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ENERGY AND PROTEIN REQUIREMENTS FOR MAINTENANCE AND GAIN OF NELLORE CATTLE IN TROPICAL CONDITIONS

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ABSTRACT

Research on the purebred Zebu cattle requirements are scarce in tropical areas. Therefore, we evaluated body composition as well as protein and energy requirements for cattle maintenance and growth. Using comparative slaughter, 30 purebred Nellore bulls non-castrated presenting initial average body weight (BW) of 380.2 ± 28.7 kg. Five animals were assigned as reference group and the remaining 25 were fed *ad libitum;* randomly distributed in 5 five treatments in completely randomized design: maintenance diet (containing 60% Tifton 85 hay and 40% concentrate, daily offer of 1.2% of BW) and increased concentrate levels: 17; 34; 51 and 68% for 112 days of confinement. The net energy demand for were obtained by the equation: RE= $0.0430*EBWeq^{0.75}*GEBW^{1.3595}$. The requirements for maintenance found out for net and metabolizable energy were 78.99 and 116.13 kcal/ kg $EBW^{0.75}$/day, respectively. The efficiency of metabolizable energy for maintenance was 68.01% and 46.50%, respectively. The demand for metabolizable protein was 4.81 g/kg $BWJ^{0.75}$/day and the efficiency of its use for weight gain was 51.5%. We found the equation: NPg = 131.8*GEBW+22.7*RE to estimate net protein daily demand for weight gain on male non-castrated Nellore bovines in tropical weather.

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INTRODUCTION

There is lack of studies about energy and protein requirements of purebred Zebu cattle in tropical conditions, as well as their interactions and the influence of treatment on energy, protein and efficiency if use. Mainly due to climate complexity, non-uniformity of food supply and different nutritional levels reflect on the success of the global production of beef cattle. Under tropical conditions Nellore cattle is used successfully in the farming chain and it is even more common due to its adaptability and productive efficiency; thus, the importance of knowing its nutritional requirements to supply its energy and protein needs accurately.

However, according to Gomes *et al*. (2017), comparative studies on nutritional demand of purebred *Bos Taurus Indicus* are scarce and they show that there are differences between the energy requirement in male purebred Taurus and Zebu cattle, therefore the need of studies about the differences on the nutritional recommendations given by international committees. In BCNRM (2016), the net energy daily demand for maintenance on male castrated bovines is 77 kilocalories on metabolic body weight unit (kcal/$BWJ^{0.75}$/day), and for non-castrated cattle it is around 88 kcal/$BWJ^{0.75}$/day, therefore, on this value it is added 15% referring to warmth production while fasting.

Nevertheless, it is recommended a 10% deduction of this value. Valadares Filho *et al*. (2016) suggest that the demand of net energy for maintenance should be 75 kcal/per empty metabolic body ($EBW^{0.75)}$ /day for confined Zebu cattle. The net energy for gain (NE_g) can be understood as all the retained energy in an empty body as protein of fat (Garrett *et al*., 1959). Therefore, the nutritional demand of protein needs to be calculated and supplied accordingly to the potential of each reality. Not only because balanced diets in tropical conditions are based on international systems, but also because of the lack of data that allows the development of a prediction equation that represents the variety of existing ecosystems, in order to obtain better reliability. It means a great advance to the production of beef cattle in these regions, given that other countries have their systems very well stablished, as reported by Olmedo *et al*. (2010). The objective was to estimate the nutritional requirements of energy and protein for maintenance and weight gain, as well as the respective usage efficiency in male non-castrated confined Nellore cattle under tropical conditions.

MATERIALS AND METHODS

The experiment with confined animals and the experimental analysis were conducted at Engenho Gambá farm, in the city of Tracunhaém and in Animal Science Department of the Federal Rural University of Pernambuco, Recife, Brazil. All procedures have been conducted in accordance with the guidelines set out by the Brazilian College of Animal Experimentation in the Code of Practice for the Care and Use of Animal for Experimental Purposes and were reviewed and approved by the Ethics Committee on Use of Animal for Research (CEUA) of Federal Rural University of Pernambuco, protocol number 23082.015634/2012-41. Thirty Nellore bovines male non-castrated, with an average initial body weight (BW) of 380.2 ± 28.7 kg were confined in individual stalls measuring 1 meter long linear feeder and water drinker. Initially, the animals were weighted, identified and prevented against ecto and endoparasites, and then treated with vitamin complex A, D and E. The experimental lineation was completely randomized design. After 30 days of adaptation period -during this time they received a feed composed by 60:40 (bulky: concentrated relation), five animals designated as reference group were slaughtered after solid fasting for 16 hours, to estimate the body composition and the initial empty body weight (EBW) of the remaining experimental units. The 25 remaining animals were randomly distributed; 5 repetitions, within the maintenance treatments, 17; 34; 51 and 68% of ration concentration. The animals within the maintenance group receive the same ingredients feed provided during the adaptation period; however, the provided feeding was limited to 1.2% of their BW per day.

We calculated the experimental diets using the recommendations on BR CORTE (Valadares Filho *et al*. (2010, to provide approximately 13.2% of BW (base percentage MS) aiming an average daily gain (ADG) of approximately 1.0 kg. The concentrate was composed by ground coin, soy bean, mineral supplement, limestone and sodium bicarbonate; while the roughage was composed by Tifton 85 hay (*Cynodon dactylon (L.)* Pers.), corrected to crude protein with a mixture of urea – ammonium sulfate (ratio 9:1) aiming to have an isoproteic diet (Tables 1 and 2). We fed the cattle twice a day (at 8 a.m. and at 4 p.m.) adjusted every 2 days allowing 10% leftovers over the total amount of food provided, aiming consumption *ad libitum*; except for the maintenance group which had feeding restrictions. Water was permanently at the animals' disposal. The experiment lasted 112 days divided into four periods of 28 days. The quantities of provided feed and leftovers were registered daily to obtain final consumption. We collected samples of the roughage weekly and then separated constituents of the concentrate and the leftovers of each animal individually. The weekly samples were pre-dried and, later, grouped proportionally, creating a composed sample for further laboratory analysis. At the end of each period all animals were weighted for monitoring their development, highlighting that on the first and last weighing, the animals went through 16 hours of solid fasting. After the experimental period the animals were then slaughtered in a commercial slaughterhouse in Maceió–AL, in which the procedures followed the recommendations for humanitarian slaughtering of animals in slaughterhouses. The numbness was made by the mechanical method of percussive penetrative type, using a pneumatic gun with a captive dart. The numbness was followed by the immediate bloodletting through the section of the jugular and carotid. During the bloodletting, the blood was collected in a previously tared and weighted container. Proceeding with the skinning, evisceration and removal of the head, limbs and tail. The components of the gastrointestinal tract – GIT (rumen/reticulum, omasum, abomasum, large and small intestines) were emptied and washed, weighed and the records summed to the weights of the heart, esophagus, trachea, industrial meat, lungs, leaver, tongue, spleen, fat, shavings, reproductive apparatus, kidneys, skin, limbs, head, tail, pancreas, blood and carcass to obtain the empty body weight (EBW).

The GIT alongside the internal fat, leaver, heart, kidneys, lungs, tongue, spleen, pancreas, industrial meat and shavings, esophagus, trachea and reproductive apparatus were grinded in an industrial mill for 20 minutes in order to retrieve a homogeneous sample of organs and entrails. The skin on the head and the limbs of each animal was removed and the other parts were grinded in a bone grinder, also retrieving samples of the head and limbs. The hide was sampled from different areas (shoulder, dorsal line, abdomen, back part, head and limbs) and they were sectioned manually in small pieces (4 cm^2) for sampling. The carcass of each animal was divided in 2 half-carcasses which were identified, weighed, and, then, cooled in a cold room to 4 ^oC, for 24 hours. After that time, the carcasses were again weighed. Then we performed a complete dissection of the left-carcass of each animal. The bones, meat and fat were grinded, bones separately, weighed and packed inside aluminum trays. The blood samples, leather, organs and entrails, head, and limbs e carcass were freeze-dried according to the method on INCT-CA.G-002/1 (Detmann *et al*., 2012). All freeze-dried samples were grinded in a ball mill and then, we evaluated the contents of dry matter, organic matter, total nitrogen and ethereal extract to estimate the EBW chemical composition according to the Association of Official Analytical Chemists (AOAC 1990), method 934.01 for DM; 930.05 for OM and 981.10for CP. Ether extract (EE) was analyzed by Soxhlet extraction with petroleum ether, according to the AOAC (1990), method 920.39. To convert BW into EBW and body weight gain (GBW) into GEBW we calculated the relation between EBW (kg) and BW (kg), GBW (kg/day) and GEBW (kg/day) of the animals kept on the experiment which were used in the conversion of the requirements of EBW or GEBW in requirements of BW or GBW, respectively.

We obtained the body energy determination from the body contents of protein and fat and their respective caloric equivalents, according to the equation recommended by ARC (1980): EC= 5.6405 X + 9.3929 Y. In which: EC = energy content (Mcal); X = body protein (kg); Y = body fat (kg). The contents of fat, protein and energy in the bodies of the animals in each treatment and for the overall studied population, the function EBW were estimated using non-linear equations relating the body contents of energy of the animals in development and in the reference group, according to the model: $Y = \beta_0 * X^{\beta 1} + e$. Where Y: body content of fat (kg), protein (kg) or energy (Mcal); X= empty body weight (EBW, kg) and 'β_0'e 'β_1' = regression parameter of the equation of the content of fat, protein and energy as a function of EBW; e= random error. By the derivate of these equations we obtained the net requirements of protein (NPg, g/GEBW) and energy (NEg, Mcal/GEBW) for each kilo gained in empty body weight and the content of fat in the empty body gain, according to the equation $Y' = ß_0 * ß_1 * X^{ß1-1}$. In which Y'= fat content (kg/GEBW), net requirement of protein (g/GEBW) or energy for gain (Mcal/GEBW) and 'β_0' e 'β_1' = regression parameters; X=EBW (kg).

We adjusted regression equations for retained energy (Mcal/day) in function of GEBW (kg/day) and $EBW_{equivalent}$ metabolic ($EBWeq^{0.75}$) of animals in development to estimate the NEg in any daily weight gain range. In order to do that, we adopted the method of non-linear models (procedure PROC NLIN do SAS, 2005), using the iteration algorithm of Gauss-Newton: $RE = ß_2 \times EBWeq^{0.75} + GEBW^{ß1}$; where, RE = retained energy (Mcal/day) and $ß_2$ e $ß_1$ are regression parameters. $EBWeq = (EBW/EBW_{mant}) * EBW_{ref}$; in which EBW_{mant} = empty body weight when mature (kg) and EBW_{ref} = reference empty body weight for non-castrated animals = 517, according to Valadares Filho *et al*. (2016). The efficiency of the metabolizable energy for weight gain (kg) was calculated using the relation between the retained energy (RE, Mcal/kg $EBW^{0.75}$) and the metabolizable energy of intake (MEI, Mcal/kg $EBW^{0.75}$), according to the model: $RE = \beta_0 + \beta_1 * MEI$; where, β is the efficiency of metabolizable energy usage for weight gain (kg). By equaling the RE on the equation to zero, we obtained the consumption of metabolizable energy in which the energy retaining is null and it represents the requirements of metabolizable energy for maintenance (MEm). The latest was obtained by the ratio of the coefficients β_0 and β_1 of the equation above ($MEm = \beta_0 / \beta_1$). The parameters β_0 and β_1 presented were estimated using the orthogonal regression method from Fuller (1987), which considers that both model variables (RE e MEI) contain random error associated to them.

The parameters of the equations were obtained the following way: $\beta_0 = \bar{Y} - \beta_1 X$; $\beta_1 = (\sigma^2_y - \sigma^2_x + ((\sigma^2_y - \sigma^2_x)^2 + 4\sigma^2_{xy}{}^2)^{\wedge 0.5}))/ 2\sigma_{xy}$; where X = average consumption of metabolizable energy, Y = average of retained energy, σ^2_x = variance of X; σ^2_y = variance of Y and σ_{xy} = covariance between X and Y.

The calculus of the requirements of net energy for maintenance (heat production while fasting) that is equivalent to the coefficient β_0 from the exponential regression equation between the warmth production (BW, Mcal/kg $EBW^{0.75}$) and the metabolizable energy intake (MEI, Mcal/kg $EBW^{0.75}$) was made according to Ferrell and Jenkins (1998): $HP = \beta_0 * e^{\beta 1 * MEI}$; β_0 = interception (NEm); e = Euler number (≈2.7183); X= MEI (Kcal/$EBW^{0.75}$).

An alternative to estimate the MEm is through the iterations method using the point where MEI and BW are equals. We obtained the efficiency energy use for maintenance (k_m) from the relation between the net and metabolizable requirements of energy. To calculate the net requirement of protein for weight gain, in any gain range, we adjusted the model that estimates the retained protein of the animals in development in function of the GEBW and the RE, according to the equation: $RP = ß_0 \times GEBW + ß_1 \times RE$. Where RP = retained protein (g/day), GEBW = empty body weight gain (kg/day), RE = retained energy (Mcal/day) and $ß_0$ and $ß_1$ are regression parameters. We calculate the requirements on metabolizable protein for maintenance (MPm) according to BR-CORTE (Valadares Filho *et al*., 2016), and obtained the regression of metabolizable protein intake (MPI, g/day) as a function of GEBW (kg/day) from the animals in development and maintenance: $IPM_t = \beta_0 + \beta_1 * GEBW$. Where MPI = intake of metabolizable protein (g/day), GEBW = empty body weight gain (kg/day) and β_0 and β_1 are regression parameters.

The relation between the interception of this regression through the average metabolic empty body weight of the animals in development and in maintenance allowed to estimate the requirements of metabolizable protein for maintenance (MPm, g/$EBW^{0.75}$) = $\beta_0/EBW^{0.75}$); afterwards we converted that result in numbers that represent body weight while fasting (g/$BWJ^{0.75}$), according to BCNRM (2016). In another alternative and using the same group of animals the protein retained was printed as a function of metabolizable protein consumption, following the model: $RP = \beta_0 + \beta_1 \times CP_{met}$; where, RP = retained protein (g/$EBW^{0.75}$/day), MPI= metabolizable protein of intake (g/$EBW^{0.75}$/day); β_0 and β_1= regression parameters, considering that β_1 is the usage efficiency of the metabolizable protein for gain. The coefficients β_0 and β_1 from the last models presented were estimated by Fuller (1987) method of orthogonal regression which considers that both variables in the model contain random errors associated to them.

The parameters of the equation were obtained the following way: $\beta_0 = \bar{Y} - \beta_1 X$; $\beta_1 = (\sigma^2_y - \sigma^2_x + ((\sigma^2_y - \sigma^2_x)^2 + 4\sigma^2_{xy}{}^{2)\wedge 0.5))} / 2\sigma_{xy}$. Where X = average consumption of metabolizable energy, Y = average of the retained energy, σ^2_x = X variance; σ^2_y = Y variance and σ_{xy} = covariance between X and Y.To estimate the demand on degradable rumen protein (DRP) and non-degradable rumen protein (NDRP) according to Valadares Filho *et al*. (2016); we used the following equation: $CPmic = -53.07 + 304.9 * CPI + 90.8 * TDNI - 3.13 * TDNI^2$ and disregarded the efficiency of degradable nitrogen conversion in the rumen into microbial nitrogen of 1.11. Considering that DRP requirements were taken as equal to the synthesis of CPmic (DRP= CPmic). The NDRP was obtained by the equation: $NDRP = [(MP - (CP_{mic} * 0.64)]/0.80$ in which PM is the total demand of metabolizable protein. The CP was estimated by the sum of the DRP and the NDRP and the percentage estimate of dry matter.We estimated the CMS as a function of BW and GBW daily (kg/day) of the animals in development, similarly to the model adopted by Valadares Filho *et al*. (2016). To develop the equations for the data relating to the nutritional requirements, we adopted the method of non-linear models, adjusted using the iteration algorithm of Gauss-Newton through the NLIN procedure from the software SAS 9.2.

RESULTS

The average relation between EBW and SBW; EBWG and ADG were 0.85 and 0.95, respectively; which means that one kilo of SBW(Figure 1). is equivalent to 0.85 kg of EBW and for an average daily gain of ADG we assume 0.95kg of EBWG. This value (0.85) is within the range of variation reported by BCNRM (2016) of 85 to 95%. The knowledge of this relation is recommended to minimize error by the farmer when weighing the animals on the estimates of nutritional requirements, because it decreases the influence of factors linked to the content of the gastrointestinal tract. Regarding the content of body energy, we obtained an equation for all treatments together, in which: BE (Mcal) = $0.2995*EBW^{1.3695}$. We observed that as the EBW raised there was an increase in the content of body energy (Table 3). The content of body protein ($Prot_c$) was estimated by the equation: $Prot_c$ (kg) = $0.269*EBW^{0.971}$. We can observe that as much the body weight increases the body protein content also does so (Table 4). We observed that the body protein decreased as the weight increases. By deriving the equation (Table 4) we obtained the demand of net energy for different body weights per kilo of empty body weight gain, according to the equation: NEg (Mcal/kg GEBW) = $0.299*1.369*EBW^{0.369}$ (Table 5).

Table 1. Proportion of ingredients and chemical composition used in the experimental diets

Item (% of DM)	Concentrate	Roughage	
Corn grain milled	83,30	-	
Soubean meal	11,50	-	
Mineral[a]	3,00	-	
Limestone	1,20	-	
Sodium bicarbonate	1,00	-	
Urea /sulfate ammonium[b]	-	1,48[b]	
Tifton 85 hay	-	98,52	
Total	100	100	
Chemical composition (g/kg of DM^{-1})	Corn	So ybean	Hay
Dry matter	815,8	823,4	837,2
Crude protein	88,7	470,5	97,2
Neutral detergent fiber[c]	119,9	134,6	708,8

[a] Nutrients/kg of product: Ca: 55 g; P: 45 g; S: 4120 mg; Na: 152 g; Co: 38,9 mg; Cu: 1050 mg; Fe: 1300 mg; I: 50,25 mg; Mn: 1000 mg; Se: 9 mg; Zn: 2520 mg; F: 450 mg. [b] nine parts of urea and one part of ammonium sulfate. [c] Corrected for ash and protein.

According to Garrett *et al*. (1959), the net energy is in function of the protein contents and fat deposited in the body; thus, we observed an increase on the NEg and in the quantities of energy and fat deposited in the empty body as the animals' body weight increase; a similar behavior was observed in other researches (Valadares Filho *et al*., 2010; Souza *et al*., 2012; Jolomba, 2015). Alternatively, aiming to estimate the NEg to any weight range the NRC (2000) suggested to adjust the RE regression in function of the metabolic empty body weight ($EBW^{0.75}$) and the GEBW of the animals in development; obtaining the equation: $ER=0.0430*EBWeq^{0.75}*GEBW^{1.3595}$. In which: RE (Mcal/day) = energy retained or net energy demand for gain; $EBW^{0.75}$ (kg) = metabolic empty body weight; GEBW (kg/day) = empty body weight gain. It is needed to evidence that the GEBW is elevated to a coefficient superior to one, that is related to the average daily gain (ADG) of the animals, meaning that in the case of the ADG observed is higher than 1 kg/day, the coefficient of the GEBW will behave the same, increasing the energy retention as the animal grows.

Table 2. Chemical composition of experimental diets

Item (g kg DM^{-1})		Concentrate levels (%)			
	Maintenance	17	34	51	68
Dry matter (g kg^{-1} fresh matter)	830.9	836.4	834.3	832.2	830.2
Organic matter	924.4	922.4	923.9	925.3	926.7
Crude protein	132.8	134.7	133.3	132.0	130.6
Ethereal extract	25.1	22.0	24.3	26.5	28.8
Neutral detergent fiber[a]	465.1	599.2	500.1	401.0	301.9
Acid detergent fiber	233.5	311.8	253.9	196.0	138.1
Calcium	5.2	4.9	5.3	5.8	6.2
Phosphorus	3.1	2.1	2.5	2.9	3.3

[a] Corrected for ash and protein.

Table 3. Body energy contents (BEc) in different weights and concentrate levels

		Concentrate levels (%)			
BW (kg)	EBW (kg)	17	34	51	68
		Energy (Mcal/kg EBW)			
300	254.91	2.49	2.44	2.37	2.37
350	297.39	2.52	2.51	2.52	2.51
400	339.88	2.54	2.58	2.64	2.65
450	382.36	2.55	2.64	2.76	2.77

Table 4. Content of retained protein in different weight and concentrate levels

	Concentrate levels (%)			
BW (kg)	17	34	51	68
	Body protein (g/kg EBW)			
300	232.93	232.64	231.68	229.83
350	230.82	230.42	228.98	229.37
400	229.01	228.52	226.67	228.98
450	227.42	226.86	224.65	228.63

BW = body weight; EBW (empty body weight) = 0.85*BW; $Prot_c$ (17%) = $(0.323*EBW^{0.941})$ /EBW*1000; $Prot_c$(34%) = $(0.338*EBW^{0.938})$ /EBW *1000; $Prot_c$(51%) = $(0.353*EBW^{0.924)}$ /EBW *1000; $Prot_c$(68%) = $(0.247*EBW^{0.987)}$ /EBW *1000.

Table 5. Net requirements of energy for gain (NEg) in different weight levels and concentrated

		Concentrate levels (%)			
BW (kg)	EBW (kg)	17	34	51	68
		NEg (Mcal/kg GEBW)			
300	254.91	2.65	2.91	3.26	3.29
350	297.39	2.67	2.99	3.46	3.50
400	339.88	2.69	3.07	3.63	3.69
450	382.36	2.71	3.14	3.80	3.86

BW = body weight; EBW (empty body weight) = 0.85*BW; NEg (17%) = $(1.767*1.062*EBW^{0.062})$; NEg(34%) = $(0.847*1.191*EBW^{0.191)}$; NEg(51%) = $(0.299*1.374*EBW^{0.374)}$; NEg(68%) = $(0.268*1.393*EBW^{0.393)}$.

Using the exponential relation of warmth production (Hpro, $kcal/EBW^{0.75}$/day) as a function of the metabolizable energy consumption (MEI, $kcal/EBW^{0.75}$/day) of the animals in the maintenance group plus the animals in development, we obtained the equation: Hpro = $78.987*e^{0.0035*MEI}$ (Figure 2). From the interception of this equation we found out net energy requirement to maintenance of 78.99 $kcal/kgEBW^{0.75}$/day. The demand of metabolizable energy for maintenance (MEm) were estimated using the exponential equation between Hpro and MEI(Figure 2), by iterations, which represents the point where the MEI and the Hpro are equal. We found a MEm of 116 $kcal/EBW^{0.75}$/day. The efficiency of metabolizable energy usage for maintenance (k_m), of 68.1% were estimated using the relation between the net and metabolizable energy requirements for maintenance (NEm/MEm).

Table 6. Total requirements (maintenance and gain) of net energy, metabolizable energy and total of digestible nutrients of Nellore cattle with different body weights (BW) and average daily gain (ADG) of 0.5; 1.0 and 1.5 kg/day

Weight gain (kg/day)	Body weight (kg)			
	350	400	450	500
Net energy requirements (Mcal/day)				
Maintenance	6.39	7.06	7.72	8.35
0.50	1.10	1.22	1.33	1.44
1.00	2.90	3.21	3.50	3.79
1.50	5.10	5.64	6.16	6.67
Metabolizable energy requirements (Mcal/day)				
Maintenance	9.40	10.39	11.35	12.28
0.5	2.38	2.63	2.87	3.10
1.0	6.24	6.89	7.53	8.15
1.5	10.97	12.13	13.25	14.34
Total of metabolizable energy requirements (Mcal/day)				
0.5	11.77	13.02	14.22	15.38
1.0	15.64	17.28	18.88	20.43
1.5	20.37	22.52	24.60	26.62
	Digestible nutrients requirements (kg/day)			
Maintenance	2.82	3.10	3.38	3.65
0.5	1.03	1.14	1.24	1.34
1.0	2.05	2.26	2.46	2.65
1.5	3.25	3.58	3.89	4.20
Total of digestible nutrients requirements (kg/day)				
0.5	3.85	4.24	4.62	4.99
1.0	4.87	5.36	5.84	6.30
1.5	6.07	6.68	7.27	7.85

EBW (empty body weight) =0.85*BW; GEBW(empty body weight gain)=0.95*GBW; EBWeq = equivalent empty body weight; EBW0.75 = metabolic empty body weight; DMI = dry matter intake; NEm (net energy for maintenance) =78.99 kcal/EBW0.75; MEm (metabolizable energy for maintenance) = 116.13 kcal/EBW0.75; km (efficiency of energy usage for maintenance) = 68.01%; kg(efficiency of energy usage for gain) = 46.5%; RE (retained energy)= 0.0430*EBWeq0.75*GEBW1.3595; TDN and DE = estimated according to BR CORTE (2016): TDN (total digestible nutrients) = (DE)/4.4; DE (digestible energy) = [((MEtotal /DMI) + 0.3032)/0.9455]*DMI.

Table 7. Net requirements of protein for weight gain (NPg) of animals with different body weights

	Concentrate levels (%)				
BW (kg)	17	34	51	68	
	NPg (g/kg GEBW)				
300	219.19	218.21	214.07	226.85	
350	217.20	216.14	211.58	226.39	
400	215.50	214.36	209.44	226.00	
450	214.01	212.80	207.58	225.65	

BW = body weight; EBW (empty body weight) = 0.85*BW; *NPg (17%)= 0.3233*0.9416*$EBW^{0.9416-1}$*1000; NPg (34%) = 0.3280*0.9387*$EBW^{0.9387-1}$ *1000; NPg (51%) =0.3538*0.9246*$EBW^{0.9246-1}$ *1000; NPg (68%) = 0.2478*0.9879*$EBW^{0.9879-1}$ *1000.

Table 8. Requirements of Net protein and metabolizable protein for weight gain, maintenance and total in male, non-castrated Nellore cattle, in different body weight ranges and daily weight gain

	BW (kg)				
Weight gain	300	350	400	450	500
Net protein requirements (g/day)					
Maintenance	109.98	123.46	136.47	149.07	161.33
0.5	82.33	84.76	87.10	89.37	91.58
1.0	177.05	183.43	189.58	195.54	201.33
1.5	279.04	290.25	301.07	311.55	321.75
Metabolizable protein requirements (g/day)					
Maintenance	306.88	344.50	380.78	415.95	450.15
0.5	159.87	164.58	169.13	173.54	177.82
1.0	343.80	356.17	368.11	379.69	390.94
1.5	541.83	563.60	584.60	604.96	624.75
Total metabolizable protein requirements (g/day)					
0.5	466.75	509.08	549.91	589.49	627.97
1.0	650.68	700.67	748.89	795.64	841.09
1.5	848.71	908.10	965.38	1020.91	1074.90

BW= body weight; EBW (empty body weight) = 0.85*BW; $EBW^{0.75}$ = metabolic empty body weight; GEBW (empty body weight gain) =0.95*GBW; $BWJ^{0.75}$ = metabolic body weight while fasting; NPm (net protein for maintenance) = 1.72 g/$EBW^{0.75}$/day; NPg (net protein for weight gain) = 131.8*GEBW+22.7843*ER; RE (retained energy) = 0.0430*$EBW^{0.75}$*$GEBW^{1.3595}$; MPm (metabolizable protein for maintenance) = 4.81 g/$BWJ^{0.75}$/day; efficiency of metabolizable protein usage for weight gain of 51.5%.

Table 9. Total requirements (maintenance and gain) of degradable protein in the rumen, non-degradable protein in the rumen and crude protein

Weight Gain (kg/day)	Body weight (kg)									
	300		350		400		450		500	
	Degradable protein in the rumen									
	g/day	%CP	g/day	%CP	g/day	%CP	g/day	%CP	g/day	%CP
0.5	227.00	40.24	275.15	43.34	323.78	45.89	372.73	48.05	421.87	49.90
1	384.99	44.19	444.61	46.64	503.95	48.73	562.74	50.52	620.73	52.06
1.5	504.08	44.26	573.71	46.62	641.72	48.62	707.67	50.32	771.09	51.76
	Non-degradable protein in the rumen									
	g/day	%CP	g/day	%CP	g/day	%CP	g/day	%CP	g/day	%CP
0.5	401.84	63.90	416.23	60.20	428.37	56.95	438.67	54.06	447.47	51.47
1	505.35	56.76	520.14	53.91	532.96	51.40	544.35	49.17	554.78	47.20
1.5	657.63	56.61	676.16	54.10	693.36	51.93	710.00	50.08	726.76	48.52
	Crude protein									
	g/day	%CP	g/day	%CP	g/day	%CP	g/day	%CP	g/day	%CP
0.5	628.84	13.92	691.38	13.58	752.15	13.32	811.41	13.13	869.35	12.97
1	890.35	13.87	964.76	13.79	1036.91	13.74	1107.10	13.69	1175.51	13.66
1.5	1161.71	15.72	1249.86	15.69	1335.08	15.67	1417.67	15.65	1497.85	15.64

EBW=0.85*BW; GEBW=0.95*GBW; RE = 0.0430*EBW$^{0.75}$*GEBW$^{1.3595}$;NPg = 131.8*GEBW+22.7843*RE - MPm = 4.81 g/BWJ$^{0.75}$/day; efficiency of metabolizable protein usage of weight gain of 51.5%; DMI = 3.002+0.065*BW$^{0.75}$+6.5966*ADG-1.8606*ADG^2; g/day = g/day; %CP= percentage of crude protein; CPmic = -53.07+304.9*CPI+90.8*TDNI-3.13*TDNI$^{2;}$ DRP = CPmic; NDRP= [(MPtotal- ((CPmic*0.64)]/0.80. EBW= empty body weight, BW= body weight; BWJ= body weight while fasting; GEBW= empty body weight gain; GBW= body weight gain; RE= retained energy; NPg= requirements of net protein for weight gain; MPm= requirements of metabolizable protein for maintenance; DRP= degradable protein in the rumen, NDRP= non degradable protein in the rumen; MPtotal= total requirements of metabolizable protein (maintenance more gain).

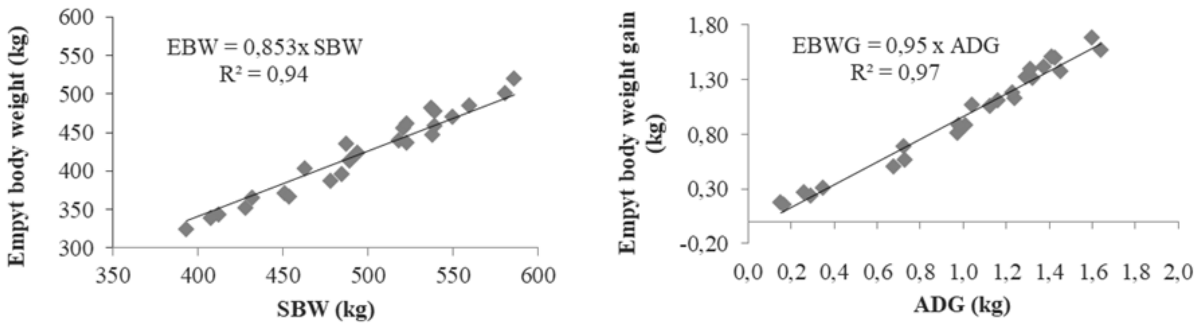

Figura 1. Relationship between the SBW and empty body weight EBW; relationship between average daily gain ADG and EBWG

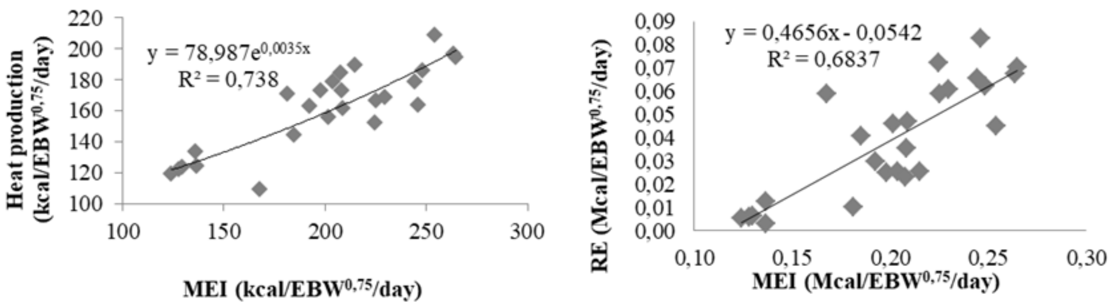

Figura 2.Relationship between heat production (Hpro) and metabolizable energy consumption in Nellore cattle; relationship between retained energy (RE) and intake metabolizable of energy (MEI)

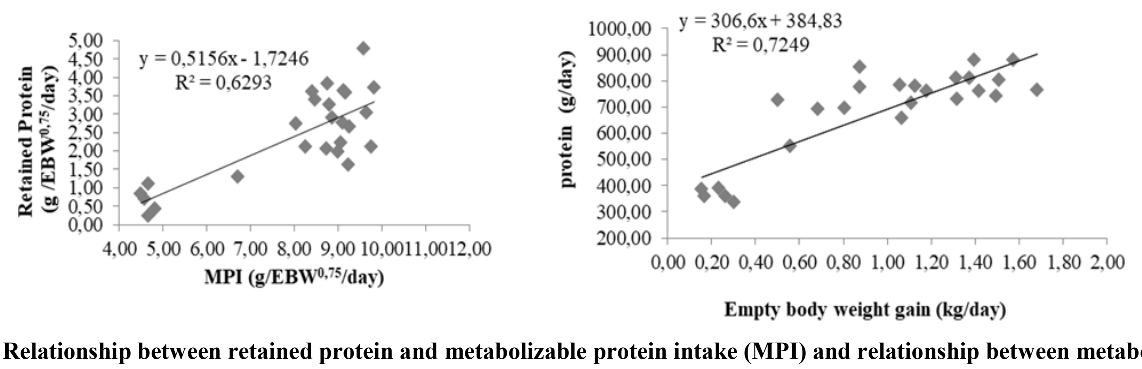

Figura 1. Relationship between retained protein and metabolizable protein intake (MPI) and relationship between metabolizable protein consumption and empty body weight gain (GEBW)

Using the inclination coefficient from the linear regression equation between RE and MEI(Figure 2), we found the efficiency of metabolizable energy usage for gain (k_g) which was 46.56%. From the results obtained we estimated the energy requirements on male, non-castrated Nellore cattle with different BW and ADG (Table 6). We observed that in every way the energy requirements were expressed, the requirements increased as the body weight did. This behavior was expected due to the fact that the energy demand grows as the animal grows; such fact is characterized metabolically by the deposition of tissues in the animal's body derived from the synthesis and degradation of protein and fat. After deriving the joined equation of the amount of body protein (Table 4) we obtained the equation to estimate the demand of net protein per kilo gained of EBW (GEBW) for different body weights: NPg = (0.2612*EBW$^{-0.029)}$ *1000) (Table 7). There was a decrease on the net demand of protein for weight gain with an increase of body weight on the animals. Therefore, the NPm of the animals in this experiment was 1.72 g/EBW$^{0.75}$/day; and the efficiency of metabolizable protein usage for weight gain was 51.5%. We obtained the equation: MPI = 384.8 + 306.6*GEBW(Figure 3; b) by dividing the interception of this regressing by the average of the empty body weight of the animals in development and maintenance obtaining MPm of 4.58 g/EBW$^{0.75}$/day (384.83/83.96). This value is a little above the values fixed by NRC (2000), of 3.8 g/BW$^{0.75}$/day for Taurus cattle and Valadares Filho *et al*. (2010) of 4.0 g/BW$^{0.75}$/day, for Zebu cattle. When converting the MPm expressed in g/EBW$^{0.75}$/day to g/BWJ$^{0.75}$/day, we found the value 4.81 g/BWJ$^{0.75}$/day). From the regression between the RP (g/EBW$^{0.75}$/day) and the MPI (g/EBW$^{0.75}$/day) we obtained the requirements of net protein for maintenance (PLm) represented by the modulus of the interception of the equation (β_0) and the efficiency of the net protein conversion into metabolizable for weight gain which is the coefficient of inclination (β_1)(Figure 3;a). To calculate the net protein, metabolizable protein and total metabolizable protein (maintenance + gain) requirements in different weight and growth rates we used the equation obtained in this study to estimate the net demand of protein for weight gain (NPg = 131.8*GEBW+22.7843*ER), where we considered the equation obtained from the data in this experiment to estimate the retained energy in Nellore cattle Nellore (RE = 0.0430*EBW$^{0.75}$*GEBW$^{1.3595}$), a PLm of 1.72 g/EBW$^{0.75}$/day, MPm of 4.81 g/BWJ$^{0.75}$/day and the efficiency of the metabolizable protein for weight gain of 51.5% (Table 8).

The requirements of protein for weight gain increases as the body weight increased and also as the desired gains increased. It is important to highlight that the gain is characterized by the increase in tissue deposition. The requirements on MPm increase as the body weight did so, considering that the requirements for maintenance are a function of the body weight. On the other hand, the metabolizable protein for weight gain and total raised as the body weight increased. From the requirements of metabolizable protein described on Table 8 and the consumption of total digestible nutrients we calculated the daily needs of crude protein (CP), degradable protein in the rumen (DRP) and non- degradable protein in the rumen (NDRP), according to the protocol suggested by Valadares Filho *et al*. (2016) (Table 9). The DMI equation as a function of BW$^{0.75}$ and ADG, estimated from the data of the animals in development (DMI=-3.002+0.065* BW$^{0.75}$+ 6.5966* ADG-1.8606*ADG^2) was used to predict the requirements of CP and the percentage of dry matter.

We observed and increase of the DRP and the NDRP as the body weight and the daily weight gain increase. However, the DRP participation in the CP was bigger as the body weight increased which reduces the need of NDRP in the diet to supplement the total requirements of CP and, consequently, the cost of production. The requirements of NDRP increased as the DRP decreased with the raise in the desired ADG.

DISCUSSION

The proportion of content in the gastrointestinal tract (GIT) can explain the difference between conversion factors, because Taurus beef cattle have greater development of the GIT when compared to the Zebu cattle which gives them a smaller conversion rate. According to Owens *et al*. (1995), higher accuracy on the estimate of the body composition can be reached when calculated from the EBW. According to Valadares Filho *et al*. (2016) the GEBW is considered the actual body weight gain obtained during a determined period of evaluation divided by the evaluation period (days), this variable is important in the calculation of the nutritional requirements for considering the ingestion of food. To convert the GEBW into GBW, Marcondes *et al*. (2011) found the rate 0.92; but Costa e Silva *et al*. (2012) found it as 1.01 for Nellore cattle. Valadares Filho *et al*. (2016) recommend 0.96 for Nellore cattle raised in confinement, in accordance to the value obtained in the present work. Considering that these indexes are essential to calculate the nutritional demand, which allows us to calculate the nutritional requirements in practical conditions of feeding management.

The NRC (2000) suggests 88.55 (77+15%) kcal/kg EBW$^{0.75}$/day as a net requirement of energy for maintenance for non-castrated animals, while the value found in this work is below the proposed by such system. On the other hand, the NEm of 78.99 kcal/kgEBW$^{0.75}$/day was close to the recommended by Valadares Filho *et al*. (2016), of 75 kcal/kgEBW$^{0.75}$/day for confined animals. Marcondes *et al*. (2011) working with crossbred and Nellore, and Costa e Silva *et al*. (2012), working with confined Nellore cattle (75.8 e 76.5 kcal/kgEBW$^{0.75}$/day, respectively) presented similar values. According to Ferrell e Jenkins (1984) the maintenance requirements are influenced by race, sex, age, body composition and nutritional level to which the animals are submitted. So, it is necessary to consider such factors to minimize errors on the estimates, choosing, then, a more representative and adequate model. Valadares Filho *et al*. (2010) found a MEm value of 112.4 kcal/EBW$^{0.75}$/day for confined Nellore cattle, which is below the one found in this work. Now, in Valadares Filho *et al*. (2016), regarding confined Nellore cattle, they found a value closer to the one found in the present work (118 kcal/EBW$^{0.75}$/day). MEm requirements obtained (116 kcal/EBW$^{0.75}$/day) is close to the one found in Marcondes *et al*. (2011) for Nellore and crossbred, castrated animals (112.82 kcal/EBW$^{0.75}$/day) and by Costa e Silva *et al*. (2012) for Nellore bovines non-castrated (113.84 kcal/EBW$^{0.75}$/day); it is evident that, possibly, these differences are related to the genetic group and the sexual condition of the animals used. The great challenge is to determine the factors that affect the MEm requirements and could cause these differences mainly by the complexity of the physiological and metabolic alterations for bigger understanding. The value (k_m) of 68.1% is in accordance to the one found in Valadares Filho *et al*. (2010), who recommended a k_m of 68% for Zebu cattle with a BW of 450 and ADG of 1 kg. Lately, Marcondes *et al*. (2011), found for male non-castrated and confined Nellore cattle a value of 67%. On the other hand, NRC (2000) and Valadares Filho *et al*. (2006) suggested a k_m of 65 and 63%, respectively; Garrett (1980) observed that the (k_m)can be affected by the weight gain composition and the nutritional plan, besides other factors such as sex, race, age, environment, creation system and the metabolizable energy concentration in the diet (CSIRO, 2007; NRC, 2000) which could justify the differences found for efficiency of metabolizable energy usage for maintenance. ARC (1980) suggests a k_g between 50 and 59%, which is above the result obtained in this work (46.6%). On the other hand, values closer to the one found on this work were also found by Tedeschi *et al*. (2002) and Valadares Filho *et al*. (2006), of 45.9 and 47%, respectively. The energy requirements estimated in this work, using the models obtained from the experimental data, were smaller than the ones found by Valadares Filho *et al*. (2010) and Marcondes *et al*. (2010a) for non-castrated Nellore bovines in confinement; which shows the necessity of further research aiming to determine the nutritional requirements of purebred Zebu cattle in confinement. We observed a reduction of approximately 2% on the NPg with an increase of bodyweight from 250 to 450 kg. Tedeschi *et al*. (2013) reported the protein proportion in the empty body is inversely proportional to the body condition, physiological state and strongly linked to the flux of the body's reserve of fat. Goulart *et al*. (2008) observed the same behavior with Nellore bovines of 450kg, ended in confinement.

Chizzotti *et al*. (2008) e Marcondes *et al*. (2010) followed the model proposed by the NRC (2000), however using the GEBW instead of GBW, and suggested the equations: NPg = GEBW × (217 – 12.8 × ER/GEBW); NPg = GEBW × (140 – 0.70 × RE/GEBW), respectively. Valadares Filho *et al*. (2010), through data combination from various experiments, obtained the equation: NPg = 238.79* GEBW-15.68*RE, for non-castrated Nellore male cattle. The efficiency of MPg usage of 51.5% is close to the reported by NRC (2000), of 49.2% for animals with a bodyweight above 300 kg, and above the recommended by Valadares Filho *et al*. (2010), of 46.9%; and Marcondes *et al*. (2010b), of 37%. These changes in protein requirements can be associated to differences on the genetic group of the experimental subjects used. Age, body composition, nutritional level, physiological state and environmental conditions; it is a challenge to stablish the protein requirements due to the complexity of the factors involved. Thus, it is essential that new research is conducted using the management conditions and environmental conditions in tropical weather areas (Valadares Filho *et al*. 2016; Marcondes *et al*. 2013). Probably, because of differences on the nutritional plan or quality adopted to formulate the feed. The quality of the food as well as the protein source and biological value of the microbial protein affect the efficiency of PMg usage (Souza *et al*., 2012). While balancing the ration, the higher the desired weight gain, the higher is the need to provide NDRP on the diet increasing the demand for protein sources coming from better amino acid profiles to the animal. The MPm is well above the reported by BCNRM (2016) and Valadares Filho *et al*. (2016) of 3.96 e 3.80 g/BWJ$^{0.75}$/day, respectively; probably because of the higher BWJ present on the animals for this experiment. According to Marcondes *et al*. (2010) there is a higher demand for net protein for weight gain in non-castrated animals, with bigger bodyweight. That is probably due to the effect of testosterone which promotes higher anabolism of nitrogen when gaining weight, resulting in more growth and efficiency in gain, however, increasing the requirements of NPg. Furthermore, Silva *et al*. (2017) reported that the study conditions, number of samples and evaluation periods can denote said differences; therefore, it is important new researches to validate the results for the estimate nutritional demand of purebred Zebu cattle in tropical regions.

Conclusion

The equation RE= $0.0430*EBWeq^{0.75}*GEBW^{1.3595}$ is recommended to predict the energy net demand for weight gain on confined Nellore cattle. The confinement Nellore cattle presented 51.5% weight gain efficiency of metabolizable protein. It is recommended to use the equation: NPg = 131.8*GEBW+22.7843*RE to predict the daily net demand of protein for weight gain of male non-castrated Nellore bovines in confinement in tropical climate conditions. Studies on nutritional requirements of purebred confined Zebu cattle are important in tropics and must be encouraged in order to gather precise information on their protein and energy requirements.

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