

HEMOGLOBIN D TRAIT WITH ALPHA THALASSEMIA IN A SAUDI FAMILY

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In 1951, Itano¹ encountered a new hemoglobin variant, which co-migrated with hemoglobin S (Hb S) at alkaline pH, but failed to sickle. It was labeled as hemoglobin D (Hb D), and was subsequently found to have the structure α -2- β -2-121 Glu→Gln, where glutamine replaced the normal β -121 glutamic acid.²

The electrophoretic mobilities of Hb D and Hb S are identical at alkaline pH, however, Hb S can be differentiated from Hb D by its decrease solubility in the reduced state and its associated presence with sickling. Furthermore, Hb D can be separated from Hb S on citrate agar gel electrophoresis at pH 6.2.³ It was later found that the Hb D gene can be detected by DNA amplification and globin chain analysis.³⁻⁵ Several cases of Hb D have been reported and were labeled as Hb D Punjab or Hb D Los Angeles.⁶⁻¹⁰

Biochemically, Hb D occurs in four forms: heterozygous Hb D trait, Hb D-thalassemia, Hb S-D disease, and the rare homozygous Hb D disease, which is associated with a clinical disorder similar to, but less severe than, sickle cell anemia.¹¹ Hb D has been reported in association with hematological malignancy such as leukemia and Hodgkin's lymphoma.¹²

In Saudi Arabia, the α -thalassemia gene is highly prevalent, with an incidence of more than 50% in certain areas of the Eastern Province, although Hb D, which is quite prevalent in other ethnic groups, has not been reported in the region.¹³

Materials and Methods

Blood samples were collected in 5 mL tubes containing EDTA as anticoagulant. Red cell indices were determined by Coulter S Plus III (Coulter Electronics, Luton, England), and blood smears were assessed using Wright stain. Sickling test was investigated using sickle quick test (General Diagnostic, North Carolina, U.S.A.).

BCB stain was used to look for hemoglobin H. Serum iron and total iron-binding capacity (TIBC) were determined

by ACA (U.S.A), and serum ferritin was quantified radioisotopically. Hb A₂ was estimated by microcolumn chromatography. Hemoglobin electrophoresis was obtained using cellulose acetate at pH 8.6 and citrate agar at pH 6.2. Globin chain biosynthesis was performed at King Fahad Medical Research Center, Jeddah, Saudi Arabia, according to previously reported methods and by standard techniques.¹⁴

Case Report

The propositus was a four-year-old Saudi girl from the Eastern Province of Saudi Arabia. She was investigated in our clinic for her lifelong microcytic hypochromic anemia, which was refractory to the iron therapy prescribed at several hospitals. There was no history of blood transfusion at any time during her life.

Physical examination revealed pallor of the mucous membranes but no jaundice. Her weight was 11.9 kg and height 94 cm, and within the 50th percentile. The liver and spleen were not enlarged and there was no skeletal deformity. She had two sisters who showed similar clinical findings, while her parents, who are cousins, were asymptomatic.

Results

The laboratory results are shown in Table 1. The propositus and her sisters had mild microcytic hypochromic anemia, and their peripheral blood smears, together with those of the father, showed microcytic hypochromic red cells with a few target cells. Erythrocytosis was observed in the propositus and her father. Iron deficiency anemia was excluded by the normal serum iron, normal TIBC and ferritin concentrations.

The propositus, her mother and sisters were of the Hb D Punjab trait, and they, with the exception of one, demonstrated measurable Hb A₂. On the other hand, Hb A was seen in the father's specimen. α -thalassemia was confirmed by the increased non-alpha to alpha chain ratios in the propositus and her sisters. Gene analysis to determine the type of α -thalassemia is being carried out and will be reported when available. No hemoglobin H was detected electrophoretically, and no Hb H inclusions were seen in red cell smears.

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TABLE 1. *Clinical laboratory data of the study family.*

| | Age | Hb g/L | RBC 10 ¹² /L | Hematocrit | MCV fL | MCH pg | RDW | Retics % | Non-alpha to alpha ratio | HbA % | HbA ² % | HbD % | Iron μmol/L | TIBC μmol/L | Ferritin μg/L |
|------------|-----|-----------|----------------------------|------------|-----------|-----------|------|-------------|-----------------------------|----------|-----------------------|----------|----------------|----------------|------------------|
| Father | 27 | 13.8 | 6.36 | 0.44 | 68.6 | 21.8 | 13.9 | 0.6 | — | 97 | 3 | 0 | 14.7 | 45.7 | 124.4 |
| Mother | 22 | 12.5 | 5.3 | 0.39 | 80.9 | 28.6 | 13.3 | 1.7 | — | 66.1 | 2 | 31.9 | 15.9 | 52.7 | 9.5 |
| Propositus | 4 | 9.9 | 7.1 | 0.28 | 56.0 | 20.3 | 14.4 | 0.8 | 2.82 | 62 | 2 | 36 | 17.2 | 44.6 | 11.23 |
| Sister (1) | 2 | 11.7 | 5.73 | 0.37 | 63.7 | 20.7 | 14.1 | 1.9 | 2.23 | 65 | 2 | 33 | 21.7 | 57.5 | 9.1 |
| Sister (2) | 7 m | 10.5 | 5.53 | 0.33 | 58.9 | 19.0 | 14.6 | 1.2 | 3.4 | 69 | 0 | 31 | 20.9 | 49.4 | 26.1 |

Discussion

Overlooking nearly 700 km of the coastline along the Arabian Gulf, Eastern Arabia could well have played a great role in trade traffic among ancient civilization, particularly from the North and the East. During the Islamic period, after about 630 AD, the area witnessed active trade with different regions and territories, including India and Persia. There was also a period of Turkish rule in this region. Abnormal hemoglobin has been reported frequently from these countries,^{6,8,15,16} It is, therefore, not surprising that Hb D, Hb S, and other abnormal hemoglobin yet to be discovered, are found in this part of Saudi Arabia. The origin of the family in this study is the Al-Hasa area in the Eastern Province of Saudi Arabia. There is no ethnic relationship with Persia or the Indian subcontinent.

The unusual combination of Hb D and α -thalassemia in our Saudi family have a similarly unknown historical background. It is interesting that both clinical and hematological abnormalities observed in our patient, as well as her sisters, are consistent with previous observations reported in double heterozygosities for Hb D-thalassemia.¹⁷⁻¹⁹ Although the non-alpha to alpha ratio was not performed on either parent, the laboratory data suggested that the father was responsible for transmitting the alpha gene to the α -thalassemia genes in his three daughters, while the mother proved to be the source of the Hb D gene (32%). With normal MCV, however, α -thalassemia was difficult to rule out in her case. As there were no other abnormalities to which we could attribute the mild nature of anemia in the patient, the compound heterozygosity of α -thalassemia and Hb D remains the only explanation for this.

A community survey of the hemoglobin patterns in this region is needed to assess the incidence of abnormal hemoglobins. The history of interaction of the population of this Province with other countries in the region, and the prevalence of consanguinity, may be responsible for the occurrence of several inherited problems seen in the population.

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