

as those that encode the enzymes for heme biosynthesis. Normal red blood cell (RBC) differentiation requires the coordinated expression of the globin genes with the genes responsible for heme and iron metabolism. RBC precursors contain a protein, α -hemoglobin-stabilizing protein (AHSP), that enhances the folding and solubility of a globin, which is otherwise easily denatured, leading to insoluble precipitates. These precipitates play an important role in the thalassemia syndromes and certain unstable hemoglobin disorders. Polymorphic variation in the amounts and/or functional capacity of AHSP might explain some of the clinical variability seen in patients inheriting identical thalassemia mutations.

CLASSIFICATION OF HEMOGLOBINOPATHIES

There are five major classes of hemoglobinopathies (Table 127-1). *Structural hemoglobinopathies* occur when mutations alter the amino acid sequence of a globin chain, altering the physiologic properties of the variant hemoglobins and producing the characteristic clinical abnormalities. The most clinically relevant variant hemoglobins polymerize abnormally, as in sickle cell anemia, or exhibit altered solubility or oxygen-binding affinity. *Thalassemia syndromes* arise from mutations that impair production or translation of globin mRNA, leading to deficient globin chain biosynthesis. Clinical abnormalities are attributable to the inadequate supply of hemoglobin and the imbalances in the production of individual globin chains, leading to premature destruction of erythroblasts and RBC. *Thalassemic hemoglobin variants* combine features of thalassemia (e.g., abnormal globin biosynthesis) and of structural hemoglobinopathies (e.g., an abnormal amino acid sequence). *Hereditary persistence of fetal hemoglobin* (HPFH) is characterized by synthesis of high levels of fetal hemoglobin in adult life. *Acquired hemoglobinopathies* include modifications of the hemoglobin molecule by toxins (e.g., acquired methemoglobinemia) and clonal abnormalities of hemoglobin synthesis (e.g., high levels of HbF production in preleukemia and α thalassemia in myeloproliferative disorders).

TABLE 127-1 CLASSIFICATION OF HEMOGLOBINOPATHIES

- I. Structural hemoglobinopathies—hemoglobins with altered amino acid sequences that result in deranged function or altered physical or chemical properties
 - A. Abnormal hemoglobin polymerization—HbS, hemoglobin sickling
 - B. Altered O₂ affinity
 1. High affinity—polycythemia
 2. Low affinity—cyanosis, pseudoanemia
 - C. Hemoglobins that oxidize readily
 1. Unstable hemoglobins—hemolytic anemia, jaundice
 2. M hemoglobins—methemoglobinemia, cyanosis
- II. Thalassemias—defective biosynthesis of globin chains
 - A. α Thalassemias
 - B. β Thalassemias
 - C. $\delta\beta$, $\gamma\delta\beta$, $\alpha\beta$ Thalassemias
- III. Thalassemic hemoglobin variants—structurally abnormal Hb associated with coinherited thalassemic phenotype
 - A. HbE
 - B. Hb Constant Spring
 - C. Hb Lepore
- IV. Hereditary persistence of fetal hemoglobin—persistence of high levels of HbF into adult life
- V. Acquired hemoglobinopathies
 - A. Methemoglobin due to toxic exposures
 - B. Sulfhemoglobin due to toxic exposures
 - C. Carboxyhemoglobin
 - D. HbH in erythroleukemia
 - E. Elevated HbF in states of erythroid stress and bone marrow dysplasia

EPIDEMIOLOGY



Hemoglobinopathies are especially common in areas in which malaria is endemic. This clustering of hemoglobinopathies is assumed to reflect a selective survival advantage for the abnormal RBC, which presumably provides a less hospitable environment during the obligate RBC stages of the parasitic life cycle. Very young children with a thalassemia are *more* susceptible to infection with the nonlethal *Plasmodium vivax*. Thalassemia might then favor a natural protection against infection with the more lethal *Plasmodium falciparum*.

Thalassemias are the most common genetic disorders in the world, affecting nearly 200 million people worldwide. About 15% of African Americans are silent carriers for a thalassemia; a thalassemia trait (minor) occurs in 3% of African American and in 1–15% of persons of Mediterranean origin. β Thalassemia has a 10–15% incidence in individuals from the Mediterranean and Southeast Asia and 0.8% in African Americans. The number of severe cases of thalassemia in the United States is about 1000. Sickle cell disease is the most common structural hemoglobinopathy, occurring in heterozygous form in ~8% of African Americans and in homozygous form in 1 in 400. Between 2 and 3% of African Americans carry a hemoglobin C allele.

INHERITANCE AND ONTOGENY

Hemoglobinopathies are autosomal codominant traits—thus, compound heterozygotes who inherit a different abnormal mutant allele from each parent exhibit composite features of each. For example, patients inheriting sickle β thalassemia exhibit features of β thalassemia and sickle cell anemia. The α chain is present in HbA, HbA₂, and HbF; α -chain mutations thus cause abnormalities in all three. The α -globin hemoglobinopathies are symptomatic in utero and after birth because normal function of the α -globin gene is required throughout gestation and adult life. In contrast, infants with β -globin hemoglobinopathies tend to be asymptomatic until 3–9 months of age, when HbA has largely replaced HbF. Prevention or partial reversion of the switch should thus be an effective therapeutic strategy for β -chain hemoglobinopathies.

DETECTION AND CHARACTERIZATION OF HEMOGLOBINOPATHIES—GENERAL METHODS

Electrophoretic techniques are still widely used for hemoglobin analysis. Electrophoresis at pH 8.6 on cellulose acetate membranes is especially simple, inexpensive, and reliable for initial screening. Agar gel electrophoresis at pH 6.1 in citrate buffer is often used as a complementary method because each method detects different variants. Some important variants are electrophoretically silent. These mutant hemoglobins can usually be characterized by more specialized techniques such as mass spectroscopy, which is rapidly replacing electrophoresis for initial analysis.

Quantitation of the hemoglobin profile is often desirable. HbA₂ is frequently elevated in β thalassemia trait and depressed in iron deficiency. HbF is elevated in HPFH, some β thalassemia syndromes, and occasional periods of erythroid stress or marrow dysplasia. For characterization of sickle cell trait, sickle thalassemia syndromes, or HbSC disease, and for monitoring the progress of exchange transfusion therapy to lower the percentage of circulating HbS, quantitation of individual hemoglobins is also required. In most laboratories, quantitation is performed only if the test is specifically ordered. Complete characterization, including amino acid sequencing or gene cloning and sequencing, is readily available from several reference laboratories.

Because some variants can comigrate with HbA or HbS (sickle hemoglobin), electrophoretic assessment should always be regarded as incomplete unless functional assays for hemoglobin sickling, solubility, or oxygen affinity are also performed, as dictated by the clinical presentation. The best sickling assays involve measurement of the degree to which the hemoglobin sample becomes insoluble, or gelled, as it is deoxygenated (i.e., sickle solubility test). Unstable hemoglobins are detected by their precipitation in isopropanol or after heating to 50°C. High-O₂ affinity and low-O₂ affinity variants are detected