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HUMAN RED CELL ENZYMES DISTRIBUTION IN A POPULATION SAMPLE FROM BAMBUI (MINAS GERAIS)

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ABSTRACT

Genetic polymorphism of four red cell enzymes (ACP1, ADA, PEPA and ESA) was examined in a sample of 390 individuals from a trihybrid population. Only common alleles were detected, except the ESA system which presented polymorphic frequency in a variant that may be Tashian's *AC* variant (*Am. J. Hum. Genet. 17*: 257-272, 1965) or the ESA^*D_{MAC1} variant (Neel *et al.*, Hum. Genet. 36: 81-107, 1977). Gene and phenotype distributions are presented. They were found at a frequency similar to those of other populations.

INTRODUCTION

Few studies about the distribution of genetic variability have been performed on Brazilian populations except for the Indians. The characterization of gene and genotype frequencies made in a tri-racial sample from Bambui, Minas Gerais state, provided the elements for further analyses that will likely give some indication of the nature of the variability observed in four enzymatic systems: erythrocyte acid phosphatase, esterase A, peptidase A and adenosine deaminase.

Polymorphism in human red cell acid phosphatase (ACP1, EC 3.1.3.2) was described by Hopkinson *et al.* (1963) and its different types are determined by three common alleles, ACP1* *A,B* and *C*. There also exists a series of other alleles reported in different individuals or populations, identified by differences in electrophoretic or isoeletric focusing mobilities (Miller *et al.*, 1987).

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Esterase A (ESA, EC 3.1.1.1) seems to be specific to the erythrocyte. Tashian and Shaw (1962), Tashian (1965) described the common type A1 and the variant AC. Other allelic variants are rare and some have private polymorphism, as for example the *ESA*D$_{MAC1}$* among the Macushi Indians as described by Neel *et al.* (1977).

Peptidase A (PEPA, EC 3.4.11 or 13) occurs practically in all tissues, with two common alleles found with relatively high frequency (Rapley *et al.*, 1971).

Adenosine deaminase (ADA, EC 3.5.44) is observed in all tissues with different electrophoretic patterns. Its polymorphism was described by Spencer *et al.* (1968) and the common phenotypes are due to two alleles, *ADA*1* and 2. Rare variants are described in different populations (Weissmann *et al.*, 1982) and homozygosity for the silent gene *ADA*0* is responsible for the severe combined immunodeficiency disease described in 1972 by Giblett *et al.*

MATERIALS AND METHODS

The population studied is from a Brazilian area endemic for Chagas' disease, Bambui, Minas Gerais state (Dias, 1979). A multidisciplinary study on this American trypanosomiasis was carried out by Krieger and collaborators (1978) in order to study genetic variability to susceptibility to *Tripanosoma cruzi*. This study included the dynamics of biochemical polymorphisms. Families were selected through the records of Fundação Oswaldo Cruz (Bambui Center) by selecting 25-40 year old patients in the chronic phase of the disease. For population details and blood samples see Barbosa *et al.* (1981).

The samples for this work were processed from stored blood cells within six years after bleeding. The red cell electrophoretic patterns were determined following some adaptations (Mestriner, unpublished) of the methods of Harris and Hopkinson (1978), for ACP1, Spencer *et al.* (1968), for ADA, Lewis and Harris (1967) for PEPA and Harris and Hopkinson (1978) for ESA.

The Hardy-Weinberg formula, in spite of some factors limiting gene frequency analyses, was employed and the significance level adopted was 5%.

RESULTS AND DISCUSSION

The four enzymes tested had the usual isozyme patterns encountered in other studies. Table I gives the phenotype numbers observed as well as the χ^2 values calculated on the assumption of the Hardy-Weinberg equilibrium. There were no significant deviation expectations for any system. The corresponding gene frequencies determined by gene counting, as well as the system phenotype distribution according to racial categories of the Bambui population are also presented.

Comparison with other populations yields the following comments:

Table I - Gene frequencies and phenotype distribution of 4 red cell enzymes in Bambui, and gene and phenotype distribution among racial categories in this same population. W: White; B: Black; M: Mixed.

System	Phenotype	Obs	X^2	Gene frequency	Race distribution			Total	Race (%) distribution		
					W	M	B		W	M	B
ACP_1	AA	22	2.43	0.203	20	2	1	23	21	20	21
	AB	112	1.24		82	23	6	111	-	-	-
	BB	251	0.16	0.797	193	42	12	247	79	80	79
Total		385	3.83		295	67	19	381	-	-	-
ESA	A_1A_1	385	0	0.991	293	69	19	381	99	99	100
	A_1AC	7	0	-	6	1	0	7	-	-	-
	ACAC	0	0.04	0.009	0	0	0	0	1	1	0
Total		392	0.04	-	299	70	19	388	-	-	-
PEPA	1-1	383	0	0.994	293	67	19	379	99	99	100
	2-1	5	0	-	4	1	0	5	-	-	-
	2-2	0	0.04	0.006	0	0	0	0	1	1	0
Total		388	0.04	-	297	68	19	384	-	-	-
ADA	1-1	359	0	0.958	272	65	19	356	95.5	96.5	100
	2-1	33	0.06	-	27	5	0	32	-	-	-
	2-2	0	0.71	0.042	0	0	0	0	4.5	3.5	0
Total		392	0.77	-	299	70	19	388	-	-	-

ACP1 system

In our sample only two genes were present: *ACP1*A* and *ACP1*B* (Table I), with frequencies of 0.203 and 0.797, respectively. We noted the absence of the *C* allele, characteristic of Caucasian populations and the *R* allele, wich is a Black racial marker. However, the sample from three individuals presented uncertain phenotypes (B or BC) and after testing them several times, they were considered as B phenotype and as such were included in the analyses. If these subjects had a BC phenotypes, the gene frequencies would be: $A = 0.203$, $B = 0.793$ and $C = 0.004$. The *C* allele does not occur in Brazilian Indian populations, with a rare exception in the Guaymi tribe (Chakraborty *et al.*, 1988), is absent among Blacks and its frequency is 0.03 among Whites of Aracaju, in Northeastern Brazil (Conceição *et al.*, 1987). According to Tashian *et al.* (1967) its presence among individuals of Southeastern Africa reveals Caucasian ascendency. Its frequency is 0.10 in Sweden (Beckman *et al.*, 1971) and 0.11 among Armenians of Iran (Akbari *et al.*, 1986).

The gene frequencies in this system among White, Mixed and Black individuals of Bambui (Table I) differs from that of Aracaju (Conceição *et al.*, 1987) and from parental gene frequencies (reported by Santos *et al.*, 1987) for an Amazonian population. Table II gives these frequencies as percentages.

ESA system

The *A1* gene frequency was 0.991 and that of a variant, 0.009 (Table I). All the variant phenotypes were from unrelated individuals.

The rare variant was less anodal than the common type. Similar electrophoretic variants, the $ESA_{1,2,3}$ $*D_{MAC1}$ variants, respectively, had been described by Tashian (1965) and Neel *et al.* (1977). The characteristics of these variants are:

a. both exhibit slower moving components;

b. the activity of some bands seems to be increased in both variants;

but:

1. in Tashian's variant the Alb band is more active, whereas
2. in Neel *et al.*'s variant the highest intensity is observed in the A2a band.

c. Tashian's variant presents additional slow moving bands in the A2 region, not observed in that of Neel's *et al.*

Our variant did not present visible differences of intensity of the bands, nor were the additional A2 bands of the AC type observed. This may be due to problems of storage, because all the normal A2 bands were reduced or not visible in our sample.

Table III compares our results with those reported by Tashian (1965) and Chakraborty *et al.* (1988) in a paper about genetic variation among tribal populations

Table II - ACP_1 gene frequency distribution among different populations and racial categories.

Population		Gene frequency			B Allele freq.: (X100)-Found in			Reference
Indian	Other	A	B	C	White	Mixed	Blacks	
_	Bambui (tri-hibrid)	0.20	0.80	-	79	80	79	This work
Xavante	-	0.19	0.81	-	-	-	-	Tashian *et al.* (1967)
Amazonian	-	0.09	0.98	-	-	-	-	Salzano *et al.* (1986)
Pano	-	0.06	0.94	-	-	-	-	Mohrenweiser *et al.* 1979)
Yanomana	-	0.01	0.99	-	-	-	-	Tanis *et al.* (1973)
-	Aracaju (white)	0.25	0.72	0.03	72	74	64	Conceição *et al.* (1987)
-	Amazonian (mixed)	0.16	0.79	0.04	65	-	82	Santos *et al.* (1987)
-	Amazonian (white)	0.17	0.81	0.02	-	-	-	Santos *et al.* (1987)

of Central and South America. We note that the *ESA*C* variant was found in a Black population. When we associated the ESA system with the racial composition of the Bambui sample we noted, however, a relatively high concentration of our variant among Caucasian individuals (6 ESAC). But the Black component could exist in individuals of caucasoid phenotype: the Bambui White racial class presents 20% Black genes in its genetic composition (Nogueira, 1981).

Table III - ESA variant distribution among different populations.

Population	Total	Variant alleles	Reference
Caucasian	2600	1 ESA*B	Tashian (1965)
Black	600	1 ESA*B	Tashian (1965)
		1 ESA*C	
Amerindian	11340	85 ESA*D MAC-1	Chakraborty *et al* (1988)
		1 ESA*D GUA-1	
Tri-Racial:	397	7 ESA*C variants	This work
Black	0		
Mixed	1		
Caucasian	6		

$ESA^{*}D_{Macushi}$ variant is restricted to some Indian tribes of Northwestern Brazil and Central America, very distant from Bambui (Eastern Brazil), restricting the gene flow.

We may therefore suppose that our variant is the same *ESA*C* described by Tashian (1965).

PEPA system

We observed no polymorphic frequency of *PEPA*2* (0.064) in Bambui, (Table I). Only the usual alleles *PEPA*1* and *PEPA*2* were observed in this population, and four blood samples without activity were found. Two hypotheses can be made to explain this:

1. These individuals were homozygous for *PEPA*8* gene, whose presence causes enzymatic inactivity in the red cell (Lewis, 1973).

2. The individuals were homozygous for *PEPA*1*, an unstable enzyme (Malcolm *et al.*, 1982) that had lost its activity due to storage problems, transport conditions, etc.

It was not possible to distinguish the true hypothesis through familial distribution, and the individuals were eliminated from the gene and phenotype frequencies calculations. But since the samples were stored for some years, the second hypothesis is more probable than the first one.

This system is monomorphic among some racial groups: Mongolic (Brequet *et al.*, 1982, and Chakraborty *et al.*, 1988) and Caucasians (Lewis and Harris, 1967; Fox *et al.*, 1981, etc.). All Black individuals of the Bambui sample presented the *PEPA*1* gene only, but the PEPA system is polymorphic among other Black populations (Lewis and Harris, 1967; Ojikutu *et al.*, 1977).

ADA system

The frequency of the *ADA*1* gene was 0.958 and of the *ADA*2*, 0.042 (Table I). This is close to that observed in other populations: 0.04 in Bogalusa, USA (Fox *et al.*, 1981); 0.037 among the adult Black population of Porto Alegre, Brazil (Franco *et al.*, 1982) and 0.052 among the Caucasian population of Porto Alegre, Brazil (Weimer *et al.*, 1981).

According to Weissman *et al.* (1982) a pattern of distribution of ADA genes is impossible to determine due to fluctuations observed in gene frequencies between and within populations.

As we could observe, gene and phenotype frequencies of ACP1, ESA, PEPA and ADA systems in Bambui follow the same kind of distribution observed in other populations. Chagas's disease seems to have no influence on the gene and phenotype frequency patterns (data to be presented in a future publication).

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RESUMO

Trezentos e noventa indivíduos de uma população tri-híbrida brasileira foram estudados em relação a 4 sistemas genéticos (ACP1, ESA, PEPA and ADA). Somente os alelos comuns foram detectados nesta população, com exceção do sistema ESA que apresentou frequência polimórfica numa variante rara. Esta variante pode ser classificada como sendo *ESA*D$_{MAC1}$* (Neel *et al.*, 1977) or *ESA*C*

(Tashian, 1965). São apresentadas as freqüências gênicas e fenotípicas e estas são encontradas em distribuição semelhante a de outras populações descritas.

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