Hemoglobin E

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Abstract
Hemoglobin E (HbE) is a variant hemoglobin with a mutation in the β globin gene causing substitution of glutamic acid for lysine at position 26 of the β globin chain. HbE is the second commonest abnormal hemoglobin after sickle cell hemoglobin (HbS). HbE is common in South-East Asia, where its prevalence can reach 30-40% in some parts of Thailand, Cambodia and in Laos. Hb E is also found in Sri Lanka, North Eastern India, Bangladesh, Pakistan, Nepal, Vietnam, Malaysia. The β chain of HbE (β^E) is synthesized at a reduced rate compared with that of normal adult hemoglobin (HbA), as the mutation creates an alternate splicing site within an exon. Consequently heterozygotes AE, compound heterozygotes SE and homozygotes EE show some β thalassemic features. Subjects heterozygous for HbE (AE) have an asymptomatic condition with no clinical relevance, except for the risk of transmitting E/β thalassemia if the other parent carries β thalassemia. The severity of these E/β thalassemia forms is very variable, the clinical picture ranging from that of β thalassemia minor through thalassemia intermedia to thalassemia major. Subjects homozygous for HbE (EE) are asymptomatic. Management concerns exclusively E/β thalassemia forms. During childhood, regular follow-up of growth and facial deformities, hemoglobin level, prophylaxis of infections source of worsening of anemia with vaccines, treatment of potential infectious sites are essential. Daily oral penicillin is recommended. Transfusions are indicated in case of bad tolerance of anemia and /or facial deformities. The main complication of regular transfusions is iron overload, source of hepatopathy, cardiac failure, endocrinopathies requiring thus iron chelation. The most widely used chelator is deferoxamine (Desféral ®). Splenectomy can diminish or suppress transfusion requirements in intermediate forms and should not be performed before 5 years. Pregnancy requires regular antenatal care by obstetricians, in collaboration with hematologist. Transfusions are often required in the 3rd trimester in women with β thalassemia intermedia, to reduce anemia and low weight births. Treatment of infections as well of thrombosis prophylaxis are requested. Bone marrow transplantation has very rare indications. Recombinant erythropoietin alone or associated with hydroxyurea may be useful in reducing transfusions requirements, in improving quality of life and in diminishing hemopoietic ectopic extra-medullary masses.

Keywords
Hemoglobin E, *β globin* gene, hemoglobin E trait, hemoglobin E disease, hemoglobin E/βthalassemia, sickle cell/hemoglobin E disease

Disease name
Hemoglobin E

Included diseases
Hemoglobin E trait, hemoglobin E disease, hemoglobin E/β thalassemia, sickle cell/hemoglobin E disease

Definition
Hemoglobin E =HbE ($\alpha 2\beta 2^{26\ Glu\_Lys}$) is a variant hemoglobin with a mutation in the *β globin* gene causing substitution of glutamic acid for lysine at position 26 of the β globin chain. HbE may be present in the heterozygous state (genotype AE or hemoglobin E trait), the homozygous state (EE or hemoglobin E disease) and a variety of compound heterozygous states such as hemoglobin E/βthalassemia (E/βthal), sickle cell/hemoglobin E disease (SE genotype).
The β chain of HbE (β^E) is synthesized at a reduced rate compared with that of normal adult hemoglobin (HbA). This is because the mutation creates an alternate splicing site within an exon. This results in reduced rate of synthesis of β^E chain and therefore of HbE, and consequently heterozygotes, compound heterozygotes and homozygotes show some β thalassemic features. HbE may therefore be regarded as a β^+thalassemic hemoglobinopathy.

Diagnostic criteria and clinical description
Hemoglobin E trait
HbE trait is defined by the heterozygous condition associating with one normal *adult hemoglobin (HbA) β* gene and one variant *hemoglobin E β* gene.

Clinical features
HbE trait is an asymptomatic condition with no clinical relevance, except for the risk of compound heterozygous states with βthalassemia in the offspring.

Laboratory features
Most of individuals with HbE trait have reduced mean corpuscular volume (MCV) and mean cell hemoglobin (MCH), with or without mild anemia. The red blood cell indices resemble those of thalassemia trait. Individuals with HbE trait may carry α thalassemia conditions of varying severity according to the number of non-functional α genes 1,2,3. However even those with a full complement of α genes may be microcytic and mildly anemic.
The blood film may be normal or may show hypochromia, microcytosis, target cells, irregularly contracted cells, basophilic stippling or any combination of these features.

Electrophoretic profile
Hemoglobin electrophoresis at alkaline pH on cellulose acetate shows that the variant HbE has the same mobility than that of the variant HbC ($\alpha 2\ \beta 2^{6glu\_lys}$) and the adult hemoglobin HbA2 ($\alpha 2\ \delta 2$). On citrate agar or agarose gel at acid pH, the mobility of HbE is the same than that of HbA and HbA2. HbE has a characteristic mobility on isoelectric focusing, being well separated from HbA and focusing like HbA2, and thus unlike HbC. On HPLC, HbE is easily separated from HbA and HbC, but may co elute with HbA2.
In HbE heterozygotes, the variant usually comprises 33 % or less of total HbA. Individuals with less than 30% of HbE almost always have co existing α thalassemia trait.
HbE is slightly unstable in heat and isopropanol stability tests.

Hemoglobin E disease
Hemoglobin E disease is defined by the coexistence of two βE alleles (homozygous state EE). At birth, differential diagnosis is E/β^Othalassemia, which is always symptomatic after disappearance of HbF. Study of both parents (heterozygotes AE in case of homozygous EE in the child) is mandatory.

Clinical features
Individuals with the genotype EE are usually completely asymptomatic. There is usually no anemia and rarely any evidence of hemolysis. The spleen is not usually enlarged. Otherwise a coexisting HbH disease (α_3 thalassemia) must be considered.

Laboratory features
Blood count. The blood count often ressembles that of βthalassemia trait, with a normal hemoglobin concentration or very mild anemia and increased red blood cell (RBC), reduced MCV and MCH. MCHC is usually normal.

Blood film. The blood film usually shows hypochromia and microcytosis with variable numbers of target cells, basophilic stippling and irregularly contracted cells.

Electrophoretic profile
Hemoglobin electrophoresis shows the major hemoglobin to be HbE, with HbE plus HbA_2 constituting 95-99 % of total hemoglobin.

Hemoglobin E/βthalassaemia
Hemoglobin E trait may be co inherited with either β^0 or β^+ thalassemia. The compound heterozygous state is quite common in Thailand and occurs throughout a large part of South-East

Asia, stretching form Indonesia to Sri Lanka, North-East India and Bangladesh, making this the most common β thalassemia phenotype in the world and one of the most prevalent genetic disease in the world.

Clinical features
The severity of compound heterozygotes for HbE and β thalassemia is very variable, the clinical picture ranging from that of βthalassemia minor through thalassemia intermedia to thalassemia major. Most patients have a disease that is at least moderately severe.
The most severely affected individuals are transfusion dependent and have liver enlargement and splenomegaly, intermittent jaundice, growth retardation and over expansion of the bone marrow cavity leading to facial deformity and defective tooth implantation. Less severely affected individuals may have splenomegaly and facial deformity but do not require regular transfusions to maintain life. Hypersplenism may occur. Splenectomy is usually efficient in reducing transfusion requirements. During pregnancy, patients may temporarily become transfusion dependant. Extramedullary hemopoiesis can occur and has sometimes led to compression of the spinal cord or brain by tumour like masses of hemopoietic tissue.

Blood count
The hemoglobin concentration is lower than in HbE sease. Hydroxyurea therapy alone or with recombinant erythropoietin may induce a slight rise in hemoglobin concentration, but sufficient to avoid transfusion need.

Other investigations
Hemoglobin electrophoresis and HPLC show the presence of HbE, HbA2 and HbF in the case of HbE/β^0 thalassemia, and HbE, HbA, HbA2 and HbF in the case of HbE/β^+ thalassaema. When HbA is present, it usually represents around 10 % of total hemoglobin.

Compound heterozygotes SE
The coinheritance of *βS* and *βE* genes leads to a sickling disorder similar to sickle cell β^+ thalassemia, with mild chronic haemolytic anemia, reduced MCV and MCCH. Vaso-occlusive crises are usually rare, more frequent during pregnancy. Recurrent splenic infarctions during air travel have been reported.

Laboratory features
HbE concentration may be normal or reduced (9-14 g/dL) and reticulocyte count is usually mildly elevated. MCV is often reduced. The blood film shows target cells, sickle cells are uncommon. HbS represents a larger proportion of total hemoglobin than HbE (around 65%). HbF may be normal or slightly elevated.

Epidemiology
HbE is common in South-East Asia. The highest prevalence of carriers is in some parts of Thailand, in Cambodia and in Laos. Thailand and Myanmar (previously Burma) have an overall prevalence of around 14-15 %. Gene frequency in Thailand varies from 8 to 50-70 %, being highest in North-Eastern Thailand. Hemoglobin E is also found in Sri Lanka, North Eastern India, Bangladesh, Pakistan, Nepal, Vietnam, Malaysia, the Philippines, Indonesia and Turkey.

Management
SE forms
Management is comparable to that of sickle cell form, although vasoocclusive symptomatology is reduced, except in rare situations (air travel) and pregnancy.

E β thalassemia forms
During childhood, regular follow-up of growth and facial deformities, hemoglobin level, prophylaxis of infections source of worsening of anemia with vaccines, treatment of potential infectious sites are essential. Daily oral penicillin, like in sickle cell anemia is recommended. Transfusions are indicated in case of bad tolerance of anemia and /or facial deformities. The main complication of regular transfusions is iron overload, source of hepatopathy, cardiac failure, endocrinopathies requiring thus iron chelation. The gold standard for chelation is deferoxamine (Desféral®), which is administrated subcutaneously daily during 10 hours. It should be started after about 20 units transfused, to maintain a ferritin level under 1500 mcg/l. Deferiprone is an oral chelator, less efficient than Desféral® and has side effects requiring often interruption of therapy: nausea, vomiting, agranulocytosis in 1,5% of cases (twice monthly minimal follow-up of hematological parameters is required). Oral chelator ICL 670 seems promising, with efficacy comparable to Desféral R, and few tolerable side effects. Results of phase III studies will be known in 2005. Splenectomy can diminish or suppress transfusion requirements and should not be performed before 5 years. When performed in adults, there is increased risk of portal thrombosis. Pregnancy requires regular antenatal care by obstetricians, in collaboration with hematologists. Transfusions are often required in the 3rd trimester in women with thalassemia intermedia, to reduce anemia and low weight births. Treatment of infections as well

of thrombosis prophylaxis are requested. Bone marrow transplantation has very rare indications. Recombinant erythropoietin alone or associated with hydroxyurea may be useful in reducing transfusions requirements, improve quality of life and diminish hemopoietic ectopic extra-medullary masses.

References

Weatherall DJ. Thalassemia: the long road from bedside to genome. Nat Rev genet 2004; 8:625-31.

Bain B. Hemoglobinopathy diagnosis. Blackwell Science, Oxford, 2001.