Haemoglobin Lepore_{Boston} in a Turkish family

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Summary. Haemoglobin Lepore was demonstrated in four members of a Turkish family. It was found in the heterozygote state and was associated with erythrocyte morphology similar to that observed in the β thalassaemia trait. The average concentration of haemoglobin Lepore was 8.1% of the total haemoglobin. Structural analysis showed that the Lepore haemoglobin was the Lepore_{Boston} type. This is the first reported instance of the occurrence of haemoglobin Lepore in Turkey.

Haemoglobin Lepore was first discovered in an Italian family by Gerald and Diamond in 1958. Several Lepore type haemoglobinopathies have been seen within the past 17 years (Ostertag and Smith, 1969; Pearson, Gerald, and Diamond, 1959; Pearson *et al*, 1962).

Haemoglobin Lepore is now accepted as an abnormal haemoglobin made up of two Lepore ($\delta\beta$) chains and of two normal α chains (Ostertag and Smith, 1969). The Lepore chain is made up of the N-terminal end of part of the δ chain joined to a portion of the C-terminal end of the β chain (Ohta *et al*, 1971). Baglioni (1962) showed that Lepore haemoglobins were the fusion products of unequal crossing over between δ and β structural genes.

Three different types of haemoglobin Lepore have now been identified each resulting from genetic crossovers at different sites; namely haemoglobin Lepore_{Boston}, haemoglobin Lepore_{Hollandia}, and haemoglobin Lepore_{Baltimore} (Ostertag and Smith, 1969; Williams *et al*, 1972). The fusion in haemoglobin Lepore_{Boston} has occurred somewhere between residue 87 of the δ chain and 116 of the β chain (Ostertag and Smith, 1969; Rahbar, Golban-Moghadam, and Saoodi, 1974). The crossing over in haemoglobin Lepore_{Hollandia} is between the nucleotides coding for $\delta^{22} \text{Ala}$ and $\beta^{50} \text{Thr}$ (Barnabas and Müller, 1962). In haemoglobin Lepore_{Baltimore}, the crossover is between residue 50 of the δ chain and residue 86 of the β chain (Ostertag and Smith, 1969).

It is of considerable interest that the Lepore haemoglobins have been found almost exclusively in

Mediterranean, Papuan, and Negro populations (Barnabas and Müller, 1962; Beaven *et al*, 1964; Fessas, Stamotoyannopoulos, and Keraklis, 1962; Jonxis, 1962; Ostertag and Smith, 1969; Quattrin *et al*, 1967; Ranney and Jacobs, 1964). However, no case of haemoglobin Lepore has yet been shown in Turkey. A description of haemoglobin Lepore occurring in a Turkish family forms the basis of this report.

Subject and methods

Blood counts, haemoglobin, and packed cell volume were measured by conventional methods. Starch block and starch-gel electrophoreses and the determination of haemoglobin F were performed by previously described techniques (Çavdar and Arcasoy, 1971). A solubility test was carried out according to Itano's method (Lehmann, Huntsman, and Young, 1966) and sodium metabisulfite was used for a sickling preparation (Pearson *et al*, 1959).

The structural identification of the abnormal haemoglobin was performed at the MRC Abnormal Haemoglobin Unit, Cambridge, by previously described methods (Arcasoy *et al*, 1974).

Case report

The proband was a 10-year-old boy who was first brought to our attention about a year ago because of fever and a swollen neck. Physical examination showed a slight pallor, marked bilateral cervical lymphadenopathy, and tonsillitis. A differential white cell count suggested the diagnosis of infectious mononucleosis. The blood counts were repeated after the patient recovered from the infection, because of an alteration in red cell morphology noticed on the initial blood film. The haematological findings were as follows: Hb,

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11.2 g/dl; RBC, $4.3 \times 10^{12}/l$; PCV, 31%, WBC $6.52 \times 10^9/l$; platelets, $350 \times 10^9/l$; MCV, 74 fl; MCH, 26 pg; MCHC, 28%. The blood film again showed hypochromia, anisopoikilocytosis, elliptocytosis and a few target cells and basophilic stippling. These findings were considered to be compatible with heterozygous β thalassaemia and further haematological studies were performed. On starch block electrophoresis at pH 8.6, an abnormal fraction was observed in the position of haemoglobin S (Fig. 1). Starch gel electrophoresis also confirmed this finding. However, sickling and solubility tests were both negative. Furthermore the abnormal haemoglobin accounted for 8% of the total haemoglobin of the patient. The haemoglobin A₂ level was 2.1% (normal mean: $2.4 \pm 0.4\%$) and the haemoglobin F was slightly raised at 4.7%. The serum iron level was a little raised to 175 $\mu\text{g}/\text{dl}$, and TIBC was decreased to 220 $\mu\text{g}/\text{dl}$. These findings were similar to those found in previously described haemoglobin Lepore heterozygotes.

Family study

We investigated the family over three generations and three additional cases were also found to be the carriers of haemoglobin Lepore, namely the father, the paternal uncle, and the paternal grandfather (Fig. 2). The

FIG. 2. Pedigree of the family with Hb Lepore.

father aged 42 years was not anaemic. His blood count was as follows: Hb 13.0 g/dl; RBC, $4.74 \times 10^6/\mu\text{l}$; PCV, 40%; MCV, 85 fl; MCH, 27.5 pg; the red cells were hypochromic and there was anisopoikilocytosis, target cells, and ovalocytes on the film. The serum iron was 180 $\mu\text{g}/\text{dl}$ and TIBC 330 $\mu\text{g}/\text{dl}$. The paternal grandfather aged 91 years was anaemic. His blood count was as follows: Hb, 10.2 g/dl; RBC, $3.58 \times 10^{12}/l$; PCV, 35%. His red cells showed anisopoikilocytosis, hypochromia, elliptocytosis, and target forms. The mother was not anaemic and had no abnormal haemoglobin. The electrophoretic analysis of the family is shown in Table I.

The structural analysis of abnormal haemoglobin fraction showed the characteristic changes of haemoglobin Lepore_{Boston}, with the fusion of δ and β chains between residues 87 and 116.

Discussion

The family described here represents the first example of heterozygote haemoglobin Lepore_{Boston} in a Turkish family. This haemoglobinopathy was characterized by erythrocyte morphology similar to that observed in β thalassaemia trait and by the presence of an abnormal haemoglobin with the same mobility as haemoglobin S on starch block and starch gel electrophoresis. The Lepore haemoglobin occurred in a concentration of average 8.1% of the total haemoglobin in 4 members of this family. It is interesting to note that the oldest person carrying haemoglobin Lepore in the family was the paternal grandfather, aged 91, who appeared to be more anaemic than the other haemoglobin Lepore carriers. Old age could have contributed to the severity of his anaemia.

Haemoglobin Lepore_{Boston} has been observed among Italians, Rumanians, Yugoslavians, Greeks, Cypriots, and Iranians, since the original description

FIG. 1. Starch block electrophoresis at pH 8.6 (from bottom); 1—Normal; 2—the propositus, Lepore trait; 3— β -thalassaemia trait with raised Hb A₂; 4— β -thalassaemia trait with raised Hb A₂; 5—chronic myelocytic leukaemia with slight increase in Hb F.

TABLE
ELECTROPHORETIC ANALYSIS IN 'LEPORE' FAMILY

Case	Hb Lepore (%)	A ₂ Hb (%)	F Hb (%)
Propositus	8.0	2.1	4.7
Father	8.5	2.2	4.6
Grandfather	5.8	1.2	2.2
Uncle	10.2	2.2	2.2
Mother	Absent	2.8	1.5

in an Italian family in Boston (Beaven *et al*, 1964; Duma *et al*, 1968; Fessas *et al*, 1962; Gerald and Diamond, 1958; Quattrin *et al*, 1967; Rahbar *et al*, 1974; Ranney and Jacobs, 1964; Rawly *et al*, 1972). The interaction of haemoglobin Lepore and β thalassaemia to produce a disease resembling thalassaemia major has also been described (Beaven *et al*, 1964; Gerald and Diamond, 1958). The homozygous state of haemoglobin Lepore has been observed among Italians, Yugoslavian, and Iranian families (Rahbar, Azizi, and Nowzari, 1975; Weatherall and Clegg, 1972). Furthermore, the patients doubly heterozygous for one of the haemoglobin Lepore variants and haemoglobin C, S, and haemoglobin Peterbrough, have been seen in Afro-American, Greek, and Jamaican families (Ranney and Jacobs, 1964; Weatherall and Clegg, 1972).

Although the haemoglobin Lepore abnormality has been previously reported in Turkish Cypriots, this is the first observation of haemoglobin Lepore_{Boston} among the Turks living in the mainland of Turkey (Beaven *et al*, 1964).

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REFERENCES

- Arcasoy, A., Casey, R., Lehmann, H., Cavdar, A. O., and Berki, A. (1974). A new hemoglobin J from Turkey—Hb ANKARA (β 10 (A 7)→Asp). *FEBS letters, Federation of European Biochemical Societies*, **42**, 121–123.
- Baglioni, C. (1962). The fusion of the peptides chains in hemoglobin Lepore and its interpretations as a genetic deletion. *Proceedings of the National Academy of Sciences of the United States of America*, **48**, 1880.
- Barnabas, J. and Müller, J. C. (1962). Hemoglobin Lepore Hollandia. *Nature (London)*, **194**, 931.
- Beaven, G. H., Gratzner, W. R., Stevens, B. P., Shooter, B. M., Elis, M. J., White, J. C., and Gillespie, J. E. O. N. (1964). An abnormal hemoglobin (Lepore-Cyprus) resembling hemoglobin Lepore and its interaction with thalassemia. *British Journal of Haematology*, **10**, 159.
- Cavdar, A. O. and Arcasoy, A. (1971). The incidence of thalassemia and abnormal hemoglobins in Turkey. *Acta Haematologica*, **45**, 312–318.
- Duma, H., Efremov, G., Sadikario, A., Teodosijev, D., Mladenovski, B., Vlaski, R., and Andreeva, M. (1968). Study of nine families with hemoglobin-Lepore. *British Journal of Haematology*, **15**, 161–172.
- Fessas, P. I., Stamotoyannopoulos, G., and Karaklis, A. (1962). Hemoglobin 'Pylos', study of hemoglobinopathy resembling thalassemia in the hetero-homo and double heterozygotes states. *Blood*, **19**, 1.
- Gerald, P. and Diamond, L. K. (1958). A new hereditary hemoglobinopathy (the Lepore trait) and its interaction with thalassemia. *Blood*, **13**, 835.
- Jonxis, J. H. D. (1962). A thalassemia like picture in a Papuan family. In *Hemoglobin Colloquium*, p. 60. Ed. by H. Lehmann and K. Betke.
- Lehmann, H., Huntsman, R. G., and Young, F. G. (1966). *Man's Hemoglobins*, p. 291. North-Holland, Amsterdam.
- Ohta, Y., Yamoka, K., Sumida, I., and Yenase, T. (1971). Hemoglobin Miyada, A β - δ fusion peptide (Anti-Lepore) type discovered in a Japanese family. *Nature (London)*, **234**, 218–220.
- Ostertag, W. and Smith, E. W. (1969). Hemoglobin Lepore Baltimore: a third type of δ - β crossover (δ 50- β 86). *European Journal of Biochemistry*, **10**, 371–376.
- Pearson, H. A., Gerald, P., and Diamond, L. K. (1959). Thalassemia intermedia due to interaction of Lepore Trait with thalassemia. *American Journal of Diseases of Children*, **97**, 464–472.
- Pearson, H. A., *et al* (1962). Erythokinetics in thalassemia II. Studies in Lepore trait and hemoglobin H disease. *Journal of Laboratory and Clinical Medicine*, **59**, 147–157.
- Quattrin, N., *et al* (1967). Studies on 9 families with hemoglobin Lepore in Campania. *Acta Haematologica*, **37**, 266.
- Rahbar, S., Azizi, M., and Nowzari, G. (1975). A case of Homozygous Haemoglobin Lepore_{Boston} in Iran. *Acta Haematologica*, **53**, 60–64.
- Rahbar, S., Golban-Moghadan, N., and Saoodi, H. (1974). Hemoglobin Lepore Boston in two Iranian families. *Blood*, **42**, 79–83.
- Ranney, H. M. and Jacobs, A. S. (1964). Simultaneous occurrence of hemoglobins C and Lepore in an Afro-American. *Nature (London)*, **204**, 463.
- Rawly, P. T., Barnes, F., and Williams, E. (1969). A Lepore hemoglobin in a Rumanian family. *Human Heredity*, **19**, 48.
- Weatherall, D. J. and Clegg, J. B. (1972). *The Thalassemia Syndromes*, 2nd ed., pp. 178. Blackwell, Oxford.
- Williams, W. J., Beutler, E., Erslev, A. J., and Rundles, R. W. (1972). *Hematology; The Lepore Hemoglobins*, p. 341. McGraw-Hill, New York.