Introduction

More than 800 hemoglobin variants are known to this day. Most of them have no clinical relevance but a great number of hemoglobin variants are responsible for many symptoms, such as jaundice, leg ulcers, erythrocytosis, hematuria, cerebrovascular incidents, cyanosis, retinal and vitreous hemorrhages, priapism and many others.

It was a study of hemoglobin S by Pauling and co-workers that led to the concept of “molecular disease” and the subsequent development of the field of molecular biology. Studies of hemoglobin S also led to the elucidation of the double helix structure of DNA (Watson and Crick).

Structure, Function and Nomenclature

The various hemoglobins (Hb) present in normal red cells are tetramers of subunits called α, β, γ, δ:

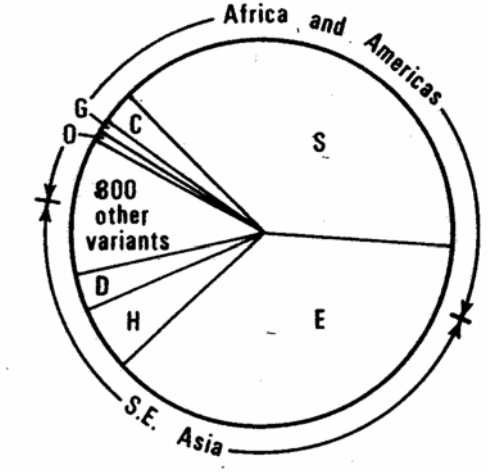
Hb A	α_2, β_2
Hb F	α_2, γ_2 (fetal hemoglobin)
Hb A_2	α_2, δ_2

The chains of 141 or 146 amino acid residues that constitute the primary structure of hemoglobins form seven or eight helical regions. The heme group, composed of a porphyrin ring with an iron atom in the centre, is tucked into a crevice between two helices. The oxygen enters the heme pocket and is bound between the distal histidine and the iron atom of heme.

The designation of the first known normal and abnormal hemoglobins were named according to the alphabet. But later on, based on the increasing number of new variants, it was recommended to name hemoglobin variants after the community, state, county or nationality of the index case, the hospital or medical centre in which the discoverer of the variant works (Huisman et al., 1998).

Indication of Testing for Hb Variants

Clinically significant Hb variants are usually first observed by routine hematological procedures. A low Hb level, microcytosis, hypochromia, reticulocytosis, bilirubinemia may suggest the possible presence of an α- or β-thalassemia, or an unstable hemoglobin type. Furthermore, a high Hb level (erythrocytosis) together with appropriate clinical observations may suggest a hemogobin variant with an increased oxygen affinity.



Worldwide distribution of Hb variants

Bioscientia´s Testing Programs

For many years we performed, as a basic method, the electrophoresis on cellulose acetate. In the meantime, the chromatographic techniques, especially HPLC are so highly developed that we changed our testing program from electrophoretic to chromatographic methods. We therefore proceed as follows:

1. HPLC chromatography and evaluation of the chromatogram with the different peaks and for the results for HbA2, HbF and HbA. Most Hb variants can easily be detected by this method.

If there is an abnormal chromatogram, we continue with:

2. Isoelectric Focussing (IEF) to confirm or to get a more precise differentiation between different suspected variants.

In all cases of hemoglobin variants we routinely perform the:

3. Electrophoresis in acidic milieu: Citrate-Agar-Gel Electrophoresis (CAG) at pH 5,9–6,1 with reference hemoglobins.

Using these different steps and methods, usually more than 95% of all abnormal hemoglobins are detectable. For all additional structure analyses with different techniques (such as DNA analysis) we collaborate with colleagues from the Department of Biochemistry of the Hôpital Henri Mondor at Creteil near Paris, France.

Bioscientia: New Variants Detected

During the last 20 years in performing hemoglobin studies we have detected in our lab in collaboration with our colleagues from Creteil 4 new variants:

1. Hb Ingelheim (also known as Hb Coimbra)
   β99 (G1)Asp→ Glu
   (Wajcman, Behnken, 1991)
   Symptoms: Cyanosis with erythrocytosis

2. Hb Mainz, β98 (FG5) Val→ Glu
   (Wajcman, Behnken, 1994)
   Unstable hemoglobin causing severe hemolytic anemia

3. Hb Frankfurt, α50 (CE8) His→ Gln
   (Wajcman, Behnken, 1998)
   Hemoglobin variant without any clinical relevance, found during HbA1c studies

4. Hb Ilmenau (C7) β41 Phe→Cys
   (Prehu, Behnken, 2002). Unstable hemoglobin with low oxygen affinity causing hemolytic anemia and cyanosis.

In our cooperation with many hospitals in the Middle East we have detected patients with Hb S, Hb D (Hb D Punjab and Hb D Iran), Hb E, Hb O Arab, and furthermore we were able to detect carriers of:

Hb E-Saskatoon, β22 Glu → Lys

Hb Fontainebleau, α21 Ala → Pro

Hb Handsworth, α18 Gly → Arg

Hb Okayama, β2 His → Gln

Hb Setif, α94 Asp → Tyr

Further Hemoglobin variants detected in our Laboratories:

Hb Name	Substitution	Hemato-logy
Hb J-Wenchang-Wuming	α11 Lys→Gln	normal
Hb Shaare Zedek	α56 Lys→Glu	Anemia
Hb O Indonesia	α116 Glu→Lys	normal
Hb Sassari	α126 Asp→His	Polyglobuly
Hb Okayama (18Families)	β2 His→Gln	normal
Hb Freiburg	β23 Val fehlt	Anemia
Hb J Auckland	β25 Gly→Arg	normal
Hb Rothschild	β37 Trp→Arg	Anemia
Hb Seattle	β70 Ala→Asp	Anemia
Hb Köln (5 Families)	β98 Val→Met	Anemia
Hb Camperdown	β104 Arg→Ser	Anemia
Hb Presbyterian	β108 Asn→Lys	normal
Hb Andrew Minneapolis	β144 Lys→Asn	Polyglobuly

References

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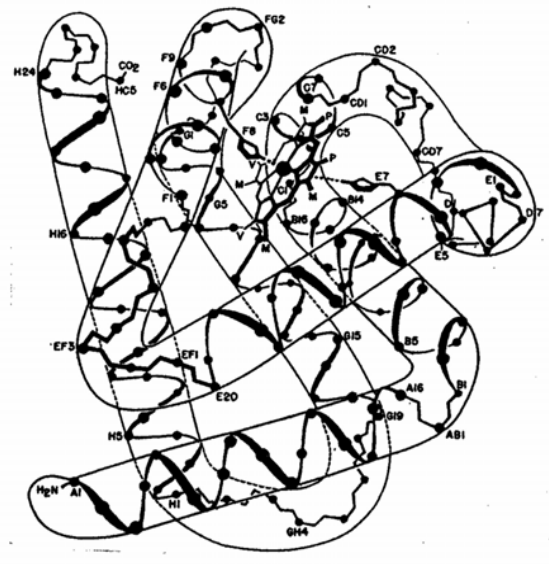
Hemoglobinopathies and Thalassemias



Hemoglobin molecule

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