Maple Syrup Urine Disease, Type III (E3 Deficiency): *DLD* Gene Sequencing

<table>
<thead>
<tr>
<th>Test Code:</th>
<th>HI</th>
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<tbody>
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<td>Turnaround time:</td>
<td>4 weeks</td>
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<tr>
<td>CPT Codes:</td>
<td>81406 x1</td>
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**Condition Description**

The *DLD* gene (7q31-q32) encodes the protein dihydrolipoamide dehydrogenase, sometimes called E3. The DLD protein is a component of the pyruvate dehydrogenase complex, the alpha-ketoglutarate dehydrogenase complex, and the branched-chain alpha-keto acid dehydrogenase complex (BCKD), all of which are mitochondrial multienzyme complexes.

Mutations in *DLD* are responsible for a very rare variant form of the recessive condition maple syrup urine disease (MSUD). MSUD type III (or E3 deficient MSUD) initially presents in newborns with symptoms common to intermediate MSUD, but also includes severe lactic acidosis, which is potentially life-threatening. Persistent lactic acidosis occurs between eight weeks and six months of life. In addition to lactic acidosis, patients with E3-deficient MSUD can display neurologic deterioration, hypotonia, developmental delay, and movement disorders. Laboratory findings include elevated blood pyruvate, lactate, alpha-ketoglutarate, branched-chain amino acids, alpha-hydroxyisovalerate, and alpha-hydroxyglutarate. Dietary therapy, vitamin therapy with thiamine and biotin, and lipoic acid therapy have all been attempted without success.

Sequencing of the *DLD* gene is recommended after a biochemical diagnosis of E3-deficient MSUD, and provides a complementary method to confirm the presence of mutations in a proband, identify carriers among the proband's relatives, and provide prenatal diagnosis in families with known mutations.

For patients with mutations not identified by full gene sequencing, a separate deletion/duplication assay is available using a targeted CGH array (HJ).

**Genes**

*DLD*

**Indications**

This test is indicated for:

- Confirmation of a clinical/biochemical diagnosis of E3-deficient MSUD
- Carrier testing in adults with a family history of E3-deficient MSUD

**Methodology**

**Next Generation Sequencing:** In-solution hybridization of all coding exons is performed on the patient's genomic DNA. Although some deep intronic regions may also be analyzed, this assay is not meant to interrogate most promoter regions, deep intronic regions, or other regulatory elements, and does not detect single or multi-exon deletions or duplications. Direct sequencing of the captured regions is performed using next generation sequencing. The patient's gene sequences are then compared to a standard reference sequence. Potentially causative variants and areas of low coverage are Sanger-sequenced. Sequence variations are classified as pathogenic, likely pathogenic, benign, likely benign, or variants of unknown significance. Variants of unknown significance may require further studies of the patient and/or family members.

**Detection**

Clinical Sensitivity: Among 13 affected patients from 7 Ashkenazi Jewish families, mutations were identified in 12 of 14 DLD alleles. The other 2 alleles had a previously identified insertion mutation. Mutations in the promoter region, some mutations in the introns, and other regulatory element mutations cannot be detected by this analysis. Large deletions will not be detected by this analysis. Results of molecular analysis should be interpreted in the context of the patient's biochemical phenotype.

Analytical Sensitivity: ~99%

**Specimen Requirements**

Submit only 1 of the following specimen types

* Preferred specimen type: Whole Blood

**Type: Whole Blood**

Specimen Requirements:

- In EDTA (purple top) tube:
  - Infants (2 years): 3-5 ml
  - Older Children & Adults: 5-10 ml

Specimen Collection and Shipping: Refrigerate until time of shipment. Ship sample within 5 days of collection at room temperature with overnight delivery.

**Type: Saliva**
Specimen Requirements:

Oragene™ Saliva Collection kit (available through EGL) used according to manufacturer instructions.

Specimen Collection and Shipping: Store sample at room temperature. Ship sample within 5 days of collection at room temperature with overnight delivery.

Special Instructions

Submit copies of diagnostic biochemical test results with the sample. Contact the laboratory if further information is needed.

Related Tests

- Plasma Amino Acid Analysis (AA)
- Urine Organic Acid Analysis (OA)
- Custom Diagnostic Mutation Analysis (KM) is available to family members if mutations are identified by sequencing.
- Prenatal testing is available to couples who are confirmed carriers of mutations. Please contact the laboratory genetic counselor to discuss appropriate testing prior to collecting a prenatal specimen.