SAMPLE SUBMISSION FORM

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			CLINIC INFORMATION							PATIENT I	NFORMA [*]	ΓΙΟΝ
Veter	/eterinarian:							Owr	er:			
Clinic Name:							Pet:					
Clinic Account No.:							Pet ID No.:					
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[^]	900 Complete muscle profile (first muscle)			first muscle)	\$124.00	[V]		7.1	Dystrophy pa			\$290.00
	901	· ·	Each additional muscle or nerve					907.2	Dystrophy immunoblot			\$216.00
	902	Peripheral nerve profile only			\$66.00 \$124.00			7.3		I (CDs, MHCs)		\$290.00
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	905		ry Muscle Myos ne receptor an	\$110.00		91			ote special handling.			
	-	(Myastheni	ia Gravis) Note special ha					Price is per sample.)			\$66.00	
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907 Special immunohistochemistry				\$87.00			5.1		on and storage		\$71.00	
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SAMPLE SUBMISSION INSTRUCTIONS

900. Complete Muscle Profile

Useful links: How to take a muscle biopsy, What is the complete muscle profile?

The Complete Muscle Profile includes the histopathological and histochemical evaluation of a single muscle biopsy specimen utilizing 10-12 different stains and enzyme reactions including fiber typing. A biopsy approximately 0.5 X 0.5 X 1.0 cm should be taken by from an affected but not end-stage muscle by an open biopsy procedure. Wrap the muscle specimen in a saline dampened (not dripping, just moist) gauze sponge, place into a dry watertight container (5 or 10 ml red top tube, dry urine cup) and keep refrigerated until shipped to the laboratory on cold packs. Collect a second smaller biopsy and immersion fix in 10% neutral buffered formalin. For optimal results the specimens need to stay chilled and be received by the laboratory within 24-48 hours of collection.

- Do not ship specimens on Friday for Saturday delivery as the laboratory is closed.
- Please call with questions PRIOR to collecting biopsy specimens.
- Within the USA, Federal Express and UPS Red Label provide dependable overnight service and deliver directly to the laboratory. DO NOT USE USPS FOR SHIPPING BIOPSY SAMPLES. For international shipments Federal Express is the recommended courier.
- Please note the following University holidays for 2019. Do not ship packages for arrival during these days, as the laboratory will also be closed. January 1st, January 21st, February 18th, March 29th, May 27th, July 4th, September 2nd, November 11th, 28th & 29th, December 24th, 25th, & 31st

901. Each additional muscle or nerve

Additional charges apply if biopsies are submitted from more than 1 muscle or peripheral nerve specimen.

902. Peripheral Nerve Profile

Useful reference: Muscle and Nerve Biopsy. Vet Clinics North America (2002) 32: 63-102.

The Peripheral Nerve Profile includes evaluation of resin embedded 1 µm thick sections for determination of axonal degeneration, demyelination, and nerve regeneration. The fixed nerve specimens can also be further processed for ultrastructural analysis and teased nerve fibers on a case-by-case basis. Frozen nerve biopsy sections are used for immunohistochemical analysis on a case-by-case basis. A biopsy specimen approximately 2.5 cm (1 inch) in length should be divided with 2/3 placed on a tongue depressor to maintain length (do not stretch) and immersion fixed in 10% neutral buffered formalin. 1/3 of the biopsy specimen should also be placed a tongue depressor, wrapped in a saline dampened gauze sponge, placed into a watertight container, and kept chilled during shipping.

• Peripheral nerve biopsy should only be performed by a veterinarian experienced in this procedure

903. Muscle and Peripheral Nerve Profile

A special rate applies to a single muscle and peripheral nerve submitted together from the same case. Additional fees will apply if multiple muscles and nerves are submitted (901).

904. 2M Antibody Test (Masticatory Muscle Myositis)

Useful links: Commonly asked questions about masticatory muscle myositis and featured cases.

Useful reference: Masticatory Muscle Myositis: Pathogenesis, Diagnosis and Treatment. Comp Contin Ed Pract Vet 2004;26:590-605.

The 2M antibody test is a serum assay for the detection of autoantibodies against canine masticatory muscle type 2M fibers or proteins. A positive antibody titer is diagnostic of masticatory muscle myositis. Since this is an antibody based test, collect the serum sample PRIOR to initiation of corticosteroid or other immunosuppressive therapy. Submit 1-2 ml of serum via standard overnight or second day service using Federal Express or UPS. Packages shipped via second day service are delivered to a central receiving warehouse, sorted, and delivered by a university courier the following day to the laboratory.

905. Acetylcholine Receptor Antibody Test (Myasthenia Gravis)

Useful links: Commonly asked questions about acquired myasthenia gravis and featured cases.

The acetylcholine receptor (AChR) antibody test is a serum assay for the detection of autoantibodies against nicotinic AChRs at the neuromuscular junction. A positive antibody titer is diagnostic of acquired myasthenia gravis in both dogs and cats. Since this is an antibody based test, collect the serum sample PRIOR to initiation of corticosteroid or other immunosuppressive therapy. Submit 1-2 ml of serum via standard overnight or second day service using Federal Express or UPS. Packages shipped via second day service are delivered to a central receiving warehouse, sorted, and delivered by a university courier the following day to the laboratory.

906/911. Plasma Lactate and Pyruvate (NOTE: Special collection and handling required)

To obtain the most information, paired samples (collected at rest and following 10 minutes of strenuous exercise) for lactate and pyruvate determination should be submitted. These tests are indicated for detection of lactic acidemia and abnormalities of oxidative metabolism resulting in exercise intolerance.

<u>Lactate</u> – Plasma samples should be collected at rest and following 10 minutes of strenuous exercise. Collect blood samples in sodium fluoride/potassium oxalate (grey top) tubes at both time-points and label appropriately. Mix the samples well, centrifuge, then separate the plasma and place into labeled red top tubes. Send immediately to the laboratory by an overnight service on cold packs (if lactate alone) or on dry ice if paired with samples for determination of pyruvate concentrations.

Pyruvate – As pyruvate is unstable unless collected appropriately, please carefully follow instructions. Plasma samples should be collected at rest and following 10 minutes of strenuous exercise. Make up an 8% solution of perchloric acid (PCA, Perchloric acid 60% Fisher fA-228-6) by diluting 13.3 ml of 60% PCA to 100 ml of distilled water. This solution is stable refrigerated for 6 months. An 8% solution of trichloroacetic acid (TCA) can be substituted if PCA is not available. Immediately after blood collection, add exactly 2 ml of blood to 2 ml of 8% PCA or TCA. Mix and place on ice for 10 minutes to insure complete precipitation of protein. Centrifuge, pull off clear supernatant, place into separately labeled red top or plastic tubes and freeze. The frozen samples should be shipped on dry ice by an overnight service.

907. Special Immunohistochemistry

Useful links: Commonly asked questions and featured cases.

Immunohistochemical testing procedures are performed on fresh frozen muscle biopsy sections, usually following the complete muscle profile. Individual antibody staining can be chosen from the dystrophy panel (907.1). Staining for CD8+ T cells/MHC Class I can be performed to support a diagnosis of polymyositis.

907.1 Dystrophy Panel: This is an expanded immunohistochemistry panel for detection of many proteins that result in muscular dystrophy including but not limited to dystrophin, sarcoglycans, laminin α 2, dysferlin, α - and β - dystroglycans, utrophin and spectrin.

907.2 Dystrophy Immunoblotting: Protein extracts from submitted muscle biopsies are probed with various monoclonal and polyclonal antibodies against dystrophy associated proteins to detect an absence or abnormality in size or amount. This testing procedure further confirms the results of the dystrophy panel by immunohistochemistry

907.3. Myositis Panel: This panel is useful in characterizing cellular infiltrates in frozen muscle biopsy specimens from cases of myositis. Determination of B cell, T cell, and macrophage populations may aid in choosing specific therapies. In addition, up-regulation of Major Histocompability (MHC) antigens on muscle membranes can aid in the diagnosis of immune-mediated polymyositis when only minimal cellular infiltrations are present in the biopsy specimen.

907.4 Sarcolemmal Antibody: Sarcolemmal staining of muscle fibers using specific immunoreagents can be found in cases of immune-mediated myositis. Antibodies can be detected in the serum (2ml shipped by an overnight service) or directly within frozen muscle biopsy specimens (Vet Immunol Immunopath 2006:116;113;1-10).

907.5 Antistrial Antibody: Antibodies against muscle striational proteins can have diagnostic usefulness in cases with a cranial mediastinal mass and paraneoplastic associated muscle weakness from either myasthenia gravis or polymyositis. Identification of antistrial antibodies is thymoma associated. This serum test requires 2 ml of serum shipped by an overnight service. Useful link: What are antistrial antibodies and when is testing indicated?

915.1. DNA Purification and storage

Submit 5 ml of EDTA whole blood packaged with a cold pack and shipped by a service such as FedEx or UPS. **DO NOT USE USPS**. Ship samples between **Monday and Wednesday**.