**Original Article** 

## PERFORMANCE, BODY COMPOSITION AND NET REQUIREMENTS OF PROTEIN AND ENERGY FOR WEIGHT GAIN IN STEERS SUPPLEMENTED WITH OR WITHOUT ADDITION OF LIPIDS

DESEMPENHO, COMPOSIÇÃO CORPORAL E EXIGENCIAS LÍQUIDAS DE PROTEÍNA E ENERGIA PARA GANHO DE PESO EM NOVILHOS SUPLEMENTADOS COM OU SEM ADIÇÃO DE LIPÍDIOS

## Tiago Cunha ROCHA<sup>1\*</sup>; Carlos Augusto de Alencar FONTES<sup>2</sup>; Elizabeth Fonsêca PROCESSI<sup>2</sup>; Laila Cecília Ramos BENDIA<sup>2</sup>; Clóvis Carlos SILVEIRA FILHO<sup>2</sup>; Cláudio Teixeira LOMBARDI<sup>2</sup>; Paulo Roberto Silveira PIMENTEL<sup>3</sup>; Ronaldo Lopes OLIVEIRA<sup>3</sup>; Leilson Rocha BEZERRA<sup>4</sup>

1. State University of the Tocantina Region of Maranhão, Imperatriz, Maranhão, Brazil; 2. State University of North Fluminense Darcy Ribeiro, Department of Animal Science, Campos dos Goytacazes, Rio de Janeiro, Brazil; 3. Federal University of Bahia, Department of Animal Science, Salvador, Bahia, Brazil; 4. Federal University of Piauí, Campus Professora Cinobelina Elvas, Department of Animal Science, Bom Jesus, Piauí, Brazil. tiagoticuro@yahoo.com.br

**ABSTRACT:** This research aimed to evaluate the performance, body composition and net requirements for protein and energy in grazing Nellore steers supplemented with or without addition of lipids. Twenty-eight steers,  $301 \pm 5.8$  kg BW, were used in the experiment. The comparative slaughter method was used, with four steers used as references. The remaining 24 steers were randomly distributed into four groups, during 180 days, according to treatment: only Panicum maximum cv. Mombasa grass; Mombasa grass with concentrated supplementation based on soybean meal; Mombasa grass with concentrate containing lipids from soybean oil; and Mombasa grass with concentrate containing lipids derived from soybean grains. The total DMI of the steers fed only the pasture did not differ (P > 0.05) from the average intake of the treatments with or without addition of lipids. There were no differences (P > 0.05) between treatments for total daily gain, carcass and non carcass for protein, energy and fat. The requirements of protein and energy did not differ (P > 0.05) among the treatments. When the pasture is of good quality, supplementation does not alter the body composition or the net requirements of protein and energy for weight gain. The supplementations with higher concentrations of lipids (oil and grain) were not enough to promote greater gains in fat and energy in carcass.

KEYWORDS: Carcass. Gastrointestinal tract. Panicum maximum. Ruminants.

## **INTRODUCTION**

The NRC (1984, 1996, 2000), ARC (1980) and CSIRO (1990, 2007) are commonly used to determine nutritional requirements in beef cattle systems. However, inadequate nutritional requirements of animals and differences in the nutritional values of feed are easily observed when the results provided by these systems feeding standards are compared to those observed in Brazilian production conditions (GONZAGA et al., 2011).

Additionally, in animal production systems in subtropical and semiarid regions, research is increasingly being conducted to identify auxiliary models to assist in decision making, especially with regard to the use of concentrated supplementation because of the high costs of food such as soybeans (BEZERRA et al., 2013). Soybean is one of the vegetables that contain protein with reasonably balanced amino acid composition, and it is a protein source used to balance diets for cattle (TEDESCHI et al., 2015). Many soybean byproducts are used in ruminant diets, such as the bran, meal, cake and oil. Thus, the comparison of these products becomes necessary because the byproduct used, as well as the amounts to be used in the diet, significantly influences the performance of animals and the final weight to be sent to the market, and supplementation of soybeans byproducts is a cheaper approach to reduce production cost (RODRIGUES FILHO et al., 2013; SILVA-MARQUES et al., 2015).

Thus, a comparison between protein and energy requirements in animals fed on tropical pastures supplemented with no lipids may help in selecting the best byproduct to be used, providing valuable information on the characteristics of greatest concern for this production system, where soybean protein concentrate has been supplied to beef cattle for a long time.

Almost all studies on the nutritional requirements of cattle are conducted under constrained conditions. Therefore, information on pasture-raised animals is needed (CSIRO, 1990, 2007). Thus, to optimize animal performance with balanced diets, information on the nutritional requirements for the various functions and the various levels of performance are needed so that the animals can have their nutritional needs met and can achieve their genetic potential for production.

The objective of this study was to evaluate the performance, body composition and net requirements of protein and energy for weight gain in Nellore steer supplemented with or without addition of lipids.

## MATERIAL AND METHODS

## **Ethical considerations**

This study was performed in strict accordance with the recommendations in the Guide for the National Council for Animal Experiments Control. The protocol was approved by the Committee on the Ethics of Animal Experiments of the State University of North Fluminense, Rio de Janeiro State, Brazil (Permit Number: 168-2012).

## **Experimental area**

The research was conducted in Campos dos Goytacazes, Rio de Janeiro (Brazil, 21°42′ S, 41°19′ W, 13 m alt) in the transition from the dry to rainy period of the year, in an area of 9.0 ha of *Panicum maximum* cv. Mombasa grass. The experimental area was divided into two blocks (replicate areas), each 4.5 hectares, equipped with an irrigation sprinkler system grid and divided into 18 paddocks of 0.25 ha.

Due to the reduced rainfall in the period, pasture irrigation was performed at intervals of seven days. The pastures were fertilized by the application of 100 kg N and 50 kg  $K_2O$ /ha/year was applied in each paddock before the mowing of vegetation, which occurred early in the growing season (first grazing cycle); this was repeated fertilization outside of the bulls of each paddock before the third grazing cycle. The animals were adapted to the experimental procedures and supplementation during a period of 36 days, which represented a grazing cycle.

The pasture was managed under a rotational stocking system, with periods of occupation and rest of two and 34 days, respectively. The duration of the rest period was established based on previous results from the same site and according to the production of biomass of Mombasa grass during the dry season (PALIERAQUI et al., 2006). The trial included five complete grazing cycles, totaling 180 days. A variable stocking rate (*put* and *take*) was adopted to maintain the supply of biomass of green leaf blades at 4 kg DM 100 kg<sup>-1</sup> body weight of the animals. Eventually regulator animals (put and take) that were selected in advance in order to be similar to the experimental animals according to age, sex and body weight were used.

## Animals and treatments

The experimental group included 28 Nellore steers, with an initial average weight of  $300.6 \pm 5.8$  kg; four of these served as the reference group and were slaughtered at the beginning of the experiment to allow estimates of the empty body weight and the initial body composition for the others.

The remaining 24 animals were randomly distributed into four groups of six animals. The groups were allocated at random to each replicate area of each of the following treatments: control only Mombasa grass; soybean meal (animals Mombasa grass with concentrate receiving supplementation based on soybean meal); soybean oil (animals receiving Mombasa grass with concentrate supplementation based on soybean oil); and soybean grain (animals receiving Mombasa grass with concentrate supplementation based on soybeans). Different sources of soybeans were provided (meal, oil and grain) for these have different concentrations of lipids and degradation rates.

## Experimental feeds and chemical analyses

*Panicum maximum* intake was *ad libitum*. The supplements were provided in the proportion of 0.75 kg concentrate 100 kg<sup>-1</sup> of BW in DM. Assuming ad libitum forage intake and the total DMI corresponding to 2.5 kg 100 kg<sup>-1</sup> BW day<sup>-1</sup> (NRC, 1996), the diet would consist of 0.75 kg of concentrate and 1.75 kg of roughage, per 100 kg BW, corresponding to the ratio of forage: concentrate of 70:30 in DM. The steers received supplements in individual stalls at 0900 to 1300 h and after returned to the pasture where they remained the rest of the day. Animals in the control treatment were subjected to the same handling, receiving only mineral supplement during the period of stay in the stall.

At the end of each grazing cycle, the animals were weighed, fasted for 16 hours with access to water, and the amount of individually supplied supplement was adjusted based on the Performance...

refused feed to adjust of the intake. The mineral mixture was made available in individual stall daily.

Samples of ingredients, refused feed and feces were pre-dried at 55 °C for 72 hours, ground with a Willey mill (Tecnal, Piracicaba City, São Paulo State, Brasil) with a 1 mm sieve, stored in air-tight plastic containers (ASS, Ribeirão Preto City, São Paulo State, Brasil), and sealed properly until the laboratory analysis for determining the DM (Method 967.03 – AOAC, 1990), ash (Method

942.05 – AOAC, 1990), CP (Method 981.10 – AOAC, 1990), and EE (Method 920.29 – AOAC, 1990). To determine the NDF content, the method of Van Soest et al. (1991) was used with the modifications that were proposed in the Ankon device manual (Ankon Technology Corporation, Macedon, New York, US). The proportion of ingredients and the chemical composition are shown in Table 1.

	Treatments								
Ingredients g kg <sup>-1</sup>	Mombasa grass	Soybean Meal	Soybean Oil	Soybean grain					
Soybean grain	-	-	-	548					
Corn bran	-	563	437	434					
Soybean Oil	-	-	103	-					
Mixed minerals <sup>a</sup>	19.4	19.7	19.0	18.3					
Soybean Meal	-	418	440	-					
Chemical composition (g k	$g^{-1}$ DM)								
Dry Matter <sup>b</sup>	231	284	285	285					
Crude Protein	100	146	146	146					
Ether Extract	09.9	16.0	45.2	45.0					
Neutral detergent fiber	660	526	522	523					
Total digestible nutrients	510	606	640	609					
Calcium	06.8	07.4	07.4	07.3					
Phosphorus	02.1	02.5	02.7	02.8					

<sup>a</sup>Composition (/kg): calcium - 11 g; phosphorus - 80 g; sodium - 195 g; magnesium - 17.3 g; sulfur - 20 g; iodine - 301 mg; iron - 487 mg; selenium - 40 mg; cobalt - 100 mg; manganese - 1.000 mg; fluoride - 800 mg; copper - 1500 mg; and zinc - 3000 mg. <sup>b</sup>Organic matter basis

#### Feed intake and digestibility determination

For the second and fourth grazing cycles, the intake was estimated using double-indicator method (chromium oxide  $(Cr_2O_3)$  and indigestible neutral detergent fiber – iNDF).

The estimated grass intake was calculated according to the following equation, DMI g day<sup>-1</sup>= (FE iNDFfecal DMsupl × \_ х iNDFsupl)/iNDFforage + DMsupl where, DMI = dry matter intake (g day<sup>-1</sup>); FE = fecal excretion (g/day); iNDF<sub>fecal</sub> is fecal content of indigestible NDF (g g<sup>-1</sup>); DM<sub>Supl</sub> is intake of supplements (g DM day<sup>-1</sup>); iNDF<sub>Sup1</sub> supplement content of indigestible NDF (g/g); and iNDFforage is content of indigestible NDF (g g<sup>-1</sup>) in feces (iNDF feces), supplemets (iNDF supl), and forage (i NDF forage), respectively.

To estimate the fecal output, chromic oxide was used as an external indicator. The chromic oxide was provided in a 10 g dose, two portions of 5 g, wrapped in paper cartridges and administered by gavage at 0900 and 1700 hours for 13 days; seven days were used for the adaptation and regulation of the excretion of the flow marker, and six days were used for the collection of stool. The feces were collected directly from the rectum once per day during indicator management and stored in a cold chamber at  $-10^{\circ}$ C. The stool samples were analyzed with colorimetric method, which included nitroperchloric digestion (KIMURA; MÜLLER, 1957) and a UV-visible spectrophotometer reading at 440 nm (SPEKOL UV device visible).

For the determination of the internal indicator, indigestible neutral detergent fiber (iNDF), samples of fodder concentrates and feces were incubated in the rumens four fistulated steers for 240 hours, and the residue was assumed to be indigestible.

The individual supplement intake was determined daily, with the quantities offered and the leftovers being weighted and noted. The supplements were sampled weekly, and proportional samples of individual remains were collected daily. Nutrient intake was estimated from the chemical composition of the refused supplement and feces and the dry matter intake and fecal excretion.

## Performance, slaughter and carcass

The steers were weighed at the beginning and end of the experiment to determine the average daily weight gain (ADG). After deprived of solid feed for a period of 16 hours the animals were weighted and slaughtered by captive bolt and exsanguination, followed by hide removal and evisceration, without electrical stimulus. The carcasses were identified, washed, and divided into halves, according to the federal standards prescribed for the humane slaughter of cattle (BRAZIL, 2000). After slaughter, the gastrointestinal tract (GIT) of each animal was emptied and washed, and like the other organs, weighed individually. The weights of the GIT were added to the weights of the other body parts (carcass, head, leather, tail, feet and blood) to determine the empty body weight (EBW). The relation obtained between the EBW and shrunk body weight (SBW) and carcass yield of the reference animals were used to estimate the EBW, initial weight of the carcasses experimental animals, and their daily weight gain empty body (DWGEB) and carcass. The tissue components (muscle, fat and bone) were determined in the sections taken from the left half carcass (HH section). The proportions of muscle, adipose and bones tissue were estimated based on the proportions of these components in the section HH according to equations proposed by Hankins and Howe (1946): Muscle Y = 16.08 +0.80 X; Fat Y = 3.54 + 0.80; and Bone Y = 5.52 X + 0.57 X wherein: X is percentage of the component in HH section. Energy content in the empty body of steers was estimated from the body mass of protein and fat and their caloric equivalents (ARC, 1980): 5.6405 kcal  $g^{-1}$  (protein) and 9.3929 kcal  $g^{-1}$  (fat).

For the animals slaughtered at the beginning of the experimental period (reference group), the mean ratio of slaughtered body weight SBW EBW<sup>-1</sup> and body energy content (kcal g EBW<sup>-1</sup>) were calculated within each treatment, which were used to predict the EBW and energy content and body of animals at the beginning of the experimental period. The energy retained in each animal during the experimental period was calculated as the difference between the final body energy and initial body energy, which was predicted from the reference group.

In the analysis of the chemical composition of body tissue from each animal, the samples were first dried at 105 °C until constant weight was obtained to determine the fat dry matter (FDM). FDM was treated with petroleum ether to extract part of the fat, thus yielding a pre-defatted dry matter (PFDM), as described by Kock and Preston (1979). Subtracting the PFDM of FDM yielded fat extracted in the pre-degreasing. The total fat content of the sample was obtained by adding the fat removed in the pre-degreasing with residual ether extract. From the ground PFDM, were analyzed nitrogen, ether extract and ashes, using the methods 981.10, 920.29, 942.05, respectively (AOAC, 1990). Once the PFDM content of the body tissues was determined, the determination of the composition of natural matter was possible.

Due to the influence of the internal organs of the carcass on the requirements of maintenance and gain efficiency, the weights of these components were determined to quantify the weight gain and fat partition energy and protein deposited on them compared to the carcass and EBW. The weight of the GIT was determined by adding up the weights of its components, stomachs and intestines. Weight changes and protein, fat and energy content in the metabolically active organs such as the liver, spleen, kidneys, lungs and heart, were also determined.

According to the comparative slaughter methodology (LOFGREEN; GARRET, 1968), initial EBW of each animal and the initial protein and fat contents of the experimental animals were estimated from the body composition of the reference animals slaughtered at the beginning of the experiment.

Allometric regression equations were set up to describe the nonlinear increase in the contents of the body components: fat, protein, energy, carcass, noncarcass, GIT and organs due to the increase of weight of the animals of each treatment; these depended on the following model (ARC, 1980): Yij =  $\square * Xij^{\square} + \epsilon ij$ , where Yij represents the total contents of the different body parts (kg or Mcal) in the empty body of the animal j, which belongs to treatment i; Xij is the EBW;  $\square$  and  $\square$  are equation parameters; and  $\epsilon ij$  is the random error with zero mean and variance equal to  $\square^2$ 

Derivations of the regression equations of body fat, protein, energy, carcass, noncarcass, GIT and organs content, depending on the EBW, yielded the gain of the prediction equations of the aforementioned components for the weight gain unit, EBW:  $Y' = \square * \square * X^{(\square -1)}$ , where Y' is gain (kg or Mcal) of the body component per unit of EBW gain;  $\square$  and  $\square$  are parameters of the prediction equations of body content; and X = EBW kg.

Based on the equations obtained, were estimated from the different body parts, and weight gains were estimated for each component per kg of EBW gain for animals of different EBW. For several components, EBW were used to calculate the corresponding values of shrunk body weight (SBW), which were estimated using the regression equation proposed by Fontes et al. (2005), namely: SBW = 1.08 \* (EBW + 19.61).

#### **Statistical Analysis**

Statistical analyses were performed using the following model:  $Y_i = \mu + T_i + \varepsilon_i$ , using the steer Nellore as the experimental unit, where  $Y_{ij}$  = observation concerning the animal, receiving treatment i in replicate (n = 6);  $\mu$  is average;  $T_i$  = treatment effect i, where i is Control (Mombasa grass), Mombasa grass (MG) + Soybean Meal Concentrate (SM), Mombasa grass + Soybean Oil Concentrate (SO) and Mombasa grass + Soybean Grain Concentrate (SG); and  $\varepsilon_i$  = random error.

In the statistical analysis, we used the PROC MIXED procedure of SAS (2014). Treatments were compared using three orthogonal contrasts. The first comparison made among the three treatments involved supplementation with sources of soybeans and the Mombasa grass. The second comparison was between the treatment supplemented with soybean meal versus those with soybean oil and soybeans (SM versus SO and SG), and the third comparison was between the supplementation with soy oil versus soybeans (SO versus SG).

PROC NLIN (SAS) was used to fit the allometric equations of the type  $Y = \Box * X^{\Box}$ , which described the relationship between the protein, fat and energy contents as functions of EBW and body

components (carcass, noncarcass, GIT and organs) as functions of EBW.

For all variables, testing was performed to verify the equality of the parameters and the identity of the adjusted models for the four treatments as follows: 1) without limitation, assuming different values for the four treatments for  $\alpha$  and  $\Box$  parameters; 2) with restriction, while assuming, for the four treatments, different values for the parameter d; 3) with restriction, while assuming, for the four treatments, different values for the four treatments, different values for the parameter d and a common value for the parameter d and a common value for the parameter d and a common value for a parameter; and 4) with restriction, while assuming, for the four treatments, only common values for a and d parameters;

The test likelihood ratio was used to assess the identity of regression models and to verify equal parameters, by using an F-test approach, according to Bates and Watts (2007) and Regazzi and Silva (2004), to provide a lower type I error than achieved with the chi-square approach. When there is identity between templates or equal parameters, the model will always choose the least number of parameters.

#### RESULTS

The total dry matter intake of the animals fed only the Mombasa grass did not differ (P = 0.624) from the average intake of the animals supplemented with or without addition of lipids (8.19 kg day<sup>-1</sup>) (Table 2).

**Table 2.** Means, standard deviations (±) and *P*-values of total intake, pasture intake, concentrate intake, body weight (fasting), body weight (empty) carcass, gastrointestinal tract (GIT) and metabolically active organ (liver, spleen, kidneys, lungs and heart) weight of beef cattle fed supplementation with soybean byproduct concentrates

byproduct	byproduct concentrates									
	Treatments				<i>P</i> -value	P-value				
Variables	Mombasa grass	Soybean Meal	Soybean Oil	Soybean grain	MG vs S	$\frac{\text{SMC}}{\text{SO} + \text{SG}}$	SO vs SG			
Dry matter intake (k	(g)			*						
Total	7.78±0.6	8.79±0.6	7.82±0.6	7.96±0.6	0.62	0.31	0.89			
Pasture	7.78±0.6	5.82±0.6	5.73±0.6	6.08±0.6	0.03*	0.92	0.73			
Concentrate	-	2.97±0.17	2.09±0.17	$1.88 \pm 0.17$	-	<0.01*	0.44			
Body weight (fasting) (kg d <sup>-1</sup> )	0.74±0.07	0.73±0.07	0.73±0.07	0.77±0.07	1.00	0.77	0.66			
Body weight (empty) (kg d <sup>-1</sup> )	0.68±00.6	0.74±0.06	0.73±0.06	0.73±0.06	0.43	0.87	1.00			
Carcass weight (kg d <sup>-1</sup> )	0.44±0.04	$0.49 \pm 0.04$	0.47±0.04	0.47±0.04	0.44	0.66	0.93			
Gastrointestinal tract (g d <sup>-1</sup> )	3.56±0.40	2.83±0.40	2.46±0.40	2.91±0.40	0.11	0.79	0.46			
Organs (g d <sup>-1</sup> )	2.45±0.40	2.36±0.40	2.75±0.40	2.59±0.40	0.79	0.54	0.78			

P - value (<0.05) indicates the probability of obtaining the observed difference between feeding strategy if the null hypothesis is true

As might be expected, the animals that were fed exclusively on Mombasa grass + showed higher pasture intake (P = 0.031). Animals that received soybean meal showed an increase in intake (P = 0.001) of 34% compared with animals receiving supplementation with soybean oil concentrate and soybeans. There were no differences in the mean weight gain of the gastrointestinal tract (P = 0.110) and the mean gain for metabolically active organs (P = 0.790) among supplementation with or without addition of lipids. With regard to the forage intake of animals, supplementation with any of the concentrates reduced (P > 0.05) forage intake.

The orthogonal contrasts revealed no differences (P > 0.05) between treatments in the average daily body weight gain while fasting, EBW gain, and carcass weight gain and average daily carcass gain. There were no differences (P > 0.05) between treatments for total daily gain, carcass and noncarcass for protein, energy and fat (Table 3). The animals had total average daily gains of 100 g day<sup>-1</sup> of protein, 210 g day<sup>-1</sup> of fat and 2.63 Mcal day<sup>-1</sup> of energy.

**Table 3.** Means, standard deviations  $(\pm)$  and *P*-values of protein, fat, energy gain total, in carcass and noncarcass of beef cattle fed supplementations with soybean byproduct concentrates

	Treatments		, end	V I	<i>P</i> -value			
Variables	Mombasa grass	Soybean Meal	Soybean Oil	Soybean grain	MG vs S	SMC vs SO + SG	SO SG	VS
Total								
Protein gain (kg day <sup>-1</sup> )	0.11±0.01	0.10±0.01	0.10±0.01	0.10±0.01	0.43	0.95	0.60	
Fat gain						0 - 4	0.0 <b>7</b>	
$(\text{kg day}^{-1})$	$0.20 \pm 0.03$	$0.22 \pm 0.03$	$0.22 \pm 0.03$	0.21±0.03	0.50	0.74	0.85	
Energy gain (Mcal day <sup>-1</sup> )	2.41±0.31	2.69±0.31	2.72±0.31	2.72±0.31	0.33	0.92	0.99	
Carcass Protein gain (kg day <sup>-1</sup> )	0.07±0.01	0.06±0.01	0.06±0.01	0.06±0.01	0.39	0.77	0.60	
Fat gain (kg day <sup>-1</sup> )	0.13±0.02	0.15±0.02	0.14±0.02	0.13±0.02	0.64	0.66	0.72	
Energy gain (Mcal day <sup>-1</sup> )	1.58±0.24	1.76±0.24	1.76±0.24	1.73±0.24	0.45	0.94	0.90	
Noncarcass Protein gain (kg day <sup>-1</sup> )	0.04±0.00	0.04±0.00	0.04±0.00	0.04±0.00	0.66	0.67	0.72	
Fat gain (kg day <sup>-1</sup> )	$0.06 \pm 0.00$	$0.07 \pm 0.00$	$0.07 \pm 0.00$	$0.07 \pm 0.00$	0.35	0.97	0.79	
Energy gain (Mcal day <sup>-1</sup> )	0.82±0.09	0.93±0.09	0.96±0.09	0.99±0.09	0.27	0.71	0.84	
P - value (<0.05) indicates	the probability o	f obtaining the ob	served difference	between feeding s	trategy if the	null hypothe	sis is true	

P-value (<0.05) indicates the probability of obtaining the observ In order to assess the changes in carcass and noncarcass weight, the weights of the appropriate components in the allometric regression equations

were adjusted with the increase in body weight equations due to the EBW, and these are shown in Table 4.

 Table 4. Estimated parameters ± confidence interval (CI) for allometric equations and prediction equations of carcass, noncarcass, gastrointestinal tract (GIT) and metabolically active organ (liver, spleen, kidneys, lungs and heart) weight of beef cattle fed supplementation with different soy byproduct concentrates

Component	Estimated para	ameters ± CI	- Equation	Prodiction equations
	$\alpha \pm CI$	$\delta \pm CI$	— Equation	Prediction equations
Carcass	$0.62 \pm 0.21$	1.01±0.06	$Y = 0.6211 \times X^{1.0072}$	$Y = 0.6255 \times X^{0.0072}$
Noncarcass	$0.39 \pm 0.24$	0.99±0.12	$Y = 0.3809 \times X^{0.9866}$	$Y = 0.3757 \times X^{-0.0134}$
Gastrointestinal tract	0.44±0.31	0.61±0.12	$Y = 0.4356 \times X^{0.6117}$	$Y = 0.2664 \times X^{-0.3883}$
Organs	0.24±0.16	0.64±0.11	$Y = 0.2397 \times X^{0.6385}$	$Y = 0.1530 \times X^{-0.3615}$

Performance...

No significant differences (P > 0.05) between the equations adjusted for the four treatments, describing the increase in carcass weight and noncarcass due to the EBW. Therefore, equations for the four treatments were adopted only, and these were used to estimate the weight of the carcass and noncarcass, depending on the EBW.

The estimated weights and gains for carcass and noncarcass for animals of different EBW showed an increase that was expected with the development of animals (Table 5). However, there was a steady increase in weight and in the proportion of the carcass and noncarcass, with the increased EBW more easily observed when the carcass and noncarcass weights were expressed as kg EBW. Carcass and noncarcass weight were 645 g kg<sup>-1</sup> and 354 g kg<sup>-1</sup>, respectively, for animals with 200 kg EBW, and these approached those values of 648 g kg<sup>-1</sup> and 351 g kg<sup>-1</sup>, respectively, for animals of 400 kg EBW.

**Table 5.** Estimated weight mean and weight gain of carcass (C), noncarcass (NC), gastrointestinal tract (GIT) and metabolically active organs (liver, spleen, kidneys, lungs and heart) weight of beef cattle fed supplementation with soy byproduct concentrates.

	suppementation with soy byproduct concentrates.									
Weight/kg EBW			Gain/kg	Gain/kg PCV		Weight/kg EBW		EBW		
BW	EBW	С	NC(lrg)	NC/C	С	NC	GIT	ORG	GIT	ORG
(kg)	(kg)	(kg)	NC (kg)	NC/C	(g/kg)	(g/kg)	(g/kg)	(g/kg)	(g/kg)	(g/kg)
237	200	129	71	0.55	650	350	56	35	34	22
291	250	161	88	0.54	650	349	51	33	31	20
345	300	194	105	0.54	651	348	48	30	29	19
399	350	226	123	0.54	652	347	45	29	27	18
453	400	259	140	0.54	653	346	43	27	26	17
EDW										

EBW - empty body weight

However, there was a steady increase in the weight and the proportion of the carcass and noncarcass, with the increased EBW, and these were more easily observed when the carcass and noncarcass weight were expressed as kg EBW.

The ratios between the carcass and noncarcass did not change with increasing empty body weight, illustrating that the two components grew at similar rates close to the EBW as a whole. The estimated weights and gains for the gastrointestinal tract and organs in g per kg EBW for animals of different EBW showed reduction in the relative size of the gastrointestinal tract and organs with increasing body weight, as expected from the equation in Table 4, where the  $\Box$  value was less than one.

The identity test of nonlinear models proposed by Bates and Watts (2007) and Regazzi and Silva (2004) showed no differences (P > 0.05) between the models and set parameters for the four treatments for protein content, fat and energy due to the EBW. Therefore, equations were adopted for the four treatments to estimate the protein, fat and energy contents of empty body, carcass and noncarcass, depending on the EBW (Table 6).

**Table 6.** Allometric regression equations for protein, fat and energy content in empty body, carcass, and noncarcass as a function of the empty body and weight gain

Nutrient	Estimated parameters Confidence interval (CI)		Component	Empty body equation	Weight gain equation
	$\alpha \pm CI$	$\delta \pm CI$	-		
	0.81±0.47	0.75±0.10	Empty body	Y=0.8113×X <sup>0.7516</sup>	Y=0.6097×X <sup>-0.2484</sup>
Protein	0.50±0.31	$0.76 \pm 0.11$	Carcass	Y=0.4978×X <sup>0.7550</sup>	Y=0.3758×X <sup>-0.2450</sup>
	0.31±0.21	$0.75 \pm 0.12$	Noncarcass	Y=0.3138×X <sup>0.7460</sup>	Y=0.2340×X <sup>-0.2540</sup>
	$0.001 \pm 0.001$	1.91±0.27	Empty body	Y=0.00076×X <sup>1.9116</sup>	$Y=0.0014 \times X^{0.9116}$
Fat	$0.001 \pm 0.001$	$1.86 \pm 0.32$	Carcass	$Y=0.00066 \times X^{1.8641}$	$Y=0.0012 \times X^{0.8641}$
	$0.001 \pm 0.001$	2.01±0.24	Noncarcass	Y=0.00014×X <sup>2.0076</sup>	$Y=0.00028 \times X^{1.0076}$
	$0.24 \pm 0.21$	$1.40 \pm 0.15$	Empty body	Y=0.2432×X <sup>1.4034</sup>	Y=0.3413×X <sup>0.4034</sup>
Energy	$0.16 \pm 0.17$	$1.40 \pm 0.18$	Carcass	Y=0.1639×X <sup>1.3959</sup>	Y=0.2287×X <sup>0.3959</sup>
	$0.08 \pm 0.07$	1.42±0.14	Noncarcass	$Y=0.0796 \times X^{1.4174}$	$Y=0.1128 \times X^{0.4174}$

514

For the evaluation of the body contents of protein, fat and energy, as well as the aforementioned contents in the carcass and noncarcass, the allometric regression equations were adjusted in the empty body, carcass and noncarcass, according to the EBW (Table 7). Derivations of the estimated protein gain (Table 7) showed a decrease with the increase of body weight, corresponding to 163 g kg EBW<sup>-1</sup> in animals of 200 and 137 g kg EBW<sup>-1</sup> in animals 400 kg due to the reduced muscle development and further development of adipose tissue in heavier animals.

ROCHA, T. C. et al.

**Table 7.** Content and estimated weight gain relative to protein (g kg<sup>-1</sup>), fat (g kg<sup>-1</sup>) and energy (Mcal kg<sup>-1</sup>) of carcass (C) and noncarcass (NC) to differences in body weight (BW) and empty body weight (EBW)

$\frac{1}{2} = \frac{1}{2} \sum_{i=1}^{n} \frac{1}{2} \sum_{i=1$											
BW (kg)	EBW (kg)	Protein (g EBW <sup>-1</sup> )				Fat (g kg EBW <sup>-1</sup> )			Energy (Mcal kg EBW <sup>-1</sup> )		
D W (Rg) = LD W (Rg)	EW	С	NC	EW	С	NC	EW	С	NC		
Content in EBW											
237	200	218	136	82	96	65	31	2.06	1.33	0.73	
291	250	206	129	77	118	79	39	2.25	1.45	0.8	
345	300	197	123	74	139	92	47	2.42	1.56	0.86	
399	350	189	119	71	160	105	55	2.58	1.66	0.92	
453	400	183	115	69	180	118	62	2.72	1.75	0.97	
Estimated v	weight gain in l	EBW									
237	200	163	102	61	183	121	62	2.9	1.9	1.0	
291	250	154	97	57	224	146	78	3.1	2.0	1.1	
345	300	147	92	55	265	171	94	3.4	2.2	1.2	
399	350	142	89	53	305	196	109	3.6	2.3	1.3	
453	400	137	86	51	344	220	124	3.8	2.4	1.4	

Empty body weight (EBW)

#### DISCUSSION

The average dry matter intake of forage was 7.78 kg for animals not supplemented and 5.87 kg for animals supplemented with or without addition of lipids. This phenomenon is referred to as a substitute or pasture replacement effect because the concentrate is a higher quality of food, especially with the increased protein (HILLS et al., 2015).

The animals that received soybean meal showed an increase in concentrate intake (P = 0.001) by 34% compared with steers receiving supplementation soybean oil and soybean grain. The soybean oil and soybean grain animals exhibited restricted intake due to the higher lipid concentration in their supplements.

There was no improvement in body weight and carcass gains with the addition of the concentrations, and notably there was not much difference between these characteristics in cattle grazed perhaps treatment duration was short

The increase in speeds relative growth of the carcass and noncarcass remained similar to the rate of increase in body mass as indicated by the two-parameters d allometric equation of the type Y =  $\alpha * X^{\Box}$ , which yielded values of 1.0072 and 0.9866 for carcass and noncarcas. The equality between the values of growth of carcass and noncarcass with 1, as shown by the confidence

intervals, revealed development close to the growth rate of the EBW as a whole.

To derive the regression equations (Table 4), which estimate the carcass weight, noncarcass weight and TGI organs in animals due to the EBW, the gain prediction equations were obtained per kg of the carcass and noncarcass gains; these gains remained almost unchanged with the increase in the EBW, as expected because they were obtained by derivation of equations whose values of d were close to unity.

Notably, the weights of the gastrointestinal organs decreased from 56 to 43 g kg EBW<sup>-1</sup> in the gastrointestinal tract and from 35 to 27 g kg EBW<sup>-1</sup>, when the weight empty body increased from 200 to 400 kg.

An animal that reaches maturity shows decline in weight and proportion of visceral organs, resulting in reduced energy requirement for maintenance per unit of body weight (MURRAY; SLEZACEK, 1988). This decline occurs because the viscera and internal organs (heart, kidney, digestive tract and liver) reach the maximum development in cattle at around two years of age (BERG; BUTTERFIELD, 1976).

Although the viscera and internal organs account for approximately 6-15% of body mass, these components consume approximately 55% of the animal maintenance energy, while the muscles, which account for over 40% of the EBW, require

only 21% (15 to 26%) of the energy expended by the body as a whole (CATON et al. 2000). Reducing the mass of bodies by unit of body weight, with increasing body weight, explains in part the relative decline of the verified maintenance requirements of a growing animal.

Similar results to those observed in this study were reported by Peron et al. (1993) who compared the proportion of internal organs in animals slaughtered at the beginning and end of the experiment. The cited authors attributed these results to the fact that the vital organs are relatively more developed at the earliest stage of the animal's life.

The animals of this study were aged 2.5 to 3 years, so the organs would be close to their maximum sizes. As animals age, there is more intense growth of muscle and adipose tissue. Thus, the organs will represent a smaller proportion of EBW (BERG; BUTTERFIELD, 1976).

Knowledge of changes in body composition of the animal with increasing body weight is necessary when seeking to produce castings that meet the demands of the consumer market. In turn, knowledge of the composition of factors associated with weight gain is essential for establishing the nutritional requirements of the animals because the content of each nutrient in the gain corresponds to the net weight of the nutrient requirements for weight gain. Analyzing the parameter values d of the regression equations, the protein body content increased at a lower rate than the increase of EBW as a whole ( $\delta = 0.75$ ). On the other hand, the fat and energy contents increased more rapidly than the body weight ( $\delta = 1.91$  and 1.40, respectively). These results are corroborated by the results of Berg and Butterfield (1976), which showed that with increased weight, there is a gradual decrease in the deposition of muscle tissue and a rapid increase for adipose tissue.

With the increase of animal weight, body fat mass increases quadratically whereas the protein mass increases linearly. Thus, the decrease of the proportion of body protein with increased body weight is due to a decrease in muscle development as well as to the dilution effect of the increased fat deposition (OWENS et al. 1995). The concentration of energy varies in a similar manner to that of fat, which is due to the higher energy content of the fat (9.39 kcal g<sup>-1</sup>) in relation to protein (5.64 kcal g<sup>-1</sup>) (NRC, 2000).

By deriving the regression equations of body content of fat, protein and energy, depending on the EBW, the contents of the prediction equations (gain) of fat, protein and energy per kg of EBW gain, carcass and noncarcass were obtained. The protein and energy gains per kg EBW gain are the net requirements of protein and energy for gain of 1 kg EBW.

The estimated values of protein kg<sup>-1</sup> EBW for the different body weights decreased with the increase in animal weight ranging from 218 g kg<sup>-1</sup> for animals of 200 kg EBW to 183 g kg<sup>-1</sup> for animals with 400 kg EBW, which is in agreement with the reports of Owens et al. (1995). Berg and Butterfield (1976) also showed that even after birth, the deposition of lean meat is proportionally greater than that of fat. Once a certain stage of growth is reached, however, fat deposition becomes proportionally greater than that of lean meat.

The values of protein content per kg EBW were similar to those found by other authors in cattle in the same weight range (SANT'ANA et al., 2011; TEDESCHI et al., 2015).

Regarding the fat content per kg EBW, as expected, the opposite was observed for protein. With the increasing weight of the animal, the fat content is increased. In animals of 200 kg EBW, the estimated value was 96 g kg EBW<sup>-1</sup>, while in animals of 400 kg this amount was 180 g kg EBW<sup>-1</sup>. As mentioned above, this tendency reflects the deceleration of the growth of muscle tissue in contrast to the faster development of adipose tissue with increase in EBW, as adipose tissue has the highest growth boost at higher ages (BERG; BUTTERFIELD, 1976).

The concentration of energy per kg EBW increased with increasing weight of the animal; this behavior was expected because the concentration of energy is very influenced by the fat content that has higher energy content than protein. In animals of 200 kg EBW, the estimated value was 2.06 Mcal kg EBW<sup>-1</sup>, and in animals of 400 kg, this value was 2.72 Mcal kg EBW<sup>-1</sup>.

The contents of protein, fat and energy in the carcass and the noncarcass do not show the same trend for the EBW as a whole; instead, these contents hold the same proportions relative to the weight of each component. As shown previously (Table 5), the ratio of carcass weight and noncarcass tended to remain constant with increasing body weight. Most of the protein, fat and energy deposited in the body was located in the carcass, and the proportion of protein deposited on the substrate, relative to the total deposited in the empty body remained substantially constant (62.3%, in animals of 200 kg of EBW, and 62.8% in animals of 400 kg EBW). The proportion of fat deposited on the substrate, however, decreased from 67.7% EBW in animals of 200 kg to 65.5% in animals of 400 kg of EBW, with no increase in carcass.

The net requirements of gain values for protein were similar to those values found by other authors (COSTA and SILVA, 2015; PRADOS et al., 2015). The differences are mainly due to differences in fat content in the weight gain observed in several experiments. Due to the increase in fat weight gain in proportion to the increase in animal weight, net energy requirements are increased. In animals with 200 kg EBW, the estimated net requirement is 2.9 Mcal kg<sup>-1</sup> and in animals with 400 kg EBW, the energy requirement is 3.8 Mcal kg<sup>-1</sup>.

#### CONCLUSIONS

The use of supplements, with or without the addition of lipids, does not modify the EBW gain and other body components, when steers have access to good quality pastures.

Supplementation with soybean grain to present similar gains is a lower cost alternative compared to other sources of soy. The supplementations with higher concentrations of lipids (oil and grain) were not enough to promote greater gains in fat and energy in carcass.

**RESUMO:** Esta pesquisa teve como objetivo avaliar o desempenho, composição corporal e requisitos líquidos para proteínas e energia de novilhos Nelore em pastejo suplementados com ou sem adição de lipídios. Foram utilizadas no experimento vinte e oito novilhos,  $301 \pm 5,8$  kg PC. O método comparativo de abate foi utilizado, com quatro novilhos utilizados como referências. Os 24 restantes novilhos foram distribuídos aleatoriamente em quatro grupos, durante 180 dias, de acordo com o tratamento: apenas Panicum maximum cv. Grama Mombasa; Grama de Mombasa com suplementação concentrada à base de farelo de soja; Grama de Mombasa com concentrado contendo lipídios de óleo de soja; e Mombasa com concentrado contendo lipídios de óleo de soja; e Mombasa com a pastagem não diferiu (P> 0,05) da ingestão média dos tratamentos com ou sem adição de lipídios. Não houve diferenças (P> 0,05) entre os tratamentos para o ganho diário total, carcaça e não carcaça para proteínas, energia e gordura. Os requisitos de proteína e energia não diferiram (P> 0,05) entre os tratamentos. Quando a pastagem é de boa qualidade, a suplementação não altera a composição corporal ou os requisitos líquidos de proteína e energia para ganho de peso. As suplementações com maiores concentrações de lipídios (óleo e grão) não foram suficientes para promover maiores ganhos de gordura e energia na carcaça.

PALAVRAS-CHAVE: Carcaça. Panicum maximum. Ruminantes. Trato gastrointestinal.

**Abbreviations:** ADG, average daily weight gain; BW, body weight; C, carcass; CP, crude protein, DM, dry matter; EE, ether extract; EBW, empty body weight; GIT, gastrointestinal tract; NC noncarcass; NDF, neutral detergent fiber; TDN, total digestible nutrients; iNDF, indigestible neutral detergent fiber; ORG, metabolically active organs; SBW, shrunk body.

#### REFERENCES

AOAC - ASSOCIATION OF OFFICIAL ANALYTICAL CHEMISTRY. Official methods of analysis. 15th ed. Washington: AOAC International, 1990. 1298 p.

# AGRICULTURAL RESEARCH COUNCIL (GREAT BRITAIN); COMMONWEALTH AGRICULTURAL BUREAUX. The Nutrient requirements of ruminant livestock: technical review. CAB Intl, 1980.

BATES, D. M.; WATTS, D. G. Nonlinear Regression Analysis and its application. New York: John Wiley, 2007. 392 p.

BERG, R. T.; BUTTERFIELD, R. M. New concepts of cattle growth. Sydney: University Press, 1976. 240 p.

BEZERRA, L. R.; NETO, S. G.; DE MEDEIROS, A. N.; DE ALBUQUERQUE MARIZ, T. M.; OLIVEIRA, R. L.; CÂNDIDO, E. P.; DE AZEVEDO SILVA, A. M. Feed restriction followed by realimentation in

Performance...

prepubescent Zebu females. **Tropical animal health and production**, v. 45, n. 5, p. 1161-1169, 2013. https://doi.org/10.1007/s11250-012-0341-8

CATON, J. S.; BAUER, M. L.; HIDARI, H. Metabolic components of energy expenditure in growing beef cattle-review. Asian-Australasian Journal of Animal Sciences, v. 13, n. 5, p. 702-710, 2000. https://doi.org/10.5713/ajas.2000.702

COSTA E SILVA, L.; ENGLE, T. E.; VALADARES FILHO, S. C.; ROTTA, P. P.; VALADARES, R. F. D.; SILVA, B. C.; PACHECO, M. V. C. Intake, apparent digestibility, and nutrient requirements for growing Nellore heifers and steers fed two levels of calcium and phosphorus. **Livestock Science**, v. 181, p. 17-24, 2015. https://doi.org/10.1016/j.livsci.2015.09.024

Commonwealth Scientific and Industrial Research Organization. Feeding Standards for Australian Livestock Ruminants. CSIRO, Commonwealth Scientific and Industrial Research Organization, Melbourne, Australia; 1990.

Commonwealth Scientific and Industrial Research Organization. Nutrient Requirements of Domesticated Ruminants. CSIRO, Commonwealth Scientific and Industrial Research Organization, Collingwood, Australia; 2007.

FONTES, C. A. D. A.; OLIVEIRA, R. C. D., ERBESDOBLER, E. D. Á.; QUEIROZ, D. S. D. Use of comparative slaughter to set maintenance energy requirements of beef cattle grazing elephantgrass: description of methodology and results. **Revista Brasileira de Zootecnia**, v. 34, n. 5, p. 1721-1729, 2005. https://doi.org/10.1590/S1516-35982005000500034

GONZAGA NETO, S.; BEZERRA, L. R.; MEDEIROS, A. N.; FERREIRA, M. A.; EC FILHO, P.; CÂNDIDO, E. P.; OLIVEIRA, R. L. Feed restriction and compensatory growth in Guzera females. Asian-Australasian Journal of Animal Sciences, v. 24, n. 6, p. 791-799, 2011. https://doi.org/10.5713/ajas.2011.10417

HANKINS O. G, HOWE, P. E. Estimation of the composition of beef carcasses and cuts. Washington, D.C (Tech. Bulletin - USDA, 926); 1946.

HILLS, J. L.; WALES, W. J.; DUNSHEA, F. R.; GARCIA, S. C.; ROCHE, J. R. Invited review: An evaluation of the likely effects of individualized feeding of concentrate supplements to pasture-based dairy cows. **Journal of Dairy Science**, v. 98, n. 3, p. 1363-1401, 2015. https://doi.org/10.3168/jds.2014-8475

KIMURA, F. T.; MILLER, V. L. Chromic oxide measurement, improved determination of chromic oxide in cow feed and feces. **Journal of Agricultural and Food Chemistry**, v. 5, n. 3, p. 216-216, 1957. https://doi.org/10.1021/jf60073a008

KOCK, S. W.; PRESTON, R. L. Estimation of bovine carcass composition by the urea dilution technique. **Journal of Animal Science**, v. 48, n. 2, p. 319-327, 1979. https://doi.org/10.2527/jas1979.482319x

LOFGREEN, G. P.; GARRETT, W. N. A system for expressing net energy requirements and feed values for growing and finishing beef cattle. **Journal of animal science**, v. 27, n. 3, p. 793-806, 1968. https://doi.org/10.2527/jas1968.273793x

MURRAY, D. M.; SLEZACEK, O. The effect of weight stasis on the non-carcass components of crossbred sheep. **Australian Journal of Agricultural Research**, v. 39, n. 4, p. 653-658, 1988. https://doi.org/10.1071/AR9880653

NRC. National Research Council. Nutrient requirements of beef cattle. 6th ed, NRC National Academy Press, Washington, DC, USA; 1984.

NRC. National Research Council. Nutrient requirement of beef cattle. 7th ed, NRC National Academic Press, Washington, D.C., USA; 1996.

NRC. National Research Council. Nutrient requirements of beef cattle. 7th ed, NRC, National Academy Press, Washington, D.C., USA; 2000.

OWENS, F. N.; GILL, D. R.; SECRIST, D. S.; COLEMAN, S. W. Review of some aspects of growth and development of feedlot cattle. **Journal of animal science**, v. 73, n. 10, p. 3152-3172, 1995. https://doi.org/10.2527/1995.73103152x

PALIERAQUI, J. G. B.; FONTES, C. D. A.; RIBEIRO, E. G.; CÓSER, A. C.; MARTINS, C. E.; FERNANDES, A. M. Influência da irrigação sobre a disponibilidade, a composição química, a digestibilidade e o consumo dos capins mombaça e napier. **Revista Brasileira de Zootecnia**, v. 35, n. 6, p. 2381-2387, 2006. https://doi.org/10.1590/S1516-35982006000800026

PRADOS, L. F.; VALADARES FILHO, S. C.; DETMANN, E.; ZANETTI, D.; SANTOS, S. A.; SATHLER, D. F. T MARIZ L. D. S.; BORGES A. L. C. C.; NUNES A. N.; RODRIGUES F. C.; AMARAL, P. M. Energy and protein requirements of 3/4 Zebux 1/4 Holstein crossbreds fed different calcium and phosphorus levels in the diet. **Arquivo Brasileiro de Medicina Veterinária e Zootecnia**, v. 67, n. 2, p. 555-563, 2015. https://doi.org/10.1590/1678-6905

REGAZZI, A. J.; SILVA, C. H. O. Teste para verificar a igualdade de parâmetros e a identidade de modelos de regressão não-linear. I. dados no delineamento inteiramente casualizado. **Revista de Matemática e Estatística**, v. 22, n. 3, p. 33-45, 2004.

RODRIGUES FILHO, M.; ANDRADE, I. F;LADEIRA, M. M.; RODRIGUES, N. E. B.; LOPES, L. S. Characteristics of carcass and commercial meat cuts of Red Norte young bulls supplemented with fry and soybeans oil finished in feedlot. **Revista Brasileira de Saúde e Produção Animal**, v. 14, n. 1, p. 54-66, 2013. https://doi.org/10.1590/S1519-99402013000100006

SANT'ANA N. F.; FONTES, C. A. A.; SILVA, R. S. T.; GARCIA, C. S.; VIEIRA, R. A. M.; ROCHA, T. C. Body composition and net requirements of protein and energy for weight gain of Nellore and crossbreed steers finished under grazing. **Revista Brasileira de Zootecnia**, v. 40, n. 4, p. 912-921, 2011. https://doi.org/10.1590/S1516-35982011000400028

SILVA-MARQUES, R. P.; ZERVOUDAKIS, J. T.; HATAMOTO-ZERVOUDAKIS, L. K.; DA SILVA CABRAL, L.; ALEXANDRINO, E.; NETO, A. J.; SOARES J. Q.; MELO, A. C. B. Multiple supplements for beef heifers on pasture during the dry season: nutritional characteristics. **Semina: Ciências Agrárias**, v. 36, n. 1, p. 509-524, 2015. https://doi.org/10.5433/1679-0359.2015v36n1p509

TEDESCHI, L. O.; FOX, D. G.; FONSECA, M. A.; CAVALCANTI, L. F. L. Models of protein and amino acid requirements for cattle. **Revista Brasileira de Zootecnia**, v. 44, n. 3, p. 109-132, 2015. https://doi.org/10.1590/S1806-92902015000300005

VAN SOEST, PJ; ROBERTSON, JB; LEWIS, BA Methods for dietary fiber, neutral detergent fiber, and nonstarch polyssacharides in relation to animal nutrition. **Journal of Dairy Science**, v. 74, p. 3583–3597, 1991. http://dx.doi.org/doi::10.3168/jds.S0022-0302(91)78551-2