Origin of the hemoglobin S gene in a northern Brazilian population: the combined effects of slave trade and internal migrations

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ABSTRACT

We analyzed DNA polymorphisms in the β-globin gene cluster of 30 sickle cell anemia patients from Belém, the capital city of the State of Pará, in order to investigate the origin of the βS mutation. Sixty-seven percent of the βS chromosomes were Bantu type, 30% were Benin type, and 3% were Senegal type. The origin of the βS mutation in this population, estimated on the basis of bS-linked haplotypes, contradicts the historical records of direct slave trade from Africa to the northern region of Brazil. Historical records indicate a lower percentage of people from Benin. These discrepancies are probably due to domestic slave trade and later internal migrations, mainly from northeastern to northern regions. Haplotype distribution in Belém did not differ significantly from that observed in other Brazilian regions, although historical records indicate that most slaves from Atlantic West Africa, where the Senegal haplotype is prevalent, were destined for the northern region, whereas the northeast (Bahia, Pernambuco and Maranhão) was heavily supplied with slaves from Central West Africa, where the Benin haplotype predominates.

INTRODUCTION

Analysis of polymorphic restriction sites in the beta-globin gene cluster has revealed five geographically specific haplotypes linked to βS. These haplotypes are identified by numbers (Antonarakis *et al*., 1984) and also named in accordance with the region where they predominate: haplotype 3 or Senegal type, haplotype 20 or Central African Republic (Bantu) type, haplotype 19 or Benin type (Pagnier *et al*., 1984), haplotype 31 or Arab-India type (Kulozik *et al*., 1986) and haplotype 17 or Cameroon type (Lapouméroulie *et al*., 1992).

The sickle cell mutation is absent among native American populations and was introduced into the American continent by gene flow from Africa during the Atlantic slave trade from the 16th to the 19th century. Thus, analysis of βS-associated haplotypes is a useful tool to investigate the origin and spread of the βS gene in the Americas.

The population of Belém, capital city of the State of Pará, is the result of an intense process of admixture among Caucasian, represented by Portuguese, Amerindians and blacks, represented by African slaves. From 1775 to 1815 the Atlantic slave trade brought about 53,000 slaves imported directly from Africa to the northern region of Brazil, most of them from Bantu Africa (Angola, Congo and Mozambique), with a small percentage from Senegambia, Guine-Bissau and Cape Verde (Curtin, 1969; Vergolino-Henry and Figueiredo, 1990). Another important source of the βS mutation in Belém was internal migration from the northeastern region of Brazil. These people headed towards Amazonian region for rubber tapping or to escape from prolonged droughts in the northeast. Aproximately 300,000 Northeasterners are believed to have entered Amazonian region between 1872 and 1910 (Carneiro, 1980; Oliveira, 1984). The frequency of the bS gene in the Belém population is 0.016 (Ayres *et al.*, 1976), and the proportion of black admixture, estimated using different genetic markers and methods, is from 15 to 33% (Guerreiro and Chautard-Freire-Maia, 1988).

The first study on the origin of black migration to Brazil on the basis of βS haplotypes demonstrated that the Bantu haplotype predominates in the southeast (Zago *et al*., 1992). The Bantu predominance and the scarce contribution of the Senegal haplotype was later confirmed in all other studies (Figueiredo *et al*., 1994; Gonçalves *et al*., 1994).

In a previous study we described the distribution of haplotypes linked to βS mutation in 15 sickle cell anemia patients from Belém (Figueiredo *et al.*, 1994). The three most common African haplotypes were present, with a predominance of the Bantu haplotype (10/15). In this paper we extend this analysis to 15 other patients from Belém. The data are pooled with those previously reported and compared with results obtained in other populations from Brazil, the Caribbean and North America.

MATERIAL AND METHODS

The sample investigated consisted of 30 unrelated sickle cell anemia patients from the Hemotherapy and Hematology Center of the State of Pará, in Belém.

Genomic DNA was isolated from whole blood by phenol-chloroform extraction and ethanol precipitation (Old and Higgs, 1993). Haplotypes were determined by analysis of the following polymorphic restriction sites: 1) *Xmn*I-5'γG; 2) *Hin*dIII-γG; 3) *Hin*dIII-γA; 4) *Hin*cII-Ψβ; 5) *Hin*cII-3'Ψβ and 6) *Hin*fI-5'β. Typing was made by PCR amplification followed by restriction digestion and agarose gel electrophoresis, according to Sutton *et al*. (1989).

RESULTS

The three major African haplotypes (Bantu, Benin and Senegal) were identified among the 30 sickle cell disease patients studied. The haplotype combinations showed: Bantu/Bantu 43%, Bantu/Benin 47%, Benin/Benin 7% and Senegal/Senegal 3%. Sixty-seven percent of the bS chromosomes analyzed were of the Bantu type, 30% of the Benin type, and 3% of the Senegal type.

The βS haplotype frequency distribution observed in Belém did not differ significantly ($\chi^2 = 13.8$; d.f. = 8; P = 0.07) from those observed among patients from other Brazilian regions (Table I). These other regions also show the predominance of the Bantu haplotype, 55 to 79%, followed by Benin, 18 to 45%, and Senegal, 0 to 2% (Zago *et al.*, 1992; Figueiredo *et al.*, 1994; Gonçalves *et al.*, 1994; Wagner *et al.*, 1996).

Table I - βS-linked haplotype frequency distribution (%) in Belém and other Brazilian populations, excluding atypical haplotypes.

Population	No. of chromosomes	Haplotypes		
		Bantu	Benin	Senegal
Belém (PA) [1,2]	60	66.7	30.0	3.3
Salvador (BA)[2]	42	55.0	45.0	0.0
Ribeirão Preto (SP)[3]	67	73.0	25.5	1.5
Campinas/São Paulo (SP)[4]	142	64.7	35.2	0.0
Porto Alegre (RS) [5]	49	79.6	18.4	2.0
Overall	360	67.8	31.1	1.1

1. Present study; 2. Figueiredo *et al.* (1994); 3. Zago *et al.* (1992); 4. Gonçalves et al. (1994); 5. Wagner *et al.* (1996).

DISCUSSION

Data from Curtin (1969) about the African slave trade revealed that 2,876,800 slaves were imported into Brazil during 1701-1810 and 1817-1843, 73% of whom were from Bantu-speaking Africa (Angola, Congo and Mozambique), 26% from Central West Africa (Bight of Benin and Bight of Biafra), and a small group from Atlantic West Africa (Senegambia and Guine-Bissau). Historical data include more detailed information about slaves brought directly from different parts of Africa to different regions of Brazil. These data suggest that about 90% of the slaves imported into northern Brazil were from Angola, Congo and Mozambique, where the Bantu haplotype predominates (Öner *et al.* 1992, Lavinha *et al.* 1992), whereas about 10% of them were from Atlantic West Africa (Senegambia, Guine-Bissau, and Cape Verde), a region where the Senegal haplotype is prevalent (Lavinha *et al.*, 1992; Sow *et al.*, 1995). Moreover, there is evidence that the northeastern region of Brazil (Bahia, Pernambuco and Maranhão) was heavily supplied with slaves from Central West Africa until the middle of the 19th century, and that region probably became the most concentrated area

of the Gold Coast (region between the Bight of Biafra and the Windward Coast) culture in America (Klein, 1986).

The expected distribution of bS haplotypes in Belém is about 86% Bantu, 9% Benin and 4% Senegal on the basis of historical data about the direct importation of slaves from Africa, and considering the distribution of bS haplotypes among populations from Bantu-speaking Africa and Atlantic West Africa (Table II). This does not agree with the observed frequencies (67% Bantu, 30% Benin and 3% Senegal). Nevertheless, there is evidence that slaves were also brought from the Brazilian States of Bahia, Maranhão, Pernambuco, and Rio de Janeiro, and that a small number of slaves from French Guyana and Surinam were introduced into the region, some of whom by the illegal slave trade, while others came as fugitives. Thus, these secondary sources of slaves, as well as later internal migrations, particularly from the northeastern region, may explain the observed frequency of the Benin haplotype in the Belém population. On the other hand, considering that the Senegal haplotype has been associated with a more benign clinical course of sickle cell anemia, having less anemia and potential for less vaso-occlusion, because of higher HbF levels (Powars *et al.*, 1990; Steinberg *et al.*, 1995), it is possible that this haplotype is under-represented in the sample studied, because it was obtained from a reference hospital.

Table II - βS haplotype frequency distribution in various African populations.

Place	βS chromosomes	Haplotype			
		Bantu	Benin	Senegal	Other
Bantu-speaking Africa					
Central African Republic[1]	28	24	2	0	2
Kenya[2,6]	227	223	3	0	1
Tanzania[6]	41	41	0	0	0
Angola[5,7]	60	56	4	0	0
Southern Africa[3]	23	20	0	0	3
Mozambique[7]	4	2	2	0	0
Central West Africa					
Benin[1]	20	0	20	0	0
Nigeria[4]	34	0	33	0	1
Cameroon[5]	80	0	67	0	13
Atlantic West Africa					
Senega[1]	56	0	8	46	2
Cape Verde[7]	11	0	2	9	0
Guine-Bissau[7]	4	0	1	2	1
Republic of Guinea[8]	47	0	2	45	0

1. Pagnier *et al.* (1984); 2. Ojwang *et al.* (1987); 3. Ramsay and Jenkins (1987); 4. Kulozik *et al.* (1986); 5. Lapoumeroulie *et al.* (1992); 6. Öner *et al.* (1992); 7. Lavinha *et al.* (1992); 8. Sow *et al.* (1995).

The haplotype distribution observed among sickle cell anemia patients from Bahia (Figueiredo *et al.*, 1994) certainly reflects the significant presence of slaves from Central West Africa, where the Benin haplotype predominates, since the percentage of the Benin haplotype is only 18% lower than the Bantu haplotype, whereas in other regions the differences range from 46 to 76%. However, regional differences with respect to the place of origin of the slaves brought directly from Africa were probably changed by the domestic trade of slaves, as well as internal migrations, since the βS haplotype frequency distribution observed in Belém and other Brazilian regions did not differ significantly. The interprovincial slave trade was an activity that supplied regional demands for slaves, and redistributed a significant number of them, particularly after the end of the Atlantic slave trade in 1850 (Conniff and Davis, 1994).

When data for the different Brazilian regions are pooled, βS haplotype frequencies (68% Bantu, 31% Benin and 1% Senegal) agree with that expected based on the distribution of haplotypes among the African regions from which slaves were brought to Brazil by the Atlantic slave trade: 70% Bantu, 25% Benin and 1% Senegal (Table II).

On the other hand, the data presented (Table III) show that the haplotype distribution for Brazil is clearly different from those observed among populations from North America (USA and Canada), Caribbean (Jamaica, Cuba and Guadeloupe), and Surinam, where, with the exception of Cuba, the Benin haplotype is the most common (Öner *et al.*, 1992; Wainscoat *et al.*, 1983; Antonarakis *et al.*, 1984; Muniz *et al.*, 1995; Kéclard *et al.*, 1996).

Table III - βS-linked haplotype frequency distribution (%) in Brazil and other populations from the Americas.

Place	No. of chromosomes	Haplotype			
		Bantu	Benin	Senegal	Other
Brazil[1,2,3,4,5]	380	63.9	29.7	1.1	5.3
Canada[6]	61	11.5	49.2	13.1	26.2
U.S.A.[6,7]	447	19.7	56.4	13.6	10.3
Jamaica[7,8]	338	11.5	74.0	2.4	12.1
Surinam[6]	77	29.9	53.2	2.6	14.3
Guadeloupe[9]	266	12.0	73.0	8.0	7.0
Cuba[10]	210	48.1	38.6	7.6	5.7

1. Present study; 2. Zago *et al.* (1992); 3. Figueiredo *et al.* (1994); 4. Gonçalves *et al.* (1994); 5. Wagner *et al.* (1996); 6. Öner *et al.* (1992); 7. Antonarakis *et al.* (1984); 8. Wainscoat *et al.* (1983); 9. Kéclard *et al.* (1996); 10. Muniz *et al.* (1995).

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RESUMO

Com o objetivo de investigar a origem da mutação βS na população da região norte do Brasil, foram analisados polimorfismos de DNA no complexo de genes β da hemoglobina em 30 pacientes com anemia falciforme na população de Belém, a capital do Estado do Pará. Sessenta e sete por cento dos cromossomos βS analisados apresentaram o haplótipo Bantu, 30% o haplótipo Benin e 3% o haplótipo Senegal. A origem da mutação βS na população de Belém, estimada de acordo com a distribuição de haplótipos, não está de acordo com a esperada com base em dados históricos sobre o tráfico de escravos para a região norte, os quais indicam uma reduzida contribuição de escravos da região do Benin. Essas diferenças podem ser atribuídas ao tráfico interno de escravos, bem como ao posterior fluxo de populações imigrantes, particularmente de nordestinos. A distribuição de haplótipos em Belém não difere significativamente da observada em outras regiões brasileiras, muito embora os dados históricos sugiram que a maioria dos escravos procedentes da região do Atlântico-Oeste africano, onde predomina o haplótipo Senegal, foi trazida para o norte do Brasil, enquanto que o nordeste (Bahia, Pernambuco e Maranhão) recebeu o maior contingente de escravos oriundos da região centro-oeste africana, onde o haplótipo Benin é o mais comum. Nós sugerimos que as diferenças regionais quanto à procedência dos escravos africanos também foram modificadas pelo tráfico de escravos estabelecido entre as diferentes regiões brasileiras e posteriormente pelos movimentos migratórios.

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