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# Assessment of *DGAT1* and *LEP* gene polymorphisms in three Nelore (*Bos indicus*) lines selected for growth and their relationship with growth and carcass traits<sup>1</sup>

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**ABSTRACT:** The aim of this study was to analyze *LEP* and *DGAT1* gene polymorphisms in 3 Nelore lines selected for growth and to evaluate their effects on growth and carcass traits. Traits analyzed were birth, weaning, and yearling weight, rump height, LM area, backfat thickness, and rump fat thickness obtained by ultrasound. Two SNP in the *LEP* gene [*LEP* 1620(A/G) and *LEP* 305(T/C)] and the K232A mutation in the *DGAT1* gene were analyzed. The sample consisted of 357 Nelore heifers from 2 lines selected for yearling weight and a control line, established in 1980, at the Estação Experimental de Zootecnia de Sertãozinho

(Sertãozinho, Brazil). Three genotypes were obtained for each marker. Differences in allele frequencies among the 3 lines were only observed for the *DGAT1* K232A polymorphism, with the frequency of the A allele being greater in the control line than in the selected lines. The *DGAT1* K232A mutation was associated only with rump height, whereas *LEP* 1620(A/G) was associated with weaning weight and *LEP* 305(T/C) with birth weight and backfat thickness. However, more studies, with larger data sets, are necessary before these markers can be used for marker-assisted selection.

**Key words:** *Bos indicus*, molecular marker, productive trait, quantitative trait locus

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## INTRODUCTION

In dairy and beef cattle, the K232A mutation in the *DGAT1* gene has been associated with greater milk fat content and greater fat content of semitendinosus muscles, respectively (Winter et al., 2002; Thaller et al., 2003; Grisart et al., 2004). It is possible that this mutation is directly responsible for the QTL variation at the centromeric end of BTA14 (Coppieters et al., 1998; Thaller et al., 2003).

Several polymorphisms in the *LEP* gene have been described in cattle, including *LEP* 1620(A/G) (GenBank: Y11369) located in intron 2 of the *LEP* gene (Lien et al., 1997) and *LEP* 305(T/C) (GenBank: AJ236854) located in exon 2 (Buchanan et al., 2002). For *LEP* 305(T/C), the cysteine allele has been associated with greater leptin mRNA levels, increased carcass fat content, and less marbling in beef cattle (Buchanan et al., 2002; Di Stasio et al., 2007). For *LEP* 1620(A/G), there have not been any association studies with carcass traits performed thus far.

Selection for growth traits can affect allele frequencies of a variety of genes involved in the expression of these traits. A selection experiment, with a control population of Nelore cattle, has been conducted in Brazil for about 25 yr. This population offers an opportunity to study the consequences of selection for yearling weight on the allele and genotypic frequencies. Moreover, although there are many studies associating *LEP* and *DGAT1* genetic polymorphisms with production traits in *Bos taurus* (Buchanan et al., 2002; Liefers et al.,

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2003; Thaller et al., 2003; Nkrumah et al., 2004), this association is not well-established in the Nelore breed because only 1 other study has addressed this subject (Silva, 2008).

The aim of the present study was to analyze the pattern of the *DGAT1 K232A*, *LEP 1620(A/G)*, and *LEP 305(T/C)* polymorphisms in 3 lines of Nelore cattle selected for growth and to evaluate their effects on growth and carcass traits.

## MATERIALS AND METHODS

The experiments were carried out in accordance with humane animal care and handling procedures according to the guidelines of the state of São Paulo, Brazil.

### Animals

A total of 357 animals originating from 3 Nelore (*Bos indicus*) selection lines reared at the research unit of Instituto de Zootecnia, Estação Experimental de Zootecnia de Sertãozinho (21°10' south latitude and 48°5' west longitude, São Paulo, Brazil) since 1981 were used in this study. This region is characterized by a wet tropical climate, with average annual temperature and rainfall of 24°C and 1,312 mm, respectively. Pastures mainly consist of *Panicum maximum* and *Brachiaria brizantha*, which are common tropical grasses in Brazil.

The founder sires were from private breeders, as well as from the existing experimental herd. In 1980 separate lines were established and from the 350 heifers available for reproduction, the youngest 180 were randomly assigned to the control Nelore (**NeC**; n = 60 animals) and selected Nelore (**NeS**; n = 120 animals) lines. The remaining 170 oldest females composed the traditional Nelore (**NeT**) line.

Bulls were selected based on a feedlot performance test at 378 d of age and heifers on pasture, at 550 d of age. For the control line (NeC), parents were selected to maintain a selection differential of approximately zero, and for the selection lines (NeS and NeT) animals were selected to maximize the selection differential for heavier yearling weights. The NeS and NeC are closed lines. The NeT line is characterized by a more flexible cow and bull replacement scheme. The NeT line initially included bulls originating from outside the station herds and eventually those culled from the NeS line (Mercadante et al., 2003).

Direct genetic trend for yearling weight for the selected lines was 0.72% of the phenotypic average/year. Correlated annual responses, represented as percentage of the phenotypic means, were 0.67% for birth weight, 0.38% (direct effect) and 0.14% (maternal effect) for weaning weight, and 0.22% for rump height. Nowadays, average yearling weight breeding values for heifers are 1.9, 42.2, and 44.6 kg for NeC, NeS, and NeT, respectively (Razook and Mercadante, 2007).

All females born in 2003, 2004, and 2005 (54 from NeC, 124 from NeS, and 179 from NeT) were included

in this study. Heifers were raised on pasture, without supplementation, except NeS and NeC females born from 2004 to 2005 that participated in an individual feed intake experiment and were supplemented from weaning to 365 d of age. This effect was taken into account in the model of analysis by the contemporary group definition.

### Traits Evaluated

Birth weight, weaning weight adjusted to 210 d of age (**W<sub>210</sub>**), yearling weight adjusted to 550 d of age (**W<sub>550</sub>**), and yearling rump height (**RH**) were obtained as part of the breeding program. The carcass traits were measured at 22 ± 2.5 mo of age.

Longissimus muscle area (**LMA**) and backfat thickness (**BF**) were obtained from a cross-sectional image on the LM measured between the 12th and 13th ribs. The rump fat thickness (**RF**) was measured at the intersection between the gluteus medius 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 LM using the cross-sectional ribeye image. Real-time ultrasound images were collected using 2 types of devices depending on the occasion: Aloka 500V (Corometrics Medical Systems Inc., Wallingford, CT) equipped with a linear probe of 17.2 cm and a 3.5-MHz transducer (Aloka Co. Ltd., Tokyo, Japan), and PIE Medical 401347-Aquila (Esaote Europe B.V., Maastricht, the Netherlands) equipped with a linear probe of 18 cm and a 3.5-MHz transducer. The images were stored and subsequently interpreted using the Echo Image Viewer 1.0 program (Esatoe Europe B.V.), with 1 decimal place.

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 LMA and BF images, a standoff pad was used to guarantee acoustic contact between the linear probe and the natural body shape of the animal. Transducer placement was first determined by palpating the left side of the animal between the 12th and 13th ribs. The ultrasound probe was moved toward the midline between and parallel to the 12th and 13th rib bones and then laterally until the LM came into full view on the screen. The RF images were obtained by placing the transducer at the insertion of the gluteus medius and biceps femoris muscles without the use of the acoustic coupler.

### Genotyping and Sequencing

Blood (5 mL) was collected from a jugular vein of each animal into vacuum tubes containing 7.5 mg of EDTA. Genomic DNA was extracted using the standard techniques described by Zadworny and Kuhlmeier (1990).

The primers used for amplification of the *DGAT1 K232A* fragment (CAC CAT CCT CTT CCT CAA G

and AAG GAA GCA AGC GGA CAG) were based on the sequence described by Winter et al. (2002) and flank the region containing the polymorphism that originates the K232A mutation. The primer sequences described by Lien et al. (1997) were used for amplification of the *LEP 1620(A/G)* fragment (GTC TGG AGG CAA AGG GCA GAG T and CCA CCA CCT CTG TGG AGT AG). Primers based on the sequences described by Buchanan et al. (2002) were used for amplification of the *LEP 305(T/C)* fragment (TGT AAA ACG ACG GCC AGT CCA TGG CAG ACA GCA AAT CTC GT and CAG GAA ACA GCT ATG ACC TGG TGT CAT CCT GGA CCT TCC).

Approximately 100 ng of genomic DNA was mixed with 15 pmol of each primer in a total volume of 25  $\mu$ L containing 1 $\times$  PCR buffer, 0.75 mM MgCl<sub>2</sub>, 100  $\mu$ M of each dNTP, and 1 unit *Taq* DNA polymerase (Fermentas International Inc., Burlington, Ontario, Canada). Polymerase chain reaction was performed in a Whatman Biometra Thermocycler (Biometra Biomedizinische Analytik GmbH, Goettingen, Germany) under the following conditions: denaturation at 95°C for 5 min, followed by 35 cycles of denaturation at 95°C for 45 s, annealing at 57, 66, and 54°C for 40 s [*DGAT1 K232A*, *LEP 1620(A/G)*, and *LEP 305(T/C)*, respectively], extension at 72°C for 45 s, and a final extension step at 72°C for 5 min.

For RFLP analysis, 5  $\mu$ L of the PCR product was mixed with 1 $\times$  enzyme buffer and 2 units of the enzyme in a final volume of 15  $\mu$ L. The following enzymes (all from New England Biolabs Inc., Ipswich, MA) were used for detection of the RFLP: *Cfr*I for *DGAT1 K232A*, and *Bsa*AI and *Kpn*2I for *LEP 1620(A/G)* and *LEP 305(T/C)*, respectively. The solutions were incubated according to manufacturer instructions for each enzyme, and the fragments were separated by electrophoresis on 2% agarose gels stained with ethidium bromide. The fragments were photographed under UV light and their sizes were estimated using a 100-bp DNA ladder (Fermentas International Inc.).

For characterization of the regions studied in Nelore cattle, the PCR fragments were submitted to DNA sequencing. The PCR products were sequenced in a MegaBACE 1000 Analysis System (Amersham Biosciences, Pittsburgh, PA) using the DYEnamic ET Dye Terminator Cycle Sequencing kit and Thermo Sequenase II DNA polymerase. The sequences were analyzed using the Cimarron 3.12 base caller implemented in the Sequence Analyzer (Amersham Biosciences). The purification and sequencing reactions were performed by the Human Genome Research Center (CEGH, São Paulo, Brazil). Chromas software (<http://www.techne-lysium.com.au>) was used to determine the homology with other sequences deposited in GenBank.

### Statistical Analysis

Allele and genotype frequencies and heterozygosity were estimated as described by Weir (1996). The Fisher

exact test (*F*-test) was applied to compare allele frequency among the 3 populations using the population differentiation module of the GENEPOP program, version 3.4 (<http://genepop.curtin.edu.au>), which applies Markov chain algorithms. Probability values less than 0.05 were considered to be significant.

Statistical analysis and evaluation of additive and dominance effects were performed using the MIXED procedure (SAS Inst. Inc., Cary, NC). The general mixed model can be described as

$$y = X\beta + Zu + e,$$

where  $y$  is a vector of phenotypic observations;  $X$  and  $Z$  are incidence matrices associating the fixed and random effects, respectively, to  $y$ ;  $\beta$  is a vector of fixed effects;  $u$  is a vector of sire effects; and  $e$  is a vector of residuals. It was assumed that  $E[y] = X\beta$ ;  $E[u] = E[e] = 0$ ;  $V[u] = I_{N_u}\sigma_u^2$  e  $V[e] = I_N\sigma_e^2$ , where  $E$  = expected value,  $V$  = variance,  $\sigma_u^2$  = sire variance,  $\sigma_e^2$  = residual variance,  $I$  = identity matrix, and  $N_u$  and  $N$  are the numbers of sires and animals, respectively.

At first the model included as fixed effects the genotype marker [*DGAT1 K232A*, *LEP 1620(A/G)*, or *LEP 305(T/C)*]; contemporary group (1, ..., 9), defined as line and year of birth, and month at birth (September, October, November); and the linear effects of dam age and age at measurement (for RH, LMA, BF, and RF) as covariates; and sire as a random effect (1, ..., 41). Later, instead of the genotypic marker effects, the additive effects and dominance deviations were included in the model as in Esmailzadeh et al. (2008), maintaining all of the other effects as described above. The additive covariates were 0, 1, and 2 to account for the number of alleles for *DGAT1 K232A* (KK, KA, and AA), *LEP 1620(A/G)* (AA, AG, and GG), and *LEP 305(T/C)* (CC, CT, and TT). Another covariate was added to test for dominance, where 0 = homozygous and 1 = heterozygous. Significant results for this covariate were interpreted as evidence of a dominance effect. Probability values were nominal and do not correct for multiple testing.

## RESULTS AND DISCUSSION

The sample means and SD for each trait, according to the line of selection, are shown in Table 1. In general, selected lines presented greater means ( $P < 0.01$ ) for all traits but BF and RF ( $P < 0.05$ ).

The *DGAT1 K232A*, *LEP 1620(A/G)*, and *LEP 305(T/C)* amplified fragments had sizes of 491, 522, and 280 bp, respectively. Two digestion patterns were obtained for each marker, resulting in 3 genotypes. For *DGAT1 K232A*, the intact 491-bp fragment was observed, which corresponds to allele K, in addition to fragments of 317 and 174 bp that correspond to allele A. For *LEP 1620(A/G)* the intact 522-bp fragment (allele A) was detected, in addition to fragments of 441 and 81

**Table 1.** Means and SD (in parentheses) of the traits analyzed in the 3 selection lines

Trait <sup>1</sup>	NeC <sup>2</sup>	NeS <sup>2</sup>	NeT <sup>2</sup>
Birth weight, kg	25.5 (0.7)	31.1(0.5)	30.7 (0.41)
W <sub>210</sub> , kg	151.8 (2.9)	190.0 (1.9)	186.4 (1.6)
W <sub>550</sub> , kg	259.6 (7.3)	325.3 (5.2)	295.3 (4.5)
RH, cm	127.4 (0.8)	135.9 (0.6)	131.7 (0.5)
LMA, cm <sup>2</sup>	45.1 (1.0)	48.5 (0.7)	45.6 (0.6)
BF, mm	2.1 (0.2)	1.7 (0.1)	1.6 (0.1)
RF, mm	3.7 (0.3)	3.6 (0.2)	3.5 (0.2)

<sup>1</sup>W<sub>210</sub>; W<sub>550</sub>: BW at 210 and 550 d of age; RH: rump height; LMA: LM area; BF: backfat thickness; RF: rump fat thickness.

<sup>2</sup>NeC: control line; NeS: selected line; NeT: traditional line.

bp (allele G). For *LEP 305(T/C)*, the 280-bp fragment (allele C) and a fragment of 250 bp (allele T) were obtained. Sequencing of the PCR products obtained for 1 animal of each homozygous genotype revealed that the 3 RFLP variants are caused by the same mutations as described for *Bos taurus* (GenBank accession numbers: EU348566, EU348567, FJ626852, FJ626853, FJ626854, FJ626855, FJ626856).

In the present study, the frequency of the K allele of the *DGAT1 K232A* mutation was greater than that of the A allele. Separate analysis of the 3 selection lines showed that the A allele was more frequent in the NeC line than in the NeS and NeT lines. The homozygous AA genotype was only found in the NeC line (Table 2). The *F*-test showed a significant difference in the frequency of the A allele between the NeC line and the lines selected for growth (NeS and NeT; Table 3). This difference may be due to the selection scheme effect in the NeC line. These results suggest that the K232A mutation occurred after the separation of the *Bos taurus* and *Bos indicus* breeds and that the presence of

variant A in *Bos indicus* breeds is probably due to the introgression of *Bos taurus* genes into *Bos indicus* cattle during the selection process of *Bos indicus* in Brazil. This is the first report demonstrating the presence of the A allele of the K232A mutation of the *DGAT1* gene in Nelore cattle (*Bos indicus*). The *DGAT1 K232A* mutation has frequently been described in *Bos taurus* breeds. Lacorte et al. (2006), studying this mutation in *Bos indicus* breeds, reported low frequencies of the A allele in Gyr and Red Sindhi animals and its total absence in Nelore cattle.

Choudhary et al. (2005) were the first to describe the *LEP 1620(A/G)* polymorphism in *Bos indicus* cattle, with the frequency of the G allele ranging from 0.66 in Hariana cattle to 0.75 in Gyr cattle. Almeida et al. (2003), studying Nelore × Aberdeen Angus crossbreds, reported a frequency of the G allele of 0.58. In *Bos taurus* breeds, the frequency of the G allele ranges from 0.82 to 0.76, indicating that, in contrast to *DGAT1*, this mutation might have occurred far back in evolution before the separation into taurine (*Bos taurus*) and

**Table 2.** Genotype and allele frequencies obtained for the *DGAT1 K232A*, *LEP 1620(A/G)*, and *LEP 305(T/C)* markers in 3 lines of Nelore cattle selected for growth

Marker/herd <sup>1</sup>	Genotype frequency			Allele frequency	
	<i>KK</i>	<i>KA</i>	<i>AA</i>	<i>K</i>	<i>A</i>
<i>DGAT1 K232A</i>					
Total	0.94	0.05	0.01	0.97	0.03
NeC	0.74	0.22	0.04	0.85	0.15
NeS	0.95	0.05	0	0.97	0.03
NeT	0.98	0.02	0	0.99	0.01
<i>LEP 1620(A/G)</i>					
Total	0.26	0.49	0.25	0.50	0.50
NeC	0.26	0.41	0.33	0.46	0.54
NeS	0.23	0.52	0.25	0.49	0.51
NeT	0.28	0.49	0.23	0.53	0.47
<i>LEP 305(T/C)</i>					
Total	0.76	0.22	0.02	0.88	0.12
NeC	0.74	0.24	0.02	0.86	0.14
NeS	0.76	0.22	0.02	0.88	0.12
NeT	0.77	0.21	0.02	0.88	0.12

<sup>1</sup>NeC: control line; NeS: selected line; NeT: traditional line; *DGAT1*: acyl CoA:diacylglycerol acyltransferase 1; *LEP* = leptin.

**Table 3.** *P*-values obtained by Fisher exact test for comparison of allelic frequencies among the 3 selection lines

Pair of populations <sup>1</sup>	<i>P</i> -value <sup>2</sup>		
	<i>DGAT1 K232A</i>	<i>LEP 1620(A/G)</i>	<i>LEP 305(T/C)</i>
NeC and NeS	<0.0001	0.73	0.31
NeC and NeT	<0.0001	0.26	0.25
NeT and NeS	0.07	0.37	0.90

<sup>1</sup>NeC: control line; NeS: selected line; NeT: traditional line.

<sup>2</sup>*DGAT1*: acyl CoA:diacylglycerol acyltransferase 1; *LEP* = leptin.

indicine (*Bos indicus*) cattle. In the present study, considering the 3 lines altogether, the frequencies of the A and G alleles were practically the same. Heterozygosity was high, with the heterozygous AG genotype being the most frequent genotype in all 3 selection lines. The difference in the frequency of the GG and AA genotypes between these lines was small (Table 2). Interestingly, Choudhary et al. (2005) reported low frequencies of the AA genotype in Holstein Friesian, Jersey, Hariana, Sahiwal, Gyr, and Nimari cattle. In contrast to the *DGAT1* marker, the *F*-test revealed no significant difference in the allele frequencies among the 3 selection lines (Table 3).

For *LEP 305(T/C)*, Choudhary et al. (2005) reported fixation of the C allele in *Bos indicus* of the Hariana, Sahiwal, Gyr, and Nimari breeds, and low frequencies of the T allele in crossbred populations. In *Bos taurus* breeds, the frequency of the T allele is 0.40 in Holstein Friesian, 0.56 in Jersey, 0.58 in Angus, 0.34 in Charolais, 0.55 in Hereford, and 0.32 in Simmental cattle (Buchanan et al., 2002; Choudhary et al., 2005). In Australian cattle, Barendse et al. (2005) found frequencies of the T allele of 0.48 for Angus, 0.18 for Shorthorn, 0.44 for Belmont Red, 0.78 for Hereford, 0.26 for Murray Grey, and 0.27 for Santa Gertrudis. In Brazilian Nelore cattle, frequencies of 0.93 for the C allele and of 0.06 for the T allele have been reported (Silva, 2008), which are close to those found in the present study (0.88 for allele C and 0.12 for allele T). In all 3 lines, the frequency of the C allele was greater than 0.85 and the frequency of the T allele was less than 0.15 (Table 2). These results indicate that, as is the case of the *DGAT1* polymorphism, the presence of the T allele might be due to introgression of *Bos taurus* genes into *Bos indicus* breeds during the selection process of these subspecies in Brazil. The *F*-test showed that the differences in frequencies among selection lines were not significant ( $P = 0.31$  and  $0.25$ ; Table 3).

Considering that the sires and dams from the founder herd were chosen to represent the main Nelore families at the time and that these animals have been submitted to the most common selection procedure in Brazil, we can assume that the selected lines are representative of the Nelore breed. Therefore, one could expect that allele frequencies we have observed for the 3 markers are indicative of these frequencies in the Nelore breed

in Brazil. However, the same may be not true for the control line because it is under a kind of stabilizing selection.

The *DGAT1 K232A* polymorphism has frequently been associated with milk composition traits in dairy cattle. Several studies have reported an association of the K allele with greater milk fat content and less protein yield (Spelman et al., 2002; Winter et al., 2002; Grisart et al., 2004; Naslund et al., 2008), probably due to greater triglyceride production. However, few studies have investigated the association of this polymorphism with growth and carcass traits (Thaller et al., 2003; Casas et al., 2005). Thaller et al. (2003) observed a significant effect of the K allele on semitendinosus intramuscular fat deposition in Holstein and Charolais (*Bos taurus*) cattle. Casas et al. (2005) found no association between the *DGAT1 K232A* polymorphism and carcass composition traits in the Brahman breed (*Bos indicus*). In the present study, there was a significant effect of this polymorphism on RH ( $P < 0.05$ ). However, no significant additive or dominance effect of the K variant was observed for this trait. Carcass traits evaluated by ultrasound showed no influence of the *DGAT1 K232A* polymorphism (Table 4). The low frequency of these genotypes is likely interfering with the accurate assessment of their association.

Regarding the effects of the leptin polymorphism, several investigations have shown an association with important economic traits, but this is the first association study between the *LEP 1620(A/G)* polymorphism and growth and carcass traits in cattle. Significant effects ( $P = 0.03$ ) of this polymorphism were observed for  $W_{210}$ , with the demonstration of greater mean values for AA animals compared with AG and GG animals. Birth weight and  $W_{550}$  presented *P*-values close to significance (0.08 and 0.06, respectively; Table 4). There was an additive effect of the A variant, with the presence of this allele being related to increases of  $0.610 \pm 0.270$  kg in BW,  $4.080 \pm 1.570$  kg in  $W_{210}$ , and  $0.54 \pm 0.27$  cm in RH. In addition, a dominance effect of the A allele on LMA ( $1.12 \pm 0.56$  cm<sup>2</sup>) was observed.

The *LEP 305(T/C)* mutation results in an AA substitution of cysteine to arginine in the  $\alpha$ -helix of the leptin polypeptide. In *Bos taurus* breeds, associations between the T allele and greater levels of leptin expression, fatter carcasses, and greater food intake have

**Table 4.** Nominal *P*-values, least squares means, and SE (in parentheses) for the effects of *DGAT1 K232A*, *LEP 1620(A/G)*, and *LEP 305(T/C)* on growth and carcass traits

Marker <sup>1</sup>	Trait <sup>2</sup>						
	Birth weight	W <sub>210</sub>	W <sub>550</sub>	RH	LMA	BF	RF
<i>DGAT1 K232A</i>							
<i>P</i> -value	0.22	0.92	0.56	0.04	0.54	0.32	0.93
KK (335)	29.21 (0.33)	176.17 (1.43)	293.84 (1.97)	131.85 (0.32)	46.29 (0.44)	1.88 (0.09)	3.68 (0.13)
AK (20)	28.37 (0.93)	178.17 (5.12)	290.25 (6.31)	129.99 (0.89)	44.96 (1.29)	1.59 (0.28)	3.82 (0.38)
AA (2)	25.05 (2.78)	173.73 (15.98)	276.68 (18.72)	127.64 (2.64)	44.50 (3.90)	2.84 (0.85)	3.83 (1.14)
<i>LEP 1620(A/G)</i>							
<i>P</i> -value	0.08	0.03	0.06	0.12	0.13	0.24	0.48
AA (93)	29.67 (0.46)	180.78 (2.29)	298.22 (2.88)	132.23 (0.44)	46.85 (0.63)	1.91 (0.14)	3.78 (0.19)
AG (174)	29.10 (0.39)	175.89 (1.79)	290.77 (2.37)	131.55 (0.37)	45.59 (0.51)	1.78 (0.11)	3.59 (0.15)
GG (90)	28.44 (0.47)	172.62 (2.29)	292.36 (2.91)	131.16 (0.44)	46.55 (0.64)	2.03 (0.14)	3.79 (0.19)
<i>LEP 305(T/C)</i>							
<i>P</i> -value	0.03	0.85	0.06	0.10	0.90	0.03	0.65
CC (274)	29.38 (0.34)	176.40 (1.48)	294.90 (1.99)	131.85 (0.33)	46.25 (0.46)	1.95 (0.10)	3.72 (0.14)
CT (79)	28.21 (0.48)	175.58 (2.48)	287.51 (3.05)	130.89 (0.46)	45.94 (0.67)	1.60 (0.15)	3.57 (0.20)
TT (4)	27.28 (1.85)	181.30 (10.62)	297.36 (12.40)	131.91 (1.78)	45.79 (0.64)	2.54 (0.57)	4.07 (0.76)

<sup>1</sup>*DGAT1*: acyl CoA:diacylglycerol acyltransferase 1; *LEP* = leptin.

<sup>2</sup>W<sub>210</sub>, W<sub>550</sub>: BW at 210 and 550 d of age; RH: rump height; LMA: LM area; BF: backfat thickness; and RF: rump fat thickness.

been reported (Buchanan et al., 2002; Liefers et al., 2003; Nkrumah et al., 2004). However, Barendse et al. (2005) reported no association between this SNP and fatness traits in 3,129 animals from 2 independent Australian populations. In Nelore cattle, previous studies have shown that this polymorphism affected LMA, BF, and BW gain from weaning to yearling ages. The T allele was favorable for BF and yearling weight, whereas the C allele was favorable for LMA (Silva, 2008). In the present study, the *LEP 305(T/C)* polymorphism exerted significant effects on BW and BF. Homozygous CC animals presented a greater mean BW, followed by CT and TT animals. For BF, heterozygous animals presented decreased mean values compared with the 2 homozygous genotypes (Table 4). However, no significant additive or dominance effects were found, probably because of the small number of animals with the TT genotype.

Analysis showed that the *LEP 1620(A/G)* marker was associated only with W210 and selecting for increasing W210 will also increase BW means (*P*-value = 0.08). Birth weight means in the Nelore population are around 30 kg, and the frequency of calving difficulty is extremely low. Medium-term selection for BW at any age will increase birth weight, and monitoring of increases in BW is advised in any breeding program favoring growth traits (Albuquerque et al., 2006). The *LEP 305(T/C)* marker had an effect on BW and BF, and increasing BW will decrease BF and vice versa. In Brazil, Nelore animals are raised on pasture and time in the feedlot is restricted to a short period (60 to 120 d) only for finishing. In this kind of production system, animals able to deposit fat on low energy diets could be desirable. However, more studies with larger data sets are necessary before these markers could be used for marker-assisted selection. There was no evidence that

selection for yearling weight, for about 25 yr, altered the allele frequencies of the 3 markers.

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