Exploration of Tongue Muscle Pathology in Canine Degenerative Myelopathy

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BACKGROUND

- Canine degenerative myelopathy (DM) is a neurodegenerative disease characterized by late onset and progressive degeneration of motor neurons.
- DM is divided into 4 stages based on severity:

<table>
<thead>
<tr>
<th>Stage</th>
<th>Neurologic Signs</th>
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<tr>
<td>Early</td>
<td>UMN Paraparesis</td>
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<tr>
<td></td>
<td>Asymmetric ataxia</td>
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<td></td>
<td>Intact reflexes</td>
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<td>Late</td>
<td>LMN Paraparesis</td>
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<td></td>
<td>Flaccid paralysis</td>
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<td>Reduced to absent sensory reflexes in pelvic limbs</td>
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<td>Urinary and fecal incontinence</td>
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- DM is a potential disease model to human amyotrophic lateral sclerosis (ALS)
  - Both often occur via a mutation in the superoxide dismutase 1 gene (SOD1)
  - Both cause progressive motor neuron degeneration, resulting in paralysis of muscles involved with limb movement, swallowing, vocalization, and respiration
- DM has previously been shown to cause alterations in muscle fiber composition, size and shape in intercostal muscles, but very little work has been done to determine if histologic changes occur in the orofacial muscles.
- Dysphagia and respiratory distress are two of the major clinical signs reported in ALS. Therefore, it is essential to study how canine DM impacts these vital biological functions.
- The genioglossus is the most affected oral muscle in ALS
  - Its degeneration impedes respiration and swallowing, and increases the likelihood of aspiration.

GENIOGLOSSUS

- Major dilator of the pharynx for inspiration
- Primary muscle for tongue protrusion during inspiration, swallowing & speech
- Composed of 2 major compartments: horizontal & oblique
  - Horizontal: ~2/3 type 1 fibers
  - Oblique: ~1/3 type 1 fibers
- Type 2 myofibers are fast twitch, and are therefore innervated by larger axons, which are more prone to degeneration in ALS
- Type 1 myofibers are slow twitch, and more resistant to degeneration

OBJECTIVE & HYPOTHESIS

The purpose of this study is to investigate the genioglossus of canine DM-affected dogs for evidence of muscle denervation, compared to age- and breed size-matched controls.

We hypothesize the genioglossus will display pathologic changes characteristic of neuromuscular degeneration, with degree of severity correlating to disease stage.

SIGNIFICANCE

- It is difficult to study ALS progression in humans, so a naturally-occurring animal disease model may aid in understanding specific pathogenic mechanisms for ALS. Dogs affected with DM are euthanized at various disease stages based on the pet owner’s decision.
- Donation of tissue from these dogs by the owners can help with research on canine DM and on human ALS.
- If the genioglossus of DM-affected dogs is found to have a similar pattern of degeneration as reported for human ALS, this research will provide additional support for canine DM as a disease model for human ALS.

METHODS

Tissue Processing & Slide Preparation:

- Biopsies were stored in fresh 10% NBF for 3-5 days before paraffin processing and embedding
- Each paraffin block contained 4 samples (2 horizontal & 2 oblique) from either the anterior or posterior region of the genioglossus.
- Paraffin blocks were cut via microtome into 10 μm thick sections and mounted onto silane-coated slides
- Slides were baked in 55°C for 2 hours before staining
- Staining methods are currently in process, including hematoxylin and eosin (H&E) and immunohistochemistry (IHC) targeting various proteins.

Tissue Examination:

- H&E stained slides are currently being examined by light microscopy to identify obvious signs of neuromuscular degeneration

PRELIMINARY RESULTS

H&E: general observations:

- Sections from DM-affected dogs appear to have more condensed myofiber bundles than controls.
- Vacuoles are present to some degree in all examined slides (DM-affected and control), but appear to be more abundant in samples from older dogs.
- No apparent nuclear internalization was found.
- Hyalin fibers were identified in all inspected slides, but were noticed more commonly in older and DM-affected dogs.

REFERENCES


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