

# Inborn Errors of Metabolism Diagnostic Testing Program

## Description:

Our laboratory partners with our expert clinical and biochemical geneticists to offer comprehensive diagnostic testing for a number of inborn errors of metabolism (IEOM). Our panels are designed to assist in the diagnosis of patients with unexplained hypoglycemia and/or muscle weakness. Our testing includes the sequencing panels below. Single gene analysis is available for any gene on a panel, and targeted deletion/duplication is available for many genes. Additionally, we offer ExomeSeq, a clinical whole exome sequencing test for patients with complex phenotypes.

## Panels Offered:

- Metaboseq Gene Sequencing Panel (56 genes)
- Glycogen Storage Disease Gene Sequencing Panel (19 genes)
- Riboflavin Disorders Gene Sequencing Panel (5 genes)
- Elevated C16 Gene Sequencing Panel (2 genes)
- LCHAD/TFP Gene Sequencing Panel (2 genes)
- GSD type I Gene Sequencing Panel (2 genes)

## Indications:

### Panels:

- Confirmation of genetic diagnosis in a patient with unexplained hypoglycemia and/ or metabolic myopathy

### Gene Specific Sequencing:

- Confirmation of genetic diagnosis in a patient with IEOM and in whom a specific genetic diagnosis is suspected

### Deletion/duplication analysis:

- Completion of the diagnostic evaluation in a patient with IEOM who has had a negative NGS panel or who is heterozygous for a variant in a gene associated with an autosomal recessive condition

## Variant Specific Analysis:

- Carrier testing of parents and other relatives for recurrence risk assessment
- Pre-symptomatic testing of at-risk siblings and parents for medical management
- Prenatal diagnosis of an at-risk fetus, after confirmation of variant(s) in the parent(s) and by prior arrangement only

## Metaboseq Panel by NGS:

The Metaboseq panel is designed for patients with unexplained hypoglycemia, rhabdomyolysis, and other features of IEOM. It includes glycogen storage disorders, fatty acid oxidation disorders, and a number of other genes that can present in a similar manner.

### Genes:

*ACAD9, ACADM, ACADS, ACADVL, ACAT1, AGL, ALDOA, ALDOB, CPT1A, CPT2, DECR1, ENO3, ETFA, ETFB, ETFDH, FBP1, G6PC, GAA, GBE1, GLUD1, GYS1, GYS2, HADH, HADHA, HADHB, HMGCL, HSD17B10, LAMP2, LPIN1, MLYCD, MPI, NADK2, OXCT1, PC, PCK1, PCK2, PFKM, PGAM2, PGK1, PGM1, PHKA1, PHKA2, PHKB, PHKG2, PPARG, PRKAG2, PYGL, PYGM, SLC22A5, SLC25A20, SLC2A2, SLC37A4, SLC52A2, SLC52A3, TANGO2, TAZ*

### Indications:

- Unexplained hypoglycemia
- Rhabdomyolysis and skeletal myopathy
- Metabolic acidosis
- Hepatomegaly, liver dysfunction and cirrhosis
- Hypotonia, muscle cramps/ pain
- Cardiomyopathy/ arrhythmias
- Respiratory distress
- Hepatic encephalopathy
- Growth retardation
- Fatigue

## Glycogen Storage Disease (GSD)

### Panel by NGS:

Glycogen storage diseases (GSDs) result due to errors in glycogen metabolism leading to its improper storage in different tissues. The incidence of GSDs is about 1 in 20-25,000 and the most common types are I, II, III and IV. About 90% of patients with GSDs have types I through IV, however, types 0, VI and IX have mild symptoms and may be underdiagnosed. Symptoms vary based on the type of GSD, some GSDs primarily affect liver, whereas others affect muscles.

#### Genes:

*AGL, ALDOA, ENO3, G6PC, GAA, GBE1, GYS1, GYS2, PFKM, PGAM2, PHKA1, PHKA2, PHKB, PHKG2, PRKAG2, PYGL, PYGM, SLC2A2, SLC37A4*

#### Indications:

- Hypoglycemia
- Hepatomegaly, liver dysfunction and cirrhosis
- Hypotonia and muscle cramps
- Cardiomyopathy
- Respiratory distress
- Growth retardation
- Fatigue

## Riboflavin Disorders Panel by NGS:

Brown-Vialetto-Van Laere syndrome and multiple acyl-CoA dehydrogenase deficiency (MADD) can have overlapping clinical and biochemical presentations (elevated C4 and C5 acylcarnitine profile). Brown-Vialetto-Van Laere syndrome (also known as riboflavin transporter deficiency) is caused by pathogenic variants in *SLC52A2* and *SLC52A3*, and may present from infancy through adolescence with respiratory insufficiency, sensorineural deafness and ponto-bulbar palsy. MADD (also known as glutaric acidemia type II) is caused by pathogenic variants in *ETFA*, *ETFB*, and *ETFDH*. MADD is a disorder of fatty acid, amino acid, and choline metabolism. Patients can present with a severe neonatal form with metabolic acidosis, cardiomyopathy and liver disease or with a mild childhood/adult disease with episodic metabolic decompensation, muscle weakness and respiratory failure.

For both of these conditions, some patients may benefit from high-dose riboflavin supplementation.

#### Genes:

*ETFA, ETFB, ETFDH, SLC52A2, SLC52A3*

#### Indications:

- Metabolic acidosis
- Nonketotic hypoglycemia
- Hepatomegaly
- Elevated levels of amino acids and fatty acids in urine
- Hypotonia and muscle weakness
- Cardiomyopathy

## Elevated C16 Gene Sequencing Panel by Sanger:

Pathogenic variants in *CPT2* and *SLC25A20* cause fatty acid oxidation disorders that can present in a similar manner in infancy with hypoketotic hypoglycemia, hepatic dysfunction, cardiomyopathy, and other features. *CPT2* encodes carnitine palmitoyltransferase II (CPT II) and *SLC25A20* encodes carnitine-acylcarnitine translocase (CACT). Both of these are part of the carnitine cycle, which is necessary for transport of long-chain fatty acids from cytosol to mitochondria for beta-oxidation. Both CPT II deficiency and CACT deficiency can cause increased concentrations of 16-2 palmitoylcarnitine on acylcarnitine profile, making it difficult to differentiate between the two conditions. For both conditions, presentations at a later age with a milder phenotype have been reported.

#### Genes:

*CPT2, SLC25A20*

#### Indications:

- Hypoketotic hypoglycemia
- Hyperammonemia
- Rhabdomyolysis
- Hepatic dysfunction
- Cardiomyopathy/ arrhythmias
- Encephalopathy
- Elevated 16-2 palmitoylcarnitine on acylcarnitine profile

## LCHAD/TFP Gene Sequencing Panel by Sanger:

*HADHA* and *HADHB* encode the alpha (enoyl-CoA hydratase and long chain 3-hydroxyacyl-CoA dehydrogenase (LCHAD)) and beta (acetyl-CoA acetyl transferase) subunits of the mitochondrial trifunctional protein (TFP), a multienzyme complex of the fatty acid beta-oxidation cycle. Pathogenic variants in *HADHA* and *HADHB* can cause autosomal recessive LCHAD /TFP deficiency. LCHAD deficiency / TFP deficiency prevent the body from converting certain fats to energy, particularly during periods of fasting. Presentation begins in infancy or early childhood with hypoketotic hypoglycemia, metabolic acidosis, liver disease, hypotonia, and cardiomyopathy and arrhythmias. TFP deficiency can also present with neuropathy, and variants in *HADHB* can cause a milder phenotype including peripheral polyneuropathy, episodic rhabdomyolysis and pigmentary retinopathy.

### Genes:

*HADHA*, *HADHB*

### Indications:

- Hypoketotic hypoglycemia
- Metabolic acidosis
- Liver disease
- Hypotonia
- Cardiomyopathy and arrhythmias
- Neuropathy
- Rhabdomyolysis and skeletal myopathy

## Glycogen Storage Disease type I (GSD1) Gene Sequencing Panel by Sanger:

Glycogen Storage Disease type 1a and 1b (also known as von Gierke disease) can be clinically indistinguishable. Pathogenic variants in *G6PC* or *SLC37A4* lead to autosomal recessive glycogen storage disease types 1a or 1b, respectively (GSD I). *G6PC* encodes for glucose-6-phosphatase, which catalyzes the terminal step of gluconeogenesis and glycogenolysis. *SLC37A4* encodes for glucose-6-phosphate transporter into endoplasmic reticulum, where G6PC catalyzes the next step. The incidence of GSD I is 1 in 100,000. GSD I may have neonatal onset, but typically presents at 3-4 months

of age with hypoglycemia. Additional features can include hepatomegaly, renomegaly, growth retardation, osteopenia, round face, epistaxis, delayed puberty, and polycystic ovaries. GSD type 1b can also cause infections and inflammatory bowel disease due to neutropenia/neutrophil dysfunction.

### Genes:

*G6PC*, *SLC37A4*

### Indications:

- Hypoglycemia
- Hepatomegaly and renomegaly
- Elevated levels of lactate, fats and uric acid in blood
- Delayed growth and delayed puberty
- Bone thinning from osteoporosis
- Increased mouth ulcers and infections

## Specimen:

At least 3 mLs whole blood in a lavender top (EDTA) tube or saliva in an Oragene saliva kit. Please call 513-636-4474 for a free saliva collection kit.

## Testing Methodology:

**NGS Panels:** This test is performed by enrichment of the coding exons, flanking intronic and untranslated regions (5' and 3'), as well as known pathogenic variants (HGMD 2017.3) in the promoter and deep intronic regions of the genes specified above using oligonucleotide probe hybridization followed by next-generation sequencing with >50X coverage at every target base. All pathogenic and novel variants, as well as variants of unknown (indeterminate) significance, as determined bioinformatically, are confirmed by Sanger sequencing. Regions with <50X will be filled in by Sanger sequencing. A detailed non-coding variant list is available upon request.

**Gene Specific Sequencing:** PCR-based sequencing of the entire coding region and intron/exon boundaries of the specified gene.

**Deletion/duplication Analysis:** Copy number variant analysis of the gene by comparative genomic hybridization.

**Variant Specific Analysis:** Sanger sequencing following PCR amplification of the targeted variant(s) of the specified gene.

## Sensitivity and Limitations:

**Analytical Sensitivity:** The sensitivity of DNA sequencing is over 99% for the detection of nucleotide base changes, small deletions and insertions in the regions analyzed.

**Limitations:** Variants in regulatory regions and non-reported variants in untranslated regions may not be detected by this test. Large deletions/ duplications, large insertions and other complex genetic events will not be identified using sequencing methodology.

**Note:** Single gene sequencing is available for all genes on the panel. Deletion/duplication analysis is available for all genes listed except *DECRI*, *ENO3*, *GLUD1*, *GYS1*, *LPIN1*, *NADK2*, *OXCT1*, *PCK1*, *PCK2*, *PGAM2*, *PGM1*, *PHKA1*, *PYGL*, *SLC2A2*, *SLC52A2*, *SLC52A3*, *TANGO2*, and *PRKAG2*.

## Turn-Around Time:

Metaboseq panel, Glycogen Storage Diseases panel, and Riboflavin Disorders panel: 28 days

Elevated C16 gene sequencing panel, LCHAD/TFP gene sequencing panel, GSD type I gene sequencing panel: 7 days

## CPT Codes:

- Metaboseq Gene Sequencing Panel: 81443
- Glycogen Storage Diseases Gene Sequencing Panel: 81443
- Riboflavin Disorders Gene Sequencing Panel: 81479 x3
- Elevated C16 Gene Sequencing Panel: 81405
- LCHAD/TFP Gene Sequencing Panel: 81406
- GSD type I Gene Sequencing Panel: 81406
- Single gene testing, targeted variant analysis, and deletion/duplication analysis: call for information

Please call 1-866-450-4198 for current pricing, insurance preauthorization or with any billing questions.

## Results:

Results will be reported to the referring physician or health care provider as specified on the requisition form.

## Shipping Instructions:

Please enclose **test requisition** with sample.

**All information must be completed before sample can be processed.**

Place samples in styrofoam mailer and ship at room temperature by overnight Federal Express to arrive Monday through Friday.

## Ship to:

Cytogenetics and Molecular Genetics Laboratories  
3333 Burnet Avenue NRB 1042  
Cincinnati, OH 45229  
513-636-4474

## References:

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- Kanungo, S, Wells K, et al. (2018) Glycogen metabolism and glycogen storage disorders. Ann Transl Med. 6(24):474.*
- Longo, N, di San Filippo, CA, et al. (2006) Disorders of carnitine transport and the carnitine cycle. Am. J. Med. Genet. 142C: 77-85.*
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- Sun, B, Brooks, ED, et al. (2015) Preclinical Development of New Therapy for Glycogen Storage Diseases. Curr Gene Ther. 15(4):338-47.*
- Weinstein, DA, Steuerwald, U et al. (2018) Inborn Errors of Metabolism with Hypoglycemia: Glycogen Storage Diseases and Inherited Disorders of Gluconeogenesis. Pediatr Clin North Am. 65(2):247-265.*