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BIOCHEMICAL POLYMORPHISMS AND GENETIC RELATIONSHIPS AMONG LANDRACE, LARGE WHITE AND DUROC PIGS FROM SOUTHERN BRAZIL

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ABSTRACT

Fourteen protein systems encoded by 15 structural loci were used to investigate genetic variability in three swine breeds (Landrace, Large White and Duroc), reared in Southern Brazil. The degree of genetic variability was similar in the three breeds (Landrace, $\overline{He}$ = 0.116; Large White, $\overline{He}$ = 0.119; Duroc, $\overline{He}$ = 0.095). These values are close to those computed for other populations of these breeds and higher than those obtained for wild pig populations. The gene frequencies at the polymorphic loci were employed to evaluate the usefulness of these systems for parent identification. The combined probabilities of paternity exclusion were estimated at 59% for Landrace, 54% for Large White and 50% for Duroc animals. Analysis of genetic relationships revealed that Landrace and Large White are the most similar breeds (D = 0.044), while the Duroc breed presents lower levels of genetic similarity to the other two breeds (Landrace/Duroc: D = 0.084; Large White/Duroc: D = 0.106). These findings are in agreement with the historical development of these breeds.

INTRODUCTION

Domestic pigs have been developed on all continents to provide animal protein for human nutrition. The swines of the most important breeds developed in Brazil were imported from several European and American countries.

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Biochemical polymorphisms can be used to characterize populations, to investigate the levels of genetic variability exhibited by breeds and to verify the relationships among them. Within this context, several studies have been performed on pig populations of various origins (Oishi and Tomita, 1976; Oishi *et al.*, 1980; Tanaka *et al.*, 1983; Van Zeveren *et al.*, 1990). Moreover, the gene frequencies of various polymorphic biochemical loci have been used for paternity control (Oishi and Abe, 1970). The establishment of this procedure is important for the prevention of erroneous paternity on excellent boars.

Considering the economic importance of the hog-raising industry in Brazil and the lack of studies about biochemical polymorphisms in the herds reared in this country, the objectives of the present investigation were: a) to compare gene frequencies among the samples; b) to estimate the degree of genetic variability among and within breeds; c) to evaluate the usefulness of protein polymorphisms for parentage control; d) to evaluate the genetic relationships among breeds.

MATERIAL AND METHODS

In order to study the genetic variability of Landrace, Large White and Duroc swines, blood samples were obtained from 282 animals at two localities in Rio Grande do Sul: the Boar Test Stations of Venâncio Aires and Estrela. At these Stations the pigs are tested to select the best animals for the Swine Artificial Insemination Center of Rio Grande do Sul. The pigs that were tested came from several farms and were not directly related. The criterions used to preselect the animals were to choice healthful animals and the biggest pigs from the litters.

Blood samples from 109 Landrace, 116 Large White and 57 Duroc animals were investigated for 15 protein loci: malate dehydrogenase (Mdh), isocitrate dehydrogenase (Idh), phosphogluconate dehydrogenase (Pgd), superoxide dismutase (Sod), esterase D (EsD), acid phosphatase (Acp), amylase 1 (Amy1), amylase 2 (Amy2), glyoxalase I (GloI), phosphohexose isomerase (Phi), albumin (Al), ceruloplasmin (Cp), hemoglobin (Hb), hemopexin (Hpx), and transferrin (Tf).

The laboratory work was carried out at the Departamento de Genética of the Instituto de Biociências of the Universidade Federal do Rio Grande do Sul.

The blood protein types were examined by starch and/or agarose gel electrophoresis. The techniques of electrophoresis and protein staining were as follows: Hpx, Amy1 and Amy2 were performed according to Kristjansson (1963); Tf and Cp were typed in accordance with Oishi and Tomita (1976); the procedures used for the study of Pgd and Phi were those employed by Gahne and Juneja (1985); the methods described by Spencer *et al.* (1964) and Harris and Hopkinson (1976) were used for EsD, Mdh, Idh,

Sod and Acp; GloI, Al and Hb were typed according to the methods described by Franco *et al.* (1986), Tucker (1968) and Naoum (1987), respectively. The allele frequencies were calculated by the gene counting method and Hardy-Weinberg equilibrium was verified by the classical X^2 test or by using the method described by Smith (1986) for small numbers. Genetic variability was estimated by the expected average heterozygosity, which is the mean of expected frequencies of heterozygotes at each locus calculated from Hardy-Weinberg equilibrium ($\overline{He}$; Nei, 1975). Genetic distances (the differences between populations as expressed by function of gene frequencies) were estimated according to Rogers (1972) and the dendrogram (the patterns of genetic relationships among samples), based on genetic distances, was constructed by the Unweighted Pair Group Method Analysis, UPGMA (Sneath and Sokal, 1973), using the Biosys program of Swofford and Selander (1981). The combined probabilities of paternity exclusion were estimated by the formulas of Oishi and Abe (1970).

RESULTS AND DISCUSSION

Protein variability

Of the 15 loci investigated, eight were monomorphic (Mdh, Idh, Sod, Acp, Amy2, GloI, Al and Hb), with the same allele fixed in the three breeds.

Table I shows allele frequencies estimated for the systems that were polymorphic in at least one of the samples under consideration.

At the EsD, Amy1, Cp and Tf loci, the same allele was fixed or presented frequencies higher than 83% in all populations. However, some inter-breed differences in the values estimated for the other alleles were detected. The prevalence of *EsD*B* was about twice higher in Duroc than in Landrace pigs and was not found in Large White animals. On the other hand, *Amy1*A* observed in the last two breeds was not found in Duroc. Additionally, a rare variant (*Amy1*C*) was detected only in the Landrace population.

For the Pgd and Hpx systems (Table I), a general resemblance in the frequency distributions of the most common alleles (*Pgd*A* and *Hpx*1*) was observed between Landrace and Large White pigs. In contrast to these two breeds, the Duroc sample presented predominance of *Pgd*B* and of *Hpx*3*, in addition to a higher frequency of the *Hpx*2* allele.

On the other hand, a great similarity was found in the gene frequency distributions of the Phi system between the Landrace and Duroc populations. In both breeds the values of *Phi*B* were higher than 81%, while in Large White animals the prevalence of the two alleles detected at this locus was similar.

Table I - Frequencies of various blood protein alleles in populations of Landrace, Large White and Duroc breeds.

Locus	Allele	Allele frequencies		
		Landrace (N = 109)	Large White (N = 116)	Duroc (N = 57)
Pgd	*Pgd*A*	0.628	0.621	0.228
	*Pgd*B*	0.372	0.379	0.772
EsD	*EsD*A*	0.931	1.000	0.851
	*EsD*B*	0.069	0.000	0.149
Amy1	*Amy1*A*	0.133	0.090	0.000
	*Amy1*B*	0.862	0.910	1.000
	*Amy1*C*	0.005	0.000	0.000
Phi	*Phi*A*	0.156	0.444	0.184
	*Phi*B*	0.844	0.556	0.816
Cp	*Cp*A*	0.018	0.000	0.000
	*Cp*B*	0.982	1.000	1.000
Hpx	*Hpx*0*	0.064	0.004	0.000
	*Hpx*1*	0.624	0.746	0.070
	*Hpx*2*	0.046	0.000	0.140
	*Hpx*3*	0.266	0.250	0.790
Tf	*Tf*A*	0.037	0.168	0.096
	*Tf*B*	0.963	0.832	0.904

In all systems, except Tf, the observed distributions of phenotype frequencies were as expected by Hardy-Weinberg equilibrium. A significant deviation ($P < 0.05$) in the Tf system was detected in the Large White sample, with an excess of Tf AA individuals.

The values obtained for the expected average heterozygosity, considering the 15 loci (polymorphics and monomorphics), were 0.116 for Landrace, 0.119 for Large

White and 0.095 for Duroc pigs. Considering the common loci, the values were similar to those reported for other populations of these breeds (Van Zeveren *et al.*, 1990) and higher than some estimates obtained for wild pigs (Smith *et al.*, 1980). This suggests that the extent of genetic variability, at least at the loci investigated in this and other studies, have not been affected by the selection methods commonly used by breeders.

Parentage control

The theoretical combined probabilities of excluding the paternity of one of two possible boars, using the seven polymorphic systems, were estimated according to the formulas described by Oishi and Abe (1970). Table II shows the values obtained at each locus, as well as, the combined probabilities which ranged from 59% for Landrace to 50% for Duroc. These values reflected the efficiency of the systems investigated for exclusion paternity in these breeds.

Table II - Estimate of the average probability of excluding one of two possible boars in a parentage case, with the aid of seven different polymorphic loci.

Locus	Breeds		
	Landrace	Large White	Duroc
Pgd	0.179	0.180	0.145
EsD	0.060	0.000	0.111
Amy1	0.108	0.075	0.000
Phi	0.114	0.186	0.128
Cp	0.018	0.000	0.000
Hpx	0.287	0.158	0.180
Tf	0.034	0.120	0.080
All loci	0.587	0.542	0.500

For parentage control tests, it is very important to know which systems are the most informative in a given population. For the samples studied here, and considering the loci examined, the best systems to be used are Hpx, Pgd and Phi. These results agree with those obtained by Oishi and Abe (1970) and by Oishi *et al.* (1979) who reported

values higher than 11% for these systems for the same breeds. On the other hand, in addition to the monomorphic loci, Cp was found to be of no use in this analysis due to the low degree of variability observed in these samples.

In the absence of exclusion, knowing the phenotypes of the animals investigated and the gene frequencies of the population to which they belong, it is possible to calculate a paternity index (Lee, 1980).

Genetic relationships among breeds

Table III gives the genetic distances and similarities obtained according to Rogers (1972), and Figure 1 shows the phenogram constructed from the genetic distance estimates, using the UPGMA method. The patterns of inter-racial genetic relationships revealed that Landrace and Large White pigs are similar (D = 0.044), while Duroc animals presented a genetic distance average from the other two breeds of 0.095.

Table III - Genetic distances and similarities among three swine breeds (x 1000).

Breed	Landrace	Large White	Duroc
Landrace	****	44	84
Large White	956	****	106
Duroc	916	894	****

Below the diagonal: Rogers (1972) Genetic Similarity.
Above the diagonal: Rogers (1972) Genetic Distance.

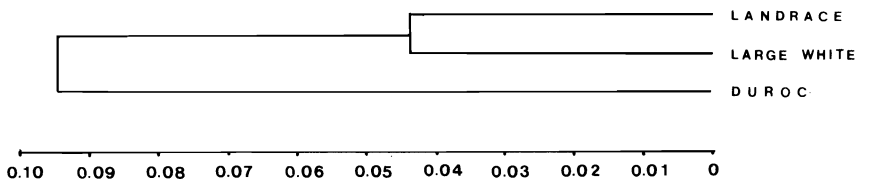

Figure 1 - Cluster analysis by the UPGMA using the Rogers Distance Matrix (1972) calculated for three swine breeds (Cophenetic Correlation: 0.941).

The results obtained in the present study do not agree with those reported by Tanaka *et al.* (1983). These authors, using 13 biochemical systems, found more similarity between Landrace and Duroc pigs than between Landrace and Large White animals. However, the results reported here are in accordance with the historical fact that the Landrace breed was originally developed from Large White swines and native pigs from Denmark (Machado, 1973).

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RESUMO

Quatorze sistemas protéicos, codificados por 15 locos estruturais foram usados para investigar a variabilidade genética em três raças suínas (Landrace, Large White e Duroc) criadas no sul do Brasil. O grau de variabilidade genética varificado foi similar nas três raças (Landrace, $\overline{He}$ = 0,116; Large White, $\overline{He}$ = 0,119; Duroc, $\overline{He}$ = 0,095). Estes valores foram próximos aos computados para outras populações destas raças e maior do que aqueles verificados para populações de suínos selvagens. As freqüências gênicas dos locos polimórficos foram empregadas para avaliar a utilidade destes sistemas para o controle de paternidade. As probabilidades de exclusão de paternidade combinadas foram estimadas em 59% para Landrace, 54% para Large White e 50% para Duroc. A análise do relacionamento genético revelou que Landrace e Large White são as raças mais similares (D = 0,044), enquanto que a Duroc apresentou níveis mais baixos de similaridade genética com as outras duas raças (Landrace/Duroc: D = 0,084; Large White/Duroc: D = 0,106). Estes achados estão de acordo com o desenvolvimento histórico destas raças.

REFERENCES

Franco, M.H.L.P., Moreira, D.M., Salzano, F.M., Santos, S.E.B., Conceição, M.M. and Schneider, H. (1986). New data on the association between the Glioxalase I and Haptoglobin loci. *Hum. Hered. 36*: 126-128.

Gahne, B. and Juneja, R.K. (1985). Prediction of the halothane (Hal) genotypes of pigs by deducing Hal, Phi, Po2, Pgd haplotypes of parents and offspring results from large scale practice in Swedish breeds. *Anim. Blood Groups Biochem. Genet. 16*: 265-283.

Harris, H. and Hopkinson, D.A. (1976). *Handbook of Enzyme Electrophoresis in Human Genetics*. North Holland, Amsterdam.

Kristjansson, F.K. (1963). Genetic control of two pre-albumins in pigs. *Genetics 48*: 1059-1063.

Lee, C.L. (1980). Numerical expression of paternity test results using predetermined indexes. *Amer. J. Clin. Pathol. 73*: 522-536.

Machado, L.C.P. (1973). *Los Cerdos*. Editorial Hemisferio Sur, Buenos Aires.

Naoum, P.C. (1987). *Diagnóstico das Hemoglobinopatias*. Sarvier, São Paulo.

Nei, M. (1975). *Molecular Population Genetics and Evolution*. North Holland, Amsterdam.

Oishi, T. and Abe, T. (1970). Studies on blood groups of pigs. VI. Usefulness of blood groups and serum protein types for parentage test. *Jap. J. Zootech. Sci. 41*: 501-506.

Oishi, T. and Tomita, T. (1976). Blood groups and serum protein polymorphisms in the Pitman-Moore and Ohmini strains of miniature pigs. *Anim. Blood Groups Biochem. Genet. 7*: 27-32.

Oishi, T., Komatsu, M. and Abe, T. (1979). Breed variations of gene frequencies in pig red cell PHI, 6PGD, PGM and ADA enzyme systems and their availability for paternity test. *Jap. J. Zootech. Sci. 50*: 879-884.

Oishi, T., Esaki, K. and Tomita, T. (1980). Genetic relationship among Göttingen Miniature, European and East Asian Pigs investigated from blood groups and biochemical polymorphism. *Jap. J. Zootech. Sci. 51*: 226-228.

Rogers, J.S. (1972). Measures of genetic similarity and genetic distance. *Univ. Texas Publ. 7213*: 145-153.

Smith, C.A. (1986). Chi-square tests with small numbers. *Ann. Hum. Genet. 50*: 163-167.

Smith, M.W., Smith, M.H. and Lehr Brisbin Jr., I. (1980). Genetic variability and domestication in swine. *J. Mamm. 61*: 39-45.

Sneath, P.H.A. and Sokal, R.R. (1973). *Numerical Taxonomy*. W.H. Freeman and Company, San Francisco.

Spencer, N., Hopkinson, D.A. and Harris, H. (1964). Phosphoglucomutase polymorphism in man. *Nature 204*: 742-745.

Swofford, D.L. and Selander, R.B. (1981). BIOSYS-1: A FORTRAN program for the comprehensive analysis of electrophoretic data in population genetics and systematics. *J. Hered. 72*: 281-283.

Tanaka, K., Oishi, T., Kurosawa, Y. and Suzuki, S. (1983). Genetic relationship among several pig populations in East Asia analysed by blood groups and serum protein polymorphisms. *Anim. Blood Groups Biochem. Genet. 14*: 191-200.

Tucker, E.M. (1968). Serum albumin polymorphism in sheep. *Vox Sang. 15*: 306-308.

Van Zeveren, A., Bouquet, Y., Van de Weghe, A. and Coppieters, W. (1990). A genetic blood marker study on 4 pig breeds. I. Estimation and comparison of within breed variation. *J. Anim. Breed. Genet. 107*: 104-112.

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