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Communication

[*Comunicação*]

SNPs rs471462296, rs456245081 and rs438495570 in the 5'UTR region of *DGAT1* gene in Nelore

[*SNPs rs471462296, rs456245081 e rs438495570 na região 5'UTR do gene* DGAT1 *em Nelore*]

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In *Bos taurus*, the diacylglycerol O-acyltransferase (*DGAT1*) gene is about 8.6Kb in length, comprise of 17 exons, spanning over 121.8bp, and it is located on BTA14 (Kaupe *et al*., 2004. *DGAT1* has been associated with phenotypes of economic interest in cattle. The gene has been traditionally known as a positional and functional marker for both dairy production and increased fat content in milk (Tabaran *et al*., 2015). *DGAT1* encodes for an enzyme, containing 489 amino acids, responsible for catalyzing the final step of triglycerides synthesis covalently linking diacylglycerol to fatty acyl-CoA substrates. This gene is expressed in many tissues, with the highest levels in the intestine, testis, and mammary glands, especially in adipose and epithelial tissues (Bovenhuis *et al*., 2016).

Association studies using different bovine populations supported the role of *DGAT1* as a functional gene candidate for traits of deposition of fat in beef cattle breeds. DGAT1 action in fat depositions is mainly inferred due to its role in triglycerides synthesis (Zhang *et al*., 2015) and its EST (expressed sequence tags) of adipose tissue (Tantia *et al*., 2006).

The current is a cross-sectional study involving 106 Nelore bulls, recorded as cattle of pure origin (PO) through the national registry from the Brazilian Association of Zebu Breeders. Peripheral blood was collected from each animal and subsequently used to isolate genomic DNA, using a commercial kit (Illustra Blood Genomic Prep Mini Spin®, GE Healthcare, UK) according to the manufacturer's instructions. DNA purity and concentration were determined in a spectrophotometer (Nanodrop®, Thermo Fisher Scientifc, USA).

Isolated DNA was used to amplify genomic regions targeting single nucleotide polymorphism (rs471462296 [A/G], rs456245081 [A/G], and rs438495570 [C/G]) in the 5'UTR of *DGAT1* gene. SNPs were amplified by qPCR using TaqMan® Assays (Applied Biosystems, USA). Thermocycling was carried out in a StepOne® Real-Time PCR System (Thermo Fisher Scientific, EUA) following the manufacturer's instructions. Amplicons were analyzed using the software StepOne® Analyzer v.2.1 (Thermo Fisher Scientific, EUA). Allele calling followed the probe signals and instructions recommended by the manufacturer.

In the current study all genotypes were monomorphic. All animals were homozygous AA for rs47146229 and rs456245081 and heterozygous CG for rs438495570 (Table 1).
Our results suggested the 5'UTR of *DGAT1* gene had low sequence diversity and low variability in Nelore breed. Most likely founder effect and artificial breeding selection are the main causes for reduced variability of the aforementioned SNPs (Anton *et al*., 2008). Although much

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interest has been associated with the role of *DGAT1* in the deposition of intramuscular fat in Nelore, the studied SNPs presented little value as molecular markers for such trait as it wasn't possible to construct useful haplotypes to guide genetic merit of the breed (Mishra *et al*., 2007). On the other had, due to the monomorphic genotypes found, the evidence of any direct effect of *DGAT1* on fat deposition cannot be ruled out in Nelore. In summary, our results were important to consolidate the knowledge about single nucleotide variation in *DGAT1* gene for different bovine breeds. Thus, allowing for the adequate selection of SNP markers to be used in association studies and in the discrimination of animals of higher genetic value for phenotypes of economic interest.

Table 1. Allele and genotypes frequencies for the SNPs for the group tested

Group	*DGAT1*/SNP	Allele frequencies		Genotypes frequencies		
		+	-	+/+	+/-	-/-
G1–Nelore (n= 73)	rs471462296	1,00	0,00	1,00	0.00	0,00
	rs456245081	1,00	0,00	1,00	0.00	0.00
	rs438495570	0.50	0.50	0.00	1,00	0.00

n number of animals; + Commum alele; - Mutant alele; +/+ Homozygous for commum alele; +/- Heterozygous for mutante alele; -/- Homozygous for mutant alele.

Keywords: bovine, phenotype, genotype, selection

RESUMO

O objetivo desta pesquisa foi avaliar os SNPs rs471462296, rs456245081 e rs438495570 do gene DGAT1 *em bovinos Nelore. Foram analisados 109 bovinos. A extração do DNA genômico foi realizada do sangue dos animais, usando-se o* kit Ilustra Blood Genomic Prep Mini Spin® *(GE Healthcare, UK). A concentração e o grau de pureza do DNA foram determinados por meio de espectrofotômetro (Nanodrop - Thermo Fisher Scientifc, USA). A genotipagem dos SNPs ocorreu mediante o emprego do ensaio Taqman® (Applied Biosystems, USA). Na análise genômica, não foram encontradas alterações nas frequências alélicas e genotípicas (P≥0,05) para os SNPs testados. Dessa forma, a região 5'UTR analisada apresentou-se monomórfica e a variação de SNPs não foi observada, o que limita seu uso como marcadores moleculares para o gene* DGAT1 *em Nelore.*

Palavras-chave: bovino, fenótipo, genótipo, seleção

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