

# Hereditary Persistence of Fetal Hemoglobin (HPFH) 8 Mutations

## *DETECTS EIGHT COMMON BETA GLOBIN GENE CLUSTER DELETIONS RESULTING IN HPFH*

### Disease Overview

- Hemoglobin (Hb) is a tetrameric molecule that reversibly binds oxygen in red blood cells. It consists of two proteins expressed from the alpha globin gene cluster and two from the beta globin cluster. The expression of genes within these clusters is developmentally regulated and results in production of embryonic, fetal, and adult hemoglobin forms.
- By 6 months of age, a shift from gamma globin to beta globin (*HBB*) gene expression occurs, reducing the amount of fetal hemoglobin (Hb F;  $\alpha 2 \gamma 2$ ) produced so that the major form of hemoglobin present is Hb A ( $\alpha 2 \beta 2$ ).
- Although residual amounts of Hb F are produced throughout life, the majority of healthy adults have less than 1 percent Hb F.
- Hereditary persistence of fetal Hb (HPFH) results from mutations within the beta globin gene cluster that alter normal hemoglobin switching.
- Delta/beta thalassemia and HPFH are inherited conditions characterized by increased Hb F production; they are distinguished using hematologic and molecular analyses. Delta/beta thalassemia and some forms of HPFH are caused by deletions within the beta globin gene cluster.
  - Heterozygotes for delta/beta thalassemia deletions have moderate elevation of Hb F (5–20 percent) and hypochromic, microcytic anemia. Non-equal distribution of Hb F among red blood cells is observed (heterocellular HPFH).
  - Heterozygotes for HPFH-associated deletions typically have high levels of Hb F (up to 30 percent) with normal red blood cell indices. Equal distribution of Hb F among red blood cells is observed (pancellular HPFH).
- Elevation of Hb F in adults can occur due to acquired conditions (e.g., pregnancy, anemias, or leukemias).
- Elevated Hb F has no clinical significance in healthy individuals; however, HPFH can be beneficial in patients with sickle cell disease or beta thalassemia, as increased Hb F leads to milder phenotypes.
- HPFH has traditionally been diagnosed hematologically by the percentage of Hb F present and the distribution among red blood cells, but molecular diagnosis is most definitive.

### Epidemiology

- Eight deletions of varying size involving the beta globin gene cluster have been reported to result in pancellular HPFH.
  - HPFH-1 (African): described in Africans and African-Americans
  - HPFH-2 (Ghanaian): described in Africans and African-Americans

- HPFH-3 (Asian Indian)
- HPFH-4 (Southern Italian)
- HPFH-5 (Italian)
- HPFH-6 (Vietnamese)
- HPFH-7 (Kenyan)
- SEA-HPFH (Southeast Asian): identified in Cambodian, Vietnamese, and Chinese populations
- The incidence of HPFH in the general population is unknown but is more frequent in the populations described above. Together, HPFH-1 and HPFH-2 are found in ~0.1 percent of African-Americans in the southeastern United States.

### Genetics

- Two different molecular mechanisms can result in HPFH:
  - Deletional: HPFH results from specific large deletions in the beta globin gene cluster involving *HBB*.
  - Non-deletional: HPFH is caused by point mutations in the promoters of the gamma globin genes (*HBG1* and *HBG2*).
- The presence of an HPFH deletion may complicate the diagnosis of sickle cell disease or beta thalassemia, especially in infancy when the major form of Hb present is Hb F.
- The presence of an HPFH deletion can also mask beta thalassemia trait by ameliorating the hematological findings typically present.
- Other genetic modifiers of Hb F levels have been identified.

### Indications for Ordering

- Diagnostic testing in individuals with elevated levels of Hb F (relative to the individual's age).
- Carrier testing for individuals with a family history consistent with HPFH.
- For optimal test interpretation, please submit a Hemoglobinopathy/Thalassemia Patient History Form detailing clinical findings, family history, and ethnicity.

### Interpretation

- Negative: If none of the eight common deletions associated with HPFH was identified, HPFH has not been excluded, as point mutations, rare HPFH deletions, and delta/beta thalassemia deletions are not identified by this assay.
- Heterozygous: When one copy of a deletion associated with HPFH is identified, this predicts the persistent elevation of Hb F in all erythrocytes.
- Homozygous or Compound Heterozygous: If two deletions associated with HPFH are identified, individuals typically have Hb F levels approaching 100 percent and mild erythrocytosis.

## Methodology

- Multiplex PCR and gel electrophoresis to detect eight common deletions associated with HPFH:
  - HPFH-1 (g.5174452\_5259368del84917)
  - HPFH-2 (g.5180404\_5263982del83579)
  - HPFH-3 (g.5215683\_5265453del49771)
  - HPFH-4 (g.5217940\_5260078del42139)
  - HPFH-5 (g.5246023\_5258951del12929)
  - HPFH-6 (g.5193975\_5273259del79278)
  - HPFH-7 (g.5247860\_5270651del22792)
  - SEA-HPFH (g.5222878\_5250288del27411)
- Clinical sensitivity and specificity are unknown.
- Analytical sensitivity and specificity for the mutations tested is over 95 percent.

## Limitations

- Only the eight targeted deletions associated with HPFH will be detected. Point mutations or rare deletions that cause HPFH or delta/beta thalassemia will not be identified.
- Other genetic modifiers of Hb F levels will not be assessed.

- This test is unable to differentiate homozygosity for an HPFH deletion from compound heterozygosity for an HPFH deletion and a rare globin gene cluster deletion.
- Rare diagnostic errors can occur due to primer-site mutations.

## Related Test

Hemoglobin Evaluation with Reflex to Electrophoresis and/or RBC Solubility ([0050610](#))

## References

1. David J. Weatherall. Disorders of globin synthesis: the thalassemias. In *Williams Hematology*, 8th ed. Kaushansky K, et al, eds. 2010; New York, NY: McGraw Hill, 675–707.
2. Bhardwaj U and McCabe ERB. Multiplex-PCR assay for the deletions causing hereditary persistence of fetal hemoglobin. *Mol Diagn* 2005;9(3):151–6.
3. Patrinos GP, et al. Improvements in the HbVar database of human hemoglobin variants and thalassemia mutations for population and sequence variation studies. *Nucl Acids Res* 2004;32 (database issue): D537–41.

## Test Information

### 2005408 Hereditary Persistence of Fetal Hemoglobin (HPFH) 8 Mutations

For specific collection, transport, and testing information, refer to the ARUP website at [www.aruplab.com](http://www.aruplab.com).

For information on test selection, ordering, and interpretation, refer to ARUP Consult® at [www.arupconsult.com](http://www.arupconsult.com).

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