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Transferrin gene frequencies in Cádiz (southern Spain)

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Summary. The genetic polymorphism of transferrin (Tf) was studied in a sample of 385 healthy unrelated subjects of both sexes resident in the province of Cádiz (southern Spain). Isoelectric focusing was carried out in polyacrylamide gels, followed by staining with Coomassie Blue R250. The gene frequencies obtained were as follows: Tf C1, 0.7922; Tf C2, 0.1883; Tf C3, 0.0195.

Key words: Transferrin – Subtypes – Population study – European distribution

Zusammenfassung. Der genetische Polymorphismus des Transferrin-Systems wurde in einer Stichprobe von 385 gesunden, unverwandten Personen beiderlei Geschlechts aus der Provinz Cádiz (Süd-Spanien) untersucht. Die isoelektrische Fokussierung wurde in Polyacrylamid-Gelen durchgeführt, die Anfärbung erfolgte mit Coomassie Blue R250. – Die erhaltenen Gen-Frequenzen waren: Tf C1 = 0,7922; Tf C2 = 0,1883; Tf C3 = 0,019.

Schlüsselwörter: Transferrin – Subtypen – Populationsgenetik – Verteilung in Europa

Introduction

Serum transferrin (Tf), sometimes also referred to as siderophilin or serotransferrin (Spik et al. 1975), is present in the β-globulin fraction of serum proteins. It is synthesized redominantly in the liver, and its vital function is to transport iron to different sites of the body.

The polymorphism of Tf was discovered by Smithies (1957) using starch gel electrophoresis for separation. The phenotype which recorded the highest frequency value in all human populations was designated as Tf C. Variants that migrated faster than C at alkaline pH were designated Tf B, and those that migrated more slowly were termed Tf D.

Recently the isoelectric focusing method (IEF) has permitted demonstration of the heterogeneity of the Tf C allele. In 1978, Kühnl and Spielmann and also Thymann observed two common subtypes of the Tf C allele (Tf C1 and Tf C2). One year later Kühnl and Spielmann (1979) described another common Tf C variant, Tf C3, among Europeans. Subsequent studies have identified a large number of additional Tf C subtypes (Constans et al. 1980; Kühnl et al. 1981; Dykes et al. 1982; Weidinger et al. 1984; Kamboh and Kirk 1983; Walter et al. 1981; Yuasa et al. 1983).

In this paper we present the results obtained from the study of the transferrin system carried out on a representative population sample in the province of Cádiz (southern Spain).

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Materials and methods

In this study we used 385 blood samples from healthy individuals resident in the province of Cádiz, randomly chosen from patients treated at the outpatients departments of different hospitals following traumatic injury. The number from each hospital was in proportion to the size of the population of its catchment area.

Sera were obtained by centrifugation and stored in Eppendorf tubes at −20°C. Some samples took 26 months to process. Before typing, the serum samples were diluted 1:4 with 0.5 *M* ferrous ammonium sulfate and incubated for 18 h at 4°C.

The IEF method used in this study is based on the condition laid down by Carracedo and Concheiro (1983). The support medium consisted of 0.4 mm thin-layer polyacrylamide gels made on a 24 × 12.5 × 0.2 cm glass plates previously treated with a solution of silane 0.2% and adjusted to pH 3.5 with acetic acid (T = 5.5% and cross-linking of C = 3%). The pH range used in this study was 5–7. Ampholine (LKB, Bromma, Sweden) was used at a concentration of 5%. Sucrose was added as a stabilizing agent at a total concentration of 12% w/v. Polymerization was carried out with 0.5% (v/v) riboflavin solution under ultraviolet light (360 nm).

The serum samples, previously treated with ferrous ammonium sulfate, were applied to the gel on Whatman 3MM paper (0.5 × 0.5 cm) 3 cm from the cathode. The electrode solutions were 1 *M* ethanolamine for the cathode and 1 *M* phosphoric acid for the anode. Focusing was carried out at a constant power of 5 W, the voltage being restricted to 2000 V and unlimited current for approximately 4 h. After focusing, the gels were stained with Coomassie Blue R250.

Results and discussion

The phenotype and gene frequencies observed for the transferrin system among the population of the province

Table 1. Tf phenotypes and gene distribution in Cádiz

Phenotypes	Observed		Expected		Allele frequencies
	n	%	*n*	%	
C1–C1	238	61.818	241.62	62.7593	
C1–C2	119	30.909	114.87	29.8364	Tf C1 = 0.7922078
C1–C3	15	3.896	11.88	3.0865	Tf C2 = 0.1883117
C2–C2	13	3.376	13.65	3.5461	Tf C3 = 0.0194805
C2–C3	0	0.000	2.82	0.7337	
C3–C3	0	0.000	0.15	0.0379	
Total	385	100.000	385.00	100.000	

$X^2 = 4.022332$; *d.f.* = 3; $0.50 > P > 0.25$

of Cádiz and the Hardy-Weinberg equilibrium established in the above-mentioned sample are shown in Table 1. Certain variants in our population for this system are presented in Fig. 1.

The estimated frequency in this study of the Tf C1 allele is very similar to those described in other European populations (Table 2). The values determined for the Tf C2 allele also do not significantly differ from those reported in different populations of southern Europe, being largely similar to those reported in the northern

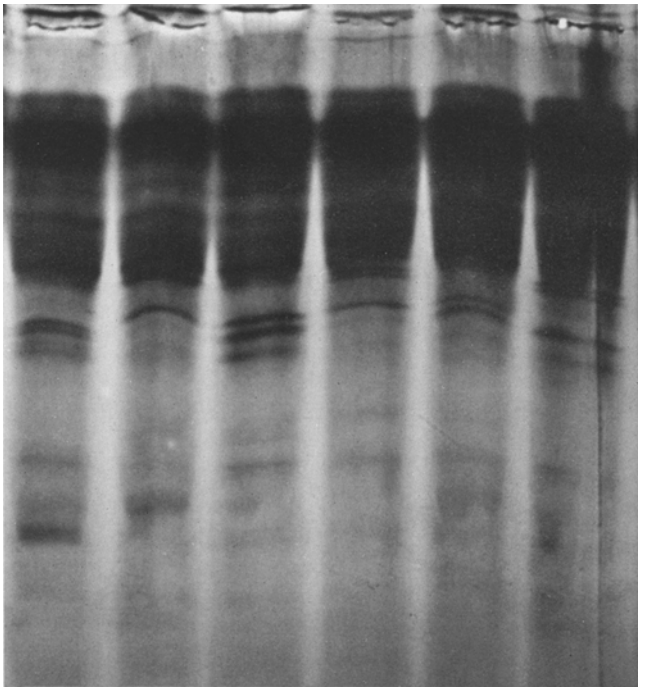

Fig. 1. Tf C variants after IEF pH 5–7. Anode at the top. *Left* to *right:* C1–C3, C1–C1, C1–C2, C1–C1, C1–C2, C2–C2

Table 2. European distribution of Tf allele frequencies

Population	Number	C1	C2	C3	Others	Reference
Germany	3.255	0.79	0.15	0.05	0.009	Rand et al. (1986)
Southern	1.108	0.79	0.13	0.07	<0.01	Weidinger et al. (1984)
Western	380	0.78	0.13	0.07	0.01	Driesel et al. (1981)
Hessen	252	0.79	0.15	0.04	<0.01	Kühnl and Spielmann (1978)
Hessen	876	0.77	0.16	0.06	<0.01	Kühnl et al. (1981)
Hessen and Baden-Württemberg	352	0.75	0.16	0.07	<0.01	Atland et al. (1980)
France	250	0.78	0.13	0.05	0.02	Constans et al. (1980)
Sweden	1.926	0.76	0.15	0.08	0.0071	Hjalmarsson (1983)
Italy						
Vicenza	595	0.78	0.16	0.04	0.001	Nagai et al. (1986)
Belluno	536	0.77	0.14	0.07	<0.01	Pascali and Auconi (1983)
Arezzo	162	0.75	0.19	0.05	–	Pascali and Auconi (1983)
Rome	1.352	0.76	0.18	0.05	<0.01	Pascali and Auconi (1983)
Marsica	126	0.78	0.15	0.05	–	Pascali and Auconi (1983)
Benevento	923	0.76	0.17	0.05	<0.01	Pascali and Auconi (1983)
Padua	618	0.78	0.18	0.04	<0.01	Cortivo et al. (1984)
Tuscany	965	0.77	0.16	0.06	<0.01	Bargagna et al. (1983)
Benevento	226	0.81	0.14	0.04	–	Parisi and Ricci (1985)
Benevento (province)	107	0.818	0.150	0.028	–	Parisi and Ricci (1985)
Siena	200	0.752	0.175	0.064	0.002	Ferretti et al. (1988)
Firenze	86	0.767	0.186	0.047	–	Lucchini et al. (1984)
Catania	84	0.744	0.173	0.065	0.018	Lucchini et al. (1984)
Spain						
Galicia	480	0.78	0.18	0.04	<0.01	Carracedo and Concheiro (1983)
Barcelona	842	0.78	0.17	0.04	<0.01	Gené et al. (1986)
Cádiz	385	0.79	0.19	0.02	–	This study
Greece	295	0.74	0.18	0.06	0.017	Kouvatsi and Triantaphyllidis (1987)

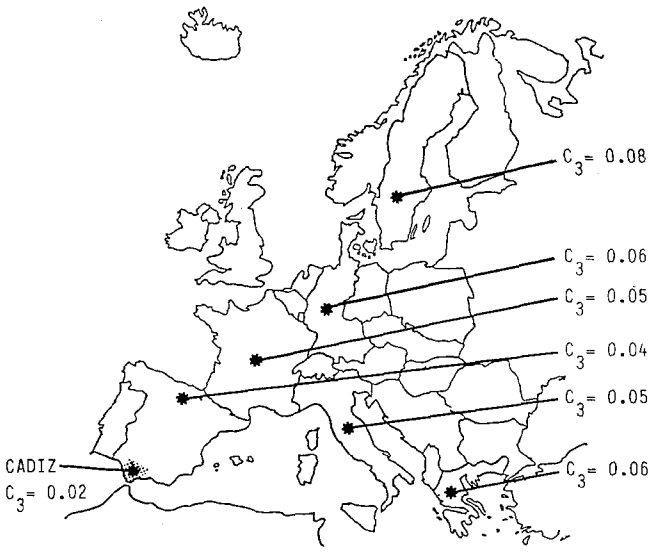

Fig. 2. Average gene frequencies of Tf C3 allele in the European population

part of the Iberian Peninsula (Carracedo and Concheiro 1983; Gené et al. 1986).

The frequency observed for the Tf C3 allele can be regarded as one of the lowest recorded in Europe. This leads us to consider the possibility of a progressive decrease in the frequency of this allele from northeast to southwest Europe, as seen in Fig. 2. Here we show the average gene frequencies of Tf C3 allele subtypes observed by different authors in other populations of the European continent (Table 2).

The a priori exclusion rate observed for this marker in the sample under study was 15.30%, which is somewhat lower than that observed by Carracedo and Concheiro (1983) and by Gené et al. (1986) in other Spanish populations.

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