PREVENTION GENETICS
DISEASE PREVENTION THROUGH GENETIC TESTING

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Myotilinopathy Testing via *MYOT* Gene Sequencing (Test #366)
Limb Girdle Muscular Dystrophy, Type 1A (LGMD1A)
Myotilin-Related Myofibrillar Myopathy

Brief Description of Clinical Features: Mutations in the myotilin gene (*MYOT*), also referred to as titin immunoglobulin domain protein gene (*TTID*), cause limb girdle muscular dystrophy type 1A (LGMD1A; OMIM 159000; Hauser et al. *Hum Mol Genet* 9:2141-2147, 2000) and myofibrillar myopathy (OMIM 609200; Selcen and Engel, *Neurology* 62:1363-1371, 2004). Patients with the LGMD phenotype exhibit distal weakness with mean age at onset of 27 years. Proximal weakness develops later in life, and approximately half of all patients exhibit a distinct nasal, dysarthric pattern of speech (Hauser at al. *Amer J Hum Genet* 71:1428-1432, 2002). CK levels are generally elevated 1.6 to 9 fold above normal range (Hauser et al. 2002). Myotilin-related MFM exhibits later onset (50 - 77 years) and has features of peripheral neuropathy with hyporeflexia, cardiomyopathy, and distal weakness greater than proximal weakness (Selcen and Engel, 2004). Stained with trichome, abnormal muscle fibers are seen containing hyaline structures and vacuoles containing membrane fragments from disintegrated sarcomeric Z disc and myofibrils (Selcen et al. *Brain* 127:439-451, 2004). With electron microscopy, affected muscle fibers reveal progressive degeneration of myofibrils beginning at the Z-disk. Immunohistochemical staining of the structurally abnormal fibers reveals abnormal expression and accumulation of several proteins, including myotilin, desmin, alpha-B crystalline, dystrophin and β-amyloid precursor protein (Selcen et al. 2004). Based upon studies of small cohorts, LGMD1A patients developed hyporeflexia later in the disease than myotilin-related MFM patients, thus tentatively differentiating these allelic disorders.

Genetics: Myotilinopathy is inherited in an autosomal dominant manner. The *MYOT* gene (OMIM 604103) encodes myotilin, a sarcomeric protein that binds actin either alone or in conjunction with alpha-actinin, an actin crosslinking protein. Thus, myotilin plays a key role in stabilization and anchorage of thin filaments, which may be a prerequisite for correct Z disc organization (Salmikangas et al. *Hum Mol Genet* 12:189-203, 2003). Nearly all *MYOT* mutations thus far reported result in amino acid substitutions in the serine rich exon 2. One missense mutation in exon 9 has been reported to be causative for LGMD1A (Shalby et al. *J Neuropath Exp Neurol* 68:701-707, 2009). Other genes involved with MFM include *DES*, *FLNC*, *LDB3*, *CRYAB*, and *BAG3*.

Description of This Particular Test: Myotilin is coded by exons 2-10 of the *MYOT* gene located on chromosome 5q31. Testing is accomplished by amplifying each coding exon and ~50 bp of adjacent noncoding sequence, then determining the nucleotide sequence using standard dideoxy sequencing methods and a capillary electrophoresis instrument.

Reference Sequences: **Genomic: NC_000005.9** **mRNA: NM_006790.2**
Protein: NP_006781.1 **mRNA and Protein: CCDS 4194.1**

Indication for Testing: Individuals with symptoms consistent with LGMD or myofibrillar myopathy and autosomal dominant inheritance. Symptomatic individuals with muscle biopsies demonstrating centrally located nuclei and fiber size variation.

Sensitivity of test: Myotilinopathy is a rare disorder (see for example, Moore et al. *J Neuropathol Exp Neurol.* 65:995-1003, 2006). Hauser et al. (*Am J Hum Genet* 71:1428–1432, 2002) screened 86 muscular dystrophy families, 44 of which demonstrated autosomal dominant inheritance, and found one with a causative *MYOT* mutation. The relative frequencies of mutations found in the Mayo Clinic MFM cohort was *LDB3* (14%), *MYOT* (13%), *DES* (8%), *FLNC* (4%), *BAG3* (4%), and *CRYAB* (3%) (Selcen and Engel, *GeneReviews* 2010).

Turnaround Time: Maximum of 40 days, although many tests are completed in 2 – 3 weeks.

Specimen Requirements: See page 4 of Requisition Form.

Price:	**Sequencing of *MYOT* Gene**		**Exons 2 – 10**		**$ 590**
CPT Codes:					
Sample Ascertainment	83890	$ 30	DNA Isolation	83891	$ 40
Amplification x 9	83898	$ 160	Sequencing x9	83904	$ 240
Separation	83894	$ 50	Interpretation/Report	83912	$ 70

Accreditation: CLIA ID:52D1027685 (expires 1/18/13) CAP ID:7185561, AU ID:1407125 (expires 12/20/12)
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