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# Genetic and environmental factors affecting ultrasound measures of longissimus muscle area and backfat thickness in Nelore cattle

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#### Abstract

The aim of the present study was to evaluate the genetic and environmental factors affecting records of longissimus muscle area (LMA) and backfat thickness (BF) obtained between the 12th and 13th ribs, and rump fat thickness (RF) between the hook and pin bones, measured by real-time ultrasound in Nelore cattle. Also, weight records of 22,778 animals born from 1998 to 2003, in ten farms across six Brazilian states were used. Carcass traits as measured by ultrasound of the live animal were recorded from 2002 to 2004 in 2590 males and females with ages varying from 450 to 599 days. Fixed models including farm, year and season of birth, sex and type of feed effects, and the covariates age of dam (AOD) and age of animal at measurement were used to study the effect of environmental factors on these traits. The genetic parameters for LMA, BF and RF were estimated with two and three-trait animal models with 120-day weights using a restricted maximum likelihood method. All environmental effects significantly affected carcass traits, with the exception of year of birth for BF and AOD for LMA. The heritability estimates for LMA, BF and RF were 0.35, 0.51 and 0.39, respectively. Standard errors obtained in one-trait analyses were from 0.07 to 0.09. Genetic correlation estimates between LMA and the two traits of subcutaneous fat were low (close to zero) and 0.74 between BF and RF, indicating that the selection for LMA should not cause antagonism in the genetic improvement of subcutaneous fat measured by real-time ultrasound.

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#### 1. Introduction

Meat production in tropical areas is dependent, mainly, on *Bos indicus* (Zebu) breeds. Currently, Brazil has the greatest commercial cattle herd in the world and the majority, about 80%, is comprised of Zebu and their

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crossbreds. Since 1995, the Brazilian Association of Zebu Breeders (ABCZ) has registered about three million animals and more than 75% of them are Nelore (www. abcz.org.br).

Over the last 15 years the interest in using genetically evaluated animals has been increasing. However, most breeding evaluation programs in tropical countries have generally been focused on weight and scrotal circumference measures at different ages, whereas carcass traits have been neglected.

Cattle breeders are faced with the challenges of producing beef to satisfy consumer demands for quantity and quality and at the same time of raising cattle that are productive and profitable under low-cost pasture systems. In terms of quality, Zebu meat is considered to be below the standards of competing markets, and without consistency. The most important factors that affect carcass value are retail cut yield and eating quality (Boleman et al., 1998), both of which are related to carcass fat (subcutaneous and intramuscular). One of the problems with Zebu breeds is their low propensity to deposit subcutaneous fat, because adequate fat cover is required to minimize evaporative weight loss in the cooler. Furthermore, subcutaneous fat thickness is related to beef tenderness through its action as an insulator to reduce the rate of chilling and muscle fiber cold-shortening (Dolezal et al., 1982; Tatum et al., 1982) that occurs during very rapid carcass cooling (Aberle et al., 2001).

One tool to evaluate carcass merit, without the need for slaughtering the animals or attribution of visual scores, is ultrasound. This technology is a rapid, noninvasive method for data collection which leaves no harmful residues in the meat of the animals studied (Perkins et al., 1992a,b). Studies have demonstrated that ultrasound is an objective and accurate tool in the selection for muscularity, subcutaneous fat thickness, intramuscular fat and boneless meat yield (Wilson, 1992; Herring et al., 1994).

To improve the quality of the meat, genetic breeding programs for carcass quality that meet consumer demands and promote genetic progress in Zebu herds will be necessary. Thus, the objective of the present study was to investigate the main environmental factors that affect carcass traits measured by real-time ultrasound, as well as their heritability and genetic correlations estimates, which are of fundamental importance for the development and implantation of genetic breeding programs for these traits.

## 2. Materials and methods

Data were provided by Nelore Breeding Program — Nelore Brazil (PMGRN). The following carcass traits as

measured by ultrasound of the live animal were evaluated: longissimus muscle area (LMA) and backfat thickness (BF) obtained from a cross-sectional image on the longissimus dorsi muscle measured between the 12th and 13th ribs, and rump fat thickness (RF) measured at the intersection between the gluteus medium and biceps femoris muscles located between the hooks and pin bones. Backfat thickness was estimated at the 3/4 position from the chine bone end of the longissimus muscle using the cross-sectional ribeye image.

Real-time ultrasound images were collected with an ALOKA 500V (Corometrics Medical Systems, Inc., Wallingford, CT, USA) apparatus equipped with a 3.5-MHz, 17.2-cm, linear probe (transducer — Aloka Co. Ltd., Tokyo, Japan) and an acoustic coupler (standoff pad) by Brazilian Ultrasound Guidelines Council-certified field technicians. The images were digitized and stored using an image capture system (Blackbox, Biotronics, Ames, IA, USA). Records were subsequently interpreted in a centralized laboratory responsible for the quality of the data (Aval Serviços Tecnológicos S/C, Uberaba, Minas Gerais, Brazil) using appropriate software with demonstrated accuracy and precision (two decimal figures).

Once the scanning area was determined, vegetable oil was applied, and the area was curried free of dirt and debris before transducer placement. For collection of the LMA and BF images, a standoff pad was used to guarantee acoustic contact between the linear probe and the animal's natural body shape. The transducer placement was first determined by palpating the left side of the animal between the 12th and 13th ribs. The ultrasound probe was placed toward the midline, between and parallel to the 12th to 13th rib bones and moved laterally until the longissimus muscle came into full view on the screen (Perkins et al., 1992a).

The animals with ultrasound measurements were born from 2000 to 2002 and recorded from 2002 to 2004 for 2590 males and females ranging in age from 450 to 599 days and belonging to ten farms in six Brazilian States. Data of 120-day standardized weights (W120) from 22,778 animals born between 1998 and 2003 in all herds were analyzed.

Data of animals with records three standard deviations above or below the mean of their contemporary group (CG) were eliminated. Contemporary groups containing progenies of only one sire and those with fewer than three animals for the carcass traits as measured by ultrasound of the live animal were excluded, whereas for W120, CGs with fewer than eight animals were eliminated. Table 1 shows a summary of the data structures.

First, to determine the effect of environmental factors on carcass traits as measured by ultrasound of the live animal, analyses of variance were performed using the least squares method, estimating adjusted means of the fixed effects and regression coefficients for the covariates using the GLM procedure of the SAS program (SAS, 2000). Fixed models considering the following variables were used: herd, sex, season of birth [SB = spring (September, October and November), summer (December, January and February), autumn (March, April and May), and winter (June, July and August)], year of birth (YB = 2000, 2001 and 2002), and

Table 1 Structure of the data set for the traits longissimus muscle area (LMA), backfat thickness (BF), rump fat thickness (RF) and 120-day standardized weight (W120) obtained for Nelore cattle

Trait	N	Mean±SD	No. of sires	No. of dams	No. of CG
LMA (cm <sup>2</sup> )	2590	48.38±8.72	244	2451	301
BF (mm)	2417	$1.93 \pm 1.36$	231	2302	288
RF (mm)	2428	$3.05 \pm 1.97$	232	2308	291
W120 (kg)	22,778	$128.78\!\pm\!19.23$	540	11,759	630

N = number of animals analyzed; No. of sires = number of sires (fathers) per trait; No. of dams = number of dams (mothers) per trait; No. of CG = number of contemporary groups per trait; SD = standard deviation.

feeding management (pasture or pasture with supplementation), in addition to the covariates age of dam at calving (AOD) in months as linear and quadratic effects (23 to 223 months) and age of animal in days as linear and quadratic effect for LMA and linear effect for BF and RF (450 to 599 days).

In order to consider all possible interactions among fixed effects, for estimation of variance components, the fixed effects were grouped for the formation of CG. The CG for the traits LMA, BF and RF were defined as herd-sex-management group-year of recording. If the range of ages in a CG were higher than 50 days, the CG was subdivided into 50-day intervals (450–499, 500 to 549, and 550 to 599 days), a practice commonly referred to as 'age slicing', limiting the difference in ages among animals which were directly compared (Meyer, 2005). The CG for W120 was defined in a similar manner, but with year and bimonthly period of birth instead of age slicing and year of recording.

In order to minimize the influence of selection and culling, the ultrasound traits were analyzed by three-trait models with W120 as anchor, since all animals were recorded at this age. The variance components were estimated using animal models and a restricted maximum likelihood (REML) method as described by Patterson and Thompson (1971). The MTDFREML (multiple trait derivative-free restricted maximum likelihood) program developed by Boldman et al. (1995) was used for REML analyses. Convergence was assumed when the variance of the simplex ( $-2\log_e$  likelihood) reached  $1 \times 10^{-9}$ . The program was reinitiated after each convergence, using the previous estimates as initial values. This procedure was repeated until the differences between the last two convergences – 2log<sub>e</sub> likelihood values were less than  $1 \times 10^{-4}$ . The relationship matrix included up to three generations back and contained a total of 36,133 animals.

The model for BF and RF included the direct genetic effect as a random, fixed effect of CG, and, as covariates, age of animal (AOA; linear effect: 450 to 599 days) and age of dam (AOD; linear and quadratic effects: 23 to 283 months). LMA was analyzed using the same model without the AOD covariate.

The model for carcass traits as measured by ultrasound of the live animal can be represented in matrix format as:

$$y = X\beta + Za + e$$

where  $\mathbf{y}$  is the vector of observations,  $\boldsymbol{\beta}$  is the vector of unknown fixed effects,  $\mathbf{a}$  is the vector of unknown random effects that represent the additive genetic values of each animal,  $\mathbf{e}$  is the vector of unknown residual random effects, and  $\mathbf{Z}$  are incidence matrices that relate the records to fixed and random genetic effects, respectively.

The assumptions regarding the distribution of  $\mathbf{y}$ ,  $\mathbf{a}$  and  $\mathbf{e}$  can be described as

$$\begin{bmatrix} \mathbf{y} \\ \mathbf{a} \\ \mathbf{e} \end{bmatrix} \sim N \left\{ \begin{bmatrix} X\beta \\ \mathbf{0} \\ \mathbf{0} \end{bmatrix}, \begin{bmatrix} \mathbf{Z}\mathbf{G}\mathbf{Z}' + \mathbf{R} & \mathbf{Z}\mathbf{G} & \mathbf{R} \\ \mathbf{G}\mathbf{Z}' & \mathbf{G} & \mathbf{\Phi} \\ \mathbf{R} & \mathbf{\Phi} & \mathbf{R} \end{bmatrix} \right\},$$

where G is the variance—covariance matrix of the random effects of vector  $\mathbf{a}$ , and R is the matrix of residual variances and covariances. The G and R matrices are described as  $G = A \otimes G_0$ , where A is the relationship matrix,  $G_0$  is the additive genetic variance—covariance matrix between records and  $\otimes$  is the direct product operator, and as  $R = I \otimes R_0$ , where I is the identity matrix of the same order as  $\mathbf{y}$ ,  $R_0$  is the matrix of residual variances and covariances between records,  $\mathbf{0}$  is the null vector, and  $\mathbf{\Phi}$  is the null matrix.

The model for W120 included direct and maternal genetic effects and maternal permanent environmental effect as random and fixed effects of CG and age of dam classes (six classes: until 35, 36–47, 48–59, 60–71, 72–119 and higher than 120 months of age). The covariance between direct and maternal genetic effects was set at zero as suggested by Meyer

Table 2 Summary of analysis of variance for ultrasound measurement of longissimus muscle area (LMA), backfat thickness (BF) and rump fat thickness (RF) in Nelore cattle

Source of	d.f.		Mean squares		
variation		LMA (cm <sup>2</sup> )	BF (mm)	RF (mm)	
Farm	9	7130.02 ***	76.53 ***	199.77 ***	
Sex	1	8122.15 ***	192.89 ***	924.03 ***	
YB	2	3776.79 ***	0.54	2.56	
SB	3	268.53 ***	2.35 ***	3.44 *	
MS	1	5662.93 ***	3.55 *	20.98 ***	
AOD	1	1.57	10.96 ***	15.75 ***	
$AOD^2$	1	0.20	7.33 ***	15.35 ***	
AOA	1	126.20 *	26.35 ***	29.55 ***	
$AOA^2$	1	216.51 *	_	_	
Residual		34.80	0.62	1.41	
Residual d.f.		2474	2397	2408	
$R^2$		0.54	0.49	0.64	

d.f. = degrees of freedom;  $R^2 =$  coefficient of determination; YB = year of birth; SB = season of birth; MS = management system; AOD = age of dam at calving (linear and quadratic effect); AOA = age of animal on ultrasound date (linear and quadratic effect); Significant: \* = P < 0.05 and \*\*\* = P < 0.0001.

(1997), Benyshek et al. (1998) and Albuquerque and Meyer (2001).

# 3. Results and discussion

Analysis of variance results are summarized in Table 2. All effects included in the model significantly affected the ultrasound measurements (P<0.05), except year of birth (effect associated with climate, management, feeding, among others) which had no significant effect on BF or RF.

Table 3 shows the least squares mean estimates obtained by analysis of variance. The LMA was on average 7.99 cm<sup>2</sup> larger in males compared to females. This difference was expected since in Bos taurus breds males are generally heavier and more muscular than females (Hassen et al., 1998; Crews et al., 2002; Meyer, 2005). On the other hand, males had lower BF and RF than females (Table 3). This finding was expected since physiologically females tend to deposit fat earlier than males as indicated by allometric growth curves (Burrow et al., 1991). Our results agree with those from Hassen et al. (1998), Crews et al. (2002) and Meyer (2005) who reported significant differences in subcutaneous fat between bulls and heifers. However, Wilson et al. (2000), working with Angus animals ranging in age from 320 to 440 days, did not observe differences for male and female subcutaneous fat. The authors suggested that this lack of difference could be explained by the feeding system to which these animals were submitted (feedlot).

Animals reared on tropical pasture with supplementation deposited 7.6% (BF) and 13.8% (RF) more subcutaneous fat and 15.3% more muscle (LMA) than those reared only on tropical pasture. These results

Table 3
Estimated means for ultrasound measurement of longissimus muscle area (LMA), backfat thickness (BF) and rump fat thickness (RF) according to sex, management system and season of birth in Nelore cattle

Source of	Variable	riable Trait		
variation		LMA (cm <sup>2</sup> )	BF (mm)	RF (mm)
Sex	Male	57.39	1.54	2.43
	Female	49.40	2.81	5.18
Management	Pasture	49.60	2.07	3.56
system	Pasture with supplementation	57.19	2.28	4.05
Season of birth	Spring	53.69	2.29	3.82
	Summer	52.81	2.20	3.73
	Autumn	51.96	2.00	3.67
	Winter	55.11	2.21	4.01

indicate that Nelore animals respond significantly to improved nutrition and these management system effects should be considered in the models for animal genetic evaluation.

With respect to season of birth, higher means were observed in animals born in winter and spring for all ultrasound measurements analyzed. Differences in season of birth are associated with climatic variations that affect the availability of feed, especially in animals reared on tropical pasture as was the case in the present study. Animals born during the winter and spring seasons are weaned during summer and autumn, respectively. Thus, these animals pass through a period of food restriction immediately after weaning. On the other hand, they find more favorable environmental conditions as they approach 18 months of age, reaching this age in better body condition. Bocchi et al. (2004) have shown that, in Brazil, Zebu animals born in winter and spring were heavier at weaning than animals born in summer and autumn. In a pasture-based production system, concentrating the calving in winter and spring could favor superior Zebu carcasses.

The covariate AOD significantly affected BF and RF but not LMA. Cows undergo morphological and physiological changes with age which affect their maternal ability, i.e. milk yield and offspring care, and, consequently, the performance of their calves. Alencar (1987) observed that milk production of dams is responsible for 26.2% and 20.3% of the variation in weight gain between birth and weaning in Canchim and Nelore animals, respectively. The amount of milk produced by a beef cow is fundamental for the calf's development until weaning. Cardoso et al. (2004) considered that post-weaning characters would be less affected by maternal effects than pre-weaning characters. Although this is clearly true, we found that postweaning traits such as BF and RF are influenced by AOD (Table 3). Probably, AOD effects on post-weaning traits are a "carry-over" of those on weaning (Meyer et al., 1993; Eler et al., 1995; Albuquerque and Meyer, 2001). On the other hand, Wilson et al. (2000) did not find a significant effect of AOD on BF or RF in Angus cattle, perhaps because the model included animal weight pre-adjusted for AOD as a covariate. Means of BF and RF as a function of AOD, as well as their respective regression equations are presented in Fig. 1. Although statistically significant, the AOD effect on subcutaneous fat was small. Calves born to younger and older dams showed RF and BF slightly higher than those born to mature cows. This effect is contrary to that found for AOD on hip height at scan date and on weaningweight (not shown). Offspring of cows that produce less

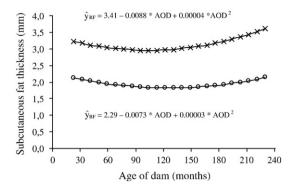


Fig. 1. Rump fat thickness (RF) and backfat thickness (BF) means as a function of age of the dam (AOD) for Nelore cattle.

milk usually present compensatory post-weaning gains, i.e., they tend to gain weight faster than those weaned by more productive dams (Muniz and Queiroz, 1999). Sainz et al. (1995) showed that steers subjected to feed restriction during the growing phase had more rapid and efficient (compensatory) growth during the finishing period than full-fed counterparts, largely due to greater feed intake. Conflicting data in the literature (e.g., Carstens et al., 1991) indicate that the influence of compensatory growth on weight gain composition is complex. Tudor et al. (1980) and Abdalla et al. (1988) observed increased fat contents in the gains of compensating animals, in agreement with the slightly fatter calves from younger or older cows observed in this study.

The covariate AOA significantly influenced (P<0.05) all ultrasound measurements. Longissimus muscle area increased almost linearly with age until 520–540 days of age (Fig. 2), followed by a slightly faster growth thereafter, suggesting that this muscle (longissimus

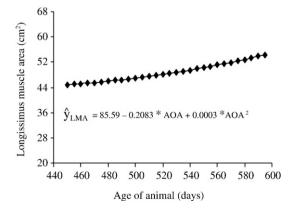


Fig. 2. Longissimus muscle area (LMA) means as a function of age of animal (AOA) for Nelore cattle.

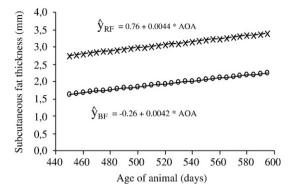


Fig. 3. Rump fat thickness (RF) and backfat thickness (BF) means as a function of age of the animal (AOA), for Nelore cattle.

dorsi) presents late maturity. Wilson et al. (1993), analyzing carcass traits in Angus, reported that age of animal significantly influenced LMA and BF only when the regression analysis was conducted for steers varying widely in age, from 360 to 660 days of age. However, Crews et al. (2002) found AOA to have significant linear effects on yearling ultrasound BF and LMA in B. taurus animals. In the current study, subcutaneous fat thickness increased linearly as a function of animal age (Fig. 3). The RF means were higher than BF means at all ages. In many cases, Zebu animals show lower subcutaneous fat means than B. taurus animals (Wilson et al., 1993; Crews et al., 2003; Sainz et al., 2003). This is not only due to genetic differences but also to differences in management, since in tropical countries Zebu cattle are raised mainly on pasture and fat deposition usually only starts again at 18 months of age, after the dry season. Rump fat was on average 58% higher than BF, with this difference being statistically significant (P<0.0001). This difference was higher than that reported by Sainz et al. (2003) for Nelore cattle and by Wall et al. (2004) and Meyer (2005) for B. taurus animals, who observed differences ranging from 20 to 46%. This finding suggests that rump fat is deposited earlier than rib fat, as reported by Tait (2002).

Table 4
Estimates of variance, heritability (diagonal), environmental (below) and genetic (above) correlations for ultrasound measurement of longissimus muscle area (LMA), backfat thickness (BF) and rump fat thickness (RF) in Nelore cattle

Trait	LMA	BF	RF
LMA	0.35	0.06	-0.04
BF	0.19	0.52	0.74
RF	0.16	0.48	0.40
$\hat{\boldsymbol{\sigma}}_{\mathrm{a}}^{2}$	$10.62 \text{ cm}^4$	$0.31 \text{ mm}^2$	$0.40 \text{ mm}^2$
RF $\hat{\boldsymbol{\sigma}}_{a}^{2}$ $\hat{\boldsymbol{\sigma}}_{r}^{2}$	19.59 cm <sup>4</sup>	$0.29~\text{mm}^2$	$0.60 \text{ mm}^2$

 $\hat{\sigma}_{a}^{2}$  = direct additive genetic variance;  $\hat{\sigma}_{r}^{2}$  = residual variance.

Table 4 shows the estimates of variance components and heritability obtained by three-trait analysis. The heritability estimates for LMA, BF and RF ranged from moderate to high. Standard errors obtained by one-trait analysis ranged from 0.07 to 0.09. Differences between estimates obtained by one- and three-trait analysis were only observed for LMA (from 0.29 to 0.35), probably because of the positive correlation between LMA and W120 (0.53). The heritability estimates obtained in the present study for LMA are close to those reported for B. taurus, Bos indicus and mixed breeds (Moser et al., 1998; Stelzleni et al., 2002; Crews et al., 2003; Sainz et al., 2003). However, for the subcutaneous fat traits (BF and RF), the heritability estimates were higher than those reported by Arnold et al. (1991), Robinson et al. (1993), Moser et al. (1998) and Kemp et al. (2002) for Angus and Hereford animals. Heritability estimates as high as those observed in the present study (higher than 0.40) were reported by Shepard et al. (1996) for Angus, by Reverter et al. (2000) for Hereford and Angus, and by Crews et al. (2003) for the Simmental breed. Heritability estimates for carcass traits in Nelore cattle are limited. with only few estimates being reported, most of them in Congress Proceedings. The estimates obtained in the present study are close to those reported by Sainz et al. (2003), except for RF (0.29, 0.44 and 0.62 for LMA, BF and RF). High heritability estimates for carcass traits as measured by ultrasound of the live animal in Nelore cattle could be expected since these traits were never considered as selection criteria.

The estimated genetic correlations between LMA and the two subcutaneous fat traits were low (close to zero), indicating that these traits are determined by different groups of genes (additive action) and the selection for LMA should not cause antagonism in the genetic improvement of BF and RF. When there is interest in selecting for subcutaneous fat as well as for LMA, both traits have to be considered as selection criteria. Appropriate selection indexes have to be developed for specific production systems and/or markets. For example, there are two types of beef carcass grades in the United States, quality grades and yield grades. Beef carcasses may carry a quality grade, a yield grade or both. Yield grades reflect the yield of boneless, closely trimmed retail cuts from the round, loin, ribs and chuck. They represent a percentage of retail cuts in the carcass and increase with the longissimus muscle area and decrease with the subcutaneous fat (May et al., 2000). In the United States, the selection could be for animals with positive genetic values for LMA and negative or next to zero for BF and RF which is possible in accordance with the genetic correlations estimated in this work. Nevertheless, quality grades in Brazil indicate meat expected palatability or eating satisfaction which depends on numerous factors such as uniform subcutaneous fat distribution over the carcass surface to protect the carcass from excessively rapid cooling. Thus the selection should be aimed at animals with positive breeding values for LMA, BF and RF.

Backfat thickness and rump fat thickness measured by real-time ultrasound are mainly determined by the same groups of genes; however the genetic correlation between them is less than 0.80. Robertson (1959) suggested that when the genetic correlation between two traits is less than 0.80, both must be included in selection programs. The selection for RF should produce a correlated genetic gain in BF that would be only 63% of the gain obtained by direct selection for BF. Therefore, selection for both traits (BF and RF) is required, particularly because subcutaneous fat has shown little uniformity in the carcass (Holloway et al., 1990; Wall et al., 2004). The estimates of genetic correlations between the carcass traits as measured by ultrasound of the live animal are in agreement with estimates reported in the literature for B. taurus (Reverter et al., 2000; Kemp et al., 2002; Stelzleni et al., 2002) and Nelore (Sainz et al., 2003).

# 4. Conclusions

Longissimus muscle area, backfat thickness and rump fat thickness measured by real-time ultrasound show a wide genetic variability in Nelore cattle. These traits should respond rapidly to individual selection if included in genetic breeding programs. There are no genetic antagonisms between longissimus muscle area and the two subcutaneous fat traits measured by real-time ultrasound.

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