

**Progress Report for the Pug Myelopathy Study
at Michigan State University College of Veterinary Medicine
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Study participants:

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Introduction

According to an ongoing survey of Pug owners and breeders, conducted by the Orthopedic Foundation for Animals, hind limb ataxia and weakness is the second most important health concern in the breed. In January 2013, a Journal of the American Veterinary Medical Association publication described 11 adult Pugs which developed progressive incoordination and weakness of the hind limbs resulting from a constriction of the spinal cord by fibrous tissue, at the level of the thoracolumbar junction. The condition, believed to be unique to Pugs, was called “constrictive myelopathy” (CM), and was associated with malformation of vertebral articulations (dysplasia of caudal articular processes) at the site, and presumed vertebral instability. Another condition, called “spinal arachnoid diverticulum” (SAD), associated with spinal cord compression by a fluid-filled pocket of cerebrospinal fluid (CSF) which often develops in association with intervertebral disc disease (IVDD) or other spinal problems, has been described in a variety of dog breeds, but results of a large study of 122 dogs, published in January 2014, indicated that the Pug was perhaps the most commonly affected breed. Since both CM and SAD are associated with thickening of the arachnoid and dura layers of the meninges which surround the spinal cord, it is possible that the 2 diseases may be related.

The **long-term objectives** of our study are to identify the underlying mechanisms (pathogenesis) behind CM and SAD, to assess the potential relationship of these 2 conditions, and to determine their potential relationship to vertebral articular malformations.

Specific aims:

1. Determine the prevalence of the described thoracolumbar vertebral malformation (hypoplasia/aplasia of caudal articular processes) by examination of imaging studies (x-rays and CT [computed tomography] scans) of Pugs seen at the Michigan State University Veterinary Medical Center (MSU VMC) for any reason since 2008.
2. Determine the prevalence of the various types of spinal cord diseases causing hind limb incoordination in Pugs (including not only CM and SAD, but also intervertebral disc disease [IVDD], degenerative myelopathy [DM], inflammatory conditions, neoplastic conditions [benign and malignant tumors], and trauma) through retrospective analysis of medical records, and prospective work-up of cases first seen at, or referred to MSU VMC.

3. Better describe the CT and MRI (magnetic resonance imaging) abnormalities in these diseases to potentially identify unique features, and improve the chances for early diagnosis and a more promising treatment outcome.
4. Analyze the chronology of neurologic signs associated with the various types of spinal cord disease in Pugs, to assess signs which are more common in some conditions than others, and compare survival times.
5. Thoroughly describe the pathologic abnormalities of CM and SAD by careful examination of biopsy and necropsy (postmortem) specimens, to better understand these conditions, their potential relationship to each other, and their potential relationships to the vertebral malformations.
6. In the future, we also plan to pursue genetic testing, to determine if there is indeed an inherited basis for CM and SAD in Pugs.

Work done to date

Clinical Diagnosis

- Thirty-three Pugs have been enrolled in our funded study since April 2014. Dogs qualify for the study if signs of hind limb ataxia and weakness can be localized to the T3-L3 (third thoracic to third lumbar) segment of the spinal cord via neurologic examination by a board-certified veterinary neurologist.
- Enrolled dogs undergo CT and MRI studies, under anesthesia, in order to examine the thoracolumbar vertebral column and spinal cord. These procedures are covered by research funds, and findings are interpreted by board-certified veterinary radiologists.
- Sites of vertebral caudal articular process dysplasia are determined from CT studies.
- Sites of spinal cord disease are determined by MRI studies, and types of disease are categorized as 1) IVDD only; 2) IVDD plus at least one other diagnosis; or 3) non-IVDD disease. Non-IVDD diagnoses may include SAD, CM, mass lesions, spinal cord atrophy, or “intramedullary hyperintensity,” which is interpreted as gliosis, edema, or myelitis.
- The clinical course of disease in each dog was analyzed for: 1) age of onset of signs of hind limb ataxia and weakness; 2) time interval between age of onset of signs and development of hind limb paralysis (paraplegia); 3) whether or not fecal incontinence and/or urinary incontinence developed over time; and 4) length of time between onset of signs and the dog’s natural death or euthanasia.
- Nine other Pugs with spinal cord disease referable to the T3-L3 segment had MRIs at MSU VMC between January 2010 and April 2014 and these data are included in our study.

Pathological Diagnosis

- Twelve of the 33 Pugs enrolled in the study have undergone postmortem examination (necropsy) following euthanasia or natural death.
- Four of the 33 enrolled Pugs were euthanized, but did not return to MSU for necropsy.
- 18 other Pugs with spinal cord disease, but not officially enrolled in our study, have also been studied at necropsy.

Summary of conclusions

1. MRI records from over 400 dogs seen at MSU VMC for spinal cord disease between January 2010 and May 2015 were examined. Thirty Pugs were included in this sample, including 17 dogs enrolled in our study. It was shown with statistical significance that among dogs with spinal cord disease, **SAD is a more frequently diagnosed in Pugs than in other breeds**. Also, **among dogs**

with IVDD, it was shown with statistical significance that Pugs more frequently than other breeds have more than one site of spinal cord compression due to disc protrusion or extrusion.

2. Radiographic and CT imaging studies which included the thoracic and lumbar vertebral column from Pugs seen at MSU VMC between November 2008 and June 2013, whether or not the dog was presented for neurologic disease, were examined. A total of 120 Pugs were included, and it was shown that greater than 27% of dogs had caudal articular process dysplasia between vertebrae T10 and T13. This portion of the vertebral column was most frequently affected, although at least one dog had dysplasia at each vertebra between T1 and L2.
3. No statistically significant correlation has been established between the site of vertebral caudal articular process dysplasia and the site of spinal cord disease. This may be due to the fact that we do not (yet) have enough dogs with spinal cord lesions at certain sites to perform statistical analysis.
4. For the 33 Pugs enrolled in our study, the **average age of onset of signs of hind limb ataxia and weakness is 9.1 years.**
5. **Fecal incontinence developed in 19 of the 33 Pugs** enrolled in the study, and **urinary incontinence developed in 6 dogs.** Four dogs became both fecal and urinary incontinent. Fecal incontinence developed, on average, 7.8 months after the onset of hind limb ataxia and weakness, and urinary incontinence also developed, on average, 7.8 months after onset of gait abnormalities.
6. **Six of the 33 dogs developed complete paralysis of the hind limbs**, on average 6 months after onset of hind limb ataxia and weakness.
7. There was no statistically significant correlation between the development of fecal incontinence, urinary incontinence, or paraplegia and the type of spinal cord disease diagnosis based on MRI findings.
8. Among 30 Pugs with spinal cord disease necropsied by Dr. Patterson, over half have fibrous thickening of the arachnoid layer of the meninges at one or more sites along the spinal cord, which is characteristic of CM and SAD. Among the 12 study dogs necropsied, only 3 had MRI findings compatible with SAD.
9. Using cheek swabs from Pugs confirmed by MRI and/or necropsy to have SAD, 2 candidate genes were tested for a possible association with the disease. The candidate genes were selected based on association with similar pathology to SAD in humans. One gene was definitively excluded as potentially associated, and one gene remains a possibility. More definite SAD cases are needed to further support or refute a genetic association for the latter gene. Testing of other candidate genes also is planned.

Data Presentation: 2015-2016

- A poster was presented by Dr. Ballegeer at the Annual Scientific Conference of the American College of Veterinary Radiology (ACVR) in Minneapolis, MN, October 7-10, 2015. The poster presented data on the vertebral anomalies of caudal articular processes in Pugs, and their possible association with spinal cord diseases.
- An oral presentation was given by Ms. Sarvenaz Bagheri, MSU CVM Class of 2018 veterinary student, at the annual MSU CVM Phi Zeta Research Day on October 2, 2015. Her talk featured the data on potential candidate genes associated with SAD in Pugs.
- A poster was presented by Mr. Michael Kluz, MSU CVM Class of 2018 veterinary student, at the national Meriel-NIH National Veterinary Scholars Symposium, Ohio State University, July 28-30, 2016. The poster presented clinical data regarding the chronologic course of disease, as well as

information on sites of vertebral caudal articular process dysplasia and MRI diagnoses. He will also give an oral presentation on this work at this year's MSU CVM Phi Zeta Research Day on October 7.

- A manuscript describing the pathology of SAD is in progress.

Funding

Michigan Animal Health Foundation (MAHF)

Pug Dog Club of America (PDCA)

Endowed funds from MSU College of Veterinary Medicine (including Detroit Kennel Club)

Communications regarding the Project

Dr. Smiler has established communication networks with Pug breeders, Pug rescue organizations, and veterinarians throughout Michigan and nationally.

Outreach has included:

- Development of a Facebook page (<https://www.facebook.com/pages/Pug-Dog-Health-Rear-AtaxiaParalysis/218123938233440>).
- Development of a website (<http://pugrearataxiaparalysis.com/>).
- Listing at the AVMA Clinical Studies page: https://ebusiness.avma.org/aahsd/study_search_detail.aspx?sid=312
- Postings on the Veterinary Information Network (VIN) that identify the study and our interest in communicating with other investigators.
- Postings in the Mid-Michigan Pug Club electronic newsletter.
- Development of a Questionnaire available online to collect information and medical records on affected Pugs.