Methylenetetrahydrofolate Reductase (MTHFR) 2 Variants

Indications for Ordering

- Determine genetic cause for hyperhomocysteinemia
- Predict sensitivity to antifolate drugs (eg, methotrexate)
- Testing not recommended for
  - Women who have recurrent pregnancy loss
  - Thrombophilia screening
  - Neural tube defect risk assessment

Test Description

- PCR followed by high-resolution melt analysis
- Hybridization probes detect
  - c.665C>T (p.Ala222Val) (previously designated c.677C>T)
  - c.1286A>C (p.Glu429Ala) (previously designated c.1298A>C)

Tests to Consider

Primary test
Methylenetetrahydrofolate Reductase (MTHFR) 2 Variants 0055655
- Determine genetic cause for hyperhomocysteinemia and potential sensitivity to antifolate drugs

Related tests
Homocysteine, Total 0099869
- Acceptable screening test for disorders of methionine metabolism (congenital hyperhomocysteinemia)
- Not recommended for risk assessment of cardiovascular disease or venous thromboembolism

Methotrexate, Sensitive 2005405
- Monitor methotrexate concentration

Thrombotic Risk, DNA Panel 0056200
- Acceptable panel to detect the 2 most common inherited thrombophilias (prothrombin related and factor V Leiden related)

Disease Overview

Prevalence
- Allelic frequency (based on data from dbSNP, February 2013)
  - c.665C>T
    - European Caucasians – 0.35
    - African Americans – 0.12
  - c.1286A>C
    - European Caucasians – 0.31
    - African Americans – 0.15
- Homozygosity for c.665C>T is 1-15% in the United States

Related conditions

Hyperhomocysteinemia
- Homozygosity for the c.665C>T variant is a risk factor for hyperhomocysteinemia
- Moderately elevated plasma homocysteine
  - May be risk factor for atherosclerotic vascular disease and venous thrombosis independent of MTHFR status
  - Affected by genetic and environmental factors (eg, dietary)
  - Folic acid supplementation reduces homocysteine levels, but effect on atherosclerosis and thrombotic risk is not clearly defined
  - Genetic testing is generally not useful for predicting risk for atherosclerosis

Thrombophilia
- Homozygosity for the c.665C>T variant and elevated homocysteine may be associated with a mild increase in risk for venous thromboembolism

Methotrexate sensitivity
- MTHFR gene variants may be associated with methotrexate toxicity, especially in individuals with
  - Family history of intolerance
  - Prolonged administration of methotrexate therapy (eg, ALL, CML, rheumatoid arthritis)
- Dose adjustment or discontinuation of therapy may be advised for individuals with high-risk MTHFR genotypes
Genetics

Gene – MTHFR

Inheritance – autosomal recessive

Structure/function – MTHFR gene variants (c.665C>T and c.1286A>C) correlate with reduced MTHFR enzyme activity
  • MTHFR enzyme is involved in folate metabolism
    o Catalyzes 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate
    o Necessary cofactor for the remethylation of homocysteine
    o Reduced enzyme function increases plasma homocysteine

Test Interpretation

Sensitivity/specificity
  • Clinical sensitivity – undefined; dependent upon multiple contributing factors
  • Analytical sensitivity/specificity – 99%

Results
  • Positive
    o Homozygosity for c.665C>T
      ▪ Associated with elevated plasma homocysteine levels and increased risk for premature cardiovascular disease
      ▪ At risk for toxicity from drugs (eg, methotrexate) that affect folate metabolism and may require dosing adjustments/discontinuation
    o Homozygosity for c.1286A>C
      ▪ Associated with intermediate enzyme activity, but not with increased plasma homocysteine levels
      ▪ No increased risk for premature cardiovascular disease/venous thrombosis
      ▪ May require lower dose requirements for methotrexate
    o Compound heterozygosity (c.665C>T/c.1286A>C)
      ▪ Associated with reduced enzyme activity, but not with increased plasma homocysteine levels
      ▪ No increased risk for premature cardiovascular disease/venous thrombosis
      ▪ May require lower dose requirements for methotrexate
    o Heterozygosity for either c.665C>T or c.1286A>C
      ▪ Associated with intermediate levels of enzyme activity, but not with increased plasma homocysteine levels
      ▪ No increased risk for premature cardiovascular disease/venous thrombosis
      ▪ May require lower dose requirements for methotrexate
  • Genotypes should be interpreted with clinical information
    o Specific genotype- or haplotype-based methotrexate dosing guidelines are not currently available
    ▪ Consultation with a clinical pharmacist is recommended
  • Negative – no variants detected
    o Neither of the common MTHFR gene variants tested, c.665C>T or c.1286A>C, was detected
    o Other causes of elevated homocysteine levels, coronary heart disease, or thrombosis are not assessed
    o Genotype is associated with a normal folate metabolism

Limitations
  • Only the 2 MTHFR gene variants (c.665C>T and c.1286A>C) will be targeted
  • Diagnostic errors can occur due to rare sequence variations

Reference