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PLASMATIC ELECTROLYTES AND WEIGHT MODIFICATIONS IN WINTERING COWS FEEDING WITH CITRUS BY-PRODUCTS

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RESUMEN: Modificaciones del peso y electrolitos plasmáticos en vacas de invernada suplementadas con residuos cítricos.
El trabajo tuvo como objetivo verificar el efecto de la suplementación con pulpa de citrus sobre el peso y los electrolitos plasmáticos de vacas de invernada cruza cebú (n = 80) mantenidas sobre pastura natural. En dos años consecutivos, 20 animales operaron como controles (C) y otros 20 fueron suplementados (S) con pulpa fresca de citrus (15 ± 3 kg/animal, durante 4 meses), efectuándose controles a los 0, 30, 60, 90 y 120 días. Con relación a C, las estadísticas finales para cada año indicaron que en S se registraron más altos niveles de magnesio (2,30 versus 2,20 y 2,05 versus 1,96 mg/dl), calcio (9,04 versus 8,35 y 8,36 versus 7,83 mg/dl), fósforo inorgánico (5,58 versus 5,04 y 4,98 versus 4,33 mg/dl), sodio (148,4 versus 144,4 y 147,0 versus 139,5 mEq/l), hierro (143,3 versus 133,8 y 143,6 versus 142,0 µg/dl) y cobre (118,1 versus 74,6 y 99,6 versus 59,6 µg/dl), así como mayores ganancias de peso (492 versus 304 y 352 g/animal/día versus *pérdida)* En varios casos las diferencias fueron significativas ($p < 0.05$) Los valores de potasio fluctuaron irregularmente. La optimización de los niveles electrolíticos y las ganancias de peso avalan la utilización de este económico residuo industrial para la suplementación invernal de vacas de descarte.

ABSTRACT: To corroborate the effect of citrus by-product on liveweight evolution and plasmatic electrolytes, 80 wintering half-bred zebu cows maintained on native grassland, were used. Two consecutive years, 20 animals operated as controls (C) and others 20 were feeding (S) with fresh citrus pulp (15 ± 3 kg/animal, during 4 months). Periodic controls at 0, 30, 60, 90 and 120 days, were made. Results were statistically processed using a repeated measures design, with means comparison by orthogonal contrasts. Every year S registered higher levels of: magnesium (2.30 versus 2.20 and 2.05 versus 1.96 mg/dl), calcium (9.04 versus 8.35 and 8.36 versus 7.83 mg/dl), inorganic phosphorous (5.58 versus 5.04 and 4.98 versus 4.33 mg/dl), sodium (148.4 versus 144.4 and 147.0 versus 139.5 mEq/l), iron (143.3 versus 133.8 and 143.6 versus 142.0 µg/dl), and copper (118.1 versus 74.6 and 99.6 versus 59.6 µg/dl), as well as body weight gain ratio (492 versus 304 and 352 g/animal/day versus *weight lost*) than C. Treatment and time effects were significant ($p < 0.05$) in some cases. Differences between C and S began to be significant by the days 60-90 of the assay. The values of potassium fluctuated irregularly. Electrolyte improvement and quite good gain ratio endorse the use of this cheap by-product in cull cows feeding.

Key words: wintering cows feeding, citrus pulp supplementation, weight, plasmatic electrolytes.

Palabras claves: engorde de vacas de invernada, suplementación con pulpa de citrus, peso, electrolitos plasmáticos.

INTRODUCTION

Energetic-proteic supplementation systematization with regional agricultural by-products which are not expensive, was identified as one of the main demands of the cattle sector in a survey conducted by INTA in the northeast of Argentina (INTA, 1992).

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This area is plenty of citrus plantations, as well as fruits juice factories. The citrus pulp (trash, residue) is a by-product of citric juices industries. It has a high energy value as ruminant food (Ammerman y Henry, 1992), with approximately 80% of total digestible nutrients in dry matter (Brown, 1990). Because of its protein concentration is limited, it is necessary to add nitrogen, when the diet is being balanced for growing animals (Pinzon and Wing, 1976; Aguilera, 1989). Nevertheless the calcium, iron and potassium content are high in citrus pulp, the phosphorous and copper concentrations are low (Morrison, 1980).

Nowadays, in that region there are a livestock of about 10 millions, half of them are bred in Corrientes Province, and the average meat production is about 30 kg/ha/year. The seasonal growth of native grazing is the main obstacle for the regional livestock development, so that, most part of the year native grazing present low nutritive values, with dry matter digestibility lower than 50% and energetic-proteic deficiencies in winter are of a great magnitude, having protein levels of 4% and metabolizable energy rates are among limits from 1.8 to 2.0 Mcal/kg DM (Peruchena *et al.*, 1992). Deficiencies of sodium (57% of analyzed samples), copper (37%), phosphorous (33%), calcium (19%), potassium (8%), iron (4%), and other minerals were detected in native grassland (Mufarrege, 1993), as well as mineral lacks (phosphorous, sodium, copper, magnesium and other) are also known in cattle (Coppo, 1985; Coppo *et al.*, 1994).

Half-bred zebu cattle are the most abundant in the area; is a rustic crossbreed which would be more efficient using food nutrients than others bovines breeds, according to many authors (Howes, 1989). Wintering cows are *cull cows*, which are destined for sale after fattening. Because of the necessity of reduce the stocking rates on native grassland, these animals are sold in late autumn after weaning the last calf, as a *conserve* category (for canned meat), which has a lower price in the market. In order to optimize the economic yield, these kind of cattle are feeding during the winter, to obtain in that way a *consumption type cow* in early spring, when the price of the finishing cattle increase (Biani y Collia, 1982).

The purpose of this work was to verify eventual rate of gain and some plasmatic electrolytes improvement in half-bred zebu wintering cows grazing native grassland and feeding with citrus by-products (without nitrogen addition), in a subtropical area characterized by scarce quality and quantity range.

MATERIAL AND METHODS

The assays were carried out in Bella Vista Department, of Corrientes Province in Argentina. Eighty wintering cows (British x Zebu cross-breed), maintained in parcels with homogeneous quantity and quality native grassland, were used two consecutive years. Every year twenty cull cows were feeding during 4 months (autumn-winter) (supplemented group, S), while the remaining 20 (control group, C) didn't receive any supplementation.

Both lots stayed on native pasture, with a stocking rate of 0.6 animal/ha in a continuous grassing system which herbaceous predominant species and brush pastures were: *Paspalum notatum*, *Paspalum dilatatum*, *Desmodium* sp., *Trifolium* sp., *Andro-pogon lateralis*, *Sorghastrum agrostoide*, *Schizachirium spicatum*, *Aristida* sp., *Vicia* sp., *Acacia coven*, *Celtis spinosa* and *Geofroea decorticans*. Native pastures had an average of 1800 kg of DM/ha during the first year, and 2200 kg of DM/ha in the second one, with DM in vitro digestibility lowest than 50%, and levels of CP swaying among 4 at 8% and from 1.8 to 2.0 Mcal of ME/kg of DM.

The available pulp corresponded to the following fruits: orange (60%), grapefruit (20%), mandarin and lemon (20%). The offered citrus pulp contained in average protein (7.6%), crude fiber (17.7%), ether extract (4.5%), ash (4.5%), nitrogen-free extract (65.7%), phosphorous

(0.17%), calcium (0.54%), sodium (0.03%), potassium (0.50%), magnesium (725 mg/kg), manganese (15 mg/kg), zinc (78 mg/kg), iron (83 mg/kg) and copper (15 mg/kg), and 3.62 Mcal per kg of dry matter (15.5%) of gross energy.

The product was given fresh, since the transport of the residuals was carried out every 3 days from the factory of juices, about 10 km distant from the farm, that's why it was not necessary the conservation of the product for any classic method. The supplementation level was 0.6 % of live weight average, what corresponded to an average of daily offer of 2.3 kg of DM/animal/day. Animals were identified with numeral tags and liveweights were individually obtained in the morning with nocturne confinement.

Individual weighing and blood extractions by jugular venepuncture were carried out in both groups, at 0, 30, 60, 90 and 120 days. Clotted blood was centrifuged (700g, 10 min) in order to obtain serum, which was kept at 4°C until assayed. Measures of magnesium (calmagite, 520 nm, Randox reagents), calcium (cresolphtaleincomplexone, 578 nm, GT-Lab reagents), inorganic phosphorous (phosphomolybdate, 620 nm, Wiener reagents), iron (PBTS, 560 nm, Wiener reagents) and copper (batocuproin, 436 nm, Boehringer reagents), were performed in a Gilford-Beckman photometer. Sodium and potassium were valued by flame photometry in apparatus Metrolab 505 (Biopur reagents) (Coles, 1989; Piquer, 1992). Daily gains rate calculation was carried out according to the following formula P2-P1/T2-T1 (P2: current liveweight; P1: previous liveweight ; T2: current date; T1: date of previous weight).

Statistical initial homogeneity was corroborated by the overlapping of confidence intervals (95% CI) and distributive normality was verified by Wilk-Shapiro test (WS). Parametric descriptive statistics included indicators of central tendency (arithmetic mean, $\bar{x}$) and dispersion (standard deviation, SD). The analysis of the variance for repeated measures (Anova), enclosed the statistical significance for the treatment (supplementation) and time (assay lapse) effects, as well as the interaction between them was released. With the aim of elucidate the very right time in which the differences between C and S began to be significant (p), post-Anova means comparison by orthogonal contrasts were carried out. An α = 5% ($p < 0.05$), were fixed for all inferences, below which the null hypothesis of equality was rejected (Steel and Torrie, 1992).

Results and discussion

Initial and final concentration of electrolytes and weight in supplemented and control cows in two consecutive years are presented in Table 1. Obtained values match with the reference interval for this cross-breed, age of the animals, feeding type and geographical area (Coppo, 2001). Initial values were statistically homogeneous (CI ±95%) and distribution was approximately normal (WS) in every evaluated parameter (Steel and Torrie, 1992).

Table 1: Initial and final concentration of electrolytes and weight in supplemented (S) and control (C) cows ($\bar{x}$ ± SD).

Parameter	Lot	Year 1		Year 2	
		initial (n = 20)	final (n = 20)	initial (n = 20)	final (n = 20)
magnesium (mg/dl)	C	2.23 ± 0.16	2.20 ± 0.12	2.06 ± 0.14	1.96 ± 0.09
	S	2.21 ± 0.18	2.30 ± 0.19	1.89 ± 0.08	2.05 ± 0.11
calcium (mg/dl)	C	8.59 ± 0.50	8.35 ± 0.46	8.30 ± 0.54	7.83 ± 0.42
	S	8.74 ± 0.58	9.04 ± 0.59	8.22 ± 0.49	8.36 ± 0.51
phosphorous (mg/dl)	C	5.34 ± 0.98	5.04 ± 0.87	4.95 ± 0.86	4.33 ± 0.78
	S	5.23 ± 0.91	5.58 ± 0.96	5.03 ± 0.91	4.98 ± 0.84
sodium (mEq/l)	C	147.9 ± 5.8	144.4 ± 4.3	140.8 ± 4.5	139.5 ± 3.9
	S	144.9 ± 6.5	148.4 ± 3.9	143.9 ± 4.8	147.0 ± 5.2
potassium (mEq/l)	C	4.35 ± 0.5	5.02 ± 0.3	4.38 ± 0.5	3.89 ± 0.3
	S	4.24 ± 0.4	4.88 ± 0.3	4.35 ± 0.4	3.97 ± 0.2
iron (μg/dl)	C	134.2 ± 13.6	133.8 ± 21.0	141.9 ± 19.4	142.0 ± 21.7
	S	129.5 ± 19.4	143.3 ± 15.4	137.5 ± 17.2	143.6 ± 22.6
copper (μg/dl)	C	77.4 ± 24.2	74.6 ± 29.2	68.1 ± 23.4	59.6 ± 20.7
	S	75.4 ± 26.0	118.1 ± 27.8	74.5 ± 19.5	99.6 ± 25.0
weight (kg)	C	406.5 ± 33.2	443.0 ± 28.5	383.8 ± 37.6	375.0 ± 35.3
	S	428.0 ± 39.3	487.1 ± 31.8	390.5 ± 30.8	432.8 ± 25.9

Magnesium values scarcely decreased in C and increased in S, in both years, without statistical significance for treatment, time effects and interaction between them. The declines rate in C are attributed to winter nutritional range deficit (Mufarrege, 1993; Piquer, 1992) and the increases rates in S to the provided citrus pulp. In dairy cows from Argentina northeastern region, magnesium decreases at late winter due to alimentary restrictions (Sandoval *et al.*, 1998). Blood magnesium rate is a good indicator of magnesium intake (Coles, 1989; Kaneko, 1989); it can also be determined in urine, cephalorachidian and ocular liquids (Corbellini, 1998).

Calcium (Fig. 1) showed a similar behavior to magnesium, but the found differences were statistically significant for treatment and time effects (interaction not significant). Orthogonal contrasts *post-hoc* comparisons revealed that C and S began to differ from day 60. Increases in S could be caused by citrus pulp high calcium content (Morrison, 1980) and falls in C would be characteristic of winter season (Mufarrege, 1993). In argentinian northeastern cattle, calcemia tends to diminish in winter, in coincidence with pasture impoverishment and the beginning of calving period (Sandoval *et al.*, 1998). Blood decreased calcium values can be due to dietary deficiencies and homeostatic regulation mechanism failures also (Corbellini, 1998; Kaneko, 1989). This electrolyte decline in cattle that is suffering malnutrition was also verified when fields are flooded (Coppo y Coppo, 1999a and b).

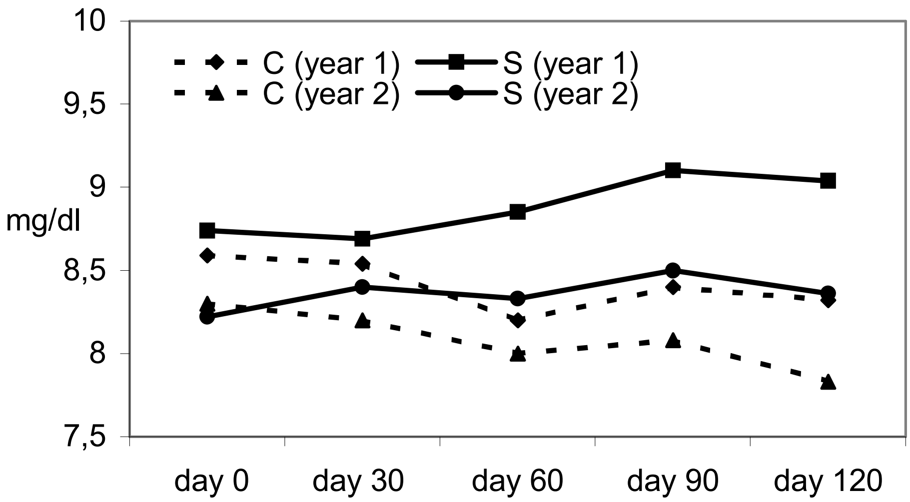

Fig. 1: Calcium evolution in supplemented (S) and control (C) animals.

Inorganic phosphorous changes (treatment and time effects) were not significant in the first year, but significant in the second. This electrolyte diminished in C (maybe for seasonal reasons) and S (in coincidence with the most rigorous winter in the second year). On the other hand, during the first year (moderate winter), inorganic phosphorous increased slightly in the supplemented animals, in spite of the fact that the citrus pulp phosphate content would not be very high (Morrison, 1980). Low plasmatic values of inorganic phosphorous indicate deficiency, but normal values not necessarily reflect enough intake, due to osseous resorption (Corbellini, 1998). In argentinian northeastern region, the lowest levels of cattle fosfatemia registers in winter, in coincidence with the impoverishment of the pastures (Sandoval *et al.*, 1998).

Inorganic phosphorus is the most important mineral lack suffered by cattle of that area (Coppo, 1985; Coppo, 2001). This deficiency conspires against the animal production by diminishing the ratio of gains; for that reason, bone meal sup-plementation (60 g/animal/day) is usual put into practice in the northeast of Argentina (Mufarrege, 1993). Administration of calcium orthophosphate during one year (3 g/animal/day) in young cattle, would have make available liveweight increases of 32 % higher than controls without supplementation (Segura-Correa y Castellanos-Ruelas, 1999).

Sodium variations were not significant in both, first and second year, neither for treatment nor time effects. Hyponatremia would not keep relationship with pasture sodium decrease; as well as potassium, sodium lack should be investigated in saliva rather than plasma (Corbellini, 1998). Because sodium is scarce in vegetables, in Argentinean northeastern, grazing cattle must be supplemented with salt (NaCl: 20-40 g/day) to avoid weight losses and reproductive alterations (Coppo, 1985; Mufarrege, 1993; Coppo, 2001).

In both groups, potassium increased during the first year, and decreased during the second year (rough winter). Deficiency of potasium is not very common in ruminants of the studied zone due to the pasture high potassium content (Coppo, 2001). At the same time, potassium food increases would not cause hyperkalemia, due to its strong feedback control (Corbellini, 1998). Potassium dietary excess would cause hypomagnesemia, with milk calcium decrease (Sandoval *et al.*, 1998).

Iron registers high concentration in citrus pulp (Morrison, 1980), which justifies the plasmatic iron increases in S, in both assays. Treatment and time effects were significant, but not their interaction. Differences between C and S began to be significant from day 60. Iron plasmatic levels vary in directly proportional way to the intake ferric content (Kaneko, 1989; Coppo, 2001). In the Argentine Mesopotamia, winter iron plasmatic concentration in heifers and cows diminish due to pasture nutrients impoverishment, reaching the lowest values in spring (Cseh *et al.*, 1998).

Copper decreased in C (winter decline) and increased in S, probably because of the citrus pulp supply, in spite of its low cupric content (Morrison, 1980), in both assays (Fig. 2). Repeated measures Anova found statistical significance for treatment and time effects (interaction was no significant). Mean comparison test revealed that differences between C and S began by day 60 (first year) and day 90 (second year). Plasmatic copper level is useful to indicate the concentration of this trace element in pastures (Kaneko, 1989; Corbellini, 1998). Although there are others methods to detect copper deficiency, they are not so practical as the blood copper determination (Quiroz-Rocha y Bouda, 2001). Lack of copper is very common in cattle from argentine northeastern (Coppo, 1985), what cause hair depigmentation, anemia, immunodeficien-cy, osseous fragility, subfertility, and lower growth rate (Coppo, 2001).

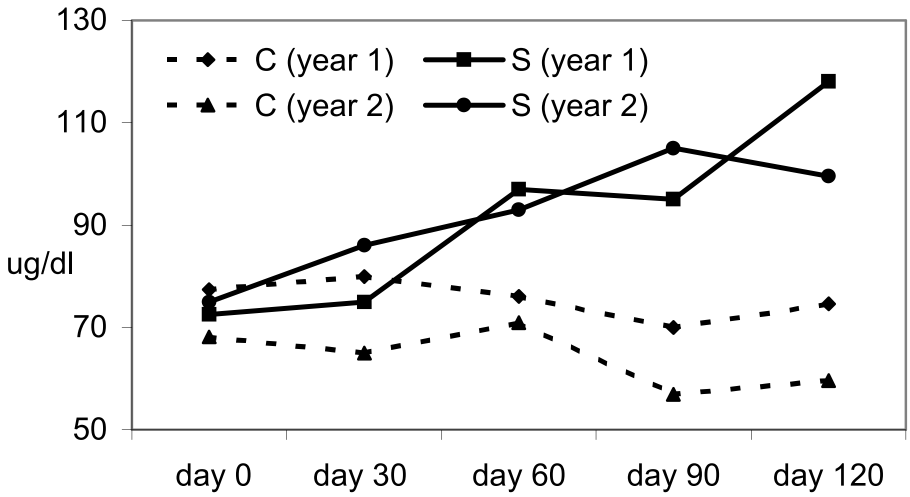

Fig. 2: Copper evolution in supplemented (S) and control (C) animals.

Final liveweight, in both assays, were higher in supplemented group than in control ones. Winter conditions were moderated the first year, and liveweight gains were 36.5 kg in C (304 g/animal/day) and 59.1 kg in S (492 g/animal/day). As a result of a rigorous cold season, in the second year, control cows lost weight (-8.8 kg) and supplemented animals showed body weight gains of 42.3 kg (352 g/animal/day). Treatment and time effects were significant in both assays, while the interaction treatment by time was not significant. Means comparisons by orthogonal contrasts revealed that liveweight differences between C and S began to be significant by day 30 (first year) and day 60 (second year). Fig. 3 shows the average body weight registered in every sampling date, standing out the highest rate of gains in S (both 1 and 2 years) and the lost of body weight in C (year 2).

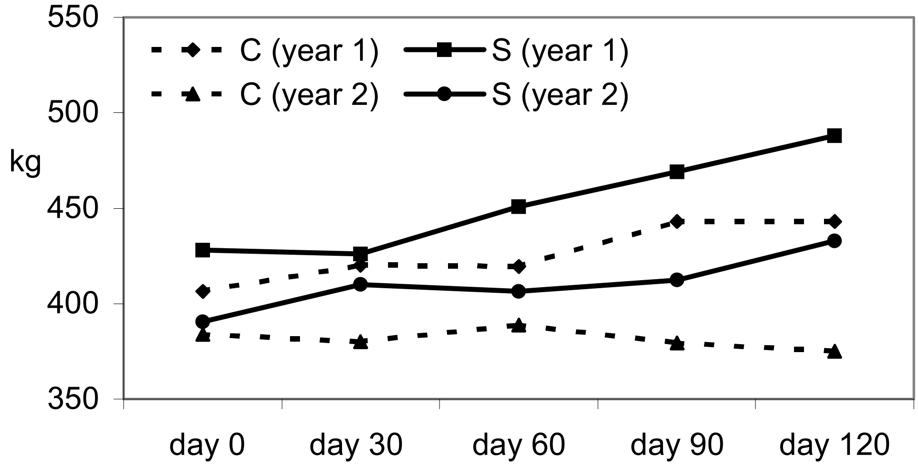

Fig. 3: Weight evolution in supplemented (S) and control (C) animals.

Supplementing wintering half-bred zebu cows with brewery residuals, produced good rates of gain in experimental groups (314 g/animal/day) and negative rates gain or lost of liveweight in control groups (-128 g/animal/day), were verified by our team-work (Capellari *et al.*, 1992). Also in argentine northeastern, the same kind of cross-breed cattle supplemented with cottonseed increased 292 g/animal/day, as well as the controls increased just 51 g/animal/day (Coppo *et al.*, 1994). By offering rising amounts of citrus pulp (Kuvera *et al.*, 1993) or citric residuals mixed with another complements (Ghisi, 1968), some researchers obtained liveweight gain rates from 236 to 1100 g/animal/day.

Citrus pulp could produce ruminal parakeratosis in calves (Santos y Aguilera, 1981); in adult ruminants could cause diarrheas (Morrison, 1980) and carry hemorrhagiparous aflatoxins like *citrinin* (Griffiths and Done, 1991), as well as viscerotropic pesticides (Nigg *et al.*, 1979). It is known that in birds it causes hepatomegalia, with percentages of mortality to 97% (Morrison, 1980). Clinical symptoms attributable to the forage supplied were not registered in present assay.

CONCLUSION

Citrus pulp revealed to be able to rise significantly ($p < 0.05$) some nutritionally important plasmatic electrolytes (Ca, P, Fe, Cu); some others increased not significantly (Mg, Na) or showed irregular behavior (K). The absence of undesirable secondary effects, added to the major rates of gain (first year: 492 versus 304 g/animal/day; second year: 352 g/animal/day versus *loss of weight in controls*), endorse the utility of this agroindustrial residual for the wintering half-bred zebu cows feeding.

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