$\delta\beta$ -Thalassaemia in two Turkish families*

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Summary. Two Turkish families with $\delta\beta$ -thalassaemia are presented. Two sibs belonging to one of these families were doubly heterozygous for both $\delta\beta$ -thalassaemia and β -thalassaemia. The levels of fetal haemoglobin in these two sibs were quite different, namely 49.5 and 75.6%, respectively. The problem of $\delta\beta$ -thalassaemia in Turkey is discussed.

If synthesis of both β - and δ -chains appears to be impaired this type of thalassaemia is called either $\delta\beta$ ($\beta\delta$) or F thalassaemia. In heterozygous $\delta\beta$ -thalassaemia, besides abnormal morphology of the red cells with or without slightly reduced haemoglobin content, an increased level of fetal haemoglobin ranging between 5 and 36% with normal amounts of haemoglobin A₂ are found. This form of thalassaemia has been reported in Italians (Zuelzer *et al*, 1961), Greeks (Fessas, 1961; Gabuzda, Nathan, and Gardner, 1964), Arabs (Ramot *et al*, 1970), American Negroes (Zelkowitz *et al*, 1972), Thais (Wasi *et al*, 1969), and Chinese (Mann *et al*, 1972).

The syndrome resulting from double heterozygosity for β - and $\delta\beta$ -thalassaemia is characterized by a haematological picture similar to that seen in the intermediate type of Cooley's anaemia. But clinical manifestations of this condition vary considerably from mild to severe forms. The doubly heterozygous states of β - and $\delta\beta$ -thalassaemia have been described in individuals of Italian (Zuelzer *et al*, 1961), Greek (Fessas, 1961; Gabuzda *et al*, 1964; Stamatoyannopoulos, Fessas, and Papayannopoulos, 1969), American Negroes (Zelkowitz *et al*, 1972), Thais (Wasi *et al*, 1969), and Chinese (Mann *et al*, 1972) extraction.

The purpose of this paper is to report five individuals with heterozygous $\delta\beta$ -thalassaemia and two sibs doubly heterozygous for both $\delta\beta$ - and β -thalassaemia in two Turkish families.

Methods

Haematological methods were all standard. Haemo-

globin analyses were performed by starch gel, cellulose acetate microzone electrophoresis, and agar gel electrophoresis at pH 8.1, 9, and 6.2 using Tris-EDTA-borate and citric acid-citrate buffers (Robinson *et al*, 1957; Jonxis and Huisman, 1968). Haemoglobin F was determined by the method of Singer, Chernoff, and Singer (1951). Haemoglobin A₂ levels were determined by starch gel electrophoresis according to the method of Aksoy and Erdem (1965). Glucose 6 phosphate dehydrogenase activity was determined quantitatively by a screening method using Dade G6PD reagents.* The heat stability of the haemoglobin in haemolysates was determined by the method of Grimes, Meisler, and Dacie (1964). Intra-erythrocytic inclusion and Heinz bodies were examined by the incubation of red cells at 37° C with cresyl blue (Cartwright, 1968). Haptoglobin types were determined by the method of polyacrylamide gel electrophoresis† (McCombs and Bowman, 1969). Distribution of fetal haemoglobin in red blood cells was determined by the acid elution method of Kleihauer, Braun, and Betke (1957).

Family studies

Family with patients doubly heterozygous for β -thalassaemia and $\delta\beta$ -thalassaemia (Family Ö)

II.1, a 5-year-old Turkish girl in whom lassitude and anaemia started two years previously. At that time, she was transfused twice. Physical examination revealed a pale, mildly icteric girl. The spleen extended 1.5 finger widths below the costal margin. Haematological findings of the patients are summarized in Table I. Skeletal radiology revealed no abnormalities.

II.2, was a 4-year-old girl. She was never transfused. Physical examination revealed a pale, mildly icteric girl. The spleen extended 1.5 finger widths below the costal

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* DADE G6PD reagents, Miami, Florida, USA.

† Beckman Modell 113 acrylamide gel electrophoresis accessory for the Microzone system.

TABLE I
HAEMATOLOGICAL DATA IN TWO TURKISH FAMILIES WITH $\delta\beta$ -THALASSAEMIA

Haematological Data	Family Ö				Family S			
	I.1	I.2	II.1	II.2	I.2	II.1	II.2	III.1
RBC ($10^6/\text{mm}^3$)	5.5	5.4	3.3	3.4		5.3	5.4	4.3
Haemoglobin (g/100 ml)	11.7	12.3	6.5	6.6		12	11.4	10.1
Reticulocytes (%)	2.5	5	7	7.5		2	2	
White blood cells (mm^3)	6200	5700	11,000	10,000		10,000	12,000	6700
Haematocrit (%)	45	45	26	26		43	41	36
MCV (μ^3)	80	83	78	76.4		80	74	83
MCH (pg)	20.8	22.7	19.7	19.4		24	20	23.4
MCHC (%)	26	29	25	25.3		29	27.8	28
Nucleated red cells/100 WBC	0	0	36	13		0	0	0
Hypochromia	+	+	+++	+++		++	++	+++
Microcytosis	+	+	+++	+++		+	+	++
Poikilocytosis	-	-	++	++		+	-	++
Basophilic stippling	-	-	++	+		+	+	++
Ovalocytosis	++	++	+	+		+	++	++
Elliptocytosis	-	-	+	+		+	+	++
Target-cells	++	++	+	+		+	+	++
Total bilirubin (mg/100 ml)	0.7	1	1.3	1.3		0.7	0.8	
Haemoglobin F (%)	0	30	49.5*	75.6	9.5	12.5	10	10.5
Haemoglobin A ₂ (%)	4.3	2.2	2.3	2.2	2.4	2.7	2.4	2.3
G6PD screening test	+	+	+	+	+	+	+	+
Haptoglobin	+	+	-	-	+	+	+	+
Distribution of Hb F in red cells	Uneven	Uneven	Uneven	Uneven		Uneven	Uneven	Uneven

* This patient was not transfused during the last two years.

† Possibly haptoglobin-0. D.: dehydrogenase.

- = absent; + = mild; ++ = moderate; +++ = marked.

FIG. 1. The pedigrees of two Turkish families with $\delta\beta$ -thalassaemia.

margin. Haematological findings are given in Table I. Skeletal radiology revealed no abnormalities.

Family studies confirmed the diagnosis of double heterozygosity for both $\delta\beta$ - and β -thalassaemia in the sibs (Fig. 1). The parents of the sibs were first cousins. The father (I.1) was 36 years old and was heterozygous for β -thalassaemia. His parents were immigrants from Yugoslavia and Crete. The mother (I.2), a 26-year-old Turkish woman, was heterozygous for $\delta\beta$ -thalassaemia. Both of her parents were immigrants from Crete.

Family with heterozygous $\delta\beta$ -thalassaemia (Family S)

II.2, a 45-year-old Turkish male came to our outpatient department for a leukocytosis of long duration. He has had diabetes mellitus for four years. For two years, his leucocyte count ranged between 10,000 and 13,000/mm³. Because of this finding he was referred to our laboratory. Physical examination was unremarkable. Haematological data are given in Table I.

II.1, a 53-year-old male, had also suffered from diabetes mellitus for 10 years. Haematological data are given in Table I.

I.2, a 73-year-old woman, was a Turkish immigrant from Milo. Haematological data are summarized in Table I. She was also diabetic.

III.1, a 4-year-old boy. Haematological data are summarized in Table I.

Discussion

In family Ö, II.1 and II.2 represent the doubly heterozygous form of both $\delta\beta$ - and β -thalassaemia in two Turkish sibs. The clinical and haematological pictures of these two sibs are moderate in degree and similar to those of some cases from Greece with this thalassaemic syndrome described by Stamatoyannopoulos *et al* (1969). According to several investigators dealing with this problem (Stamatoyannopoulos *et al*, 1969; Weatherall and Clegg, 1972), the severity of the clinical manifestations and haematological findings of the doubly heterozygous form of $\delta\beta$ -thalassaemia and β -thalassaemia varies extremely, from a mild thalassaemic syndrome to a severe form of thalassaemia resembling that of homozygous β -thalassaemia. On the other hand, the main feature of haemoglobin analyses is a very high level of fetal haemoglobin ranging between 60 and 98%, determined by the alkali denaturation method (Stamatoyannopoulos *et al*, 1969; Weatherall and Clegg, 1972). The results of haemoglobin analysis were interesting in both our patients. In II.2, the level of fetal haemoglobin, 75.6%, was high. Contrary to this, the amount of fetal haemoglobin in her sister, II.1, was compara-

tively low, 49.5%. This finding was confirmed by agar gel electrophoresis. The content of fetal haemoglobin in this patient is lower when compared to the level of alkali resistant haemoglobin reported in the literature (Stamatoyannopoulos *et al*, 1969; Weatherall and Clegg, 1972). Furthermore, the striking differences in the fetal haemoglobin levels in both sibs are difficult to explain. Apart from this, all other clinical and haematological findings were very similar in both patients. In both sibs the level of haemoglobin A₂ was normal. Although in the majority of patients doubly heterozygous for $\delta\beta$ -thalassaemia and β -thalassaemia the amount of haemoglobin A₂ is low, there are a few patients with this syndrome associated with normal or even increased content of haemoglobin A₂ (Stamatoyannopoulos *et al*, 1969; Weatherall and Clegg, 1972).

In these two Turkish families, five heterozygotes for $\delta\beta$ -thalassaemia have been found. The clinical manifestations and haematological findings of these individuals were very mild and entirely similar to that of a heterozygote for β -thalassaemia.* Haemoglobin F varied between 9.5 and 30%, mean of 14.5%. In one of these five heterozygotes for $\delta\beta$ -thalassaemia the content of fetal haemoglobin (30%) was exceptionally high. Usually, the level of alkali resistant haemoglobin in this thalassaemic syndrome varies between 10 and 20% (Stamatoyannopoulos *et al*, 1969; Weatherall and Clegg, 1972). In accordance with our finding, high amounts of fetal haemoglobin, as high as 36%, were recorded by some investigators (Gabuzda *et al*, 1964).

In respect of $\delta\beta$ -thalassaemia in Turkey, it seems that the frequency of this type of thalassaemia is low, when compared with that of β -thalassaemia or even α -thalassaemia. Although numerous studies on thalassaemic syndromes have been performed in Turkey, only the two families with $\delta\beta$ -thalassaemia presented in this paper have been found up to the present. On the other hand, in a study performed by Çavdar and Arcasoy (1971) among 1000 Turkish people, the incidence of β -thalassaemia was 1.66%. Contrary to this, there is no study showing the exact incidence of α -thalassaemia in Turkish people. But it seems that the incidence of α -thalassaemia might be low in Turkey (Aksoy and Erdem, 1968; Babacan *et al*, 1970; Aksoy *et al*, 1971; Aksoy *et al*, 1972). Furthermore, in two studies among the parents of the patients with thalassaemia

* In two of these five heterozygotes for $\delta\beta$ -thalassaemia (II.1 and II.2 in family S) a mild leukocytosis was found. This haematological abnormality was possibly due to the complications of diabetes mellitus from which both brothers had suffered for several years.

intermedia and thalassaemia major, the present author and his collaborators (Aksoy, 1970; Aksoy, Erdem, and Dinçol, 1974) showed that the frequency of the β -thalassaemia with normal levels of haemoglobin A₂ and F is not low.

The grandparents of two heterozygotes for $\delta\beta$ -thalassaemia and β -thalassaemia and the heterozygote for $\delta\beta$ -thalassaemia in family S (I.2) originated from Turkish extraction living in Greece, Crete, and Milo. Considering the comparatively frequent occurrence of $\delta\beta$ -thalassaemia in Greek people (Stamatoyannopoulos *et al*, 1969; Weatherall and Clegg, 1972) it is quite possible that a blood admixture occurred in the past. In other words, the $\delta\beta$ -thalassaemia gene was transmitted to these two families as a result of an admixture with Greek people in the past.

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