DIAGNOSTIC TESTS PERFORMED ROUTINELY OR AVAILABLE IF NEEDED

MUSCLE BIOPSY

A. Histological Evaluations

Routine stains and histochemical evaluation of sections are done on all specimens; additional Immunohistochemical or/and ultrastructural studies are done whenever needed for diagnostic evaluation.

1. Available stains on cross & longitudinal sections from paraffin tissue blocks

Histochemistry

Hematoxylin & Eosin (H&E), Masson's trichrome (MT), Verhoeff-Van Gieson (VVG), and Congo Red when amyloids are suspected.

Immunohistochemistry

Immune panel to diverse inflammatory myopathies: CD3, CD8, CD20, CD1a, CD10, CD68, CD45RO.

2. Available stains on cross & longitudinal sections from frozen tissue blocks

Histochemistry

Modified Gomori's trichrome (GT), Oil Red O (ORO), PAS (PAS +/- Diastase digestion) are run on all cases, while Phosphorylase, Phosphofructokinase , Myoadenylate Deaminase, Acid Phosphatase, Non-specific Esterase (NSE) and other useful histochemical stains are only run when specific diagnoses of glycogenosis, neurogenic myopathy or abnormal protein metabolism are suspected clinically; **Mitochondrial enzymes activity panel** (NADH-TR, COX, SDH, COX-SDH, COX-NADH, ATP synthase) are run on all cases as well as **Myosin ATPase** at pH 9.4 and 4.3 to assess differential proportions of myofiber types differentiation.

Immunohistochemistry

Antibodies to Dystrophy (congenital and adult forms) related proteins: Dystrophin (Rod, C and N terminals), Utrophin, Sarcoglycans (A, B, D & G), Dysferlin, Calpain-3, Caveolin-3, RYR1, SEPN1, Titin, Desmin, Myotilin, Lamin A/C, Emerin, Laminin, Merosin, Alpha and Beta Dystroglycans, FKRP, Collagen IV and VI, Actin, Alpha-Actinin.

Immune panel: MHCI, C5b-9, CD3, CD8, IgG, IgA, IgM, kappa & lambda light chains.

Morphometric evaluation of myofiber size, type and distribution in plastic embedded sections, for the differential diagnosis of congenital and acquired myopathies.

Electron Microscopy

Ultrastructural Study of Epon-embedded ultra-thin sections for the diagnosis of congenital storage, and metabolic diseases, and acquired infectious and degenerative processes.

B. Mitochondrial Biochemistry

Mitochondrial respiratory chain oxidative enzymes activities, and other non-oxidative mitochondrial enzymes activities are delineated. This study involves a detailed outline of the mitochondrial respiratory chain complexes (namely, CI, CII, CIII, CIV, CV, CI+III, CI+II) as well as the Citrate Synthase activity (reference for mitochondrial mass), and activities of other enzymes useful for interpretation of the observed changes (G3PDH, IDHs (NADP), and cytosolic LDH).

All above listed enzymatic activities are routinely assessed when an added request for this **Biochemical Study of Mitochondrial Activity** is received with the fresh muscle biopsy, using a routinely rapidly frozen portion of the muscle biopsy received. Such portion of the muscle biopsy is kept in deep freeze at -80°C to enable possible later requests for Biochemical study in the diagnosis of Mitochondrial Dysfunction, or Multiplex Immunoblotting in the evaluation of Muscular Dystrophies (as detailed below).

C. Muscle Dystrophy Multiplex Immunoblotting

This electrophoretic migration procedure may be requested to further assess the steady state level of 6 skeletal muscle proteins most commonly implicated in muscular dystrophies namely, Dystrophin, Dysferlin, Calpain-3, Caveolin-3, Alpha-Sarcoglycan and Beta-Dystroglycan.

D. Molecular Analysis

Molecular analysis may be performed on the patient's DNA sample, when requested, in order to explore panels of genes adapted to the clinicopathological context. MLPA genetic testing are also provided particularly for Duchenne/Becker muscular dystrophies and spinal muscular atrophies.

NERVE BIOPSY

A. Histological Evaluations

Routine stains and histochemical evaluation of sections are done on all specimens; additional Immunohistochemical or/and ultrastructural studies are done whenever needed for diagnostic evaluation.

1. Available stains on cross & longitudinal sections from paraffin tissue blocks

Routine stains and histochemistry: H&E, Masson's trichrome (MT), LFB-PAS (Myelin stain), Alcian blue, Crystal violet, Congo red, PAS+/-D, modified Gomori's trichrome (GT) and NSE.

Immunohistochemistry (when needed): CD3, CD8, CD20, CD68, and Transthyretin (TTR).

2. Available stains on cross & longitudinal sections from Frozen tissue blocks

Routine stains and histochemistry: H&E, Modified Gomori's trichrome (GT), LFB-PAS (Myelin stain), Alcian blue, Crystal violet, PAS+/-D, and NSE.

Immunohistochemistry (when needed): CD56, C5b9, CD3, CD8, CD20, CD68, IgM, IgA, IgG, Kappa and Lambda light chains, Transthyretin (TTR), CD1a, PGP 9.5.

Morphometric Analysis performed on toluidine-stained semi thin plastic sections.

Myelinated Fibers evaluation on teased nerve fibers slides.

Ultrustructural analysis of photographs taken from ultrathin plastic sections.