

http://www.uem.br/acta ISSN printed: 1806-2636 ISSN on-line: 1807-8672 Doi: 10.4025/actascianimsci.v36i2.21345

# Body composition and deposition efficiency of protein and energy in grazing young bulls

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**ABSTRACT.** The effects of supplementation with different protein: carbohydrate ratios on body composition, carcass characteristics and protein and energy deposition efficiency of young were assessed. Twenty-four Nellore calves (132.5  $\pm$  5.5 kg and 90-150 days of age) were kept on pasture for a 430 day experimental period. The treatments were: Control = mineral mixture only; HPHC = high-protein and high-carbohydrate supplement; HPLC = high-protein and low-carbohydrate supplement; LPHC = low-protein and high-carbohydrate supplement; LPLC = low-protein and low-carbohydrate supplement. Four animals at begning and 20 animal at end of experiment were slaughtered to evaluate the carcass composition. Control bulls had the lowest (p < 0.05) intake of ME (9.8 Mcal day<sup>-1</sup>) with no difference (p > 0.05) between supplemented bulls (13 Mcal day<sup>-1</sup>). Although non-supplemented bulls had less (p < 0.05) retained protein, retained energy (RE), body weight gain and dressing percentage, differences were not observed (p > 0.05) RE (596.2 Mcal) than low-carbohydrate supplements (515.5 Mcal). Differences were not found (p > 0.05) in the energy efficiency between the groups. Therefore, supplementation increases the intake and retention of protein and energy without changing the retention efficiency.

Keywords: associative effects, beef cattle, calf, grazing, multiple supplement, production.

# Composição corporal e eficiencia de deposição de proteína e energia de tourinhos em pastejo

**RESUMO.** Foram avaliados os efeitos da suplementação com diferentes relações proteína: carboidrato sobre a composição corporal, características de carcaça e eficiência de deposição de proteína e energia em tourinhos. Utilizou-se 24 bezerros Nelore (132,5  $\pm$  5,5 kg e 90-150 dias de idade) em pastagem por um período experimental de 430 dias. Os tratamentos foram: Controle = mistura mineral ; HPHC = suplemento com alta proteína e alto carboidrato; HPLC = suplemento com alta proteína e baixo carboidrato; LPHC = suplemento com alta proteína e alto carboidrato; LPLC = suplemento com baixa proteína e alto carboidrato; LPLC = suplemento e 20 animais no final do experimento para avaliar a composição da carcaça. Os tourinhos controle apresentaram o menor (p < 0,05) consumo de EM (9,8 Mcal dia<sup>-1</sup>) não havendo diferenças (p > 0,05) entre os tourinhos suplementados (13 Mcal dia<sup>-1</sup>). Embora os tourinhos não suplementados tivessem menor (p <0,05) proteína retida, energia retida (ER), ganho de peso e rendimento de carcaça que os animais suplementados, não foram associados com maior (p < 0,05) ER (596,2 Mcal) do que os suplementos de baixo carboidrato (515,5 Mcal). Não houve diferenças (p > 0,05) na eficiência energética entre os grupos. Portanto, a suplementação aumenta a ingestão e retenção de proteína e energia, sem alterar a eficiência de retenção.

Palavras-chave: efeitos associativos, gado de corte, bezerro, pastagem, suplemento múltiplo, produção.

### Introduction

The efficiency of feed utilization by cattle has been a concern of animal nutritionists and producers for centuries due to its obvious economic and environmental impacts. The efficiency of feed use can be decreased by a lack, excess or unbalance of energy and nutrients in the feed. Although the effects of protein and energy supply on feed utilization have been of interest during the last decades, our knowledge of the response and the efficiencies of feed utilization is relatively poor (SCHROEDER; TITGEMEYER, 2008), mainly with respect to grazing conditions.

The efficiency of N utilization by ruminants is usually low, leading to high feeding costs and to large amounts of N being excreted into the environment (SCHROEDER; TITGEMEYER, 2008). In general, ruminants have low efficiency of use of nutrients, and cattle normally retain only 10 to 20% of nutrient intake (COLE et al., 2006). However, many factors may affect the energy and protein efficiency of tissue deposition in an animal body.

Differences in efficiency of energy use to growth have frequently been attributed to differences in body composition (FERREL; JENKINS, 1998). More specifically, differences in rates of water and protein accretion relative to the rate of fat accretion are thought to have a major influence on the rate and efficiency of body weight gain, primarily resulting from the lower energy content of water and protein than of fat. Conversely, higher maintenance costs have been associated with body protein than with body fat. Variations in the maintenance and efficiency of gain are frequently more associated with the weight and metabolic activity of visceral organs, such as the gut and liver, than with body protein or fat or the composition of gain (FERREL; JENKINS, 1998). Therefore, the efficiency of use feed and nutrients are affected by body composition, which in turn is affected by feed plans. Mccurdy et al. (2010) observed that the body composition of grazing cattle is changed by nutritional programs, even when daily weight gains are similar.

Protein deposition is increased by the level of dietary CP and energy until it is limited by energy or by the supply of the most limiting AA (GREENWOOD; TITGEMEYER, 2000). Thereby, tissue accretion may be increased by increasing the energy supply (SCHROEDER et al., 2006) or by increasing the AA available to ruminants (SCHROEDER et al., 2006). Bailey et al. (2008) found differences in feed efficiency and carcass characteristics with the level of dietary CP.

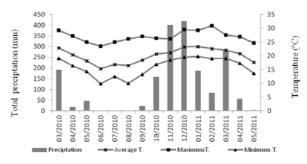
Beef cattle fed a high-fiber diet may have an improvement of energy intake with protein supplementation and greater use of dietary N when CP and carbohydrates are supplied together. Otherwise, when carbohydrates are supplied without protein, the use of fiber is reduced and energy intake may be reduced (SOUZA et al., 2010).

Although studies have demonstrated that the proportion of protein and energy in a diet affects the efficiency of tissue deposition (SCHROEDER; TITGEMEYER, 2008; REYNOLDS et al., 2011), these interactions have not been evaluated specifically for pasture conditions. Thus, the effects of supplementation with different protein: carbohydrate ratios on body composition, carcass characteristics and protein and energy deposition efficiency of young bulls from 4 until 18 months of age were assessed.

#### Material and methods

#### Animals, experimental design and diets

The experimental protocol and procedures were approved by the Universidade Federal de Viçosa Animal Care and Use Committee. This experiment was carried out at the beef cattle facility of the Universidade Federal de Viçosa, in Viçosa, Minas Gerais State, Brazil (20° 45' s 42° 52' W). The experimental area is located in a hilly area at an altitude of 670 m. This study was carried out between March of 2010 and April of 2011. The weather data is presented in Figure 1. Animals were submitted to a period of 14 days for adaptation. The experimental period was divided into four phases: phase 1 = suckling phase in rainy-dry transition season (112 days); phase 2 = post-weaning in dry season (84 days); phase3 = post-weaning in the dry-rainy transition season (84 days); phase 4 = finishing phase in the rainy season (142 days).



**Figure 1.** Precipitation, average temperature (Average T.), maximum temperature (Maximum T.) and minimum temperature (Minimum T.) in the designated experimental periods.

Twenty-four Nellore calves with average initial body weights of  $132.5 \pm 5.5$  kg and between 90 and 150 days of age and their dams were used. The animals were housed in five 10-ha plots of signal grass (Brachiaria decumbens) in phase 1 and in five 2.5 ha similar plots in other phases. The animals were randomly assigned into one of the five nutritional plans: Control: mineral mixture only; HPHC: highprotein and high-carbohydrate supplement; HPLC: high-protein and low-carbohydrate supplement; LPHC: high-carbohydrate low-protein and supplement; LPLC: low-protein and low-carbohydrate supplement (Table 1).

| Itens                  |         | Nutritional plan <sup>1</sup> |      |      |             |           |         | Pasture <sup>2</sup> |         |  |  |  |  |
|------------------------|---------|-------------------------------|------|------|-------------|-----------|---------|----------------------|---------|--|--|--|--|
| Itens                  | Control | HPHC                          | HPLC | LPHC | LPLC        | Phase 1   | Phase 2 | Phase 3              | Phase 4 |  |  |  |  |
| Corn                   | -       | 55.0                          | 0.0  | 83.5 | 53.0        |           |         |                      |         |  |  |  |  |
| Corn gluten            | -       | 3.0                           | 20.0 | 0.0  | 14.0        |           |         |                      |         |  |  |  |  |
| Soybean meal           | -       | 37.0                          | 70.0 | 12.0 | 24.0        |           |         |                      |         |  |  |  |  |
| Urea/A.S. <sup>3</sup> | -       | 1.0                           | 2.0  | 0.5  | 1.0         |           |         |                      |         |  |  |  |  |
| $MM^4$                 | 100     | 4.0                           | 8.0  | 4.0  | 8.0         |           |         |                      |         |  |  |  |  |
|                        |         |                               |      |      | Chemical co | mposition |         |                      |         |  |  |  |  |
| Dry matter             |         | 87.1                          | 89.5 | 85.8 | 87.0        | 29.6      | 42.5    | 28.0                 | 21.3    |  |  |  |  |
| Organic matter         |         | 89.3                          | 87.4 | 88.4 | 85.8        | 91.4      | 92.4    | 92.4                 | 91.5    |  |  |  |  |
| Crude protein          |         | 29.2                          | 55.3 | 15.4 | 29.5        | 8.8       | 5.5     | 12.1                 | 10.7    |  |  |  |  |
| apNDF⁵                 |         | 8.7                           | 10.2 | 7.4  | 9.2         | 65.3      | 65.0    | 61.5                 | 61.6    |  |  |  |  |
| Ether extract          |         | 2.6                           | 1.5  | 3.0  | 2.4         | 1.2       | 1.2     | 1.5                  | 1.2     |  |  |  |  |
| CNF <sup>6</sup>       |         | 46.2                          | 23.3 | 57.2 | 43.6        | 16.1      | 20.7    | 17.3                 | 17.9    |  |  |  |  |

Table 1. Ingredients and chemical composition of supplements and pasture.

<sup>1</sup>HPHC = high protein and high carbohydrate supplement; HPLC = high protein and low carbohydrate supplement; LPHC = low protein and high carbohydrate supplement; LPLC = low protein and low carbohydrate supplement; Obtained by handle plucking sample; <sup>3</sup>Urea + ammonia sulfate (9:1). <sup>4</sup>Mineral mixture; composition: calcium: 8.7 %, phosphor: 9.0 %, sulfur: 9.0 %, sodium: 18.7 %, zinc: 2400.00 mg kg<sup>-1</sup>, copper: 800.00 mg kg<sup>-1</sup>, manganese: 1600.00 mg kg<sup>-1</sup>, iodine: 40.00 mg kg<sup>-1</sup>, cobalt: 8.00 mg kg<sup>-1</sup>, selenium: 8,16 mg kg<sup>-1</sup>; <sup>5</sup>Neutral detergent fiber corrected for ash and protein; <sup>6</sup>Non-fiber carbohydrate.

Approximately 50 and 25% of the crude protein (CP) requirement was supplied by the high and lowprotein supplements respectively, and approximately 30 and 15% of the digestible energy (DE) requirement was supplied by the high and low-carbohydrate supplement, respectively. Half of the stipulated requirements were supplied by the supplements in phase 1 due to milk intake. Every 28 days, the amount of supplement was adjusted by using the estimated protein and energy requirements by BR-CORTE (VALADARES FILHO et al., 2006), considering weight gain in the adaptation period to first adjust the supplementation and the previous 28 days' weight gain to adjust requirements in other periods.

The supplements had similar proportions of CP from corn, soybean meal and urea (Table 1). Calves were supplemented once a day at 11:00 am. The animals were rotated among the five pasture paddocks every seven days, allowing each group to stay in each paddock for the same period of time with similar pasture intakes, differing only in the supplement intake. Calves were weaned at the end of phase 1 when they were approximately eight months of age, 112 days after the beginning of experimental period.

#### Experimental procedures and sampling

Every seven days, a hand-plucking sample was performed simultaneously to observe the young bulls' grazing behavior to obtain samples of forage consumed by the animals. All samples were dried at 60°C for 72 hours and ground to pass through a 1 mm screen, and proportionally sub-sampled to a composite sample per period.

To evaluate forage intake and digestibility, a digestion trial (eight days) was performed simultaneously to evaluate the performance of the animals in the middle of each production phase. Fecal dry matter excretion was determined by providing 10, 12, 14 and 16 g day<sup>-1</sup> of chromic oxide to young bulls in phases 1, 2, 3 and 4, respectively. These portions were

packaged in a paper cartridge and directly introduced into the esophagus through a rubber tube. The animals received the marker once daily at 11 am during the seven days of the digestion trial. To evaluate the individual supplement intake, 10, 12, 14 and 16 g day<sup>-1</sup> of titanium dioxide was mixed with the supplements and offered to animals in phases 1, 2, 3 and 4, respectively. The forage intake was estimated by using indigestible neutral detergent fiber (iNDF) as an internal marker (VALENTE et al., 2011a). After five days of adaptation, feces samples were collected at 3:00 pm on the 6<sup>th</sup> day, at 11 am on the 7th day, and at 7 am on the 8th day of the digestion trial period. The fecal samples were dried at 60°C for 72 hours and ground to pass through a 1 mm screen, and proportionally subsampled to make a composited sample per phase.

Urine spot samples were obtained in the middle of each experimental phase, approximately 4 hours after the feeding. The samples were filtered through a cheesecloth, and a 10 mL aliquot was separated and diluted with 40 mL of  $H_2SO_4$  (0.036 N). Blood was collected from the jugular vein on the same day as urine collection and was centrifuged at 2,700 × g, for 20 min. to obtain the serum, which was frozen at -20°C.

Milk intake by calves was estimated on days 28, 56 and 84 of the experimental period (phase 1). Cows were separated from their calves at 6:00 pm to 6:00 am of the next day, cows were milked immediately after an injection of 2 mL of oxytocin (10 IU mL<sup>-1</sup>; Ocitovet<sup>®</sup>, Brazil) in the mammary vein and the produced milk was weighed. The exact time when each cow was milked was recorded, and the milk production was converted into a 24 hours production.

#### Slaughter and body composition

Four calves were slaughtered at the beginning of the experiment and were used as reference to estimate the initial body composition of the remaining animals that were slaughtered at the end of the experimental period.

After 16 hours of fasting, the young bulls were weighed at the beginning and at the end of the experimental period. At slaughter, young bulls were stunned with a captive bolt gun and killed by exsanguination. Weights of the right and left halves of hot carcass, hide, head, shanks and tail, liver, heart, lung, kidneys, spleen, rumenreticulum, omasum, abomasum, small intestine, and large intestine as well as internal adipose tissues were recorded. Organs and viscera were ground in an industrial mill per one hour. The dressing percentage (DP) was calculated by the ratio between the hot carcass weight and fasting body weight.

After a 24 hours chill (4°C), the carcasses were weighed and the right halves were subsequently separated into lean tissue, adipose tissue and bone. The compounds from half of the animals' heads and shanks were separated and analyzed and used to estimate the composition of heads and shanks of non-sampled animals. All samples were dried at 55°C for 72 hours. Then, samples were predegreased (upon extraction with petrol ether in Soxhlet apparatuses for 6 hours) and ground into a ball mill and analyzed.

Total carcass composition was calculated as the sum of the weight of each chemical component from lean, adipose tissue and bone, and adjusted for cooler shrinkage (weight difference between hot and cold carcass weight was assumed as being water loss), and for differences in the weight of the right and left sides. Energy contents were calculated as weights of the ether-extracted material x 9.3929 kcal g<sup>-1</sup> and fat-free organic matter x 5.6405 kcal g<sup>-1</sup> (ARC, 1980).

Gross efficiency energy (GEE) was calculated as the relation between the retained energy (Mcal) in the body and the ME intake (Mcal). Gross efficiency crude protein (GECP) was calculated as the relation between the retained protein in the body (kg) and crude protein intake (CPI, kg). Gross efficiency metabolizable protein (GEMP) was calculated as the relation between the retained protein in the body (kg) and the metabolizable protein intake (MPI, kg).

#### **Chemical analysis**

Samples of carcasses, forage, feces and supplement ingredients were analyzed for dry matter (DM, index no. 920.39), crude protein (CP, index no. 954.01), organic matter (OM, index nº 942.05) and ether extract (EE, index nº 920.39) as described by AOAC (1999). Samples of forage, feces and supplements were analyzed for neutral detergent fiber (NDF), samples were treated with thermostable alpha-amylase without sodium sulfite and corrected for ash residue (MERTENS, 2002) and residual nitrogen compounds (LICITRA et al., 1996). Indigestible neutral detergent fiber (NDFi) was analyzed as described by Valente et al. (2011a). Fecal samples were evaluated concerning chromium and titanium dioxide contents by using atomic absorption (WILLIANS et al., 1962) and colorimetric (MYERS et al., 2004) methods, respectively. The milk was analyzed for protein, fat, lactose and total solids content using spectroscopy (Foss MilkoScan FT120, Hillerød, Denmark).

The fecal excretion was estimated by the ratio of marker dose (chromic oxide) and its concentration in the feces. The dry matter intake (DMI) was estimated by using iNDF as an internal marker and calculated by the following equation:

DMI (kg day<sup>-1</sup>) = [(FE x iNDF feces) – iNDF supplement)  $\div$  iNDF forage] + SI + MI,

where:

FE is the fecal excretion (kg day<sup>-1</sup>);

iNDF feces is the concentration of iNDF in the feces (kg kg<sup>-1</sup>);

iNDF supplement is the iNDF in the supplement (kg);

iNDF forage is the concentration of iNDF in the forage (kg kg<sup>-1</sup>);

SI is the supplement intake;

MI is the milk intake.

The estimation of individual intake of supplement was obtained by using the external marker titanium oxide by using the following equation:

 $SI = (FE \times MCF)/MCS,$ 

where:

SI is the dry matter supplement intake (kg day<sup>-1</sup>);

FE is the fecal excretion (kg day<sup>-1</sup>);

MCF is the marker concentration in the feces (kg kg<sup>-1</sup>);

MCS is the marker concentration in the supplement (kg kg<sup>-1</sup>).

The urea concentration in the blood serum and creatinine in the urine were obtained by colorimetric enzymatic (Bioclin<sup>®</sup> K047) and modified Jaffé (Bioclin<sup>®</sup> K016-1) methods, respectively. The urinary contents of allantoin and uric acid were estimated

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using colorimetric methods as reported by Chen and Gomes (1992).

The total urinary volume was estimated by the ratio of creatinine concentration in the urine upon excretion per unit of body weight (CHIZZOTTI et al., 2006):

CE = 32.27 - 0.01093- BW,

where:

CE is the daily creatinine excretion (mg kg<sup>-1</sup> of LW);

BW is the body weight (kg).

Purine derivative excretion was calculated by adding the quantities of allantoin and uric acid excreted in the urine. The absorbed purines were calculated from purine derivative excretion (BARBOSA et al., 2011):

 $AP = (PD - 0.301 - BW^{0.75})/0.80,$ 

where:

AP is the absorbed purines (mmol day<sup>-1</sup>);

PD is the purine derivative excretion (mmol  $day^{-1}$ ).

Microbial synthesis of nitrogenous compounds in the rumen was estimated as a function of the absorbed purines and the microorganism NRNA/NTOTAL ratio (CHEN; GOMES 1992):

NMIC =  $(70 - AP)/(0.83 \times R \times 1000)$ ,

where:

NMIC is the microbial nitrogen flow in the small intestine (g day<sup>-1</sup>);

R is the NRNA/NTOTAL ratio in the microorganisms (mg  $mg^{-1}$ ).

#### Statistical analysis

This study was conducted under a completely randomized design using a  $2 \times 2+1$  factorial arrangement (two protein levels, two carbohydrate levels, and one control). Feed intake, carcass traits, organ weights, body composition and deposition efficiency data were analyzed with the GLM procedures of SAS version 9.1 (SAS, 2004) and comparisons among treatment means were made by using orthogonal contrasts. Significant difference was considered at p < 0.05.

# Results

Non-supplemented young bulls had lower (p < 0.05) intakes of ME than supplemented. However, differences were not found (p > 0.05) between supplemented young bulls groups. The control had a lower (p < 0.05) intake of crude protein (CP) and metabolizable protein (MP) than the supplemented group. In addition, animals that received high protein supplements (HPHC and HPLC) had greater (p < 0.05) intakes of CP and MP than animals that received low-protein supplements (LPHC and LPLC) – Table 2.

The values for retained protein (RP), retained ether extract (REE) and retained energy (RE) were lower (p < 0.05) in the non-supplemented than supplemented young bulls. Although differences were not found (p > 0.05) for the RP between supplement types, animals that received the high-carbohydrate supplement (HPHC and LPHC) had greater (p < 0.05) REE and RE than those that received the low-carbohydrate supplement (HPLC and LPLC) (Table 2).

Differences were not found (p > 0.05) in the GEE between supplemented and non-supplemented animals or between supplement types. Although the gross efficiency of crude protein (GECP) and metabolizable protein(GEMP) values were similar (p > 0.05) between the control and supplemented groups (Table 2), young bulls that received high-protein supplements (HPLC and HPLC) had lower (p < 0.05) GECP and GEMP values.

The non-supplemented young bulls had lower (p < 0.05) values for urinary urea nitrogen excretion (UUN), serum urea nitrogen (SUN), production of nitrogen microbial (NMIC) and a similar (p > 0.005) ratio of urinary urea nitrogen excretion per nitrogen intake (UUN/NI) when compared with supplemented young bulls (Table 3). Although the high-protein supplements (HPHC and HPLC) had greater (p < 0.05) UUN, UUN/NI and SUN than the low-protein supplement (LPHC and LPLC), differences were not found (p > 0.05) in the NMIC and efficiency of microbial synthesis (EMS, grams of CP microbial/total digestible nutrients intake).

The control animals had lower (p < 0.05) MP: E ratios than the supplemented animals. Although supplementation with high protein was associated with a greater (p < 0.05) MP: E ratio than low protein, differences were not found (p > 0.05) between carbohydrate levels. However, there was an interaction (p < 0.05) between the protein and carbohydrate levels (Table 2). In supplements with high protein, increased carbohydrate levels were associated with an increased MP: E ratio, but low-protein supplements, with increased carbohydrate levels were associated with reduced MP: E ratios.

**Table 2.** Effect of nutritional plans on intake of metabolizable energy (MEI, Mcal day<sup>-1</sup>), crude protein (CPI, g day<sup>-1</sup>) and metabolizable protein (MPI, g day<sup>-1</sup>), retained protein (RP, kg), retained ether extract (REE, kg), retained energy (RE, Mcal), gross efficiency of energy (GEE, %), crude protein (GECP, %) and metabolizable protein (GEMP, %), and relation metabolizable protein and energy intake (MP:E, g Mcal<sup>-1</sup>).

| Items |         | Nutritional plans <sup>1</sup> |       |       |       |                 | P-value <sup>2</sup> |         |        |       |  |  |  |
|-------|---------|--------------------------------|-------|-------|-------|-----------------|----------------------|---------|--------|-------|--|--|--|
| nems  | Control | HPHC                           | HPLC  | LPHC  | LPLC  | SE <sup>3</sup> | CT                   | Р       | С      | P*C   |  |  |  |
| MEI   | 9.8     | 14.9                           | 12.0  | 12.7  | 12.5  | 1.2             | 0.032                | 0.485   | 0.209  | 0.286 |  |  |  |
| CPI   | 498.6   | 964.5                          | 857.5 | 644.6 | 684.8 | 68.9            | 0.002                | 0.003   | 0.635  | 0.302 |  |  |  |
| MPI   | 328.3   | 682.4                          | 514.9 | 415.7 | 455.3 | 49.7            | 0.004                | 0.005   | 0.2018 | 0.055 |  |  |  |
| RP    | 27.9    | 49.2                           | 44.5  | 46.8  | 45.4  | 2.7             | 0.001                | 0.777   | 0.272  | 0.536 |  |  |  |
| REE   | 22.1    | 31.9                           | 26.9  | 37.8  | 29.2  | 3.2             | 0.019                | 0.214   | 0.050  | 0.582 |  |  |  |
| ER    | 364.3   | 575.5                          | 502.0 | 616.8 | 529.0 | 31.6            | 0.001                | 0.298   | 0.022  | 0.825 |  |  |  |
| GEE   | 9.0     | 9.34                           | 10.0  | 11.5  | 10.4  | 0.8             | 0.187                | 0.143   | 0.791  | 0.308 |  |  |  |
| GECP  | 13.8    | 12.2                           | 12.5  | 17.2  | 16.1  | 1.2             | 0.609                | 0.003   | 0.740  | 0.585 |  |  |  |
| GEMP  | 21.1    | 20.5                           | 20.5  | 26.6  | 24.5  | 1.7             | 0.561                | 0.002   | 0.777  | 0.148 |  |  |  |
| PM: E | 33.3    | 45.7                           | 43.3  | 32.7  | 36.5  | 1.3             | 0.001                | < 0.001 | 0.603  | 0.031 |  |  |  |

<sup>1</sup>Control = mineral mixture only; HPHC = high protein and high carbohydrate supplement; HPLC = high protein and low carbohydrate supplement; LPLC = low protein and high carbohydrate supplement; LPLC = low protein and low carbohydrate supplement;  $^2$ CT = non-supplemented versus supplemented, P = effect of protein amount, C = effect of carbohydrate amount, P\*C = effect of interaction of protein and carbohydrate. <sup>3</sup>Standard error.

**Table 3.** Effect of nutritional plans on urinary urea nitrogen excretion (UUN, g day<sup>-1</sup>), ratio urinary urea nitrogen excretion /N intake (UUN/NI, g g<sup>-1</sup>), serum urea nitrogen (SUN, mg dL<sup>-1</sup>), production of nitrogen microbial (NMIC, g day<sup>-1</sup>) and efficiency of microbial synthesis (EMS, g kg<sup>-1</sup>).

| Itens            |         | Nutritional plans <sup>1</sup> |        |        |        |        |         | P-value <sup>2</sup> |       |       |  |  |  |  |
|------------------|---------|--------------------------------|--------|--------|--------|--------|---------|----------------------|-------|-------|--|--|--|--|
|                  | Control | HPHC                           | HPLC   | LPHC   | LPLC   | $SE^3$ | CT      | Р                    | С     | P*C   |  |  |  |  |
| UUN              | 28.40   | 55.44                          | 60.90  | 30.28  | 35.21  | 3.33   | < 0.001 | < 0.001              | 0.129 | 0.937 |  |  |  |  |
| UUN/NI           | 0.41    | 0.48                           | 0.45   | 0.27   | 0.36   | 0.03   | 0.597   | < 0.001              | 0.254 | 0.028 |  |  |  |  |
| SUN              | 6.70    | 13.28                          | 12.51  | 8.15   | 10.88  | 0.61   | < 0.001 | < 0.001              | 0.118 | 0.007 |  |  |  |  |
| NMIC             | 58.71   | 83.50                          | 78.46  | 76.55  | 70.03  | 5.19   | 0.004   | 0.158                | 0.285 | 0.890 |  |  |  |  |
| EMS <sup>4</sup> | 156.68  | 131.92                         | 131.45 | 131.75 | 128.81 | 6.38   | 0.001   | 0.827                | 0.792 | 0.847 |  |  |  |  |

<sup>1</sup>Control = mineral mixture only; HPHC = high protein and high carbohydrate supplement; HPLC = high protein and low carbohydrate supplement; LPHC = low protein and high carbohydrate supplement; LPLC = low protein and low carbohydrate supplement; <sup>2</sup>CT = non-supplemented versus supplemented, P = effect of protein amount, C = effect of carbohydrate amount, P\*C = effect of interaction of protein and carbohydrate; <sup>3</sup>standard error; <sup>4</sup>g of microbial CP kg<sup>-1</sup> NTD.

The control bulls had lower (p < 0.05) body weight gain (BWG) and empty body weight gain (EBWG) than the supplemented bulls. However, differences were not found (p > 0.05) between supplemented bulls (Table 4). Supplemented bulls had greater (p < 0.05) dressing percentages (DP) than control bulls. Bulls that received highcarbohydrate supplements (HPHC and LPHC) had greater (p < 0.05) DP. The nonsupplemented bulls had less (p < 0.05) muscle, fat and bone tissue in physical composition of the carcasses than supplemented bulls when these tissues were expressed in kg. However. differences were not observed (p > 0.05) when expressed in the percent of empty body weight (EBW). Additionally, non-supplemented bulls had greater (p > 0.05) organ-viscera proportions in the EBW (Table 4). Although highcarbohydrate supplements increased (p < 0.05) the proportion of fat tissue in the EBW, no differences (p > 0.05) were found in the proportion of muscle and bone tissue, as well as in the organ-viscera mass in the EBW between the supplements.

Differences were not found (p > 0.05) in the proportion of liver and large intestine in EBW between the nutritional plans. The non-supplemented group had a greater (p < 0.05)

proportion of abomasum and small intestine than the supplemented. Although there were no differences (p > 0.05) in the reticulum-ruminal supplemented mass between and nonanimals, high-carbohydrate supplemented supplements (HPHC and LPHC) resulted in a lower (p < 0.05) proportion of rumen-reticulum (Table 5). The bulls that received highcarbohydrate supplements had greater (p < 0.05) proportions of intermuscular fat and total fat, while differences were not observed (p > 0.05) in the proportion of visceral and subcutaneous fat (Table 5).

#### Discussion

Supplemented cattle in pasture conditions may have increased energy intakes by improving energy extraction from the pasture and/or directly from supplementation (VALENTE et al., 2011b). Although high-protein and high-carbohydrate supplements (HPHC) tended to increase the ME intake (Table 2), it is seems that energy supplementation (highcarbohydrate supplements) reduced fiber digestion in the rumen, which has more of an effect at lower levels of dietary protein (KLEVESAHL et al., 2003; SOUZA et al., 2010). Thus, energy supplementation might not necessarily be associated with significant increases in the energy supply to cattle.

Table 4. Effect of nutritional plans on body weight gain (BWG), average daily gain (ADG), empty body weight gain (EBWG), muscle tissue, fat tissue, bone tissue, dressing percentage (DP) and organ-viscera.

| Itens                      |         | Nu    | tritional plans <sup>1</sup> |       |       |                 | P-value <sup>2</sup> |       |       |       |
|----------------------------|---------|-------|------------------------------|-------|-------|-----------------|----------------------|-------|-------|-------|
| Itens                      | Control | HPHC  | HPLC                         | LPHC  | LPLC  | SE <sup>3</sup> | CT                   | Р     | С     | P*C   |
|                            |         | kg    |                              |       |       |                 |                      |       |       |       |
| BWG                        | 195.4   | 285.9 | 266.4                        | 277.4 | 281.1 | 13.7            | < 0.001              | 0.823 | 0.575 | 0.411 |
| ADG                        | 407.0   | 596.0 | 555.0                        | 578.0 | 586.0 | 28.6            | < 0.001              | 0.823 | 0.575 | 0.410 |
| EBWG                       | 163.6   | 257.6 | 232.6                        | 251.6 | 246.3 | 11.2            | < 0.001              | 0.736 | 0.200 | 0.395 |
| Muscle tissue <sup>4</sup> | 118.9   | 165.0 | 150.5                        | 156.6 | 159.2 | 10.0            | 0.004                | 0.992 | 0.562 | .0408 |
| Fat tissue <sup>4</sup>    | 19.5    | 28.0  | 21.4                         | 30.6  | 26.3  | 2.3             | 0.014                | 0.119 | 0.032 | 0.622 |
| Bone tissue <sup>4</sup>   | 36.7    | 46.3  | 44.9                         | 44.2  | 43.6  | 3.1             | 0.033                | 0.585 | 0.752 | 0.888 |
|                            |         |       | % of BW                      |       |       |                 |                      |       |       |       |
| DP                         | 53.1    | 57.9  | 55.2                         | 57.5  | 55.8  | 0.7             | < 0.001              | 0.935 | 0.011 | 0.546 |
|                            |         |       | % of EBW                     |       |       |                 |                      |       |       |       |
| Muscle tissue <sup>4</sup> | 41.5    | 43.5  | 42.7                         | 42.4  | 43.1  | 0.8             | 0.117                | 0.644 | 0.936 | 0.337 |
| Fat tissue <sup>4</sup>    | 6.8     | 7.5   | 6.1                          | 8.3   | 7.1   | 0.5             | 0.513                | 0.104 | 0.034 | 0.838 |
| Bone tissue <sup>4</sup>   | 12.9    | 12.2  | 12.7                         | 11.9  | 11.7  | 0.4             | 0.131                | 0.113 | 0.787 | 0.390 |
| Organ-Viscera              | 14.4    | 13.0  | 13.2                         | 13.3  | 13.5  | 0.3             | 0.003                | 0.306 | 0.506 | 0.960 |

<sup>1</sup>Control = mineral mixture only; HPHC = high protein and high carbohydrate supplement; HPLC = high protein and low carbohydrate supplement; LPHC = low protein and high carbohydrate supplement; LPLC = low protein and low carbohydrate supplement;  $^{2}CT$  = non-supplemented versus supplemented, P = effect of protein amount, C = effect of carbohydrate amount, P\*C = effect of interaction of protein and carbohydrate; <sup>3</sup>standard error; <sup>4</sup>in physical composition.

| Table 5. Effect of nutritiona | l plans on organs and tissues ma | ass (% of empty body weight). |
|-------------------------------|----------------------------------|-------------------------------|
|-------------------------------|----------------------------------|-------------------------------|

| Itemes            |         | Nutritional plans <sup>1</sup> |       |       |       |                 |       | P-value <sup>2</sup> |       |       |  |
|-------------------|---------|--------------------------------|-------|-------|-------|-----------------|-------|----------------------|-------|-------|--|
| Items             | Control | HPHC                           | HPLC  | LPHC  | LPLC  | SE <sup>3</sup> | CT    | Р                    | С     | P*C   |  |
| Liver             | 13.44   | 12.60                          | 12.30 | 12.40 | 13.12 | 0.4             | 0.108 | 0.496                | 0.633 | 0.251 |  |
| Abomasum          | 3.45    | 2.56                           | 2.98  | 3.04  | 3.03  | 0.2             | 0.047 | 0.267                | 0.382 | 0.360 |  |
| Reticulum-ruminal | 17.81   | 15.80                          | 17.27 | 15.97 | 17.25 | 0.5             | 0.056 | 0.890                | 0.022 | 0.866 |  |
| Small intestine   | 16.13   | 14.37                          | 13.11 | 14.53 | 13.25 | 0.8             | 0.023 | 0.855                | 0.141 | 0.991 |  |
| Large intestine   | 6.61    | 5.87                           | 6.13  | 6.10  | 6.45  | 0.1             | 0.264 | 0.462                | 0.423 | 0.909 |  |
| Total fat         | 3.65    | 4.05                           | 3.25  | 4.35  | 3.75  | 0.3             | 0.510 | 0.152                | 0.018 | 0.711 |  |
| Visceral fat      | 0.67    | 0.65                           | 0.59  | 0.63  | 0.64  | 0.7             | 0.621 | 0.789                | 0.719 | 0.617 |  |
| Subcutaneous fat  | 0.66    | 0.65                           | 0.48  | 0.74  | 0.53  | 1.1             | 0.638 | 0.563                | 0.118 | 0.828 |  |
| Intermuscular fat | 2.33    | 2.74                           | 2.19  | 2.99  | 2.58  | 1.6             | 0.111 | 0.056                | 0.008 | 0.646 |  |

 $^{1}$ Control = mineral mixture only; HPHC = high protein and high carbohydrate supplement; HPLC = high protein and low carbohydrate supplement; LPHC = low protein and high carbohydrate supplement; CT = non-supplemented versus supplemented, P = effect of protein amount, C = effect of carbohydrate amount, P\*C = effect of interaction of protein and carbohydrate; <sup>3</sup>standard error.

When protein is limiting, protein supplementation can increase the energy extraction from roughage feed, especially when associated with carbohydrates (SOUZA et al., 2010). As expected, the higher protein intake through supplementation accounted for higher daily protein intake. However, this was not observed with ME intake. There was a reduction in the ME and NE of the diet when the intake of digestible protein was in excess of what is required. The effect of excess protein has been attributed to the energy costs of urea synthesis in the liver, and energy utilization is associated with amino acid catabolism (REYNOLDS et al., 2011).

With low-protein diets, protein deposition will increase linearly with increases in protein intake, until a point is reached where energy becomes most limiting and the animal no longer responds, or responds with a very low efficiency to additional increases in protein supply (SCHROEDER; TITGEMEYER, 2008). Although high-protein supplements could increase the protein intake in supplemented animals, an increase in RP has not been observed. However, when high levels of protein and energy were used together (HPHC) in the supplement, the RP tended to increase (Table 2). Moreover, when the supply of digestible protein exceeds the animals' requirements (low energy supply) additional protein intake does not affect protein deposition (SCHROEDER; TITGEMEYER, 2008).

Studies with lambs (CHOWDHURY et al., 1997) and steers (CHOWDHURY et al., 1990) have demonstrated that N retention increases due to a rise in N supply, even when sub-maintenance levels of energy are supplied. However, in this work a linear increase in N retention with increasing N intake was not observed, suggesting that other effects besides N intake may affect N retention. Schroeder et al. (2006) observed that infusions of energy substrates increased N retention. Moreover, Raggio et al. (2006) found infusion of either glucose into the duodenum or propionate into the rumen increased milk protein output, reducing oxidation of AA and improving overall efficiency of AA use.

In general, as N intake increases, N excretion increases (COLE et al., 2005; ERICKSON; KLOPFENSTEIN, 2010). However, Firkins and Reynolds (2005) identified that N excretion is not related only to N intake, but to other parameters such as the energy content of the diet. Thus, the apparent N utilization has been reported to increase by supplementation with high digestibility feeds (COHEN et al., 2006).

The lower efficiency of protein use in high-protein supplements observed in this study might be due to an excess of protein in relation to the requirement or unbalance between protein and energy in the diet, confirmed by greater excretion of urinary urea N and serum urea N (Table 3). Archibeque et al. (2007) noted that steers receiving N levels (9-14%) had increased ME intake and retained energy, heat production and N excretion with increased N intake. Cattle can use non-limiting AAs to produce energy when protein and energy are unbalanced or there is an excess of non-limiting AAs (AWAWDEH et al., 2006). However, the use of AA energy is less efficient than carbohydrate energy (SCHROEDER; TITGEMEYER, 2008). In addition, Castillo et al. (2001) concluded that it was possible to improve the efficiency of N use by providing balanced diets to meet the animals' requirements.

Illius and Jessop (1996) described a conceptual model of metabolic constraints on intake, showing the response of an animal aiming to achieve its nutrient requirements from diets with various ratios of metabolizable protein to metabolizable energy. However, when cattle use the pasture as a basal diet, the supplement may change the effective availability of protein and energy from roughage feed by associative effects (DIXON; STOCKDALE 1999). In the present study, there was correlation between the level of protein and carbohydrate in the supplement and the ratio of metabolizable protein intake and digestible energy intake (MP: E). In lowprotein supplements, the increase in carbohydrate level decreased the MP: E ratio, but in high-protein supplements the increase in carbohydrate levels increased the MP: E ratio; this may be due to improved microbial growth and, consequently, a better protein and energy supply to the animal when young bulls are fed with supplements containing high levels of protein and energy (HPHC). Synchrony between the supply of N and energy to ruminal microorganisms should improve the capture of N, improving the efficiency of ATP use for microbial growth, increasing the nutrient supply to the host animal, and thereby improving overall animal performance (KREHBIEL et al., 2008).

The efficiency of feed use by ruminants is a function of the microbial and animal metabolism. Lazzarini (2011) studying nutritional characteristics of steers grazing on low quality pastures (8% CP) and receiving supplements observed an increase in nitrogen balance only when amide and nitrogen were supplemented together in comparison with supplementation with amide or nitrogen, even with no differences in microbial protein production because this effect is due to improved metabolizable protein use. Similar to this study, where the supplement increased the efficiency of retained energy and protein, Oliveira et al. (2011) found a greater efficiency of carcass deposition with 2% of BW concentrate diet than 1% of BW concentrate diet in the feedlot. However, animals that received highprotein supplements had higher PM: E ratios and a lower efficiency of protein use. It can be suggested that in these treatments an inappropriate PM: E ratio was less efficient for protein accretion.

The reticulo-ruminal mass may increase more from a roughage diet than a high-concentrate diet (MCCURDY et al., 2010). Therefore, as expected, high-carbohydrate supplements decreased the reticulo-ruminal mass in reason of offer an environment with lower proportion of roughage feed to rumen. Although a small intestinal mass should be related to the rate of passage and nutrient flow to the small intestine (MCCURDY et al., 2010), in the present work the greater small intestinal mass in non-supplemented bulls (Table 5) may be due to lower empty body weight gain and lower DP, and consequently a greater proportion of the gastrointestinal tract in the body (Table 4).

Diets containing concentrate increased the fat deposition in the carcasses (COSTA et al., 2013; MARTINS et al., 2011). Although highcarbohydrate supplements did not increase the MEI, an increase in fat deposition was observed. The high-carbohydrate diet resultedi in higher levels of propionate from ruminal fermentation and/or more glucose absorption in the small intestine from non-degradable starch than the low carbohydrate diet (SCHOONMAKER et al., 2010); this would, in turn, promote insulin production and therefore stimulate lipogenesis (SCHOONMAKER et al., 2003).

#### Conclusion

Supplying 25 to 50% of CP and 15 to 30% of the DE requirements by concentrate to young bulls on tropical pastures increases the intake and retention of protein and energy without changing the efficiency of retention. Although the ratio of CP and energy in the supplement does not change the CP retained, high-carbohydrate supplements (30% of DE requirement) increased fat tissue deposition and the amount of energy retained.

## Acknowledgements

The authors wish to thank the Conselho Nacional de Pesquisa e Desenvolvimento Científico e Tecnológico (CNPq) for financial support.

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Received on July 9, 2013. Accepted on January 22, 2014.

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