Alpha Thalassemia

Alpha (α) thalassemia is the most common inherited disorder of hemoglobin (Hb) worldwide and is caused by *HBA1* and *HBA2* gene variants. Decreased or absent synthesis of the hemoglobin (Hb) α chain results in clinical presentations ranging from asymptomatic silent carriers to severe anemia and fetal lethality. The two clinically significant forms of α thalassemia are Hb Bart hydrops fetalis syndrome and hemoglobin H (HbH) disease. α Thalassemia is found more often in certain ethnicities, including African, African American, Mediterranean, Middle Eastern, and Southeast Asian.

Disease Overview

Prevalence and/or Incidence

- Most common inherited disorder of Hb worldwide
- Carrier frequencies in high-risk populations:
  - African, African American: 1/3
  - Middle Eastern, Southeast Asian: 1/20
  - Mediterranean: 1/30-50
- Hb Bart hydrops fetalis syndrome and HbH disease are more frequent in Southeast Asian, Asian Indian, and Mediterranean populations than in African populations.

Symptoms

Phenotype	Associated Symptoms
α-Thalassemia silent carrier	Typically asymptomatic, though borderline anemia or mild microcytosis may be present Often misdiagnosed as iron deficiency Normal Hb electrophoresis
α-Thalassemia trait	Mild microcytic anemia may be present Often misdiagnosed as iron deficiency Normal Hb electrophoresis
HbH disease	Moderate to severe form of α thalassemia Moderate microcytic hypochromic anemia Hemolysis with Heinz bodies Splenomegaly Rare extramedullary hematopoiesis Propensity for acute hemolysis after oxidative stress, drug therapy, or infection
Hb Bart hydrops fetalis syndrome	Most severe form of α thalassemia Risk for fetus • Lethal in fetal or early neonatal period • Generalized edema, ascites, pleural and pericardial effusions • Severe hypochromic anemia • Usually detected on ultrasound at 22-28 weeks gestation Maternal complications during pregnancy • Preeclampsia • Polyhydramnios or oligohydramnios • Antepartum hemorrhage • Premature delivery

Tests to Consider

Alpha Thalassemia (HBA1 and HBA2) Deletion/Duplication with reflex to Hb Constant Spring 3003651

Method: Multiplex Ligation-dependent Probe Amplification with Sanger sequencing confirmation of HbCS

- Preferred first-tier genetic test for confirmation of suspected α thalassemia or α-thalassemia trait
- Detect common, rare, and novel deletions or duplications in the α-globin gene cluster and its HS-40 regulatory region
- Multiplex ligation-dependent probe amplification (MLPA) used to detect Hb Constant Spring (HBA2 c.427T>C; p.Ter143Gln); targeted Sanger sequencing is performed to assess HbCS copy number in absence of a concurrent HBA2 deletion

Alpha Globin (HBA1 and HBA2) Deletion/Duplication (Extended TAT as of 11/20/20-no referral available) 2011622

Method: Multiplex Ligation-dependent Probe Amplification

- First-tier genetic test for confirmation of suspected α-thalassemia or α-thalassemia trait
- Detect common, rare, and novel deletions or duplications in the α-globin gene cluster and its HS-40 regulatory region

Alpha Thalassemia (HBA1 and HBA2) 7 Deletions (Extended TAT as of 11/20/20-no referral available) 0051495

Method: Polymerase Chain Reaction/Gel Electrophoresis

- Acceptable first-tier genetic test for confirmation of suspected α-thalassemia or α-thalassemia trait
- Assesses for seven common deletions of *HBA1* and *HBA2* (-α3.7, -α4.2, -(α)20.5, --SEA, --MED-I, --FIL, and --THAI)

Alpha Globin (HBA1 and HBA2) Sequencing and Deletion/Duplication (Extended TAT as of 1/11/21-no referral available) 2011708

Method: Polymerase Chain Reaction/Sequencing./Multiplex Ligation-dependent Probe Amplification.

- Comprehensive genetic test for detection of α-thalassemia or α-thalassemia trait
- Detect deletional and nondeletional variants in *HBA1* and *HBA2*

Alpha Thalassemia (HBA1 and HBA2) Deletion/Duplication with reflex to Hb Constant Spring, Fetal 3003656

Method: Multiplex Ligation-dependent Probe Amplification with Sanger sequencing confirmation of HbCS

- Diagnostic testing for α thalassemia in fetus with suggestive clinical findings or at risk for

Pathophysiology

Typically, individuals have four functioning α-globin genes (αα/αα). Two genes, *HBA1* and *HBA2*, are present on each copy of chromosome 16, and α-globin chains function as subunits of fetal Hb (HbF: α2γ2) and adult Hb (HbA: α2β2). The number of α-globin genes deleted or inactivated correlates with different α-thalassemia phenotypes. Genotype/phenotype correlations in α thalassemia are complex and may be influenced by coinheritance of other Hb variants or α-globin gene duplications.

Phenotype	Genotype(s)
α-Thalassemia silent carrier	-α/αα
α-Thalassemia trait	-α/-α --/αα
HbH disease	--/-α
Hb Bart hydrops fetalis syndrome	--/--

α thalassemia due to familial HBA1/HBA2 deletions or Hb Constant Spring variant
- Detects common, rare, and novel deletions or duplications in the α-globin gene cluster and its HS-40 regulatory region
- MLPA used to detect Hb Constant Spring (HBA2 c.427T>C; p.Ter143Gln); targeted Sanger sequencing is performed to assess HbCS copy number in absence of a concurrent HBA2 deletion

See Related Tests

Genetics

Genes

HBA1 and *HBA2*

Inheritance

Autosomal recessive

Variants

- *HBA1* and *HBA2* large gene deletions account for approximately 90% of pathogenic α-thalassemia variants.
  - -α3.7 and -α4.2 deletions result in the deletion of a single gene.
  - -(α)20.5, --SEA, --MED-I, --FIL, and --THAI deletions result in the deletion of the *HBA1* and *HBA2* genes from the same chromosome
- Sequence variants and regulatory region variants occur mainly in *HBA2* and account for up to 15% of causative variants.
  - Nondeletional variants include:
    - Sequence variants that inactivate the gene
    - Small insertions/deletions
    - Variants that result in unstable α-globin protein (eg, Hb Constant Spring)
  - Nondeletional α-globin variants may be pathogenic or benign.
    - Both may result in an abnormal protein detectable by Hb evaluation.
    - Pathogenic nondeletional variants often have a more severe effect than single gene deletions.
- α-Globin gene duplication results in three or more active α-globin genes on a single chromosome.
  - Typically benign
  - May alter expected clinical phenotypes and hematological features when coinherited with beta (β) thalassemia

Test Interpretation

Sensitivity/Specificity

- Analytical sensitivity/specificity: 99% for both duplication/deletion analysis and sequencing
- Clinical sensitivity: most pathogenic *HBA1* and/or *HBA2* gene variants are large deletions not detectable by sequencing
  - Deletion: at least 90%, varies by ethnicity[1]
  - Sequencing: up to 15%, varies by ethnicity[1]

Results and Limitations		
Alpha Thalassemia (*HBA1* and *HBA2*) 7 Deletions	Alpha Globin (*HBA1* and *HBA2*) Deletion/Duplication	Alpha Globin (*HBA1* and *HBA2*) Sequencing

ATR-16, alpha-thalassemia-intellectual disability, chromosome 16-related; ATR-X, alpha thalassemia X-linked intellectual disability; n/a, not applicable

	Alpha Thalassemia (*HBA1* and *HBA2*) 7 Deletions	Alpha Globin (*HBA1* and *HBA2*) Deletion/Duplication	Alpha Globin (*HBA1* and *HBA2*) Sequencing
Negative result	No common α-globin gene deletions were detected • Risk for α thalassemia is reduced but not excluded	No large α-globin deletions or duplications were detected • Risk for α thalassemia is reduced but not excluded	No pathogenic variants were detected • Risk for α thalassemia is reduced • Large deletions of the α-globin genes, which account for the majority of variants, are not detected by sequencing
Positive result	Predicted genotype (-α/αα) • Individual is predicted to be a silent carrier Predicted genotype (-α/-α) or (--/αα) • Individual is predicted to have α-thalassemia trait Predicted genotype (--/-α) • Individual is predicted to be affected with HbH disease Predicted genotype (--/--) • Result is consistent with Hb Bart hydrops fetalis syndrome	Predicted genotype (-α/αα) • Individual is predicted to be a silent carrier Predicted genotype (-α/-α) or (--/αα) • Individual is predicted to have α-thalassemia trait Predicted genotype (--/-α) • Individual is predicted to be affected with HbH disease Predicted genotype (--/--) • Result is consistent with Hb Bart hydrops fetalis syndrome Predicted genotype (ααα/αα) • An extra functional α-globin gene present	1 pathogenic variant detected • Individual is predicted to be a silent carrier or carrier of α thalassemia • A more severe disorder is possible if another undetected α-globin variant is present 2 pathogenic variants detected • Individual is predicted to be a carrier of α thalassemia; mild microcytic anemia often present • Homozygosity or compound heterozygosity for nondeletional variants rarely results in HbH disease
Inconclusive result	n/a	Deletion or duplication of unknown clinical significance detected	Variant of unknown clinical significance detected
Limitations	Rare α-globin gene deletions, nondeletional variants, gene duplications and variants of the regulatory region will not be detected Diagnostic errors can occur due to rare sequence variations Rare syndromic or acquired forms of α thalassemia will not be detected	Breakpoints of large deletions/duplications will not be determined; therefore, it may not be possible to distinguish variants of similar size This assay does not assess for nondeletional variants within the coding or regulatory regions of the α-globin cluster genes Individuals carrying both a deletion and duplication within the α-globin gene cluster may appear to have a normal number of α-globin gene copies Rare syndromic or acquired forms of α-thalassemia associated with *ATRX* variants will not be detected Diagnostic errors can occur due to rare sequence variations	Large deletions/duplications and some variants of the regulatory regions will not be detected The phase of identified variants may not be determined Diagnostic errors can occur due to rare sequence variations Sequencing of both *HBA1* and *HBA2* may not be possible in individuals harboring large α-globin deletions on both alleles Rare syndromes associated with α thalassemia, such as ATR-X and ATR-16, will not be detected

ATR-16, alpha-thalassemia-intellectual disability, chromosome 16-related; ATR-X, alpha thalassemia X-linked intellectual disability; n/a, not applicable

References

1. Origa R, Moi P. Alpha-thalassemia. In: Adam MP, Ardinger HH, Pagon RA, et al, editors. GeneReviews, University of Washington; 1993-2020. [Last update: Dec 2016; Accessed: Jul 2020]

Additional Resources

Tan AS, Quah TC, Low PS, et al. A rapid and reliable 7-deletion multiplex polymerase chain reaction assay for alpha-thalassemia. Blood. 2001; 98 (1): 250-1. PubMed

Related Tests

Hemoglobin Evaluation with Reflex to Electrophoresis and/or RBC Solubility 0050610

Method: High Performance Liquid Chromatography/Electrophoresis/RBC Solubility

Hemoglobin Evaluation Reflexive Cascade 2005792

Method: High Performance Liquid Chromatography/Electrophoresis/RBC Solubility/Polymerase Chain Reaction/Fluorescence Resonance Energy Transfer/Sequencing

Familial Mutation, Targeted Sequencing 2001961

Method: Polymerase Chain Reaction/Sequencing

Familial Mutation, Targeted Sequencing, Fetal 2001980

Method: Polymerase Chain Reaction/Sequencing

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