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GENETIC CHARACTERIZATION OF A BRANGUS-IBAGE CATTLE POPULATION – BIOCHEMICAL POLYMORPHISMS AND REPRODUCTIVE EFFICIENCY

CARACTERIZAÇÃO GENÉTICA DE UMA POPULAÇÃO DE BOVINOS BRANGUS-IBAGÉ - POLIMORFISMOS BIOQUÍMICOS E EFICIÊNCIA REPRODUTIVA

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SUMMARY

Biochemical techniques were used to investigate the genetic variability in a Brangus-Ibage population by determining allele frequencies of 18 blood protein systems: Hemogloin B-Chain (Hb), Albumin (Alb), Amylase (Am), Transferrin (Tf), Carbonic Anhydrase (CA), Ceruloplasmin (Cp), Malic Enzyme (ME), Diaphorase I and II (Dia I and Dia II), Slow Alpha 2 Macroglobulin (Ap), Acid Phosphatase (ACP), Esterase B and D (EstB and EstD), Phosphogluconate Dehydrogenase (PGD), Glucose-6-Phosphate Dehydrogenase (G-6-PD), Glucose-Phosphate-Isomerase (GPI), Superoxide Dismutase (SOD) and Glyoxalase I (GLO). The percentage of polymorphic loci were estimated at 0.27, the mean number of alleles was 1.33 and the mean heterozygosity was 0.07. There was a good agreement between expected and observed heterozygosity values. The population was in agreement with Hardy-Weinberg expectations in all systems. Reproductive records allowed to estimate three parameters of reproductive efficiency: mean age at first calving $(1152.15 \pm 166.60 \text{ days})$, mean calving interval $(539.23 \pm 124.10 \pm 166.60 \text{ days})$ days) and mean weight at first calving (391.02 ± 37.59kg). No relationship was found between reproductive efficiency and genetic systems.

Key words: Brangus-Ibage cattle, genetic characterization, reproductive efficiency.

RESUMO

Técnicas bioquímicas foram utilizadas para determinar a variabilidade genética numa população de bovinos da

raça Brangus-Ibagé com relação a 18 sistemas protéicos sangüíneos: Hemoglobina - Cadeia β (Hb), Albumina (Alb), Amilase (Am), Transferrina (Tf), Anidrase Carbônica (CA), Ceruloplasmina (Cp), Enzima Málica (ME), Diaforase I and II (Dia I and Dia II), Macroglobulina o2 lenta (Ap), Fosfatase Ácida (ACP), Esterase B and D (EstB and EstD), Fosfogliconato Desidrogenase (PGD), Glicose-6-Fosfato Desidrogenase (G-6-PD), Glicose-Fosfato-Isomerase (GPI), Superóxido Dismutase (SOD) e Glioxalase I (GLO). O percentual de locos polimórficos foi estimado em 0,27, o número médio de alelos foi 1,33 e a heterozigosidade média foi de 0,07. Houve boa concordância entre a heterozigosidade média observada e a esperada. A população apresentou-se em equilíbrio de Hardy-Weinberg em todos os sistemas. Também foram determinados três parâmetros de eficiência reprodutiva: idade média ao primeiro parto (1152,15 ± 166,60 dias), intervalo médio entre partos (539,23 ± 124,10 dias) e peso médio da vaca ao primeiro parto (391,02 ± 37,59kg). Não se encontrou nenhuma associação entre os polimorfismos protéicos e os parâmetros de eficiência reprodutiva.

Palavras-chave: bovinos Brangus-Ibagé, caracterização genética, eficiência reprodutiva.

INTRODUCTION

The Brangus-Ibage is a composite beef cattle breed resulting from the crossing of Aberdeen Angus (ABG) and Nellore (NEL). This breed was

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developed as a result of a search for a beef animal, which would retain the Nellore natural ability to thrive under adverse conditions in combination with the excellent Angus meat quality. Its composition was theoretically stabilized at 3/8 NEL and 5/8 ABG (CHAGAS *et al.*, 1972). In Brazil, much of the early work in crossing Nellore and Aberdeen Angus cattle was done by the Brazilian Agricultural Research Corporation (EMBRAPA – CPPSUL). The studies involving such crosses back in 1945. At that time, similar crossings had been tested around the world, but in most of other countries, the *Bos indicus* breed was the Brahman instead of the Nellore.

Due to its hardiness under stress conditions, in the last decade the Brangus has become popular mainly in areas of high heat and humidity in Australia and America. Currently the Brangus-Ibage breed is undergoing a great expansion in Brazil, with more than 250 cattle breeders in the southern, southeastern and central western regions and more than 55,000 animals registered in a little more than two years of official registration (EMBRAPA, 1995).

Nowadays, the EMBRAPA, in association with Federal University of Pelotas (UFPEL), Federal University of Santa Maria (UFSM) and Federal University of Rio Grande do Sul (UFRGS) has initiated a program studing alternatives to improve reproductive efficiency in Brangus-Ibage. As part of this program, efforts are directed toward investigating the degree of genetic variation of the breed with the aim of characterize it and develop selection procedures to accomplish optimum use of both additive and nonadditive genetic variation. The genetic variability has been investigated in three levels: nuclear DNA, mitochondrial DNA and blood proteins. In this paper, it were investigated eighteen blood genetic systems and verified the possibility of associations among these genetic markers and reproductive performance.

MATERIAL AND METHODS

Blood samples were obtained from seventy five Brangus-Ibage cows by punction of jugular vein using ACD as anticoagulant and processed according to HENKES *et al.* (1993). The genetic systems Hemoglobin β-Chain (Hb), Albumin (Alb), Amylase (Am), Transferrin (Tf), Carbonic Anhydrase (CA), Ceruloplasmin (Cp), Malic Enzyme (ME), Diaphorase I and II (Dia I and Dia II), Slow Alpha 2 Macroglobulin (Ap), Acid

Phosphatase (ACP), Esterase B and D (EstB and EstD), Phosphogluconate Dehydrogenase (PGD), Glucose-6-Phosphate Dehydrogenase (G-6-PD), Glucose-Phosphate-Isomerase (GPI), Superoxide Dismutase (SOD) and Glyoxalase I (GLO) were carried out by horizontal starch gels electrophoresis according to HENKES *et al.* (1994). The Amylase was tested as described by GEBICKE-HÄRTER & GELDERMANN (1977).

The genetic variability was estimated through the following parameters: gene frequencies for each marker, average number of alleles at each locus, expected heterozygosity for each locus and percentage of polymorphic loci (NEI, 1987). Hardy-Weinberg equilibrium was analyzed by exact tests using GENEPOP software (RAYMOND ROUSSET, 1995). Three estimates related to reproductive performance were obtained from EMBRAPA records: age at first calving (AFC expressed in days), mean weight at first calving (MWFC - expressed in kg) and mean calving interval (CI - expressed in days). The first two estimates are related to growth rate and the other is an evaluation of reproductive performance. For this last measure only females with at least four calving records were considered. The relationship between reproduction data and genetic markers was analyzed by grouping the cows according to their protein phenotypes and calculating the average reproductive parameter for each one. The comparisons were performed through a non parametric statistic (since these variables do not have a normal distribution) of Kruskal-Wallis or Mann-Whitney U test using the SPSS[®] for Windows™ Package.

RESULTS AND DISCUSSION

Thirteen proteins (Dia II, EstB, EstD, GPI, SOD, ME, ACP, CA, Ap, Cp, GLO, G-6-PD and PGD) showed no variation and all individuals presented the same phenotype. The phenotype and gene frequencies for the five polymorphic loci (Hb, Alb, Dia I, Tf and Am) are presented in table 1. There was a good agreement between expected and observed heterozygosity values and no departures from Hardy-Weinberg equilibrium were verified for any systems. The percentage of polymorphic loci was estimated as 0.27, the mean number of alleles was 1.33, and the mean heterozygosity was 0.07.

The degree of polymorphism and the other variation parameters showed values lower than those obtained by other authors for different cattle

Table 1 - Phenotypic and Gene Frequencies of Five Protein Systems in a Brangus-Ibage Herd (n=75).

System	Phenotype frequency		Gene Frequency	ho	he
Hb	Hb AA	0.00		0.33	0.28
	Hb AB	0.33	$Hb^{B} = 0.83$		
	Hb BB	0.67			
Alb	Alb FF	0.62	$Alb^F = 0.66$	0.50	0.46
	Alb FS	0.29			
	Alb SS	0.09			
			· F		
DIA	DIA FF	0.09		0.30	0.37
	DIA FS	0.30	DIA $^{S} = 0.24$		
	DIA SS	0.61			
Tf	Tf AA	0.29	$Tf^{A} = 0.57$	0.58	0.59
	Tf AD	0.31			
	Tf AE	0.25	,		
	Tf DD	0.08	•		
	Tf DE	0.03			
	Tf EE	0.04			
Am	Am BB		$Am^{B}_{C} = 0.68$	0.53	0.50
	Am BC	0.46	$Am^{C} = 0.32$		
	Am CC	0.08			

ho - Observed heterozygosity, he - Expected heterozigosyty, $\boldsymbol{n}=$ sample size.

breeds (TEJEDOR et al., 1986). These differences are probably due to the markers analysed. Comparing only the same loci investigated in both series, the present data are similar to those obtained by these authors. In order to investigate the degree of variability, it was decided to test a random sample of loci, independently of their variability to prevent biased upward estimates of variability. Therefore, the data are consistent with results obtained for other mammalian species in which a large random set of markers were investigated (LEWONTIN, 1974; HENKES et al., 1993, 1994).

In relation to the monomorphic systems found here, studies have also failed to find variation in bovine (BAKER & MANWELL, 1980) and even in other ruminants (HENKES et al., 1993, 1994). The exception was CA that shows no variation in the present sample and has been related as polymorphic in many studies (TEJEDOR et al., 1986; PANEPUCCI, 1988), the slowest allele (CA S) ranging from 0.6 to 1.0 in taurine and Zebu cattle, this last one showing a third CA type that migrate slower than CA S and called CA Z (PENEDO et al., 1982). This result cohere with the frequencies observed in Aberdeen Angus, where CA S reaches values near 1.0 (SARTORE et al., 1969).

For Hb, two alleles were found, the rarest one, Hb A had a frequency of 0.17. Hb A had a higher prevalence in Bos taurus cattle (TEJEDOR et al., 1986; BRAEND, 1972) and also in some Indian breeds (PRASAD et al., 1983). The value obtained in the present study for Hb B is the highest so far found in bovine and cannot be explained by a specific contribution of one of the founder breeds. Notwithstanding, this herd was never been selected in function of any Hb phenotype it cannot be exclude the possibility of selection effect if some Hb phenotype has a best fitness in that environment. In sheep, there are strong evidences of physiological differences among hemoglobin types (DAWSON & EVANS, 1966, 1967; AGAR et al., 1972). The investigation of hemoglobin frequencies in other breeds reared in the same environment is important to elucidate this point. Another possibility is indirect selection if Hb is in linkage disequilibrium with some gene involved in any other trait that has been selected. The gene for globin β-chain was mapped at chromosome 15 in cattle and it is closely linked to the beta subunit of the follicle-stimulating hormone (FRIES, 1989) and the parathyroid hormone loci al.. 1988: FOREMAN (FRIES et WOMACK,1989) as well as in synteny with myogenic factor 3 (MYOD1), a muscle-specific gene (RYAN et al., 1997). These two last genes play an important role in muscular development and therefore the selection based on morphologic aspect applied to this herd could indirectly affect Hb genotype distribution.

The Alb, Tf, Am and Dia frequencies are within the range of variation observed in Zebu crossings (BORTOLOZZI, 1983; PANEPUCCI, 1988, 1989) and in European cattle (GAHNE et al., 1977; BAKER & MANWELL, 1980; TEJEDOR et al., 1986). It was found no specific allele for this breed and the gene frequencies agreed with the historical composition of the herd with a higher percentage of Aberdeen Angus component. It is also possible that these animals' environment or the selection management applied to them favor Angus genotype. The mean age at first calving, calving intervals and weight at first calving in the total sample and within each protein phenotype are presented in table 2. No differences in reproduction performance among the different genotypes were observed. Although it had been reported that transferrin polymorphisms affect fertility in dairy and beef cattle (ASHTON & FALLON, 1962), these 806 Henkes et al.

Table 2 – Productive traits averages in total sample and in relation to protein phenotypes in a Brangus-Ibage herd.

Phenotype	AFC (days) ^a	χ^2	CI (days) b	χ^2	MWFC	(kg) ^c	χ^2
Hb AB Hb BB	1126.80 ± 43.29 1172.13 ± 189.64	0,74 ^d	573.43 ± 140.39 532.19 ± 100.99	0,45 ^d	337.60 ± 4 351.90 ± 4		0,864
Alb FF Alb FS Alb SS	1129.33 ± 152.08 1164.04 ± 180.41 1193.83 ± 178.43	1,27	517.82 ± 123.43 538.73 ± 102.75 651.72 ± 150.22	5,61	353.72 ± 4 349.56 ± 4 347.83 ± 5	3.47	0,25
DIA FF DIA FS DIA SS	1204.50 ± 358.50 1194.00 ± 195.62 1154.41 ± 171.73	1,07	647.85 ± 42.21 518.70 ± 100.31 551.17 ± 138.95	3,28	347.44 ± 5 363.65 ± 4 361.46 ± 5	0.74	2,86
Tf AA Tf AD Tf AE Tf DD Tf DE* Tf EE*	1148.13 ± 160.95 1159.56 ± 162.88 1156.24 ± 184.67 1205.00 ± 212.16 1065 913	3,50	535.57 ± 160.62 524.35 ± 100.97 551.17 ± 138.95 511.58 ± 69.86 521 469	3,14	350.06 ± 4 368.31 ± 5 334.35 ± 4 362.75 ± 2	0.29 4.31	6,66
Am AA Am AB Am BB Total sample	1142.61 ± 155.56 1169.68 ± 185.56 1197.00 ± 53.03 1152.15 ± 166.60	0,04	529.88 ± 133.23 569.45 ± 113.29 435.67 ± 74.10 435.67 ± 74.10	5,47	393.87 ± 4 388.25 ± 3 391.02 ± 3	3.48	1,77

 a AFC = Mean age at first calving \pm standard deviation (days), b CI = Mean calving intervals \pm standard deviation (days), c MWFC = Mean Weight at first calving \pm standard deviation (kg) d = Z_{calc} . * Since only one animal presented Tf DE and Tf EE genotypes, no standard deviation was estimated.

data did not lent support to this hypothesis. Most of reproduction traits have low heritability estimates [AFC= 0.14±0.18, CI=0.10± 0.18 (KOOTS *et al.*, 1994)]. However, due to their high coefficient of variation it is possible to have good annual rates of response to selection, even with low heritabilities (BRADFORD, 1985).

Despite the great amount of studies reporting blood polymorphism in dairy cattle (ROCHA *et al.*, 1998) little is known regarding the genetic variation in beef cattle. Dairy cattle is commonly raised under intensive management and controlled environment, the reproductive records being taken more easily and with more confidence. Beef cattle, on the other hand, is generally raised free and under all environment challenges. Since reproduction is a multifatorial trait, any subtle genetic difference could be easily diluted by the environmental components.

Considering the lack of association verified amongst the genetic markers and reproductive parameters investigated at the present

study, it can be concluded that to search for a suitable marker as a tool for marker assisted selection for reproductive efficiency a polymorphic larger set of markers to cover all genome should be necessary (WELLER et al, 1997). Therefore, it is important to expand repertoire of known genetic marker loci in this breed in order determine appropriate implementation strategies and cost-benefit ratios for using genetic markers in animal breeding.

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