Biopsy of the middle gluteal muscle of the horse

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When is muscle biopsy relevant?

- For diseases that lack scientifically validated genetic tests and in which abnormal findings are likely, including type 2 PSSM, and myofibrillar myopathy (MFM)
- For disorders causing atrophy if it helps to establish or confirm the cause, such as immune mediated myositis, and vitamin E deficient myopathy
- For research purposes including measurement of metabolites, enzymes, contractile behavior, genomics, metabolomics, fiber typing etc. Be aware that depth of sampling can affect some of these variables, such as fiber type.

Needle biopsy samples of muscles such as the middle gluteal are excellent if small samples are acceptable, if they can be processed appropriately (ideally frozen, not formalin) and where the horse needs to return rapidly to exercise or for repeated sampling.

Technique

- Drugs and equipment:
    - Local anesthetic, sedation if needed
    - Tail wrap
    - Sterile gloves
    - Sterile gauze
    - Clippers and scrub for skin preparation
    - Scalpel blade
    - Skin suture material
    - Forceps and needle holders are optional

- Muscle biopsy needle – this consists of 3 parts - a pointed trocar (outer diameter 6 mm) with a sampling window, a cutting cannula that fits inside the trocar, and a central stylet used after collection to remove sample (see image).
- Site - 1/3 to 1/2 of the way along a line drawn from the tuber coxa to the tail head (see image).
- Technique
    - Infiltrate the subcutaneous tissue (not muscle!) with 1 to 2 ml of local anesthetic in the region to biopsy
    - Make a stab incision through the skin, subcutaneous tissue and fascia using a 22 scalpel blade
    - Pass the biopsy needle (trocar and cutting cannula seated together, stylet removed) through the incision, perpendicular to the skin, and to a depth of 6 to 8 centimeters into the middle gluteal muscle.
    - Cut out the muscle sample. Have the cutting window facing forward or toward the last rib on the other side of the horse and apply pressure to push the tip forward to help improve sample volume during cutting. Repeatedly partially withdraw and replace the cutting cannula to open and close the sample window 4 to 6 times.

- Remove the needle from the muscle with the needle closed (the cutting cannula fully inserted) to keep the sample in the needle
- Remove the sample from the biopsy needle by pointing the tip upright, and removing the cutting cannula quickly. Then point the cannula down to a piece of tin foil and pass the stylet into it to express the sample of 50 to 200 mg of muscle.
- The needle can be reinserted through the same incision if more muscle is required. Ideally for histopathology and total half inch long sample is needed and may take two to three tries. If the sample looks very bloody it can be placed onto a piece of gauze but try to minimize handling as much as possible.
- The skin can be left open or closed with a single cruciate (straight cutting needle with nylon suture) or 2 to 3 staples. Sutures or staples can be removed in 7 days.
- Update tetanus toxoid if approaching due date in the patient
- This needle technique can also be used in semitendinosus, biceps femoris, triceps brachi, longissimus lumborum and potentially other reasonably sized muscles if only a small sample is required that can be processed quickly
- After sampling, the tissue can be processed by freezing for cryosectioning if the materials and equipment are immediately available to facilitate this (ideally the sample is frozen in isopentane chilled in liquid nitrogen, then stored at – 80c). Evaluation of frozen samples greatly expands the diagnostic capabilities and stains that can be applied.
- If specialized freezing is not immediately available you can roll the sample into the tin foil, place it in a cryovial and put the cryovial into a dry shipper.
- If samples cannot be frozen, wait 3 to 5 minutes after biopsy and then place the sample carefully into a cassette and into formalin. Leave the sample in formalin for 24 to 48 hours before embedding into paraffin blocks.
- Ideally contact the receiving laboratory or diagnostic service to be used in advance of performing any muscle biopsy to determine the samples and methods they recommend.