle, P. P. Rotta, M. I. Marcondes, F. A. S. Silva, E. C. Martins, and A. T. Tokunaga. 2015a. Macro minerals and trace element requirements for beef cattle. PloS ONE. 10:1-20. doi: 10.1371/journal.pone.0144464

Costa e Silva, L. F., T. E. Engle, S. C. Valadares Filho, P. P. Rotta, R. F. D. Valadares, B. C. Silva, and M. V. C. Pacheco. 2015b. Intake, apparent digestibility, and nutrient requirements for growing Nellore heifers and steers fed two levels of calcium and phosphorus. Livest. Sci. 181:17-24. doi:10.1016/j.livsci.2015.09.024

CSIRO, 2007. Nutrient requirements of domesticated ruminants. Commonwealth Scientific and Industrial Research Organization Publications: Collingwood, AU.

Dou, Z., K. F. Knowlton, R. A. Kohn, Z. Wu, L. D. Satter, G. Zhang, J. D. Toth, and J. D. Ferguson. 2002. Phosphorus characteristics of dairy feces affected by diets. J. Environ. Qual. 31:2058-2065. doi:10.2134/jeq2002.2058

Engle, T. E., C. F. Nockels, C. V. Kimberling, D. L. Weaber, and A. B. Johnson. 1997. Zinc repletion with organic or inorganic forms of zinc and protein turnover in marginally zinc-deficient calves. J. Anim. Sci. 75:3074-3081. doi: 10.2527/1997.75113074x

Field, A. C. 1981. Some thoughts on dietary requirements of macro-elements for ruminants. Proceedings of the Nutrition Society. 40:267-272. doi: 10.1079/PNS19810042

Goff, J. P., 2000. Pathophysiology of calcium and phosphorus disorders. Veterinary Clinics of North America: Food Anim. Pract. 16:319-337. doi:10.1016/S0749-0720(15)30108-0

Greenberg, D. M., D. H. Copp, and E. M. Cuthbertson. 1943. Studies in mineral metabolism with the aid of artificial radioactive isotopes. 7. The distribution and execration, particularly by way of the bile, of iron, cobalt, and manganese. J. Biol. Chem. 147:749-756.

Hidiroglou, M. 1979. Manganese in ruminant nutrition. Can. J. Anim. Sci. 59:217-236. doi: 10.4141/cjas79-028

Horst, R. L., J. P. Goff, and T. A. Reinhardt. 1994. Calcium and vitamin D metabolism in the dairy cow. J. Dairy Sci. 77:1936-1951. doi:10.3168/jds.S0022-0302(94)77140-X

Humer, E., Q. and Zebeli. 2015. Phytate in feed ingredients and potentials for improving the utilization of phosphorus in ruminant nutrition. Anim. Feed Sci. Tech. 209:1-15. doi:10.1016/j.anifeedsci.2015.07.028

Huntington, G.B., R. A. Britton, and R. L. Prior. 1981. Feed intake, rumen fluid volume and turnover, nitrogen and mineral balance and acid-base status of wethers changed from low to high concentrate diets. J. Anim. Sci. 52:1376-1387. doi:10.2134/jas1981.5261376x

Jongbloed, A.W., N. P. and Lenis. 1998. Environmental concerns about animal manure. J. Anim. Sci. 76:2641-2648. doi: 10.2527/1998.76102641x

Kim, H., H. Y. Son, S. M. Bailey, and J. Lee. 2009. Deletion of hepatic Ctr1 reveals its function in copper acquisition and compensatory mechanisms for copper homeostasis. Am. J. Physiol. Gastrointest. Liver Physiol. 296:G356-G364. doi: 10.1152/ajpgi.90632.2008

Kume, S., K. Nonaka, T. Oshita, T. Kozakai, and H. Hirooka. 2008. Effects of urinary excretion of nitrogen, potassium and sodium on urine volume in dairy cows. Livest. Sci. 115:28-33. doi:10.1016/j.livsci.2007.06.003

Lucas, H. L. 1960. Relations between apparent digestibility and the composition of feed and feces: 1. A quantitative theory. North Caroline State College, Raleigh.

Mandal, G. P., R. S. Dass, D. P. Isore, A. K. Garg, and G. C. Ram. 2007. Effect of zinc supplementation from two sources on growth, nutrient utilization and immune response in

male crossbred cattle (Bos indicus× Bos taurus) bulls. Anim. Feed Sci. Tech. 138:1-12. doi:10.1016/j.anifeedsci.2006.09.014

Millen, D. D., R. D. L. Pacheco, M. D. B. Arrigoni, M. L. Galyean, and J. T. Vasconcelos. 2009. A snapshot of management practices and nutritional recommendations used by feedlot nutritionists in Brazil. J. Anim. Sci. 87:3427-3439. doi:10.2527/jas.2009-1880

Miller, J. K., and R. G. Cragle 1965. Gastrointestinal sites of absorption and endogenous secretion of zinc in dairy cattle. J. Dairy Sci. 48:370-373. doi:10.3168/jds.S0022-0302(65)88231-5

Miller, W. J., D. M. Blackmon, G. W. Powell, R. P. Gentry, and J. M. Hiers. 1966. Effects of zinc deficiency per se and of dietary zinc level on urinary and endogenous fecal excretion of 65Zn from a single intravenous dose by ruminants. J. Nutr. 90:335-341.

Morse, D., H. H. Head, C. J. Wilcox, H. H. Van Horn, C. D. Hissem, and B. Harris. 1992. Effects of concentration of dietary phosphorus on amount and route of excretion. J. Dairy Sci. 75:3039-3049. doi:10.3168/jds.S0022-0302(92)78067-9

NRC. 1996. Nutrient requirements of beef cattle. 7th rev. ed. Natl. Acad. Press, Washington, D.C.

NRC. 2016. Nutrient requirements of beef cattle. 8th rev. ed. Natl. Acad. Press, Washington, D.C.

Oliveira, C. A., and D. D. Millen. 2014. Survey of the nutritional recommendations and management practices adopted by feedlot cattle nutritionists in Brazil. Anim. Feed Sci. Tech. 197:64-75. doi:10.1016/j.anifeedsci.2014.08.010

Pickering, E. C. 1965. The role of the kidney in sodium and potassium balance in the cow. Proceedings of the Nutrition Society. 24:73-80. doi: 10.1079/PNS19650013

Powell, J. J., R. Jugdaohsingh, and R. P. H. Thompson. 1999. The regulation of mineral absorption in the gastrointestinal tract. Proceedings of the Nutrition Society. 58:147-153. doi: 10.1079/PNS19990020

Prados, L. F., S. C.Valadares Filho, S. A. Santos, D. Zanetti, A. N. Nunes, D. R. Costa, L. D. S. Mariz, E. Detmann, P. M. Amaral, F. C. Rodrigues, and R. F. D. Valadares. 2015. Reducing calcium and phosphorus in crossbred beef cattle diets: impacts on productive performance during the growing and finishing phase. Anim. Prod. Sci. 55:1369-1374. doi: 10.1071/AN14781

Robertson, B. M., T. Magner, A. Dougan, M. A. Holmes, and R. A. Hunter. 1996. The effect of coal mine pit water on the productivity of cattle. I. Mineral intake, retention and excretion and the water balance in growing steers. Crop Past. Sci. 47:961-974. doi:10.1071/AR9960961

Spears, J. W., and W. P. Weiss. 2014. Invited Review: Mineral and vitamin nutrition in ruminants. Profess Anim. Sci. 30:180-191. doi: 10.15232/S1080-7446(15)30103-0

Suttle, N. F. 1975. The role of organic sulphur in the copper-molybdenum-S interrelationship in ruminant nutrition. Brit. J. Nutr. 34:411-420. doi: 10.1079/BJN19750009

Symonds, H. W., and E. D. Hall. 1983. Acute manganese toxicity and the absorption and biliary excretion of manganese in cattle. Res. Vet. Sci. 35:5-13.

Taylor, D. M. 1962. The absorption of cobalt from the gastro-intestinal tract of the rat. Phys. Med. Biol. 6:445-451.

Valadares Filho S. C., M. I. Marcondes, M. L. Chizzotti, and P. V. R. Paulino. 2010. Nutrient requirements of Zebu beef cattle – BR-Corte. Suprema Gráfica e Editora, Visconde do Rio Branco, Brazil.

Valente, T. N. P., E. Detmann, A. C. D. Queiroz, S. C. Valadares Filho, D. I. Gomes, and J. F. Figueiras. 2011b. Evaluation of ruminal degradation profiles of forages using bags made

from different textiles. Braz. J. Anim. Sci. 40:2565-2573. doi:10.1590/S1516-35982011001100039

Vasconcelos, J. T., and M. L. Galyean. 2007. Nutritional recommendations of feedlot consulting nutritionists: The 2007 Texas Tech University survey. J. Anim. Sci. 85:2772-2781. doi:10.2527/jas.2007-0261

Table 1 – Experimental and chemical composition of diets (DM basis)

Item	Treatments					
	SH100[1]	**SH0[2]**	**SC100[3]**	**SC0[4]**	**CS100[5]**	**CS0[6]**
Sugarcane, dietary %	40	40	40	40	-	-
Corn Silage, dietary %	-	-	-	-	58	58
Concentrate, dietary %	60	60	60	60	42	42
Ground corn, % of the concentrate	49.74	49.74	80.37	80.37	63.49	63.49
Soybean meal, % of the concentrate	8.46	8.46	12.67	12.67	29.87	29.87
Soybean hulls, % of the concentrate	35.00	35.00	-	-	-	-
Urea, % of the concentrate	2.45	2.45	2.45	2.45	1.89	1.89
Sodium bicarbonate, % of the concentrate	1.50	1.50	1.50	1.50	1.50	1.50
Salt, % of the concentrate	1.00	1.00	1.00	1.00	1.00	1.00
Magnesium oxide, % of the concentrate	0.50	0.50	0.50	0.50	0.50	0.50
Ammonium sulfate, % of the concentrate	0.31	0.31	0.31	0.31	0.21	0.21
Dicalcium phosphate, % of the concentrate	0.61	-	0.27	-	0.63	-
Limestone, % of the concentrate	0.39	-	0.89	-	0.85	-
Micromineral premix[7], % of the concentrate	0.04	-	0.04	-	0.06	-
Sand, % of the concentrate	-	1.04	-	1.20	-	1.54
Chemical composition						
Dry matter, %	50.64	50.64	50.64	50.64	41.50	41.50
Organic matter, %	95.72	95.40	96.17	96.03	93.69	93.47
Neutral detergent fiber, %	37.60	37.60	28.91	28.91	37.37	37.37
Crude protein, %	13.95	13.95	13.67	13.67	14.05	14.05
Ether extract, %	2.95	2.95	3.23	3.23	3.55	3.55
Non fibrous carbohydrate, %	42.68	42.68	51.93	51.93	39.72	39.72
Indigestible neutral detergent fiber, %	11.62	11.62	11.15	11.15	12.19	12.19
Calcium, g/kg	3.72	2.02	3.40	1.13	3.38	1.49
Phosphorus, g/kg	2.05	1.54	2.03	1.80	2.35	1.98
Sodium, g/kg	5.61	5.60	5.47	5.46	4.02	4.00
Potassium, g/kg	6.48	6.47	4.51	4.50	10.37	10.36
Magnesium, g/kg	2.92	2.90	2.67	2.66	2.48	2.47
Sulfur, g/kg	1.14	1.05	1.06	0.97	0.83	0.74
Copper, ppm	22.06	5.24	21.34	4.67	23.27	5.70
Zinc, ppm	72.05	22.88	66.99	18.02	69.22	17.73
Manganese, ppm	60.64	36.23	57.33	35.03	51.63	27.22
Cobalt, ppm	0.50	0.42	0.48	0.39	0.57	0.48
Iron, ppm	335.38	275.03	224.24	197.80	541.99	498.12
Molybdenum, ppm	0.46	0.46	0.47	0.47	0.70	0.70

[1] Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM.

[2]Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM.
[3]Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM.
[4]Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.
[5]Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM.
[6]Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.
[7]Micromineral premix composed of zinc sulfate (56.8%), manganese sulfate (26.2%), copper sulfate (16.8%), and cobalt sulfate (0.2%).

Table 2 – Macromineral intake, and excretion of Nellore bulls fed with or without calcium (Ca), phosphorus (P), and microminerals (MM)

Item	Treatments						SEM	Contrasts			
	SH100[1]	SH0[2]	SC100[3]	SC0[4]	CS100[5]	CS0[6]		A[7]	B[8]	C[9]	D[10]
Calcium											
Intake, g/d	28.61	15.24	25.64	8.66	25.20	10.91	0.75	0.0001	0.0001	0.0001	0.0001
Fecal excretion, g/d	13.94	11.35	14.36	3.20	14.12	5.94	1.16	0.1237	0.0001	0.0001	0.0001
Urine excretion, g/d	1.00	0.48	0.59	0.65	0.32	0.37	0.22	0.1077	0.8500	0.8415	0.4658
Absorption, % intake	51.43	26.06	43.89	63.25	43.42	45.86	6.68	0.0108	0.0477	0.7913	0.8268
Retention, % intake	47.89	22.81	41.61	55.77	42.17	42.47	6.79	0.0131	0.1493	0.9744	0.5235
Phosphorus											
Intake, g/d	15.75	11.65	15.29	13.82	17.58	14.51	0.59	0.0001	0.0895	0.0006	0.0001
Fecal excretion, g/d	6.48	4.63	5.92	5.37	6.49	7.25	0.32	0.0006	0.2723	0.1206	0.0605
Urine excretion, g/d	0.26	0.16	0.21	0.18	1.32	0.58	0.20	0.7510	0.9372	0.0128	0.0956
Absorption, % intake	58.97	60.28	61.17	61.06	62.62	49.95	2.03	0.6506	0.9685	0.0001	0.0255
Retention, % intake	57.31	58.87	59.83	59.69	55.57	45.97	1.79	0.5414	0.9569	0.0004	0.0684
Sodium											
Intake, g/d	43.13	42.31	41.31	41.85	29.99	29.27	1.50	0.7038	0.8029	0.7290	0.7856
Fecal excretion, g/d	13.65	12.63	13.77	12.97	7.85	10.92	1.24	0.5667	0.6543	0.0798	0.6791
Urine excretion, g/d	18.73	13.99	11.98	10.24	14.08	9.00	1.95	0.0946	0.5334	0.0658	0.0197
Absorption, % intake	68.54	70.26	66.60	69.06	73.69	62.56	3.07	0.6949	0.5746	0.0119	0.3569
Retention, % intake	25.00	36.66	37.38	44.27	26.42	31.51	6.07	0.1827	0.4274	0.5442	0.1167
Potassium											
Intake, g/d	49.80	48.89	34.04	34.49	77.46	75.79	2.45	0.7941	0.8989	0.6223	0.7214
Fecal excretion, g/d	13.62	10.54	10.73	16.32	21.18	27.17	2.06	0.2977	0.0632	0.0407	0.0976
Urine excretion, g/d	23.71	24.02	20.85	19.68	45.93	34.13	4.27	0.9589	0.8540	0.0414	0.2366
Absorption, % intake	72.66	78.77	68.49	53.25	73.34	64.13	3.21	0.1870	0.0019	0.0433	0.0240
Retention, % intake	24.79	29.57	16.17	4.07	14.15	19.49	9.10	0.7122	0.3723	0.6706	0.9302
Magnesium											

	Intake, g/d	22.41	21.91	20.18	20.38	18.55	18.05	0.81	0.6659	0.8629	0.6572	0.6877
	Fecal excretion, g/d	10.09	9.01	9.61	8.66	9.70	11.22	0.61	0.2191	0.2820	0.0796	0.7297
	Urine excretion, g/d	5.84	3.06	3.37	4.05	3.19	2.81	0.44	0.0001	0.2821	0.5449	0.0286
	Absorption, % intake	54.89	59.09	52.37	57.46	47.42	37.52	2.76	0.2908	0.2018	0.0107	0.8929
	Retention, % intake	28.71	44.84	35.70	37.61	30.06	21.96	3.07	0.0007	0.6630	0.0622	0.1910
Sulfur												
	Intake, g/d	8.78	7.97	8.00	7.46	6.20	5.42	0.28	0.0510	0.1856	0.0549	0.0039
	Fecal excretion, g/d	1.79	1.53	1.65	1.34	1.61	1.68	0.14	0.2106	0.1356	0.7178	0.1617
	Urine excretion, g/d	2.60	3.20	2.14	3.22	3.27	2.50	0.38	0.2781	0.0546	0.1533	0.3349
	Absorption, % intake	79.57	80.96	79.46	81.99	73.96	69.13	1.66	0.5607	0.2908	0.0413	0.8219
	Retention, % intake	49.88	40.86	52.83	39.31	22.17	23.58	4.90	0.2009	0.0587	0.8343	0.0835

[1]Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM.
[2]Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM.
[3]Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM.
[4]Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.
[5]Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM.
[6]Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.
[7]A = SH100 versus SH0.
[8]B = SC100 versus SC0.
[9]C = CS100 versus CS0.
[10]D = (SH100, SC100 and CS100) versus (SH0, SC0 and CS0).

Table 3 – Micromineral intake, excretion, and apparent absorption and retention of Nellore bulls fed with or without calcium (Ca), phosphorus (P), and microminerals (MM)

Item	Treatments						SEM	Contrasts			
	SH100[1]	SH0[2]	SC100[3]	SC0[4]	CS100[5]	CS0[6]		A[7]	B[8]	C[9]	D[10]
Copper											
Intake, mg/d	169.47	39.62	161.15	35.80	173.81	41.68	4.43	0.0001	0.0001	0.0001	0.0001
Fecal excretion, mg/d	65.67	25.51	79.97	18.50	38.48	25.32	3.53	0.0001	0.0001	0.0099	0.0001
Urine excretion, mg/d	9.21	11.86	8.25	8.59	16.29	11.29	1.83	0.3111	0.8974	0.0530	0.6524
Absorption, % intake	61.32	23.23	50.22	48.65	77.59	38.64	5.57	0.0001	0.8428	0.0001	0.0001
Retention, % intake	55.87	5.68	45.00	24.44	68.11	11.00	6.81	0.0001	0.0395	0.0001	0.0001
Zinc											
Intake, mg/d	553.56	172.96	505.77	138.24	516.95	129.74	13.76	0.0001	0.0001	0.0001	0.0001
Fecal excretion, mg/d	166.15	94.25	168.86	50.67	65.51	38.78	8.53	0.0001	0.0001	0.0279	0.0001
Urine excretion, mg/d	12.47	13.85	7.90	9.56	16.94	10.80	2.69	0.7196	0.6661	0.1044	0.6372
Absorption, % intake	70.08	45.70	66.55	63.61	87.17	69.98	3.12	0.0001	0.5103	0.0003	0.0001
Retention, % intake	67.85	37.49	64.93	56.79	83.89	61.62	3.88	0.0001	0.1461	0.0002	0.0001
Manganese											
Intake, mg/d	465.93	273.81	432.8	268.65	385.61	199.12	12.88	0.0001	0.0001	0.0001	0.0001
Fecal excretion, mg/d	320.59	231.89	341.17	207.86	257.02	190.53	14.85	0.0001	0.0001	0.0023	0.0001
Urine excretion, mg/d	0.99	1.00	0.56	0.39	1.19	0.95	0.18	0.9903	0.5105	0.3330	0.3560
Absorption, % intake	31.35	15.61	21.09	22.67	33.09	3.71	3.95	0.0078	0.7800	0.0001	0.0001
Retention, % intake	31.14	15.23	20.96	22.52	32.77	3.24	3.96	0.0073	0.7826	0.0001	0.0001
Cobalt											
Intake, mg/d	3.83	3.16	3.65	3.02	4.25	3.51	0.15	0.0023	0.0037	0.0007	0.0001
Fecal excretion, mg/d	1.98	0.86	1.71	0.90	2.15	1.62	0.10	0.0001	0.0001	0.0009	0.0001
Urine excretion, mg/d	0.11	0.11	0.06	0.04	0.13	0.10	0.02	0.9387	0.5921	0.3470	0.3746
Absorption, % intake	48.33	72.73	53.34	70.14	49.26	53.46	2.52	0.0001	0.0001	0.2318	0.0001
Retention, % intake	45.36	69.18	51.63	68.57	46.15	50.50	2.61	0.0001	0.0001	0.2322	0.0001
Iron											
Intake, mg/d	2576.86	2078.70	1692.94	1517.10	4047.72	3644.38	120.20	0.0058	0.3076	0.0191	0.0007

Fecal excretion, mg/d	1739.19	1525.22	1364.6	1451.4	2452.18	2924.77	138.59	0.2820	0.6604	0.0174	0.3105
Urine excretion, mg/d	83.24	98.33	47.58	31.92	123.32	90.34	18.68	0.5712	0.5570	0.2055	0.4637
Absorption, % intake	32.60	26.08	19.53	4.19	39.39	19.34	4.67	0.3306	0.0260	0.0034	0.0007
Retention, % intake	29.36	21.2	16.68	1.99	36.39	16.96	4.72	0.2334	0.0340	0.0047	0.0007
Molybdenum											
Intake, mg/d	3.53	3.47	3.55	3.61	5.23	5.13	0.18	0.8189	0.8225	0.6620	0.8027
Fecal excretion, mg/d	2.47	2.38	2.66	2.66	2.85	3.78	0.18	0.7582	0.9997	0.0010	0.0728
Urine excretion, mg/d	0.41	0.21	0.35	0.60	0.40	0.44	0.07	0.0871	0.0273	0.7226	0.6075
Absorption, % intake	30.18	31.44	25.34	25.98	45.18	25.92	3.85	0.8181	0.9066	0.0008	0.0715
Retention, % intake	18.46	25.19	15.41	9.19	37.67	17.42	3.55	0.1888	0.2237	0.0002	0.0277

[1]Diet composed of sugarcane and concentrate based in soybean hulls, with supplementation of Ca, P and MM.
[2]Diet composed of sugarcane and concentrate based in soybean hulls, without supplementation of Ca, P and MM.
[3]Diet composed of sugarcane and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM.
[4]Diet composed of sugarcane and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.
[5]Diet composed of corn silage and concentrate based in soybean meal and ground corn, with supplementation of Ca, P and MM.
[6]Diet composed of corn silage and concentrate based in soybean meal and ground corn, without supplementation of Ca, P and MM.
[7]A = SH100 versus SH0.
[8]B = SC100 versus SC0.
[9]C = CS100 versus CS0.
[10]D = (SH100, SC100 and CS100) versus (SH0, SC0 and CS0).

Table 4 – Mineral endogenous fraction, true retention coefficients, body content, and requirements for weight gain of Nellore beef cattle

Mineral	**Endogenous fraction** mg/kgBW (macro) µg/kgBW (micro)	**True retention coefficient** %	**Body composition predict** g (macro) and mg (micro)	**Requirement predict for gain** g (macro) and mg (micro)
Calcium	4.11	48.4	$4.9014\times EBW^{1.1033}$	$5.4077\times EBW^{0.1033}$
Phosphorus	8.05	74.4	$2.6436\times EBW^{1.0084}$	$2.6658\times EBW^{0.0084}$
Sodium	11.97	44.9	$1.0766\times EBW^{1.0421}$	$1.1219\times EBW^{0.0421}$
Potassium	13.61	26.4	$2.1245\times EBW^{0.9281}$	$1.9717\times EBW^{-0.0719}$
Magnesium	8.18	48.1	$0.1382\times EBW^{1.1504}$	$0.1590\times EBW^{0.1504}$
Sulfur	8.57	80.7	$0.0626\times EBW^{1.1760}$	$0.0736\times EBW^{0.1760}$
Copper	67.20	70.9	$32.9001\times EBW^{0.5849}$	$19.2433\times EBW^{-0.4151}$
Zinc	107.40	78.9	$9.9706\times EBW^{1.0928}$	$10.8959\times EBW^{0.0928}$
Manganese	176.02	41.8	$0.00861\times EBW^{1.6264}$	$0.0140\times EBW^{0.6264}$
Cobalt	1.99	33.1	$0.0461\times EBW^{0.9770}$	$0.0450\times EBW^{-0.0230}$
Iron	1606.19	44.1	$8.4020\times EBW^{1.2382}$	$10.4034\times EBW^{0.2382}$
Molybdenum	2.70	45.1	$0.00251\times EBW^{1.4063}$	$0.00353\times EBW^{0.4063}$

Table 5 – Daily net mineral requirements of Nellore beef cattle considering variable BW and ADG = 1.0 kg

Body weight, kg	**Ca**	**P**	**Na**	**K**	**Mg**	**S**	**Cu**	**Zn**	**Mn**	**Co**	**Fe**	**Mo**
	---------- g/d ----------						---------- mg/d ----------					
200	10.2	4.4	3.8	4.1	2.0	1.9	15.6	39.3	35.6	0.4	358.0	0.6
300	11.0	5.2	5.0	5.4	2.8	2.8	22.0	50.7	53.3	0.6	522.3	0.8
400	11.7	6.0	6.2	6.7	3.7	3.6	28.5	62.0	71.0	0.8	685.8	1.1
500	12.3	6.8	7.4	8.1	4.5	4.5	35.1	73.1	88.7	1.0	848.8	1.4

Table 6 – Comparison of dietary mineral requirements predicted according to this study, Costa e Silva et al. (2015), BR-Corte (2010), and beef cattle NRC (2016), considering BW = 400 kg and ADG = 1.0 kg

Council/ study	**Ca**	**P**	**Na**	**K**	**Mg**	**S**	**Cu**	**Zn**	**Mn**	**Co**	**Fe**	**Mo**
	---------- g/d ----------						---------- mg/d ----------					
This study	24.2	8.1	13.9	25.5	7.6	4.5	40.2	78.6	169.9	2.5	1556.3	2.5
Costa e Silva et al. (2015)	11.1	17.3	7.1	20.6	7.3	5.6	77.2	334.5	54.7	27.3	1593.9	-
Beef catle BR-Corte (2010)	27.6	17.9	4.2	43.8	9.0	8.4	-	-	-	-	-	-
Beef cattle NRC (2016)	33.6	16.3	6.8	50.6	8.4	12.7	84.4	253.2	168.8	0.8	422.0	-

Chapter 4

Prediction of water intake by beef cattle raised in tropical and temperate climates

Short title: Prediction of water intake by beef cattle

D. ZANETTI[1]*, M. KISTNER[2], L. F. PRADOS[1], L. F. COSTA E SILVA[1],

E. DETMANN[1], F. A. S. SILVA[1], T. E. ENGLE[2], S. C. VALADARES FILHO[1]

[1] *Department of Animal Science, Universidade Federal de Viçosa, PH Rolfs St, 36570-900, Minas Gerais, Brazil*

[2] *Department of Animal Science, Colorado State University, 350 W Pitkin St, 80523, Colorado, United States*

* To whom all correspondence should be addressed. Email: diego.zanetti@ufv.br ; diegozanetti_@hotmail.com

Prediction of water intake by beef cattle raised in tropical and temperate climates

SUMMARY

Water is the most important nutrient in animal nutrition, and its sustainable use is relevant in animal production. The objective of this study was to validate six current water intake (WI) equations for beef cattle using water intake data from four experiments conducted in North America (n = 1 experiment; crossbred Angus beef steers) and Brazil (n = 3 experiments; Nellore beef cattle). Animal performance, diet composition, and environmental data were collected for all experiments. The prediction of WI using the current published WI equations was tested though the regression between predicted and measured WI values. All tested equations differed from the measured WI data from the four experiments. Several factors can help explain why the published equations did not predict the WI obtained from the three experiments, including that the tested equations were developed in temperate climates using predominantly *Bos taurus taurus*. From the current data, was generated new WI equations based on metabolic BW, DMI, humidity and temperature-humidity index for Nellore cattle in Brazil and metabolic BW, DMI, maximum daily temperature, and dietary concentrate level for Angus crossbred cattle in North America.

INTRODUCTION

Beef production systems require a considerable amount of water: Mekonnen & Hoekstra (2012) reported that beef cattle production systems require more water than swine or poultry production systems. Moreover, Beckett & Oltjen (1993) reported that 3,682 L of water are required to produce one kg of boneless beef, considering drinking water, water in feedstuffs, water used to produce feedstuffs, and water used in carcass processing. However, in some parts of the world,

water shortages have impacted livestock production (Ward & Michelsen 2002; Pluske & Schlink 2007).

Research aiming to increase the efficiency of water use for livestock production has been encouraged (Tilman *et al.* 2002; Rijsberman 2006); however, recent research evaluating the amount of drinking water used by beef cattle remains limited (Brew *et al.* 2011). Furthermore, several factors that affect water intake (WI) are known to exist in beef cattle, such as climatic variables, type of diet, breed, body weight, and physiological status. The most recent report about WI by beef cattle is by Sexson et al. (2012), who studied this in temperate environmental conditions. Brazil has the largest commercial cattle herd (FAS/USDA 2017), and the environmental conditions and beef cattle breeds are distinct from those described in the literature. Moreover, there is no equation to predict the water demand of beef cattle for Brazilian breeds. Based on constant changes in animal body, dietary type and environmental characteristics in beef cattle production systems, being able to accurately predict WI under different conditions is greatly important. It could help to sustainable usage of water in beef cattle production systems.

Therefore, was hypothesized that the current equations to predict WI are not applicable to beef cattle raised in North America and Brazil. Thus, the aim of this study was to evaluate the accuracy of six current water intake equations for beef cattle, and suggest new equations if the current do not predict accurately water intake.

MATERIAL AND METHODS

Water intake data collected from tropical conditions in Brazil and data collected from temperate conditions in Colorado, USA, were used in this study. This study comprised three experiments conducted in the Experimental Feedlot of the Animal Science Department at the

Universidade Federal de Viçosa, Viçosa, MG, Brazil; and one experiment conducted at the Agricultural Research Development and Education Center at the Colorado State University (ARDEC/CSU), Fort Collins, CO, USA; all experiments followed the recommendations of the Ethics Committee for Animal Use and Care from the respective institution.

Brazilian trials

Nellore is the main breed in the Brazilian beef herd. Therefore, all three experiments conducted in Brazil utilized Nellore beef cattle. One experiment utilized heifers and two experiments utilized bulls. In the Nellore bull experiments, 74 young Nellore bulls were divided into two experiments conducted in two consecutive years: 2013 and 2014. In experiment 1 (from July to December 2013; Prados *et al.* 2017), 32 bulls (274 ± 34 kg, 8 months) were randomly divided into four groups of eight animals. Each group was fed sugarcane-based diets, with 60% of concentrate on a dry matter (DM) basis. The treatments were based on the supplementation of inorganic sources of calcium (Ca) and phosphorus (P), and microminerals (MM) further to the basal diet. In experiment 2 (from July to November 2014; Zanetti *et al.* 2017), 42 Nellore bulls (270 ± 36 kg, 8 months) were randomly assigned to the following treatments: sugarcane as a forage source and a soybean meal and soybean hull-based concentrate with (SH100) and without (SH0) Ca, P and MM supplementation; sugarcane as the forage source and a soybean meal and ground corn-based concentrate with (SC100) and without (SC0) Ca, P and MM supplementation; and corn silage as the forage source with a soybean meal and corn-based concentrate with (CS100) and without (CS0) Ca, P and MM supplementation. In the Nellore heifer trial, 16 Nellore heifers (377 ± 49 kg, 16 months) were randomly divided into four groups of four animals. Each group was fed sugarcane or elephant grass for 77 days (from February to April 2015); without concentrate or with

40% concentrate on (DM) basis. Thus, the experiment was conducted as a completely randomized design in a 2 × 2 factorial arrangement (concentrate level [0% or 40% DM basis], and forage type [sugarcane or elephant grass]). Diets were formulated according to the BR-CORTE (Valadares Filho *et al.* 2010) for an ADG of 1.2, and 1.1 respectively for experiments 1 and 2, and all diets contained 13% CP on a DM basis.

For the measurements of feed and water intake, the animals were housed in a group pen that contained electronic feeders (Model AF-1000 Master, Intergado Ltd., Contagem, Minas Gerais, Brazil) and waterers (Model WD-1000 Master, Intergado Ltd., Contagem, Minas Gerais, Brazil). Prior to the beginning of the experiments, each animal was fitted with an ear tag containing a unique passive transponder (FDX – ISO 11784/11785; Allflex, Joinville, Santa Catarina, Brazil) in the left ear. The Intergado monitoring system was used to evaluate individual feed and water intake. For each visit to the feed bunk or waterer, the system recorded the animal ID, initial and final times of each feeding or drinking water event, and the weight of the amount of feedstuffs or water consumed. These data were continuously recorded and transferred via a network cable to the Intergado web software for data capture and storage (Chizzotti *et al.* 2015).

All feeds were sampled daily. The samples were oven-dried at 55°C for 72 hours, ground in a knife mill through a 1 mm screen, and were analyzed for DM content (method 934.01; AOAC 2012). Environmental variables were collected in an automatic weather station (model MAWS301, Brand Vaisala) of National Institute of Meteorology (INMET) installed 700 m far from the experimental feedlot (latitude: -20.7626^{o}, longitude: -42.8640^{o}, and elevation from sea: 698 m).

Colorado trials

Thirty steers (466 ± 89 kg, 14 months) were ranked by body weight and divided into 2 weight block replicates. Each successive weight block replicate was labeled as replicate 1 or 2, where the heaviest group of 15 steers was considered as replicate 1 and the lighter group of 15 steers was considered as replicate 2. Replicate 1 began the experiment on February and ended in July 2015. Replicate 2 began the experiment in July and ended in November 2015. In both replicates, steers were stratified by body weight (BW) and breed to one of five water treatments, so that BW and breed were equally represented within each treatment. Steers were then sorted into their respective individual pens and the experiment was initiated. Water treatments consisted of: 1) 0.0, 2) 160 3) 320 4) 480, and 5) 960 μg of supplemental Mo/L added as Na_2MoO_4 (Acros Organics, Geel, Belgium; purity: 99%) to the water. The sodium contribution from Na_2MoO_4 was balanced across water treatments by using NaCl. Each pen was 2.5 x 20.0 m and was equipped with an individual water tank, individual feed bunk, and a 3.0 m concrete bunk apron. Each animal had access to an individual 265 L Rubbermaid® structural foam stock tank (102.9 × 61 × 81 cm, length × height × width). Water intake was monitored daily at 0800 h ± 30 min by measuring the disappearance of water over a 24 h period. Since the tank was not symmetrical, the water volume for every 0.25cm on a plastic meter stick was correlated with the amount of water remaining in the tank. This calibration was accomplished by metering (TM Series Water Meter, Great Plains Industries, Inc. Wichita, KS; accuracy ± 3.0%) 0.25 cm of water into each tank, recording the volume (L) of water metered and then by weighing the amount of water as a secondary validation of water volume. Water tank calibrations were conducted approximately every two months. To account for evaporation, a separate water tank was placed in front of an empty pen and measured daily. Daily water disappearance was determined using the following equation: WD = [V1 – (V2 + evap.)] where: WD = water disappearance (assumed to be water intake), L/d; V1 = the previous

day's water volume; V2 = the current day's volume; and evap. = the amount of water disappearance due to evaporation. As an internal check, a line was placed around the inside of all tanks that corresponded to the tank containing 265 L of water.

Steers were fed a corn silage-based growing diet for 28 d and then transitioned to a high concentrate finishing diet. Diet changes during the step-up program were simultaneous for all treatments. Steers reached the finishing diet by day 47 of the experiment. Rations were formulated to meet National Research Council (NRC 2000) requirements for growing-finishing beef cattle. The basal growing, step-up, and finishing diets contained 5.4, 4.1, and 3.7 mg Cu/kg DM, respectively. Supplemental Cu as $CuSO_4 \cdot 5H_20$ was added to the supplement to supply a total mixed ration that contained NRC (2000) recommended concentrations of Cu (10.0 mg Cu/kg DM) and S (0.15%). Monensin feeding was initiated on day 1 and Tylan® was introduced into the step-1 diet. Monensin was given in feed at 28.0, 36.5, and 44.4 g/metric ton, on a dry matter (DM) basis in the growing, step-1, and finisher diets, respectively. Tylan® was fed at 90 mg $head^{-1}$ day^{-1} beginning with the step-1 diet. Optaflexx was fed to all treatments in the final 28 d of the finishing period at 27.3 g/metric ton (DM basis), providing approximately 300 mg $head^{-1}$ day^{-1}. Cattle feed bunks were observed each morning to determine the daily total feed delivery. Cattle were fed in amounts that allowed ad libitum access to feed throughout the day. Feed was delivered to pens once daily. All feeds were sampled daily. The samples were oven-dried (55°C), ground in a knife mill using a 1-mm screen and then analyzed for DM content (method 934.01; AOAC, 2012). Environmental variables were collected from the Colorado Climate Center (available at <http://ccc.atmos.colostate.edu/datarequest.php>) and National Oceanic and Atmospheric Administration (available at <http://www.ncdc.noaa.gov/data-access>). The environmental stations were located 16 and 11.2 km from ARDEC, respectively (latitude: 40.5734º, longitude: -

105.0865º, and elevation from sea: 1,525 m; and latitude: 40.5756º, longitude: -105.0236º, and elevation from sea: 1,525 m, respectively).

Current water intake predictions

In the literature, six equations or recommendations were found for WI prediction in beef cattle. The NRC (2000) recommended the equation proposed by Hicks *et al.* (1988) for WI prediction, as follows:

$$WI = -18.67 + 0.3937 \times T_{MAX} + 2.432 \times DMI + 3.87 \times PP - 4.437 \times DS$$

where WI is the water intake in L/d; T_{MAX} is the maximum temperature in ºF; DMI is the DM intake in kg/d; PP is the water precipitation in mm; and DS is the dietary salt in %.

Meyer *et al.* (2006) reported WI prediction in moderate climates based on the following model:

$$WI = -3.85 + 0.507 \times T_{AVG} + 1.494 \times DMI - 0.141 \times DF + 0.248 \times DMF + 0.014 \times BW$$

where WI is the water intake in L/d; T_{AVG} is the average temperature in ºC; DMI is the DM intake in kg/d; DF is the dietary forage content in %; DMF is the percentage of DM of forage; and BW is the body weight of the animal in kg.

CSIRO (2007) recommended different WI prediction equations for *Bos taurus indicus* and *Bos taurus taurus*, according to the product of DMI and a value fixed for WI, based on the average temperature. Estimates of water intake for cattle at lower than 15, equal to 20, 25, and 30, and greater than 35ºC, expressed as L/kg of DM intake, are 3.5, 4.0, 5.5, 7.5 and 10.0 for *Bos taurus taurus* and 3.0, 3.5, 4.5, 6.0 and 8.0 for *Bos taurus indicus,* respectively.

Arias & Mader (2011) recommended two equations (AM[1] and AM[2]), as follows:

$$WI_{AM[1]} = 5.92 + 1.03 \times DMI + 0.04 \times SR + 0.45 \times T_{MIN}$$

$$WI_{AM[2]} = -7.31 + 1.00 \times DMI + 0.04 \times SR + 0.30 \times THI$$

where $WI_{AM[1]}$ and $WI_{AM[2]}$ are the water intakes in L/d, predicted by equations 1 and 2, respectively; DMI is the DM intake in kg/d; SR is the solar radiation in W m^{-2}; T_{MIN} is the minimum temperature in ºC; and THI is the temperature humidity index, calculated according to the following equation $THI = 46.4 + 0.8 \times T_{AVG} + {HU \times (T_{AVG} - 14.4)}/{100}$ where HU is the percentage of relative humidity and T_{AVG} is the average temperature.

Sexson *et al.* (2012) proposed an equation to predict WI based on 14 parameters, as follows:

$$\begin{aligned} WI = {} & -14.07 + 0.163 \times HU - 0.0017 \times HU^2 + 0.0096 \times {T_{MAX}}^2 - 0.00032 \times {HU_{MAX}}^2 \\ & + 1.01 \times T_{MIN} - 0.026 \times {T_{MIN}}^2 + 0.038 \times HU_{MIN} + 0.396 \times SLP - 0.055 \times WS \\ & - 1.395 \times BW - 0.249 * SLP_{MAX} - 0.411 \times SLP_{MIN} + 0.0012 \times {T_{MAX.PREV}}^2 \\ & + 8.79 \times MBW \end{aligned}$$

where WI is the water intake in L/d; HU is average relative humidity in %; T_{MAX} is the maximum temperature in ºC; HU_{MAX} is the maximum relative humidity in %; T_{MIN} is the minimum temperature in ºC; HU_{MIN} is the minimum relative humidity in %, SLP is the average sea level pressure in mmHg, WS is the wind speed in km/h; BW is the body weight in kg; SLP_{MAX} is the maximum sea level pressure in mmHg; SLP_{MIN} is the minimum sea level pressure in mmHg; $T_{MAX.PREV}$ is the maximum temperature on the previous day in ºC; MBW is the metabolic body weight in kg $BW^{0.75}$.

Water intakes, estimated by the equations proposed in the literature, were compared to the actual WI values using the following regression model: $Y = \beta 0 + \beta 1 \times x$, where "Y" was the observed value; β0 and β1 were the intercept and slope of regression, respectively; and "X" was the predicted value. The regression was evaluated using the simultaneous hypothesis test (Mayer

et al. 1994): H0: $\beta 0 = 0$ and $\beta 1 = 1$ and Ha: not H0. If the null hypothesis was not rejected, was concluded that the tested equation accurately estimated water intake. Moreover, the coefficient of determination (r^2), concordance correlation coefficient (CCC), bias correction (Cb), and mean square error of prediction (RMSEP) were also computed to assess precision and accuracy of WI prediction equations. These analyses were performed using the MES (Model Evaluation System Software, version 3.1.16).

Predictor variables and proposed equations

All variables included in the current equations were measured to evaluate the equations. Also, other variables were measured in relation to water intake. In total, sixteen variables were measured and divided into two groups: (1) dietary and animal characteristics: DM content of forage (%, DMF), dietary forage content (%, DF), dietary neutral detergent fiber content (%, NDF), dietary salt (sodium chloride) content (%, DS), body weight (kg, BW), as fed intake (kg/d, FI), and DM intake (kg/d, DMI); and (2) environmental characteristics: average temperature (ºC, T_{AVG}), maximum temperature (ºC, T_{MAX}), minimum temperature (ºC, T_{MIN}), relative humidity (%, HU), rain (mm, Ra), snow (cm, Sn), temperature humidity index (THI), wind speed (m s^{-1}, WS), and solar radiation (W m^{-2}, SR).

Data from 80% of animals of each study was randomly selected to develop new predictor equations. The database from these trials conducted in temperate and tropical conditions were used separately due to the substantial differences among animals and environmental characteristics in each location. In sequence, multiple regression analyses were performed using stepwise regression in SAS (SAS Institute Inc., Cary, NC). The final number of parameters included in each model was determined based on partial r^2 values of each variable (Mader *et al.* 2006). If the inclusion of

a variable in the model produced an increase greater than 0.01 in total R^2, this variable was included in the model. The intercept was excluded from the equation if it was not significant. The equations without intercept were based on the same variables adopted in equations with a significant intercept and submitted again to stepwise selection. However, the partial R-squares were not adopted to exclude variables in the proposed equations without an intercept. To evaluate the proposed equations, data from 20% of the remaining animals were used in a similar procedure as described for the evaluation of the current equations in the literature.

RESULTS

Due to the previously found difference in water intake between *Bos taurus indicus* and *Bos taurus taurus* cattle, data for the Nellore cattle and Angus crossbred steers were evaluated separately. It is important to accurately predict different WI levels to estimate water demand for different seasons and feedlot phases. The patterns of predicted WI for each breed by these models were tested for different levels of WI (**Table 1, Figures 1 and 2**). Current equations for WI predictions do not adequately predict water intake.

The characteristics of beef cattle production are presented in **Tables 2 and 3**, showing the differences between production systems. Among all measured variables, those related to BW, DMI, temperature, and humidity were selected to predict WI for Nellore and Angus crossbred cattle (**Table 4**). Both suggested equations were validated (**Table 5, Figure 3**).

DISCUSSION

Evaluation of current equations to predict water intake

Some studies (Ittner *et al.* 1951; Winchester and Morris 1956; Valente *et al.* 2015) have reported greater WI for *Bos taurus* cattle than for *Bos indicus* cattle raised in similar conditions.

However, the experiments used in these comparisons were conducted in distinct environmental and handling conditions, using the predominant breed raised in each condition. In general, WI was overestimated by the previously published WI equations when compared to the actual WI observed in Nellore cattle raised in tropical conditions and Angus crossbred steers raised in temperate conditions. An exception was observed for the equation proposed by Sexson *et al.* (2012), which underestimated WI for Angus crossbred steers. However, these equations were developed for temperate conditions, reflecting greater differences between predicted and observed values for Nellore cattle than for Angus crossbred steers. In tropical conditions, the equation adopted by Sexson *et al.* (2012) overestimated WI by 165.2% for Nellore cattle, while in temperate conditions, the equation suggested by CSIRO (2007) overestimated WI by 39.6%. However, WI in temperate conditions as predicted by Sexson *et al.* (2012) was 19.6% lower than the observed WI values. Differences in the experimental factors between the experiments utilized in this study and the studies adopted by Sexson *et al.* (2012) could help to explain the differences in WI. Initially, these authors reported a contribution of Brahman, a *Bos taurus indicus* cattle, in the genetic composition of the animals used in the experiments adopted to develop this prediction equation.

Regarding dietary parameters, the moisture of diets used in the Colorado trials was lower than the dietary moisture utilized by Sexson *et al.* (2012). A greater dietary moisture concentration could lead to a reduction in water intake (Sun *et al.* 2014). Sexson *et al.* (2010) reported a significant influence of water quality (i.e. sulfate concentrations) on WI. In comparison with the water characteristics reported by Sexson *et al.* (2010) included in the database of Sexson *et al.* (2012), the water analyzed in the Colorado trials was of higher quality and consequently, there was a greater WI.

However, it is possible identify possible problems in these equations: the current equations (except for the CSIRO equation) presented intercepts that were not related to biological interpretations for WI. Indeed, few questions could be explained: for example, the equation proposed by Hicks *et al.* (1988) shows that DS has a negative coefficient in relation to WI; however, for dairy cattle, water intake is positively related with dietary sodium (Murphy *et al.* 1983), dietary potassium (Fraley *et al.* 2015), and dietary ash (Appuhamy *et al.* 2014). Apparently, these relationships are correlated with increases in osmotic pressure in the rumen fluid by DS (Rogers *et al.* 1979). Similarly, the addition of total dissolved salts in water increases water intake by cattle (Alves *et al.* 2017).

However, all equations adopted DMI or BW as a predictor of WI, corroborating published reports (Bond *et al.* 1976; Kramer *et al.* 2009). Moreover, all equations have in common the adoption of temperature, or parameters derived from temperature, as a WI predictor. The relationship between temperature and WI is based on the function of water for body heat homeostasis (Cunningham *et al.* 1964; Purwanto *et al.* 1996; Bewley *et al.* 2008).

Characteristics of tropical and temperate systems

Water pH and electrical conductivity were measured. While the values of pH ranged from 7.25 to 7.47 in North America, the pH ranged from 6.25 to 7.06 in Brazil. Regarding electrical conductivity, the values ranged from 150.3 to 155.6 and from 54.3 to 84.8 µS/cm in North America and Brazil, respectively. This suggests a greater ion concentration was present in water used in the North America experiment.

Initially, was observed a large difference in the sizes of the animals: the maximum BW observed for Nellore cattle was 44.2 kg less than the average of Angus crossbred steers. Regarding

dietary differences, the water content in Brazilian diets was greater than in North-American diets, and this is related to the Brazilian diets containing a greater proportion of ingredients that contain high moisture. It was also observed differences between the two systems for feed intake in as fed and dry matter basis. Water intake was also numerically greater for Angus crossbred steers than Nellore cattle. In general, the Brazilian diets presented lower starch and higher fiber contents than US feedlot diets.

In relation to environmental parameters, the prominent differences between temperate and tropical conditions were observed. In temperate conditions, greater thermal amplitude and negative temperatures were observed, with temperatures varying from -13.3 to 31.9ºC. However, in tropical conditions, temperatures varied from 8.4 to 34.6ºC. Another important parameter to describe the differences in environmental conditions was rainfall. In tropical conditions, high amounts of rain were observed. The maximum daily amount of rain was 0.5 mm of rain for North America cattle, while 27.2 mm of rain in tropical conditions was observed. Furthermore, humidity reached 95% for Brazilian cattle while relative humidity reached up to 76.8% in temperate conditions. Together, these two factors can influence animal behavior, and feed and water intake (Ali *et al.* 1994; Valente *et al.* 2015).

Prediction of water intake

The effects of the selected variables in the changes of WI have been well described in the literature. In addition, these variables are related to body temperature control mechanisms. The relationship between body thermal control and water intake is attributed to a temporary reduction of rumen temperature immediately after WI (Blackshaw & Blackshaw 1994; Bewley *et al.* 2008), and the direct effect of evaporative cooling in the reduction of the thermal load, disturbing body

water metabolism (Bernabucci *et al.* 2010; Arias & Mader 2011). After WI, a reduction in heat stress, respiration rate, and skin and rectal temperatures have been reported (Purwanto *et al.* 1996).

In both equations, DMI was the predictor with the greatest contribution to increasing WI, which agrees with the equations proposed by Arias & Mader (2011) and Meyer *et al.* (2006). The reduction of DMI is followed by a reduction in WI (Bond *et al.* 1976; Cardot *et al.* 2008; Kramer *et al.* 2009). This behavior could be explained in two ways: by the ruminal liquid dilution rate (Adams *et al.* 1981; Adams & Kartchner 1984), and by the caloric increment caused by heat from fermentation and digestion (Finch 1986). The rumen works as a fermentation chamber, and it depends on regular entrance of feed and the transit of digesta and microbes to the omasum. The digesta consists of insoluble particles and soluble fractions. However, liquids pass into the omasum two to four times faster than insoluble particles (Russell & Hespell 1981). An increase in the liquid dilution rate is generally followed by an increase in bacterial growth and the flow of nutrients through the gastrointestinal tract. Nevertheless, microbial metabolic activity dissipates free energy as heat during fermentation in the digestive tract, mainly in the rumen, as a result of inefficiencies in this system (Czerkaski 1980; Russell 1981). Bergen & Yokoyama (1977) reported that the loss of fermentation as heat varies from 3 to 12% of gross energy from the feed in ruminants, versus around 1% for non-ruminants.

Metabolic body weight was included in both equations as DMI. Initially, body weight was related to WI due to the relationship between BW and DMI. Thereby, a larger animal consumes more DM, and a greater DM intake implies a greater WI, as previously explained. Furthermore, basal metabolism is nearly proportional to animal body weight (Kleiber 1932), and the basal metabolism generates around 33% of heat in an animal (Finch 1986). In consequence, a larger animal generates more heat from basal metabolism than a smaller animal.

Also, within a given breed, a large animal tends to be fatter than a smaller animal. Body fat acts as insulation and makes it more difficult for a fatter animal to dissipate heat (Finch 1986; Gaughan *et al.* 2010). Moreover, peripheral vasodilatation helps to dissipate heat from the core region of the body. However, vasodilatation becomes more difficult with greater BW (Finch 1986; Silanikove 2000). Finally, sweat is an important pathway for heat loss in cattle (Ferguson & Dowling 1955). The rate of heat loss via sweating depends on the skin surface area for water evaporation (Hansen 2004). The relationship between surface area and BW decreases as the animal grows.

Among the environmental variables, humidity and THI were included in the equation for Nellore cattle, while the maximum temperature was included in the equation for Angus crossbred steers. Berman (2003) reported that heat exchange between the environment and the animal is the main component for body temperature maintenance. These variables have strong interactions with the previous variables described (Bernabucci *et al.* 2010).

The HU was reported by Blackshaw & Blackshaw (1994) as the major contributor to heat stress in hot climates. At a high HU, water vapor is trapped in the air space between hair and reduces water evaporation, where water diffusion depends on a water pressure gradient (Allen *et al.* 1970; Renaudeau *et al.* 2012). In consequence, in an analysis of HU separate from other aspects, in greater HU conditions, water loss via evaporation is reduced and therefore WI is reduced. For animals in pasture, Sun *et al.* (2014) reported an additional factor related to HU: higher HU caused a lower WI. Regarding temperature, non-evaporative mechanisms of heat loss decline with the increase of environmental temperature, and the animal is dependent on evaporative mechanisms (Silanikove 2000).

The influence of DMI on WI was similar for Nellore and Angus crossbred cattle. The coefficients were 0.91 and 0.87 L of water for each additional kg of DM consumed, for Nellore and Angus, respectively. However, the lower amount of forage in the diets commonly provided to Angus cattle could have increased DMI (Detmann *et al.* 2014). In addition, the greater forage content in the diet for Nellore cattle has a greater caloric increment from fiber fermentation (Fuquay 1981), justifying the greater coefficient.

However, the effect of BW on WI included in both equations indicates a greater WI for Angus crossbred steers than Nellore cattle, at a similar BW. For example, when removing the other variables, a 450 kg Nellore cattle would need 11.7 L of water per day, while a 450-kg Angus crossbred steer would need 20.0 L. This finding indicates a greater WI for *Bos taurus* than *Bos indicus* (Bianca 1965; Brew *et al.* 2011). Several factors may help to explain the greater WI for *Bos taurus*, and these are mainly related to body thermal homeostasis. Hansen (2004) reported a greater level of acclimation to heat stress for *Bos indicus*, which is probably related to the distinct climates where these species developed. This indicates that during genetic adaptations, *Bos indicus* have acquired thermoregulatory genes, reducing the dependence on mechanisms involving water to alleviate heat stress. Additionally, the darker color of the hide in Angus crossbred steers compared to Nellore cattle helps to explain this greater WI. Riemerschmid (1943) reported that dark hides can absorb approximately 80% of solar radiation, while this value for light hides is 50%. This author reported that heat from solar radiation could be three times more than heat produced by basal metabolism for animals with dark hides. The differences in environmental parameters of the locations can also be perceived in the equations.

In conclusion, current published equations do not accurately predict the water intake of Nellore cattle raised in tropical conditions and Angus crossbred steers raised in North America. In

these situations, the equations proposed here are recommended, which are based on metabolic body weight, dry matter intake, relative humidity, and temperature and humidity indexes for Nellore cattle raised in Brazil and metabolic body weight, dry matter intake, dietary concentrate, and maximum daily temperature for Angus crossbred steers raised in North America.

REFERENCES

ADAMS, D. C. & KARTCHNER, R. J. (1984). Effect of level of forage intake on rumen ammonia, pH, liquid volume and liquid dilution rate in beef cattle. *Journal of Animal Science* **58**, 708-713.

ADAMS, D. C., GALYEAN, M. L., KIESLING, H. E., WALLACE, J. D. & FINKNER, M. D. (1981). Influence of viable yeast culture, sodium bicarbonate and monensin on liquid dilution rate, rumen fermentation and feedlot performance of growing steers and digestibility in lambs. *Journal of Animal Science* **53**, 780-789.

ALI, S., GOONEWARDENE, L. A. & BASARAB, J. A. (1994). Estimating water consumption and factors affecting intake in grazing cattle. Can. *Journal of Animal Science* **74**, 551-554.

ALLEN, T. E., BENNETT, J. W., DONEGAN, S. M. & HUTCHINSON, J. C. D. (1970). Moisture, its accumulation and site of evaporation in the coats of sweating cattle. *The Journal of Agricultural Science* **74**, 247-258.

ALVES, J., ARAÚJO, G., NETO, S., VOLTOLINI, T., SANTOS, R., ROSA, P., GUAN, L., MCALLISTER, T. & NEVES, A. (2017). Effect of increasing concentrations of total dissolved salts in drinking water on digestion, performance and water balance in heifers. *The Journal of Agricultural Science*, 1-10. doi: 10.1017/S0021859617000120

AOAC. 2012. *Official Methods of Analysis*. 19th edn. Arlington, VA: Association of Official Analytical Chemists.

APPUHAMY, J. A. D. R. N., WAGNER-RIDDLE, C., CASPER, D. P., FRANCE, J. & KEBREAB, E. (2014). Quantifying body water kinetics and fecal and urinary water output from lactating Holstein dairy cows. *Journal of Dairy Science* **97**, 6177-6195.

ARAÚJO, G. G. L. D., VOLTOLINI, T. V., CHIZZOTTI, M. L., TURCO, S. H. N. & CARVALHO, F. F. R. D. (2010). Water and small ruminant production. *Brazilian Journal of Animal Science* **39**, 326-336.

ARIAS, R. A. & MADER, T. L. (2011). Environmental factors affecting daily water intake on cattle finished in feedlots. *Journal of Animal Science* **89**, 245-251.

BECKETT, J. L. & OLTJEN, J. W. (1993). Estimation of the water requirement for beef production in the United States. *Journal of Animal Science* **71**, 818-826.

BERGEN, W. G. & YOKOYAMA, M. T. (1977). Productive limits to rumen fermentation. *Journal of Animal Science* **45**, 573-584.

BERMAN, A. (2003). Effects of body surface area estimates on predicted energy requirements and heat stress. *Journal of Dairy Science* **86**, 3605-3610.

BERNABUCCI, U., LACETERA, N., BAUMGARD, L. H., RHOADS, R. P., RONCHI, B. & NARDONE, A. (2010). Metabolic and hormonal acclimation to heat stress in domesticated ruminants. *Animal* **4**, 1167-1183.

BEWLEY, J. M., GROTT, M. W., EINSTEIN, M. E. & SCHUTZ, M. M. (2008). Impact of intake water temperatures on reticular temperatures of lactating dairy cows. *Journal of Dairy Science* **91**, 3880-3887.

BIANCA, W. (1965). Sweating in dehydrated steers. *Research in Veterinary Science*. **6**, 33-37.

BLACKSHAW, J. K. & BLACKSHAW, A. W. (1994). Heat stress in cattle and the effect of shade on production and behaviour: a review. *Animal Production Science* **34**, 285-295.

BOND, J., RUMSEY, T. S. & WEINLAND, B. T. (1976). Effect of deprivation and reintroduction of feed and water on the feed and water intake behavior of beef cattle. *Journal of Animal Science* **43**, 873-878.

BREW, M. N., MYER, R. O., HERSOM, M. J., CARTER, J. N., ELZO, M. A., HANSEN, G. R. & RILEY, D. G. (2011). Water intake and factors affecting water intake of growing beef cattle. *Livestock Science* **140**, 297-300.

CARDOT, V., LE ROUX, Y. & JURJANZ, S. (2008). Drinking behavior of lactating dairy cows and prediction of their water intake. *Journal of Dairy Science* **91**, 2257-2264.

CHIZZOTTI, M. L., MACHADO, F. S., VALENTE, E. E. L., PEREIRA, L. G. R., CAMPOS, M. M., TOMICH, T. R., COELHO, S. G. & RIBAS, M. N. (2015). Technical note: Validation of a system for monitoring individual feeding behavior and individual feed intake in dairy cattle. *Journal of Dairy Science* **98**, 3438-3442.

CSIRO. (2007). *Nutrient requirements of domesticated ruminants*. Camberra, AU: CSIRO Publishing.

CUNNINGHAM, M. D., MARTZ, F. A. & MERILAN, C. P. (1964). Effect of drinking-water temperature upon ruminant digestion, intraruminal temperature, and water consumption of non-lactating dairy cows. *Journal of Dairy Science* **47**, 382-385.

CZERKAWSKI, J. W. (1980). A novel estimate of the magnitude of heat produced in the rumen. *British Journal of Nutrition* **43**, 239-243.

DETMANN, E., GIONBELLI, M. P. & HUHTANEN, P. (2014). A meta-analytical evaluation of the regulation of voluntary intake in cattle fed tropical forage-based diets. *Journal of Animal Science* **92**, 4632-4641.

FAS/USDA – *Livestock and Poultry: World Markets and Trade.* (2017). Washington, USA: Foreign Agricultural Service/ United States Department of Agriculture.

FERGUSON, K. A. & DOWLING, D. F. (1955). The function of cattle sweat glands. *Crop and Pasture Science* **6**, 640-644.

FINCH, V. A. (1986). Body temperature in beef cattle: its control and relevance to production in the tropics. *Journal of Animal Science* **62**, 531-542.

FRALEY, S. E., HALL, M. B. & NENNICH, T. D. (2015). Effect of variable water intake as mediated by dietary potassium carbonate supplementation on rumen dynamics in lactating dairy cows. *Journal of Dairy Science* **98**, 3247-3256.

FUQUAY, J. W. (1981). Heat stress as it affects animal production. *Journal of Animal Science* **52**, 164-174.

GAUGHAN, J. B., MADER, T. L., HOLT, S. M., SULLIVAN, M. L. & HAHN, G. L. (2010). Assessing the heat tolerance of 17 beef cattle genotypes. *International Journal of Biometeorology* **54**, 617-627.

HANSEN, P. J. (2004). Physiological and cellular adaptations of zebu cattle to thermal stress. *Animal Reproduction Science* **82**, 349-360.

HICKS, R. B., OWENS, F. N., GILL, D. R., MARTIN, J. J. & STRASIA, C. A. (1988). *Water intake by feedlot steers*. Miscellaneous publication - Agricultural Experiment Station, Oklahoma State University (USA).

ITTNER, N. R., KELLY, C. F. & GUILBERT, H. R. (1951). Water consumption of Hereford and Brahman cattle and the effect of cooled drinking water in a hot climate. *Journal of Animal Science* **10**, 742-751.

KLEIBER, M. (1932). Body size and metabolism. *Hilgardia* **6**, 315-332.

KRAMER, E., STAMER, E., SPILKE, J., THALLER, G. & KRIETER, J. (2009). Analysis of water intake and dry matter intake using different lactation curve models. *Journal of Dairy Science* **92**, 4072-4081.

MADER, T. L., DAVIS, M. S. & BROWN-BRANDL, T. (2006). Environmental factors influencing heat stress in feedlot cattle. *Journal of Animal Science* **84**, 712-719.

MAYER, D. G., STUART, M. A. & SWAIN, A. J. (1994). Regression of real-world data on model output: an appropriate overall test of validity. *Agricultural Systems* **45**, 93-104.

MEKONNEN, M. M. & HOEKSTRA, A. Y. (2012). A global assessment of the water footprint of farm animal products. *Ecosystems* **15**, 401-415.

MEYER, U., STAHL, W. & FLACHOWSKY, G. (2006). Investigations on the water intake of growing bulls. *Livestock Science* **103**, 186-191.

MURPHY, M. R., DAVIS, C. L. & MCCOY, G. C. (1983). Factors affecting water consumption by Holstein cows in early lactation. *Journal of Dairy Science* **66**, 35-38.

NRC (2000). Nutrient requirements of beef cattle, 7th., National research council, National Academy Sciences, Washington, D.C, USA.

PLUSKE, J. M. & SCHLINK, A. C. (2007). Managing water as a scarce resource in beef feedlots. *Australasian Agribusiness Review* **15**, 1-13.

PRADOS, L. F., SATHLER, D. F. T., SILVA, B. C., ZANETTI, D., VALADARES FILHO, S. C., ALHADAS, H. M., DETMANN, E., SANTOS, S. A., MARIZ, L. D. S. & CHIZZOTTI, M. L. (2017). Reducing mineral usage in feedlot diets for Nellore cattle: II. Impacts of calcium, phosphorus, copper, manganese, and zinc contents on intake, performance, and liver and bone status. *Journal of Animal Science* **95**, 1-11.

PURWANTO, B. P., HARADA, M. & YAMAMOTO, S. (1996). Effect of drinking-water temperature on heat balance and thermoregulatory responses in dairy heifers. *Crop and Pasture Science* **47**, 505-512.

RENAUDEAU, D., COLLIN, A., YAHAV, S., DE BASILIO, V., GOURDINE, J. L. & COLLIER, R. J. (2012). Adaptation to hot climate and strategies to alleviate heat stress in livestock production. *Animal* **6**, 707-728.

RIEMERSCHMID, G. (1943). The amount of solar radiation and its absorption on the hairy coat of cattle under South African and European conditions. *Journal of the South African Veterinary Association* **14**, 121-141.

RIJSBERMAN, F. R. (2006). Water scarcity: fact or fiction? *Agricultural Water Management* **80**, 5-22.

ROGERS, J. A., MARKS, B. C., DAVIS, C. L. & CLARK, J. H. (1979). Alteration of rumen fermentation in steers by increasing rumen fluid dilution rate with mineral salts. *Journal of Dairy Science* **62**, 1599-1605.

RUSSELL, J. B. & HESPELL, R. B. (1981). Microbial rumen fermentation. *Journal of Dairy Science* **64**, 1153-1169.

SEXSON, J. L., WAGNER, J. J., ENGLE, T. E. & EICKHOFF, J. (2012). Predicting water intake by yearling feedlot steers. *Journal of Animal Science* **90**, 1920-1928.

SEXSON, J. L., WAGNER, J. J., ENGLE, T. E. & SPEARS, J. W. (2010). Effects of water quality and dietary potassium on performance and carcass characteristics of yearling steers. *Journal of Animal Science* **88**, 296-305.

SILANIKOVE, N. (2000). Effects of heat stress on the welfare of extensively managed domestic ruminants. *Livestock Production Science* **67**, 1-18.

SUN, L. Z., AUERSWALD, K., WENZEL, R. & SCHNYDER, H. (2014). Drinking water intake of grazing steers: The role of environmental factors controlling canopy wetness. *Journal of Animal Science* **92**, 282-291.

TILMAN, D., CASSMAN, K. G., MATSON, P. A., NAYLOR, R. & POLASKY, S. (2002). Agricultural sustainability and intensive production practices. *Nature* **418**, 671-677.

VALADARES FILHO S. C., MARCONDES, M. I., CHIZZOTTI, M. L. & PAULINO, P. V. R. (2010). *Nutrient requirements of Zebu beef cattle – BR-Corte.* 2nd edn. Visconde do Rio Branco, Brazil: Suprema Gráfica e Editora.

VALENTE, É., CHIZZOTTI, M. L., DE OLIVEIRA, C. V. R., GALVÃO, M. C., DOMINGUES, S. S., RODRIGUES, A. D. C. & LADEIRA, M. M. (2015). Intake, physiological parameters and behavior of Angus and Nellore bulls subjected to heat stress. *Semina: Ciências Agrárias* **36**, 4565-4574.

WARD, F. A. & MICHELSEN, A. (2002). The economic value of water in agriculture: concepts and policy applications. *Water Policy* **4**, 423-446.

WINCHESTER, C. F. & MORRIS, M. J. (1956). Water intake rates of cattle. *Journal of Animal Science* **15**, 722-740.

ZANETTI, D., GODOI, L. A., ESTRADA, M. M., ENGLE, T. E., SILVA, B. C., ALHADAS, H. M., CHIZZOTTI, M. L., PRADOS, L. F., RENNO, L. N. & VALADARES FILHO, S. C. (2017). Estimating mineral requirements of Nellore beef bulls fed with or without inorganic mineral supplementation and the influence on mineral balance. *Journal of Animal Science* **95**, 1-12.

Table 1. *Parameters of regression between the estimates of water intake from equations in the literature and observed values of water intake and accuracy for Nellore cattle and Angus steers in feedlot**

Authors	**Regression parameters** (Estimate ± SD)		r^2	**Mayer's test**	**CCC**	**Cb**	**RMSEP**
	Intercept	Slope					
Nellore cattle							
Hicks *et al.* , 1988	15.106 ± 0.603	0.021 ± 0.014	0.006	<0.001	0.020	0.246	31.36
Meyer *et al.* , 2006	2.433 ± 1.175	0.581 ± 0.049	0.285	<0.001	0.279	0.523	8.94
CSIRO, 2007	6.059 ± 0.843	0.290 ± 0.023	0.301	<0.001	0.140	0.254	20.11
Arias and Mader, 2011 [1]	-0.120 ± 1.804	0.466 ± 0.052	0.189	<0.001	0.064	0.147	19.37
Arias and Mader, 2011 [2]	-4.037 ± 1.854	0.563 ± 0.052	0.254	<0.001	0.065	0.129	20.19
Sexson *et al.* , 2012	2.742 ± 1.291	0.314 ± 0.030	0.237	<0.001	0.061	0.156	27.10
Angus steers							
Hicks *et al.* , 1988	5.243 ± 1.150	0.710 ± 0.029	0.538	<0.001	0.654	0.892	9.83
Meyer *et al.* , 2006	-1.743 ± 1.869	0.804 ± 0.044	0.403	<0.001	0.413	0.650	13.13
CSIRO, 2007	10.058 ± 1.248	0.487 ± 0.026	0.401	<0.001	0.412	0.652	16.81
Arias and Mader, 2011 [1]	3.955 ± 2.125	0.773 ± 0.058	0.264	<0.001	0.431	0.839	10.43
Arias and Mader, 2011 [2]	1.441 ± 2.140	0.823 ± 0.057	0.296	<0.001	0.437	0.803	10.57
Sexson *et al.* , 2012	22.472 ± 1.153	0.358 ± 0.042	0.131	<0.001	0.313	0.865	13.93

* SD = standard deviation; Mayer`s test = H0: a = 0 and b = 1; CCC = concordance correlation coefficient, varies from 0 to 1; Cb = bias correction, varies from 0 to 1, 1 indicates no deviation from Y = X; RMSEP = root of mean square error of prediction.

Table 2. *Descriptive statistics of data used to develop the equations proposed here*

Parameter*	Nellore cattle (n = 276) †				Angus steers (n = 395)			
	Mean	SD	Max	Min	Mean	SD	Max	Min
WI, kg	15.7	5.5	31.3	2.3	32.0	10.8	76.3	11.8
BW, kg	352.2	59.6	525.3	216.3	569.5	102.9	826.3	340.8
DMF, %	29.3	3.1	36.8	23.4	68.8	3.6	72.0	63.7
DF, %	49.6	17.6	100.0	40.0	27.0	8.2	45.8	21.0
NDF, %	36.1	9.0	68.9	28.9	17.0	6.0	33.0	14.4
DS, %	0.3	0.2	0.6	0.0	0.1	0.1	0.2	0.0
FI, kg	15.8	3.4	31.5	8.2	17.0	4.9	34.0	6.5
DMI, kg	7.0	1.5	11.4	2.3	11.8	3.6	22.8	5.0
T_{AVG}, ºC	22.4	3.6	30.3	15.6	12.8	7.3	23.2	-6.0
T_{MAX}, ºC	26.5	2.6	34.6	18.3	20.6	8.2	31.9	-1.8
T_{MIN}, ºC	14.6	3.0	20.0	8.4	4.9	7.1	15.2	-13.3
Humidity, %	74.7	8.0	95.0	53.6	55.9	9.9	76.8	34.3
Rain, mm	2.4	4.2	27.2	0.0	0.1	0.1	0.5	0.0
Snow, cm	0.0	0.0	0.0	0.0	0.1	0.2	0.8	0.0
THI	70.2	5.5	83.5	59.7	55.5	10.2	70.1	27.9
WS, m/s	1.1	0.4	2.3	0.3	3.0	0.6	4.5	1.9
SR, W/m^2	363.6	105.0	612.9	90.8	387.9	113.6	646.0	43.4

* WI = water intake; BW = body weight; DMF = dry matter content of forage; DF = dietary forage; NDF = dietary neutral detergent fiber; DS = dietary salt; FI = feed intake; DMI = dry matter intake; T_{AVG} = average temperature; T_{MAX} = maximum temperature; T_{MIN} = minimum temperature; THI = temperature humidity index; WS = wind speed; SR = solar radiation.

† SD = standard deviation; Max = maximum; Min = minimum.

Table 3. *Descriptive statistics of data used to evaluate the equations proposed here*

Parameter*	Nellore cattle (n = 73) †				Angus steers (n = 97)			
	Mean	SD	Max	Min	Mean	SD	Max	Min
WI, kg	16.8	5.9	35.5	3.6	29.8	11.5	60.5	12.0
BW, kg	355.0	64.9	514.5	248.5	556.0	105.8	822.2	367.1
DMF, %	29.2	3.1	36.8	23.4	68.8	3.7	72.0	63.7
DF, %	50.1	17.4	100.0	40.0	27.0	8.2	45.8	21.0
NDF, %	38.7	11.9	68.9	28.9	17.0	6.1	33.0	14.4
DS, %	0.4	0.2	0.6	0.0	0.1	0.1	0.2	0.0
FI, kg	17.1	4.3	33.9	8.4	16.7	5.0	27.9	7.8
DMI, kg	7.3	1.4	12.0	3.0	11.6	3.6	21.1	5.6
T_{AVG}, ºC	22.0	3.0	29.2	15.2	12.7	7.3	22.9	-6.0
T_{MAX}, ºC	27.0	2.6	34.6	23.3	20.4	8.1	31.9	-1.8
T_{MIN}, ºC	14.9	3.2	20.2	8.0	4.8	7.0	14.9	-13.3
Humidity, %	74.9	8.4	91.0	54.9	56.0	10.0	76.8	38.8
Rain, mm	3.0	8.9	68.0	0.0	0.1	0.1	0.5	0.0
Snow, cm	0.0	0.0	0.0	0.0	0.1	0.2	0.8	0.0
THI	69.7	4.6	80.3	59.1	55.4	10.1	69.5	27.9
WS, m/s	1.1	0.3	1.9	0.4	3.0	0.6	4.6	1.9
SR, W/m^2	372.8	94.6	560.1	205.6	382.0	118.7	646.0	43.4

* WI = water intake; BW = body weight; DMF = dry matter content of forage; DF = dietary forage; NDF = dietary neutral detergent fiber; DS = dietary salt; FI = feed intake; DMI = dry matter intake; T_{AVG} = average temperature; T_{MAX} = maximum temperature; T_{MIN} = minimum temperature; THI = temperature humidity index; WS = wind speed; SR = solar radiation.
† SD = standard deviation; Max = maximum; Min = minimum.

Table 4. *Equations to predict water intake, in kg/d, for Nellore cattle in tropical conditions and Angus steers in temperate conditions*

Variable	Estimate	SE*	Partial R^2
Nellore cattle			
Intercept	-4.315	4.404	-
Metabolic body weight	0.120	0.035	0.251
Humidity	-0.178	0.034	0.088
Dry matter intake	0.913	0.232	0.034
Temperature-humidity index	0.247	0.055	0.027
Total R^2			0.400
Angus steers			
Intercept	15.005	2.689	-
Metabolic body weight	0.205	0.029	0.439
Dietary concentrate	-0.440	0.043	0.165
Maximum daily temperature	0.714	0.038	0.040
Dry matter intake	0.870	0.103	0.032
Total R^2			0.676

* SE = standard error

Table 5. *Parameters of regression between estimates of the equations proposed here and observed values of water intake and accuracy for Nellore cattle and Angus steers in feedlot*[*]

Parameter	**Regression Equation** (Estimate ± SD)		**r^2**	**Mayer's test**	**CCC**	**Cb**	**RMSEP**
	Intercept	Slope					
Nellore cattle	-0.073 ± 2.771	1.062 ± 0.169	0.356	0.243	0.500	0.838	4.803
Angus steers	-2.785 ± 2.126	1.049 ± 0.065	0.728	0.102	0.830	0.972	6.111

[*] SD = standard deviation; Mayer`s test = H0: a = 0 and b = 1; CCC = concordance correlation coefficient, varies from 0 to 1; Cb = bias correction, varies from 0 to 1, 1 indicates no deviation from Y = X; RMSEP = root of mean square error of prediction.

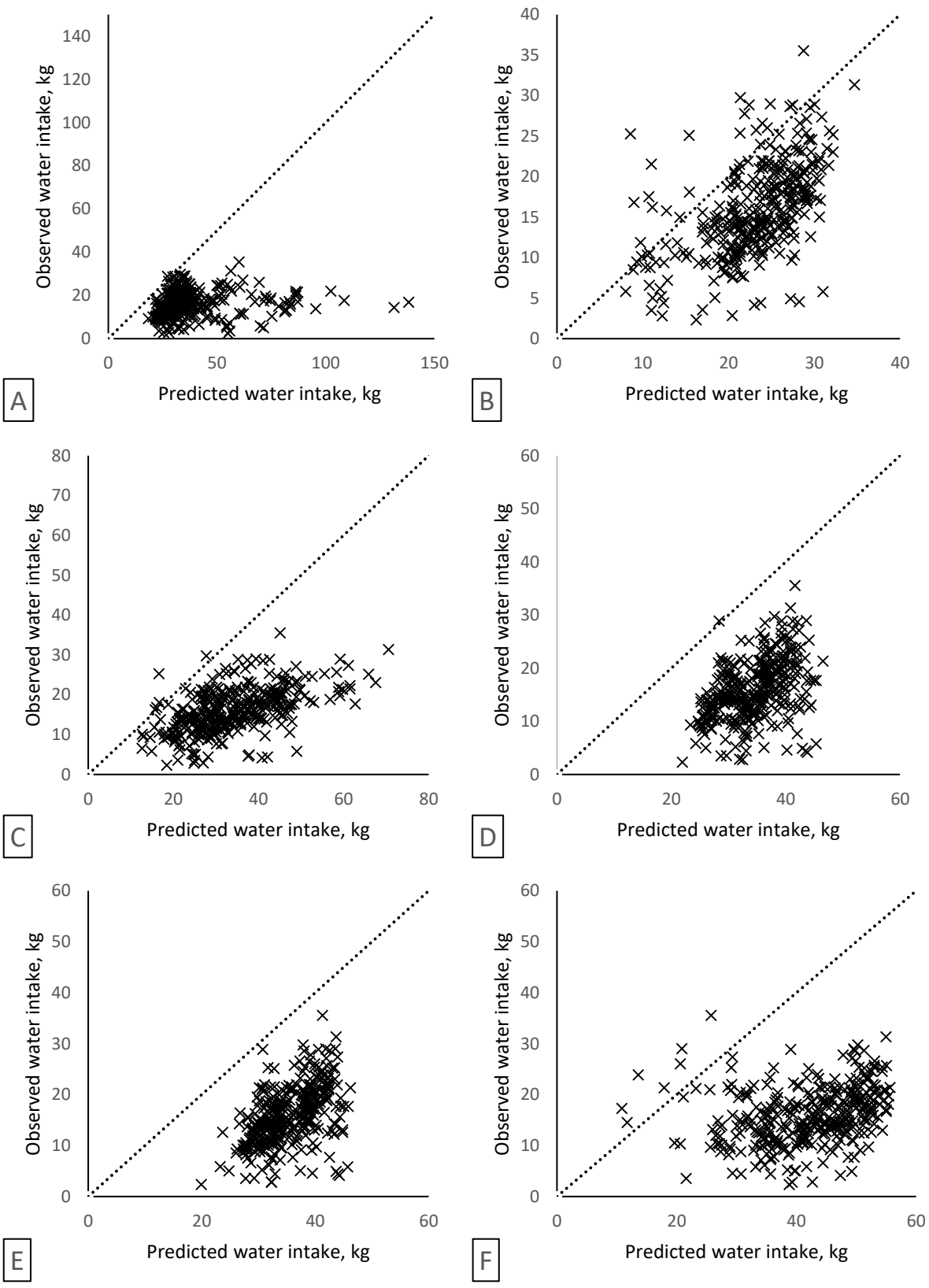

Figure 1 – Comparisons between predicted and observed water intake for Nellore cattle in tropical conditions. The dotted line represents the equality line, a point in this line represents similar values to predicted and observed water intakes. A: values predicted by Hicks *et al.*

(1988); B: values predicted by Meyer *et al.* (2006); C: values predicted by CSIRO (2007); D: values predicted by Arias and Mader (Equation 1, 2011); E: values predicted by Arias and Mader (Equation 2, 2011); F: values predicted by Sexson *et al.* (2012).

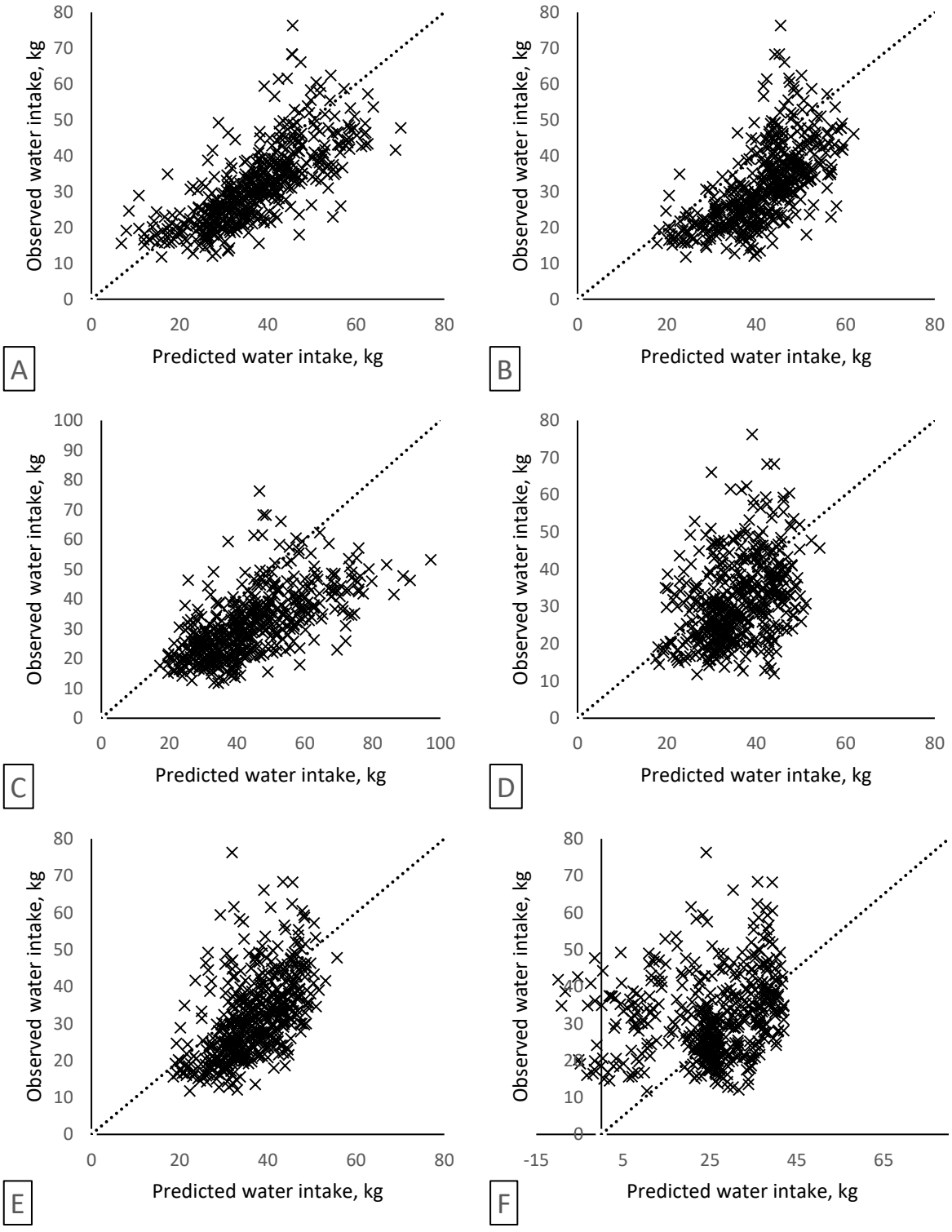

Figure 2 – Comparisons between predicted and observed water intakes for Angus steers in temperate conditions. The dotted line represents the equality line, a point in this line represents similar values to predicted and observed water intakes. A: values predicted by Hicks *et al.*

(1988); B: values predicted by Meyer *et al.* (2006); C: values predicted by CSIRO (2007); D: values predicted by Arias and Mader (Equation 1, 2011); E: values predicted by Arias and Mader (Equation 2, 2011); F: values predicted by Sexson *et al.* (2012).

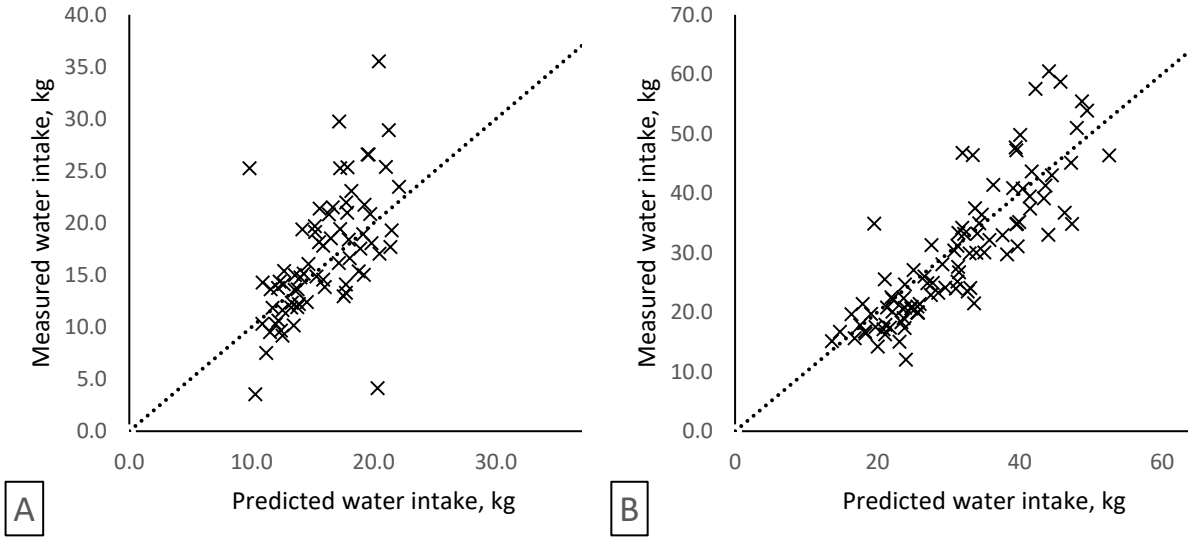

Figure 3 – Comparisons between the values predicted by equations proposed here and those observed for water intake: A: Nellore cattle raised in tropical conditions; B: Angus steers raised in temperate conditions. The dotted line represents the equality line, a point in this line represents similar values to predicted and observed water intakes.