Distribution of Haptoglobin Subtypes in Greeks

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Human haptoglobin, the serum haemoglobin-binding protein, can be differentiated into three common phenotypes (Hp 1-1, Hp 2-1, and Hp 2-2) by means of starch gel electrophoresis at pH 8.6 (Smithies, 1955). These differences were thought to be determined by a pair of allelic genes Hp^1 and Hp^2 (Smithies and Walker, 1955, 1956). However, chemical reduction of purified haptoglobin followed by electrophoresis in starch gels, prepared in formic acid buffer containing 8M urea, enabled Connell, Dixon, and Smithies (1962) and Smithies, Connell, and Dixon (1962a) to classify individuals of phenotype 1-1 into three subphenotypes, Hp^{1F} -1F, Hp^{1S} -1S, and Hp^{1S} -1S. These three subphenotypes are controlled by two alleles, Hp^{1F} and Hp^{1S} . People of haptoglobin phenotype 2-1 could also be subtyped in a similar fashion into Hp^{2F} -1F or Hp^{2S} -1S. Thus, a series of six phenotypes in the haptoglobin system can now be recognized (Hp^{1F} -1F, 1F-1S, 1S-1S, 2-1F, 2-1S, and 2-2).

Chemical reduction of purified haptoglobin separates the protein into two polypeptide chains. Only the α chain has been found to vary in the six phenotypes, and the allele designations are referred to the α chain. The β chain appears to be unaffected by the genotype of the Hp locus (Smithies, Connell, and Dixon, 1962b). Chemical studies on the α chains (Smithies *et al.*, 1962b; Dixon, Smithies, and Connell, 1962) have disclosed a single amino acid difference between the α^{1S} and the α^{1F} chain. The α^2 chain has characteristics of both α^{1S} and α^{1F} chains and appears to represent the unequal fusion of two α^1 chains. However, two α^2 chain variants were found in a population of Indians from Brazil (Nance and Smithies, 1963).

Although the distribution of the three common phenotypes in various populations is well known, very little information on the distribution of the subtypes of haptoglobin 1-1 is available. Two

interesting surveys (Giblett and Brooks, 1963; Shim and Bearn, 1964) of the distribution of haptoglobin subtypes in various populations have shown that oriental populations are restricted to the Hp^{1S} allele, whereas both Hp^{1S} and Hp^{1F} alleles are present in Caucasian and Negro populations. In Caucasians, in particular, about 30% of the Hp^1 gene frequency consists of Hp^{1F} alleles.

The purpose of this paper is to present data on the distribution of haptoglobin subtypes in Greek population.

Material and Methods

The common haptoglobin types were determined in 2026 healthy Greek adults. Sera were stored at -20°C . Of 2026 (172 of phenotype 1-1 and 430 of phenotype 2-1), 602 were examined for haptoglobin subtypes. Haptoglobin purification was performed according to the method of Smithies *et al.* (1962a). Vertical starch gel electrophoresis was carried out with the standard arrangement described by Smithies (1959), except that the electrodes were reversed and a formate bridge solution (pH 3.7) was used in the two lower and front upper electrode chambers. A voltage gradient of approximately 5 volts/cm. was applied for 18-20 hours at 18° - 20°C . Under these conditions, the resolution between the two types of α^1 polypeptide chains was satisfactory (Fig.).

Results and Discussion

The number of samples examined, the distribution of phenotypes 1-1, 2-1, and 2-2, as well as the frequencies of genes Hp^1 and Hp^2 in Greek population are summarized in Table I (Angelopoulos, Karalis, and Danopoulos, 1966).

The distribution of haptoglobin subtypes and the gene frequencies of Hp^{1F} and Hp^{1S} are shown in Table II.

Smithies *et al.* (1962a) examined a random sample of 93 White American and Canadian blood donors and estimated the Hp^{1S} and Hp^{1F} frequency to be 0.24 and 0.16, respectively. Giblett and Brooks (1963), by testing the sera of 66 White Americans, gave a frequency of 0.25 for Hp^{1S} .

FIG. Results of subtyping procedure. No. 1 = (Hp 1F-2), No. 2 = (Hp 1F-1F), No. 3 = (Hp 1F-1F) No. 4 = (Hp 1F-1S) No. 5 = (Hp 1F-1S) No. 6 = (Hp 1S-1S) No. 7 = (Hp 1S-1S) No. 8 = (Hp 1S-2) No. 9 = (Hp 1S-2).

and 0.13 for Hp^{1F} . Shim and Bearn (1964) examined a sample of 72 Italians and estimated the Hp^{1S} and Hp^{1F} frequency to be 0.25 and 0.11, respectively. The observations by Giblett and Brooks (1963), that Oriental populations have the Hp^{1S} allele whereas Caucasian and Negro populations have both Hp^{1S} and Hp^{1F} alleles, were confirmed by the study of Shim and Bearn

(1964) who extended their investigation to many Mongolian populations.

A comparison of the results obtained in the present study with those published by Giblett and Brooks (1963), Shim and Bearn (1964), and Smithies *et al.* (1962a) is illustrated in Table III.

Table III illustrates that the Hp^{1F} gene frequency in the Greek population is a little higher

TABLE I

DISTRIBUTION OF HAPTOGLOBIN PHENOTYPES AND FREQUENCIES OF GENES Hp^1 AND Hp^2 IN THE GREEK POPULATION

Phenotypes	Population Proportion (P)	Sample Proportion (P)	Gene Frequency	
			Hp^1	Hp^2
Hp^{1-1}	224	0.1096	0.339	0.661
Hp^{2-1}	928*	0.4586		
Hp^{2-2}	874	0.4318		
Total	2026	1.0000		

* 9 were 2-1 modified.

TABLE II

DISTRIBUTION OF HAPTOGLOBIN SUBTYPES AND FREQUENCIES OF GENES Hp^{1F} AND Hp^{1S} IN THE GREEK POPULATION

Total No. Tested	1-1	2-1	1F-1F	1F-1S	1S-1S	2-1F	2-1S	Gene Frequency	
								Hp^{1F}	Hp^{1S}
602	172	430	30	81	61	171	259	0.137	0.202

TABLE III

HAPTOGLOBIN GENE FREQUENCIES IN CAUCASIANS

Population	No. Tested	Gene Frequency			Reference
		Hp^{1F}	Hp^{1S}	$Hp^{1F}:Hp^1$	
American	66	0.133	0.251	0.346	Giblett and Brooks (1963)
American	93	0.16	0.24	—	Smithies <i>et al.</i> (1962a)
Italian	72	0.118	0.252	0.319	Shim and Bearn (1964)
Greek	602	0.137	0.202	0.403	Present study

than in the Italian and White American populations.

Summary

The haptoglobin subtypes after chemical purification of haptoglobin were studied in 602 healthy Greek adults.

The Hp^{1F} and Hp^{1S} gene frequencies were found to be 0.137 and 0.202, respectively. Our results are compared with the results of other published investigations.

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