Joint Pathology Center
Veterinary Pathology Services

WEDNESDAY SLIDE CONFERENCE 2023-2024

Conference #2 — 23 August 2023

CASE I:

Signalment:
2-year-old, female spayed Australian cattle dog, canine (*Canis lupus familiaris*)

History:
Unspecified clinical signs began at 11-12 months of age. An MRI performed at 21 months of age revealed brain atrophy. Clinical signs progressed to ataxia, anxiety, blindness, and aggression and the animal was euthanized at 23 months of age.

Gross Pathology:
The cerebral gyri are uniformly narrow and widely separated by deep, prominent sulci. The cerebellar folia in all lobes are more deeply incised than expected for a normal brain. There are no other significant findings.

Microscopic Description:
Cerebellar atrophy is manifest as slender folia that retain an anatomically normal branching pattern. This change is quantitatively similar throughout the cerebellum. At higher magnification, the molecular layer is narrow, and the internal granule layer is also thin and depleted of cell nuclei. Interestingly, the Purkinje cell layer is variably altered and the distance between cells is sometimes less than normal, possibly due to shrinkage in other layers. Neuronal cell bodies are multifocally swollen by variable amounts of

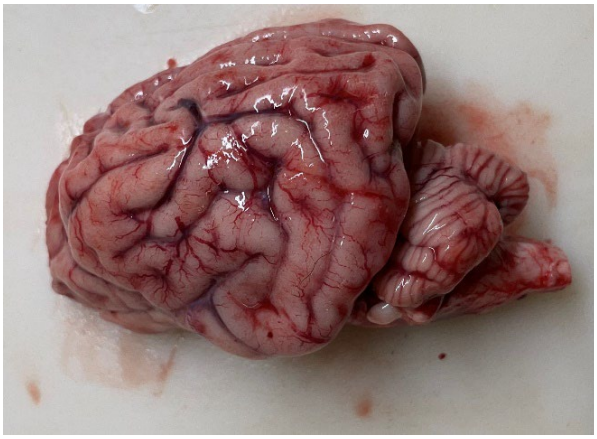

Figure 1-1. Cerebellum, dog. View of the dorsal cerebrum with the calvarium removed. The gyri are pinched and narrow, while the sulci are prominent. (Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, https://vmdl.missouri.edu/)

cytoplasmic, granular to globoid, lightly eosinophilic to brown pigment which occasionally peripheralizes the Nissl substance or nucleus. Rare individual neurons are brightly eosinophilic and shrunken with pyknotic nuclei. Sections stained with Luxol fast blue-PAS highlighted cytoplasmic inclusion bodies consistent with lipofuscin.

Contributor's Morphologic Diagnosis:
Brain: Cerebellar cortical atrophy with intracytoplasmic neuronal storage material consistent with lipofuscin.

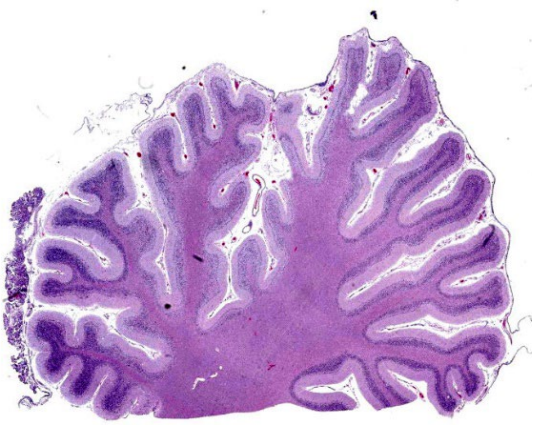

Figure 1-2. Cerebellum, dog. The cerebellar cortex is diffusely thinned across the entire section. (HE, 5X)

Contributor's Comment:
The neuronal ceroid lipofuscinoses (NCLs) are a group of inherited neurodegenerative diseases characterized by progressive decline following normal development. Characteristic of the group is accumulation of auto fluorescent pigment contained in lysosomes of neurons and other cell types.[11]

Dogs are useful models of pathogenesis for this group of diseases and can be used to test the efficacy of therapies for humans.[4] There are 13 canine sequence variants in 8 canine NCL orthologs of human NCLs that produce pathology similar to human diseases and various NCLs have been discovered in 20 different dog breeds.[4] The various subtypes of NCLs are referred to by the name of the particular mutated gene: "CLN," meaning ceroid lipofuscinosis, neuronal, followed by a number.

CLN types 1-4 are diseases of late infantile or juvenile onset, compared to CLNs 5-8 that have infantile onset.[11] There are 27 different mutations known in humans.[11] In CLN5 disease, affected dogs start showing clinical signs at around a year of age. Similar to this dog, the pathology in other breeds and humans consists of severe, generalized, progressive cerebral and cerebellar atrophy. The defective gene product is a soluble glycoprotein that is cleaved in the endoplasmic reticulum and is transported to lysosomes and cleaved by mannose-6-phosphate.[6,11] Intraneuronal storage of the subunit c of mitochondrial ATP synthase and Saponins A and D follows.[11] For all forms of CLN the end product is highly fluorescent and examination of unstained sections by fluorescence microscopy is the most sensitive form of detection.[4] Electron-dense ultrastructural deposits are similar but not identical between the different types of lipofuscinosis.[11]

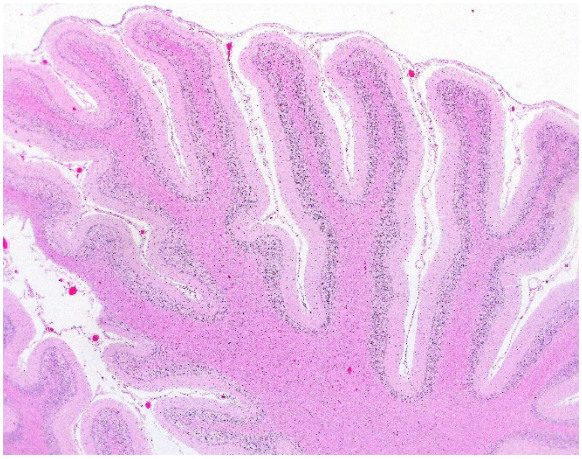

Figure 1-3. Cerebellum, dog. The midline cerebellar cortex demonstrates preservation of the architecture of the folia that are of diminished thickness. (HE, 40X) (Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, https://vmdl.missouri.edu/)

This patient was an Australian cattle dog that began showing neurological signs at 11-12 months of age. MRI imaging detected diffuse brain atrophy at 21 mos and the dog was euthanized at 23 mos. CLN5 in this breed of dog is the result of a homozygous C->T transition at position 30,574,637 on chromosome 22, reflected in the transcript (CLN5:c:619C>T). This change coverts a glutamine to a termination codon (p.Gln207TER).[5] An identical mutation has

been found in border collies and Laborador-Beagle mix dogs.[7,8,10] A second autosomal recessive mutation in the CLN5 gene (c.934_935delAG) causes disease in Golden Retrievers.[2] Purebred dogs often concentrate homozygous animals, leading to affected offspring. For instance, a high mutant allele frequency (34.8%) has been found in kennels of border collie dogs in Japan, with an allele carrier frequency of 8.1%.[7,8] Popular breeds may also develop more than one CLN mutation (e.g., CLN8 in Australian shepherd dogs and CLN12 in Australian cattle dogs).[3,9]

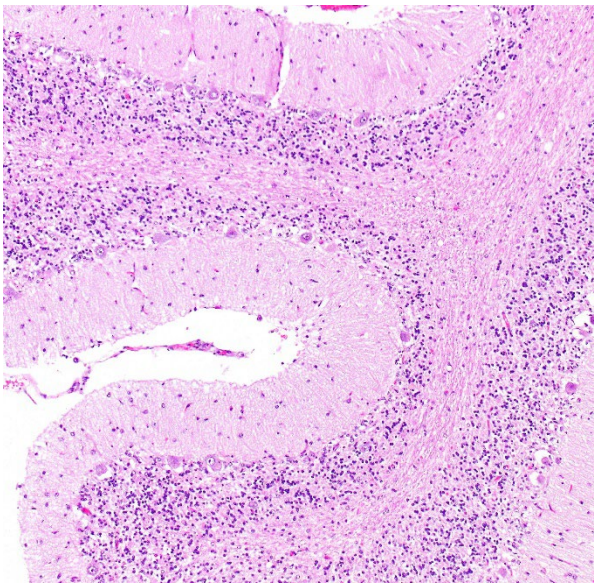

Figure 1-4. Cerebellum, dog. Higher magnification of the cortex shows relative preservation of Purkinje cells and reduced density of the internal granular layer. (HE, 100X) (Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, https://vmdl.missouri.edu/)

Increasingly, inborn errors of metabolism are being identified in mixed breed dogs in which neither parent is clinically affected. That several breeds may have a high incidence of heterozygosity and produce affected offspring may suggest that some locations are genetic hotspots in dogs. Finding a CLN5 affected Laborador-Beagle dog is presumably an example to this occurrence.[10] Recent studies of large populations of dogs suggest that some recessive mutations may occur at moderate frequencies in a number of different breeds so that inborn errors of metabolism are increasingly likely in mixed breed dogs.[1,12]

Contributing Institution:
University of Missouri
Veterinary Medical Diagnostic Lab
https://vmdl.missouri.edu/

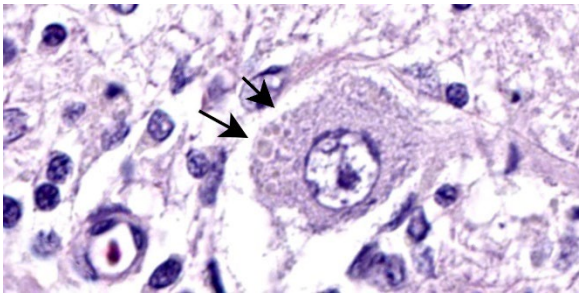

Figure 1-5. Cerebellum, dog. Purkinje cells contain large cytoplasmic vacuoles containing lipofuscin-like pigment. (HE, 1600X)

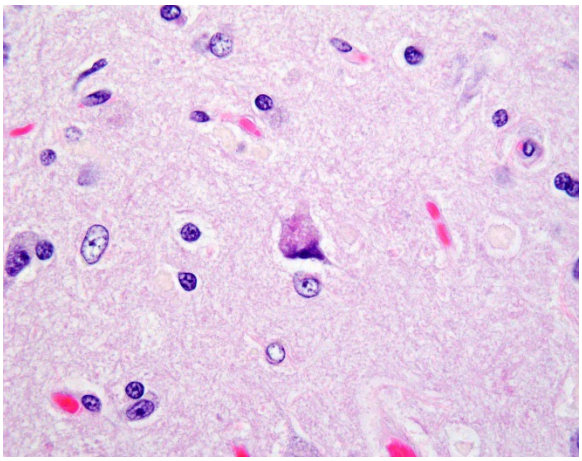

Figure 1-6. Cerebellum, dog. Occasional necrotic neurons with abundant lipofuscin-like pigment are encountered in the brainstem (as well as other sites). (HE, 100X)(Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, https://vmdl.missouri.edu/)

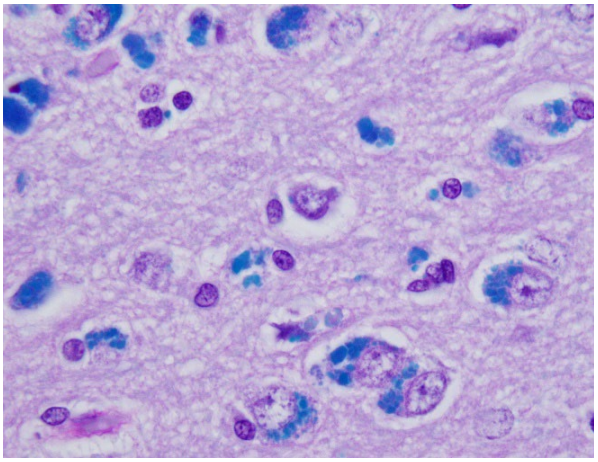

Figure 1-7. A combination Luxol fast-blue/Periodic ccid-Schiff stain demonstrates numerous lipofuscin-like granules in neurons. (LFB/PAS, 400X) (Photo courtesy of: Veterinary Medical Diagnostic Lab, University of Missouri, https://vmdl.missouri.edu/)

JPC Diagnosis:
Cerebellum: Cortical atrophy, diffuse, with neuronal ceroidosis.

JPC Comment:
The storage diseases that collectively comprise the neuronal ceroid lipofuscinoses are rather poorly named, as the storage material is neither exclusively ceroid nor lipofuscin. Instead, as the contributor notes, compounds such as subunit c of mitochondrial ATP synthase and sphingolipid activator proteins A and D may constitute the bulk of the retained material.[2]

Inherited neuronal ceroid lipofuscinoses have been described in a variety of cat, sheep, dog, and cattle breeds and are a heterogenous group of diseases. The various disease presentations reflect the diversity of genetic mutations that underly the disease entities, with most mutations occurring primarily in genes that code for lysosomal enzymes (ARSG, ATP13A2, CLN5, CTSD, PPT1, TPP1), endoplasmic reticulum proteins (CLN6), and endoplasmic reticulum-Golgi complex intermediate compartments (CLN8).[3] All mutations result in the inappropriate storage of proteins and lipofuscin-like lipopigments in multiple organs, but the most clinically significant damage occurs in the cerebral cortex, retina, and cerebellar Purkinje cells.[2] Lesions in these regions result in extensive cellular loss and atrophy with the concomitant clinical signs of dementia, blindness, and ataxia.[2]

Breed	Mutated Gene
Devon cattle	CLN5
English Setter	CLN8
Border Collie	CLN5
Australian Shepherd	CLN6
American Bulldog	CTSD
Staffordshire Terrier	ARSG
Tibetan Terrier	ATP13A2
Dachshund	PPT1
Miniature Dachshund	TPP1
Borderdale sheep	CLN5
South Hampshire sheep	CLN6

Table 1-1. Selected breeds with associated NCL-related mutations.

Disease presentation, even among species with the same gene mutation, can be subtly different. For instance, the disease presentation in Border Collies and a few other related dogs breeds are due, as the contributor discusses above, to mutation in the CLN5 gene. In these animals, there are gait and visual deficits with increasing aggression and dementia by 18-24 months of age, a case presentation that mirrors the clinical history in this case.[2] The resulting blindness is central in nature as retinal lesions are usually mild. The CLN5 gene deletion typically concentrates neuronal loss in the Purkinje cell layer and the limbic system, resulting in the ataxia and behavior changes that characterize this particular NCL.[2] By contrast, the mutated bovine CLN5 in Devon cattle causes blindness by 14 months of age due to se-

vere retinal atrophy but with only mild neuronal loss within the cerebrum and cerebellum.[2]

While the underlying pathogeneses of the NCLs are still largely unknown, diagnosis is relatively straightforward. NCL-associated storage granules are characteristically autofluorescent under ultraviolet light, have characteristic ultrastructural lamellar profiles, are PAS- and Luxol fast blue-positive, and are weakly acid fast.[2,12]

Conference discussion led by this week's moderator, MAJ Brittany Beavis, Chief of Molecular Pathology at the United States Army Medical Research Institute of Infectious Diseases, centered on the Purkinje cell layer, which appeared atrophic in some sections and relatively normal in others. Conference participants discussed how Purkinje cells can be subject to neurogenic atrophy due to loss of granule cells in addition to the direct damage caused by inappropriate retention of storage material. The variable ratio of these damaging inputs within the affected tissue could be one reason for the heterogeneity observed within the Purkinje cell layer and is illustrative of the interconnectivity among the cerebellar layers and among neural structures more generally.

References:

1. Donner J, Anderson H, Davison S, et al. Frequency and distribution of 152 genetic disease variants in over 100,000 mixed breed and purebred dogs. *PLoS Genet*. 2018;14(4):e1007361.
2. Cantile C, Youssef S. Nervous System. In: Maxie MG, ed. *Jubb, Kennedy and Palmer's Pathology of Domestic Animals*. Vol 1. 6th ed. Elsevier; 2016: 290-291.
3. Chalkley MD, Armien AG, Gilliam DH, et al. Characterization of neuronal ceroid-lipofuscinosis in 3 cats. *Vet Pathol*. 2014;51(4):796-804.
4. Gilliam D, Kolicheski A, Johnson GS, et al. Golden Retriever dogs with neuronal ceroid lipofuscinosis have a two-base-pair deletion and frameshift in CLN5. *Mol Genet Metab*. 2015;115(2-3):101-9.
5. Guo J, Johnson GS, Brown HA, et al. A CLN8 nonsense mutation in the whole genome sequence of a mixed breed dog with neuronal ceroid lipofuscinosis and Australian Shepherd ancestry. *Mol Genet Metab*. 2014;112(4):302-9.
6. Katz ML, Rustad E, Robinson GO, et al. Canine neuronal ceroid lipofuscinoses: Promising models for preclinical testing of therapeutic interventions. *Neurobiol Dis*. 2017;108:277-287.
7. Kolicheski A, Johnson GS, O'Brien DP, et al. Australian cattle dogs with neuronal ceroid lipofuscinosis are homozygous for a CLN5 nonsense mutation previously identified in border collies. *J Vet Intern Med*. 2016;30(4):1149-58.
8. Mamo A, Jules F, Dumaresq-Doiron K, et al. The role of ceroid lipofuscinosis neuronal protein 5 (CLN5) in endosomal sorting. *Molec Cell Biol*. 2012;32:1855-1866.
9. Mizukami K, Chang HS, Yabuki A, et al. Neuronal ceroid lipofuscinosis in border collie dogs in Japan: clinical and molecular epidemiological study (2000-2011). *ScientificWorldJournal*. 2012; 2012:383174.
10. Mizukami K, Chang HS, Yabuki A, et al. Novel rapid genotyping assays for neuronal ceroid lipofuscinosis in border collie dogs and high frequency of the mutant allele in Japan. *J Vet Diagn Invest.* 2011;23(6):1131-1139.
11. Schmutz I, Jagannathan V, Bartenschlager F, et al. ATP13A2 missense variant in Australian cattle dogs with late onset neuronal ceroid lipofuscinosis. *Mol Genet Metab*. 2019;127(1):95-106.

12. Vandevelde RJH, Oevermann A. *Veterinary Neuropathology: Essentials of Theory and Practice*. 1st ed. John Wiley & Sons, Ltd;2012:181.
13. Villani NA, Bullock G, Michaels JR, et al. A mixed breed dog with neuronal ceroid lipofuscinosis is homozygous for a CLN5 nonsense mutation previously identified in border collies and Australian cattle dogs. *Mol Genet Metab*. 2019; 127(1):107-115.
14. Wakley SU, Suzuki K, Suzuki, K. Chapter 6: Lysosomal Storage Diseases. In: Love S, Budka H, Ironside JW, eds. *Greenfield's Neuropathology*. CRC Press;2015:495-507.
15. Zierath S, Hughes AM, Fretwell N, et al. Frequency of five disease-causing genetic mutations in a large mixed-breed dog population (2011-2012). PLoS One. 2017;12(11): e0188543.

CASE II:

Signalment:
3-year-old, male neutered domestic short hair cat, feline (*Felis catus*)

History:
This animal was referred to University hospital with a 5-month history of progressive ataxia and a 5-day history of lethargy. Neurological abnormalities included obtundation and profound cerebellovestibular ataxia of all limbs. Proprioceptive positioning was diffusely delayed, hopping was markedly reduced to absent, and extensor postural thrust was absent. There was also positional vertical nystagmus. MRI scan revealed abnormal areas of hyperintensity in the cerebellum along with cerebellar herniation.

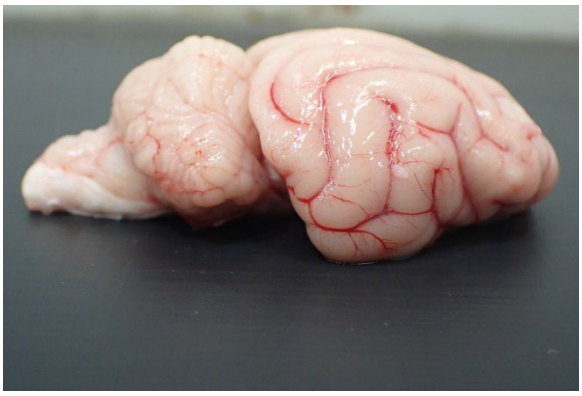

Figure 2-1. Brain, cat. The cerebellum is mildly enlarged, folia are flattened, and there is coning of the vermis. (Photo courtesy of: School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland, http://www.ucd.ie/vetmed/)

Gross Pathology:
The cerebellum is enlarged with 'coning' of the vermis. The cerebral gyri are flattened and widened. On sectioning, the cerebellar white matter was markedly expanded. Thoracic, abdominal, and pelvic organs are unremarkable.

Laboratory Results:
Serology for *Toxoplasma gondii* was negative.

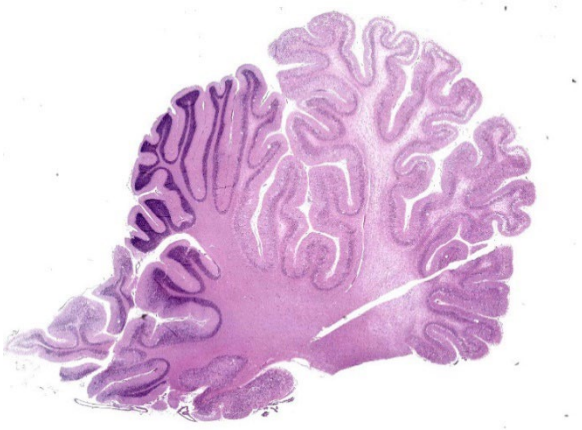

Figure 2-2. Cerebellum, cat. 80% of cerebellar folia are thickened with thinning of the granular cell layer and diminished staining of the granular cell layer within the affected region. There is coning of the posterior vermis. (HE, 6X)

Microscopic Description:
Numerous large dysplastic neurons up to approximately 60 µm in diameter populate the cerebellar Purkinje cell layer and extend into the molecular and granular layers. These pleomorphic cells are characterised by scant to abundant pale to brightly eosinophilic cytoplasm. Their nuclei are occasionally eccentrically located and up to approximately 30 µm in size varying from round to oval to reniform in shape with vesicular chromatin and a prominent nucleolus. In affected areas, the Purkinje cells are shrunken or lost and there is variable marked depletion of neurons in the granular layer. The neuropil is frequently rarefied or finely vacuolated (oedema) and contains numerous, variably-sized swollen axons (spheroids). These severe changes frequently alternate with sharply defined areas where the normal cerebellar architecture is maintained. The white matter is extensively rarefied and contains large numbers of gemistocytes. Examination with a Luxol fast blue stain confirmed marked loss of myelin in affected areas while no change in myelination was found in the cerebellar molecular layer. The meninges and perivascular spaces were multifocally infiltrated by low numbers of lymphocytes. Affected regions of cerebellum were immunohistochemically labeled for GFAP (astrocytes), NeuN (neuronal nuclei), NF clone 52 (axonal neurofilament H; heavy), and NF clone 312 (axonal panneurofilament). Immunohistochemical labeling of PTEN was also carried out to investigate the possibility of loss of PTEN protein expression as a result of PTEN gene mutation. There was consistent cell-specific labeling with each of these five antibodies. While immunolabeling with GFAP or NeuN was not found in dysplastic cells, axonal immunolabeling of NF52 and NF312 was maintained in their axons. Dysplastic neurons presented a heterogenous labeling pattern for PTEN, with the vast majority showing the absence of nuclear and reduced cytoplasmic immunolabeling. There were no abnormal findings in other brain regions.

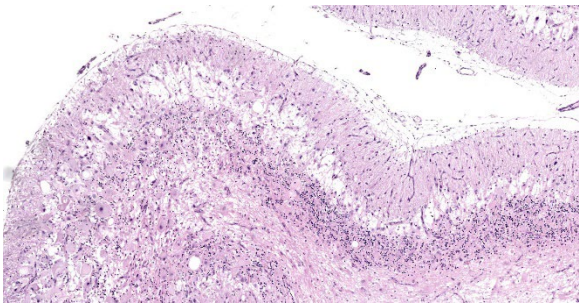

Figure 2-3. Cerebellum, cat. There is marked vacuolation of the molecular layer, loss of Purkinje cells, and marked hypocellularity of the granular cell layer. There is spongiosis of the folial white matter. (HE, 81X)

Contributor's Morphologic Diagnosis:

Brain: Dysplastic cerebellar gangliocytoma.

Contributor's Comment:
This is the first report of dysplastic gangliocytoma of the cerebellum in a cat that had presented with neurological deficits indicating cerebello-vestibular system disease. Dysplastic gangliocytoma of the cerebellum, commonly known as Lhermitte-Duclos disease, is an infrequent benign tumour described in human patients.[9] The neoplasm usually presents as a single unilateral discrete mass in the cerebellum, and bilateral involvement is very rare.[3,4,10,17] There is debate as to whether dysplastic gangliocytoma of the cerebellum represents a true neoplasm or a hamartoma.[14] Hamartomas are disorganised masses of normal to dysplastic cells which arise at sites where these cells are normally present.[8] The fact that these lesions frequently originate from clonal chromosomal aberrations makes their differentiation from benign tumours challenging and perhaps arbitrary. The World Health Organization (WHO) central nervous system confer-

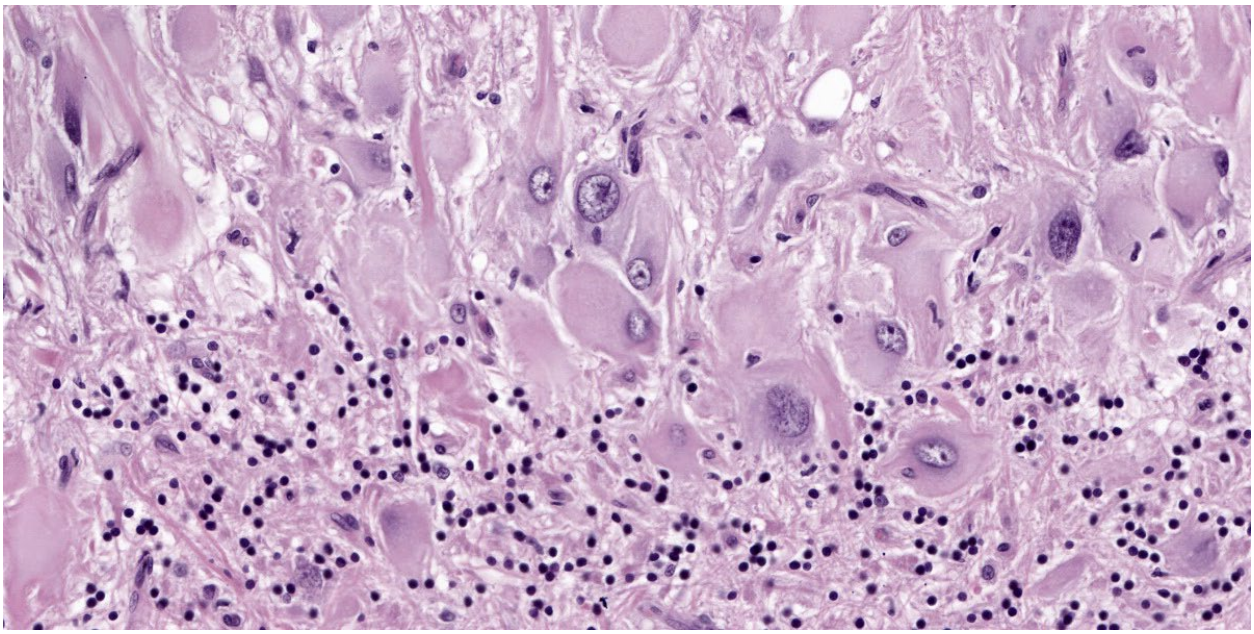

Figure 2-4. Cerebellum, cat. Affected regions of the cerebellar cortex are populated by pleomorphic ganglion cells. Cells range up to 30µm and have pleomorphic nuclei and occasionally prominent cytoplasmic vacuoles. (HE, 315X)

ence has graded dysplastic gangliocytoma of the cerebellum as a grade 1 neoplasia.[13] The classic macroscopic lesion is a unilateral enlargement of the cerebellum with maintenance of the folia.[3,10,17] Microscopic findings are variable and include replacement of the cerebellar granular layer by large, well-differentiated, dysplastic ganglion cells.[4] Dysplastic gangliocytoma of the cerebellum can occur sporadically, but has also been associated with Cowden syndrome, an autosomal dominant genetic disorder characterised by multiple hamartomas that leads to an increased risk of developing benign and malignant tumors. Cowden syndrome is associated with germline mutations of the phosphatase and tensin homolog (PTEN) gene, a tumour suppressor gene that leads to activation of the PI3K/AKT signaling pathway and uncontrolled cell proliferation.[8]

In human patients the lesion is defined by the following components: (1) variable replacement of the cerebellar internal granular layer by dysplastic ganglion cells, (2) abnormal myelinisation of the molecular layer, (3) reduced number of Purkinje cells, (4) large, bizarre neurons, and (5) vacuolisation of cerebellar white matter.[4] Each of these criteria were met in the current case except for abnormal myelinisation of the molecular layer. Interestingly, the current case posed a bilateral presentation, which is rarely described in human patients.[3,10,17] The mild inflammation, not considered to be of clinical significance, most likely occurred secondary to decreased cerebrospinal flow as a result of cerebellar enlargement and herniation.[5] Immunohistochemical labeling confirmed that the dysplastic cells were of neuronal origin; axonal neurofilament immunolabeling was present, although NeuN immunolabeling was absent. Similar results have been reported in gangliocytomas in human patients.[15]

Cowden syndrome is a multiple hamartoma syndrome associated with PTEN mutation in 80% of cases of dysplastic gangliocytoma of the cerebellum in human patients, and clini-

cal diagnosis is made by the presence of a combination of characteristic criteria including mucocutaneous lesions and a wide variety of benign and malignant tumors.[4] Major and minor diagnostic criteria for Cowden syndrome in humans include adult onset of dysplastic gangliocytoma of the cerebellum and benign lipomas.[6,10] A single case of Cowden-like syndrome has been described in the veterinary literature involving a Great Dane puppy that had colorectal hamartomatous polyposis, ganglioneuromatosis, and an associated PTEN mutation.[2] PTEN mutation resulting in downregulation/dysfunction has been reported and is suspected to contribute to tumorigenesis in cats and dogs.[12,16] In the current case, on IHC for PTEN mutations, the majority of dysplastic neurons showed loss of nuclear PTEN immunolabeling and reduced cytoplasmic immunolabeling. This may suggest that a PTEN mutation contributed to tumourigenesis in this case similar to what has been reported in Cowden syndrome. Although the 'gold standard' diagnosis of germline mutations like PTEN has been Sanger sequencing, studies have suggested the possibility of IHC detection of PTEN mutations as a superior approach as this technique also addresses potential epigenetic elements contributing to the loss of PTEN function.[7]

Contributing Institution:
University College Dublin
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JPC Diagnosis:
Cerebellum: Dysplastic gangliocytoma.

JPC Comment:
The uncontrolled growth that characterizes cancer is effected through two broad mechanisms: gain-of-function mutations in normal proteins that promote cell growth, or loss-of-function mutations that diminish the efficacy of cell growth inhibitors. The former mutations turn normal genes into oncogenes that drive cell proliferation via the production of constitutively active oncoproteins.[11] Mutations of the latter type occur in tumor suppressor genes, leading to failure of growth inhibition, a fundamental hallmark of carcinogenesis.[11]

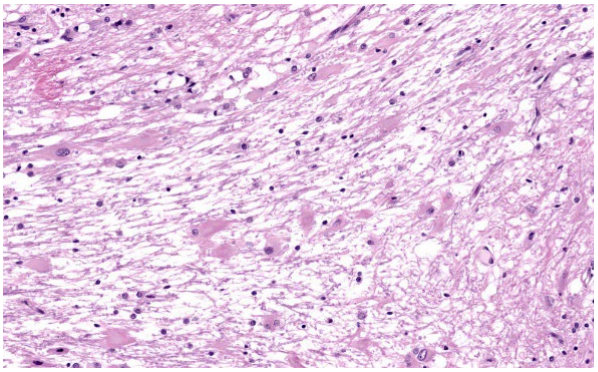

Figure 2-5. Cerebellum, cat. There is marked spongiosis of the cerebellar white matter and numerous gemistocytic astrocytes. (HE, 400X)

PTEN mutation, highlighted by the contributor as a critical factor in the development of many dysplastic gangliocytomas, is an example of a loss of function mutation in a tumor suppressor. PTEN is a participant in a canonical cell growth pathway that begins with a receptor tyrosine kinase (RTK). RTKs are transmembrane proteins with an extracellular growth factor binding domain and a cytoplasmic tyrosine kinase domain. In normal signal transduction, an extracellular growth factor binds the RTK's extracellular binding domain, transiently activating the RTK and causing it to dimerize and autophosphorylate tyrosine residues on its cytoplasmic tail. These phosphorylated tyrosine residues serve as binding and activation sites for cytoplasmic signaling molecules, most notably RAS.[11] In the activated state, RAS stimulates two downstream signaling cascades: the MAPK cascade and the PI3K/AKT pathway. The common endpoint

of both pathways is the activation or production of transcription factors that move to the nucleus and increase production of proteins that drive progression through the cell cycle. Many proteins within these pathways are encoded by proto-oncogenes; in fact, gain of function point mutations in RAS that cause constant activation of downstream, pro-growth pathways are the most common proto-oncogene abnormalities in human tumors.[11]

In this exuberant signaling milieu, PTEN's function is to inhibit PI3K, the first of a series of serine/threonine kinases in the PI3K/AKT pro-growth pathway. Regulation at this stage is critical as AKT, the next kinase in the signaling cascade, phosphorylates more than 150 proteins involved in regulating protein synthesis and apoptosis.[11] If PTEN acquires a loss of function mutation, as is the case in up to 80% of dysplastic cerebellar gangliocytomas, this cellular brake is lost, leading to unchecked, pro-growth downstream signaling and a predisposition to tumor development.[9]

The contributor alludes to the debate over the classification of this entity. A hamartoma, as stated above, is a non-neoplastic growth characterized by an abnormal growth of cells in their normal anatomic location. Some sources distinguish hamartomas from neoplasia by pathogenesis; hamartoma is usually the result of a systemic genetic abnormality while neoplasia tends to be monoclonal since it originates from a single transformed cell.[1] In this case, the eye-catching dysplastic neurons are in their native location and possibly arise from a germline mutation in PTEN, making classification as a hamartoma seem appropriate. As noted above, however, the human lesion is described by the WHO as a grade 1 neoplasm, and the literature includes a variety of terminology, including the term "hamartomatous neoplasm," which provides little clarity.

Conference participants engaged this debate without reaching general consensus, though most participants felt this entity more likely represented a hamartoma. No matter the terminology, the biological behavior of this entity is benign, though significant morbidity can accompany its growth within the confines of the skull.

References:

1. Batsakis JG. Nomenclature of developmental tumors. *Ann Otol Rhino Laryngol.* 1984;93(1):98-99.
2. Bemelmans I, Küry S, Albaric O, et al. Colorectal hamartomatous polyposis and ganglioneuromatosis in a dog. *Vet Pathol*. 2011;48(5):1012–1015.
3. Borni M, Kammoun B, Kolsi F, et al. The Lhermitte-Duclos disease: a rare-bilateral cerebellar location of a rare pathology. *Pan Afr Med J*. 2019;33:118.
4. Brat DJ, Perry A. *Pattern Recognition Series: Practical Surgical Neuropathology: A Diagnostic Approach*. 2nd ed. Elsevier; 2018.
5. Deren KE, Packer M, Forsyth J, et al. Reactive astrocytosis, microgliosis and inflammation in rats with neonatal hydrocephalus. *Exp Neurol*. 2010;226(1): 110–119.
6. Derrey S, Proust F, Debono B, et al. Association between Cowden syndrome and Lhermitte-Duclos disease. *Surg Neurol*. 2004;61(5):447–454.
7. Djordjevic B, Hennessy BT, Li J, et al. Clinical assessment of PTEN loss in endometrial carcinoma: immunohistochemistry outperforms gene sequencing. *Mod Pathol*. 2012;25(5):699–708.
8. Hargis AM, Myers S. The Integument. In: Zachary JF, ed. *Pathologic Basis of Veterinary Disease*. 6th ed. Elsevier; 2017:1009.

9. Joo G, Doumanian J. Radiographic findings of dysplastic cerebellar gangliocytoma (Lhermitte-Duclos Disease) in a woman with Cowden syndrome: a case study and literature review. *J Radiol Case Rep*. 2020;14(3):1-6.
10. Khandpur U, Huntoon K, Smith-Cohn M, et al. Bilateral recurrent dysplastic cerebellar gangliocytoma (Lhermitte-Duclos disease) in Cowden syndrome: a case report and literature review. *World Neurosurg*. 2019;127:319-325.
11. Kumar V, Abbas AK, Aster JC. Neoplasia. In: *Pathologic Basis of Veterinary Disease*. 10th ed. Elsevier; 2022: 286-292.
12. Levine RA, Forest T, Smith C. Tumor suppressor PTEN is mutated in canine osteosarcoma cell lines and tumors. *Vet Pathol*. 2002;39(3):372–378.
13. Louis DN, Perry A, Reifenberger G, et al. The 2016 World Health Organization classification of tumors of the central nervous system: a summary. *Acta Neuropathol*. 2016;131(6):803-820.
14. Nowak DA, Trost HA. Lhermitte-Duclos disease (dysplastic cerebellar gangliocytoma): a malformation, hamartoma or neoplasm? *Acta Neurol Scand*. 2002; 105(3):137–145.
15. Rainov NG, Holzhausen H-J, Burkert W. Dysplastic gangliocytoma of the cerebellum (Lhermitte-Duclos disease). *Clin Neurol Neurosurg*. 1995; 97(2):175 -180.
16. Ressel L, Millanta F, Caleri E, et al. Reduced PTEN protein expression and its prognostic implications in canine and feline mammary tumors. *Vet Pathol*. 2009;46(5):860–868.
17. Zak M, Ledbetter M, Maertens P. Infantile Lhermitte-Duclos disease treated successfully with rapamycin. *J Child Neurol*. 2017;32(3):322–326.

CASE III:

Signalment:
11-month-old, female spayed Labrador Retriever dog, canine (*Canis lupus familiaris*)

History:
The dog submitted to the CHUV (Centre Hospitalier Universitaire Vétérinaire), Faculté de Médecine Vétérinaire, University of Montreal, because of recurring respiratory difficulties (panting with its mouth closed) along with a head tilt and turning to the right. Clinical signs had been ongoing for over 9 months. Clinical signs were still present after therapeutic trials with different immunosuppressive drugs. Humane euthanasia was performed at home by the family veterinarian due to the guarded prognosis.

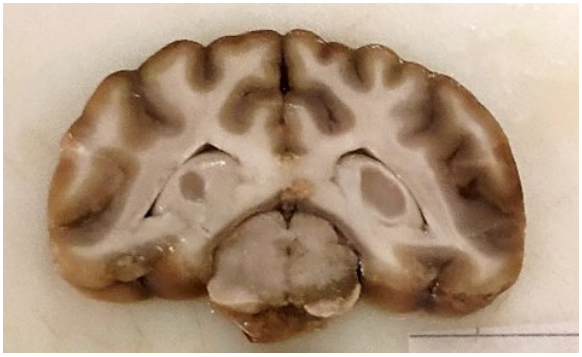

Figure 3-1. Brainstem, dog. There is an area of grey discoloration in the right portion of brain stem which extends slightly to the left of the midline. (Photo courtesy of: Centre de Diagnostic Vétérinaire de l'Université de Montréal, Faculté de Médecine Vétérinaire, Université de Montréal)

Gross Pathology:
This 24.5 kg Labrador Retriever dog is submitted frozen for necropsy examination and is in a suboptimal state of conservation following thawing. The animal has adequate muscle mass and body fat stores. The right portion of the cerebral trunk displays a 3x1x1cm area of greyish discoloration which extends slightly to the left of the mid-

line. The overlying leptomeninges have a dark discoloration. The spinal cord is unremarkable grossly. The heart weighs 221 g (0.9% of BW, within normal limits). All other internal organs are normal grossly.

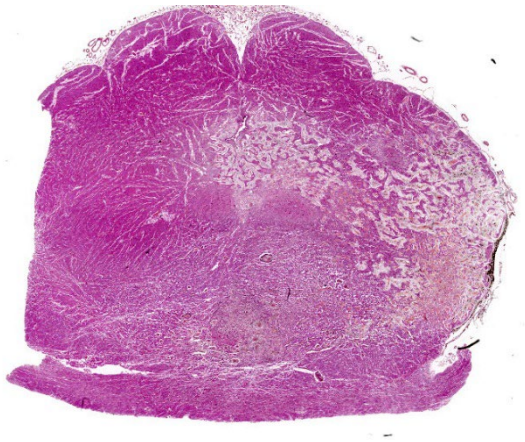

Figure 3-2. Brainstem, dog. A network of haphazardly arranged fibrovascular bundles infiltrates the brainstem. (HE, 6X)

Microscopic Description:
Brain: The observed gross changes in the area of the brain stem correspond to a poorly cellular and poorly delineated process that is continuous with the leptomeninges and multifocally infiltrates the neuroparenchyma. These changes are characterized by anastomosing fronds of spindle to stellate cells surrounded by a mostly loosely arranged fibrous stroma of variable density. This stroma is usually centered around numerous blood vessels, mostly small arterioles. The fusiform cells have a pale acidophilic cytoplasm that is finely fibrillar and moderately well delineated. The nucleus varies from elongated with rounded ends to round with a finely granular chromatin with no apparent nucleolus. Anisocytosis and anisokaryosis are minimal, atypia is unremarkable and no mitoses are observed. These spindle cells do not stain with GFAP immunohistochemistry staining. The lining of blood vessels located within that stroma is positive for Factor VIII. The leptomeninges overlying these proliferative changes are mildly to moderately thickened multifocally in some areas by similar proliferating spindle cells and contain multifocal and superficially numerous melanin-laden cells (melanocytes). In the adjacent neuroparenchyma, few neurons displaying chromatolysis and rare spheroids are observed.

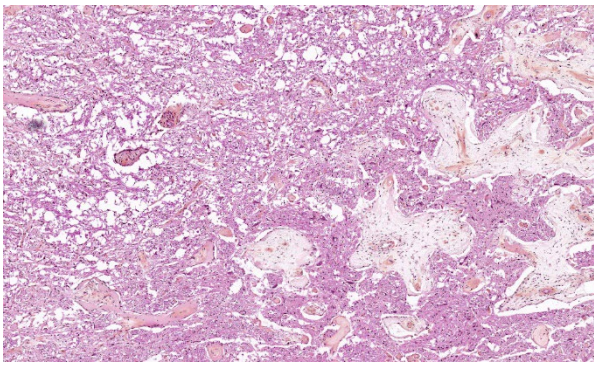

Figure 3-3. Brainstem, dog. The ingrowing bands of fibrous connective tissue contain entrapped thick-walled arterioles. (HE, 52X)

Contributor's Morphologic Diagnosis:
Brain stem: Cerebral meningioangiomatosis

Contributor's Comment:
Meningioangiomatosis (MA) is, in animals at least, a rare benign lesion, best regarded as a malformation or hamartoma producing circumscribed plaques on the surface of the brain stem and cervical spinal cord.[3] Hamartomas are mass lesions characterized by disorderly overgrowth of tissue elements. MA is seen most commonly in young dogs. Blood vessels appear in excess in these lesions and are cuffed by proliferating cells considered meningothelial in origin. The lesion usually does not extend into the underlying neural substance, which shows mixed degeneration and reactive changes, but can occasionally grow deeper along perivascular spaces.[3,19]

In veterinary medicine, this unique lesion has been described in a few dogs, one cat, one horse with associated mushroom toxicity, one cow that was part of a neuropatholo-

gy BSE survey in Scotland, and in a CD-1 mouse. [1,2,4,10,15,17,21,22,25] This lesion described by Corbett et al in a 13-y-old cat was located in the leptomeninges and outer neuroparenchyma of the right pyriform and temporal telencephalic lobes, with extensive hemorrhage.[4]

Human MA has been described as coexisting with meningiomas, arteriovenous malformations, oligodendrogliomas, meningeal hemangiopericytomas and orbital erosions.[7] An association with another nervous lesion has also been described in a 3-month-old German shepherd dog with a thalamic astrocytic hamartoma with tectal meningioangiomatosis, and in a 4-y-old Labrador retriever dog with a fibrous meningioma.[11,23]

MA typically occurs in the brain stem and cervical spinal cord of young dogs. An imaging study documented thoracolumbar spinal cord involvement in two dogs (4-y-old male Boxer and 5-month-old female Labrador), an unusual site for this condition.[12] In a 5-year-old dog, a focus of MA located in the caudal thoracolumbar spinal cord and associated with abnormal hind limb gait was successfully excised and resulted in improvement of clinical signs with a good long-term prognosis.[6] This benign lesion could then be curable with surgical resection depending on accessibility of the location, as in humans.[9]

In humans, the sporadic form of MA often results in headache and epilepsy. Focal cortical dysplasia has been described adjacent to foci of such lesions following surgical resection in cases of the sporadic form.[13] MA lesions can demonstrate variable degrees of calcification, cystic degeneration, tumor-like appearance and/or enhancement, making radiologic diagnosis a challenge. Multicystic MA is also recognized as a rare variant of the condition, in which the cysts may have resulted from the gradual accumulation of cerebrospinal fluid in the perivascular spaces of arachnoid/vascular tissue trapped in the cortical parenchyma.[16] Diagnosis of MA lesions can prove difficult but if done timely, prognosis with adequate surgical resection is typically good for seizure control in humans.[9,18,26]

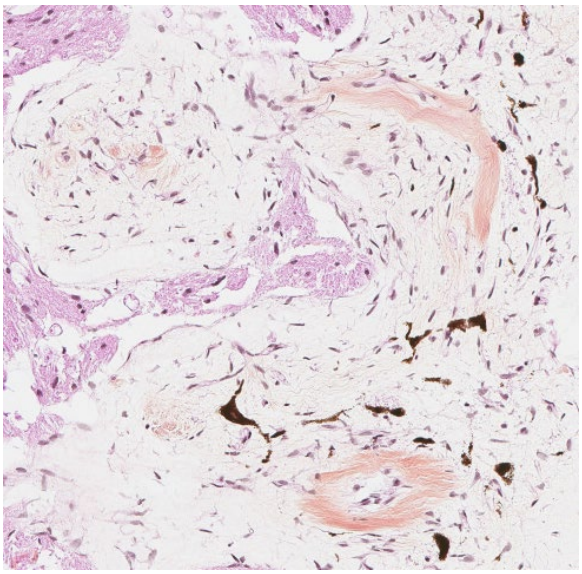

Figure 3-4. Brainstem, dog. Vessels within the fibrous bands are highlighted by a Factor VIII stain. (anti FVIIIa, 200X)

The other form of MA is, in humans, associated with neurofibromatosis type 2 (NF2). NF2-associated lesions are usually asymptomatic. The autosomal dominant mutation in NF2 in humans is located on chromosome 22q12 and encodes for the protein merlin, which is widely expressed and important for cell growth regulation. Individuals with this mutation have a predisposition to develop tumors, particularly vestibular schwannomas and meningiomas, amongst others.[5,26] MA and associated meningioma tissues were evaluated in a case series of 5 human patients using a next generation sequencing assay targeting 1425 cancer-related genes. Of the two MA cases associated with meningioma, one had deletions in the NF2 gene in both the MA and the meningioma, where-

as the other had a NF2 deletion in only the MA component. Additional mutations were identified in the MA components of these cases, suggesting that the MA arises from the meningioma rather than the opposite.[8]

In another small case series involving six human patients with MA, neurofibrillary tangles were observed in neurons present both within the MA plaques and in the surrounding cortex. Neither senile plaques nor granulovacuolar degeneration were noted. The factors stimulating the production of neurofibrillary tangles in these cases remains unknown.[14]

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JPC Diagnosis:
Brainstem: Meningioangiomatosis.

JPC Comment:
The contributor provides an excellent overview of meningioangiomatosis (MA), a rare entity in both human and veterinary medicine. Being our second hamartomatous entity of the week, it also provides an opportunity to compare and contrast the pathogeneses of NF2-associated MA and the dysplastic cerebellar gangliocytoma examined in the previous case.

Both entities are caused by loss of function mutations in tumor suppressor genes: PTEN in the case of dysplastic cerebellar gangliocytoma, and NF2 in certain cases of MA. NF2 encodes the protein product merlin, which is expressed primarily in neural tissue and mediates contact inhibition of cell growth via signals received from the extracellular matrix.[20] Merlin performs its wizardry as a cell proliferation/arrest toggle, signaling cell proliferation pathways when phosphorylated and cell growth arrest pathways when hypo-phosphorylated. At high cell density, merlin becomes hypo-phosphorylated in response to hyaluronate (HA), a component of extracellular matrix that surrounds cells, thus preventing proliferation when the cellular neighborhood becomes too crowded. Merlin's pro-growth activity is mediated by a cytoplasmic interaction with CD44, a transmembrane HA receptor; when cell density is low, merlin is phosphorylated, complexed with CD44, and growth permissive.[20]

Similar to PTEN in the previous case, merlin operates in a complex signaling environment. When not complexed with CD44, merlin is active and inhibits receptor tyrosine kinases (RTKs) and their downstream targets, including the previously discussed PI3K/AKT pro-growth pathway. In this way, active merlin functions similarly to PTEN, but at an upstream point in the signaling cascade.[24] In this state, merlin also simultaneously signals pro-apoptotic pathways and inhibits survival and growth pathways. Once complexed with CD44, however, merlin is phosphorylated, inactivated, and no longer able to inhibit RTKs, leading to the reverse effects described above: the PI3K/AKT pathway is activated, apoptotic pathways are inhibited, and survival and proliferative pathways are activated.[24] In cases of NF2-associated MA, the mutation in NF2 leads to a dysfunctional merlin, which is unable to function as a brake on the RTK, PI3K/AKT cell growth pathway, leading to a variety of neuroproliferative conditions, including MA.

Signaling pathways are complex webs, with redundant, competing inhibitory and prolif

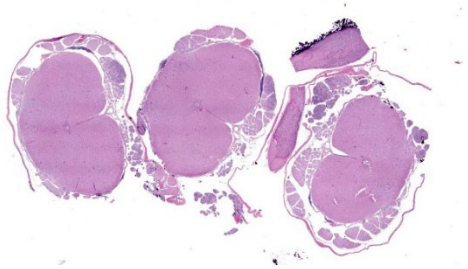

Figure 4-1. Spinal cord, cat. Multiple sections of spinal cord and spinal nerves are submitted for examination. There is scattered hypercellularity of the meninges seen at this magnification. (HE, 5X)

erative signals. This delicate balance is per turbed when any player in the process is dysfunctional. This week, cases II and III provide examples of this complexity by illustrating the effects of two tumor suppressors that are activated by different stimuli, inhibit different steps of the same canonical signaling pathway, and lead to neuroproliferative disorders when mutated.

Comparisons to the previous case brought a reprise of the hamartoma vs. neoplasia debate during the conference. Conference participants were particularly curious about the ingrowth of the abnormal tissue from a focal area of pigmented leptomeninges at the right brainstem. Participants discussed the invasive nature of this lesion and wondered if this entity might be best considered a low - grade neoplasm. With no consensus reached, the debate rages on ("rage" may be a strong word here.)

References:

1. Balne E, Roth DR, Perentes E. Cerebral meningioangiomatosis in a CD-1 mouse: A case report and comparison with humans and dogs. *Exp Toxicol Pathol*. 2008;60: 247-251.
2. Bishop TM, Morrison J, Summers BA, et al. Meningioangiomatosis in young dogs: a case series and literature review. *J Vet Intern Med*. 2014;18: 522-528.
3. Cantile C, Youssef S. Nervous System. In: Maxie MG, ed. *Jubb, Kennedy and Palmer's Pathology of Domestic Animals*. Vol 1. 6th ed. Elsevier; 2016: 404.
4. Corbett MP, Kopec BL, Kent M, et al. Encephalic meningioangiomatosis in a cat. *J Vet Diagn Invest*. 2022;34(5):889-893.
5. Coy S, Rashid R, Stemmer-Rachamimov A, et al. An update on the CNS manifestations of neurofibromatosis type 2. *Acta Neuropathol*. 2020;139: 643-665.
6. Dantio MC, Dennis AJ, Bergman RL, et al. Surgical treatment of suspected meningioangiomatosis in the thoracolumbar spinal cord. *J Am Anim Hosp Assoc*. 2020;56(4): e564-01.
7. Deb P, Gupta A, Sharma M, et al. Meningioangiomatosis with meningioma: an uncommon association of a rare entity-report of a case and review of the literature. *Childs Nerv Syst*. 2006;22: 78-83.
8. Dono A, Pothiawala AZ, Lewis CT, et al. Molecular alterations in meningioangiomatosis causing epilepsy. *J Neuropathol Exp Neurol*. 2021;80: 1043-1051.
9. Feng R, Hu J, Che X, et al. Diagnosis and surgical treatment of sporadic meningioangiomatosis. *Clin Neurol Neurosurg*. 2013;115: 1407-1414.
10. Frazier K, Liggett A, Hines M 2nd, et al. Mushroom toxicity in a horse with meningioangiomatosis. *Vet Hum Toxicol*. 2000;42(3): 166-167.
11. Ginal PJ, Blanco B, Perez J, et al. Meningioangiomatosis associated with fibrous meningioma in a dog. *Vet Rec*. 2009;164: 756-758.
12. Gonçalves R, Johnston P, Wessmann A, et al. Imaging diagnosis: canine meningioangiomatosis. *Vet Radiol Ultrasound*. 2010;51(2): 148-151.

13. Grabowski MM, Prayson RA. Focal cortical dysplasia in meningioangiomatosis. *Clin Neuropathol*. 2015;34: 76-82.
14. Halper J, Scheithauer BW, Okazaki H, et al. Meningioangiomatosis: a report of six cases with special reference to the occurrence of neurofibrillary tangles. *J Neuropathol Exp Neurol.* 1986;45(4):426-446.
15. Jeffrey M. A neuropathological survey of brains submitted under the Bovine Spongiform Encephalopathy Orders in Scotland. *Vet Rec*. 1992;131(15): 332-337.
16. Li P, Cui G, Wang Y, et al. Multicystic meningioangiomatosis. *BMC Neurology*. 2014;14:32.
17. Lorenzo V, Pumarola M, Munoz A. Meningioangiomatosis in a dog: magnetic resonance imaging and neuropathological studies. *J Small Animal Practice*. 1998;39: 486-489.
18. Makary MS, Kobalka P, Giglio P, et al. Meningioangiomatosis: clinical, imaging and histopathologic characteristics. *J Clin Imaging Sci*. 2020;10(36): 1-4.
19. Marr J, Miranda IC, Miller AD, et al. A review of proliferative vascular disorders of the central nervous system of animals. *Vet Pathol*. 2021;58 (5): 864-880.
20. Morrison H, Sherman LS, Legg J, et al. The NF2 tumor suppressor gene product, merlin, mediates contact inhibition of growth through interactions with CD44. *Genes Dev.* 2001;15(8):968-980.
21. Pumarola M, Martin de las Mulas J, Vilafranca M, et al. Meningioangiomatosis in the brain stem of a dog. *J Comp Pathol.* 1996;115: 197-201.
22. Ribas JL, Carpenter J, Hena H. Comparison of meningioangiomatosis in a man and a dog. *Vet Pathol.* 1990;27:369-371.
23. Sebastianelli M, Mandara MT, Pavone S, et al. Thalamic astrocytic hamartoma and associated meningioangiomatosis in a German shepherd dog. *Res Vet Sci*. 2013;94: 644-647.
24. Stamenkovic I, Yu Q. Merlin, a "magic" linker between extracellular cues and intracellular signaling pathways that regulate cell motility, proliferation, and survival. *Curr Protein Pept Sci*. 2010;11(6): 471-484.
25. Stebbins KE, McGrath JT. Meningioangiomatosis in a dog. *Vet Pathol*. 1988; 25: 167-168.
26. Tomkinson C, Lu JQ. Meningioangiomatosis: A review of the variable manifestations and complex pathophysiology. *J Neurol Sci*. 2018;392: 130-136.

CASE IV:

Signalment:
13-year-old, male castrated Ragdoll cat, feline (*Felis catus*)

History:
The animal presented with acute (2-3 week) onset of progressive non-painful bilateral hindlimb paraparesis that showed no improvement on corticosteroids. The lesion was localized to the L4 segment by neurology consult, but advanced imaging was declined due to cost. The animal had known chronic kidney disease that was otherwise well-managed and stable. The patient was euthanized due to poor prognosis.

Laboratory Results:
FIV/FELV testing was negative.
PARR on FFPE spinal cord tissue revealed a clonally rearranged T cell receptor gene.

Gross Pathology:
No significant gross lesions are observed in the central nervous tissues or lymphoid or-

gans. Other findings include: (1) marked cardiomegaly (32.68 g) and left ventricular wall thickening; (2) gross bilateral enlargement of the parathyroid glands; and (3) bilateral irregular kidney contours.

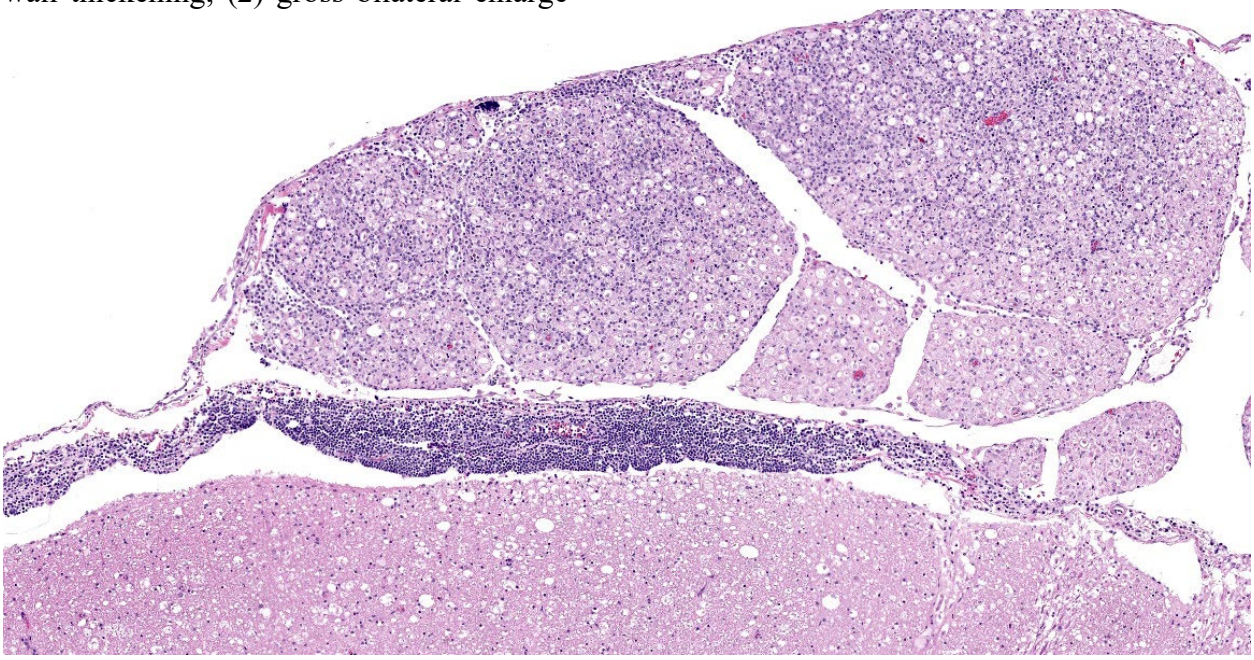

Figure 4-2. Spinal cord, cat. Neoplastic lymphocytes infiltrate and expand the meninges (center) and the adjacent spinal nerve (top). There are numerous dilated sheaths and damaged axons within the spinal nerve and underlying spinal cord. (HE, 77X)

Microscopic Description:

Spinal cord (L1-L5): Neoplastic intermediate to large (nuclei = 1.5 to 2 x RBC) round cells arranged in sheets expand the leptomeninges, infiltrate into the spinal nerve roots, and surround vessels (up to 6 cell layers thick) within the neuropil. Neoplastic cells have distinct cell borders and a small amount of amphophilic, granular cytoplasm. Nuclei are irregularly round with peripheralized chromatin and 1 variably distinct, basophilic nucleolus. There is moderate anisocytosis and anisokaryosis. There are 6 mitotic figures in 0.237 mm^2 (equivalent to 1 high power field). Separating neoplastic cells are a moderate number of small lymphocytes, histiocytes, rare plasma cells, and neutrophils. Throughout the neuropil there is increased cellularity due to neoplastic round cells and reactive glial cells. White matter myelin sheathes are occasionally dilated and contain a swollen axon (spheroid) or foamy macrophages (digestion chambers). Within the neuropil are frequent homogenous, basophilic staining, round, cytoplasmic inclusions (Lafora bodies). Occasionally neurons have peripheralized Nissl substance and yellow brown cytoplasmic pigment , interpreted as lipofuscin.

Contributor's Morphologic Diagnosis:
Spinal cord, L1-L5: Lymphoma (neurolymphomatosis), T-cell, with mild/moderate white and grey matter degeneration with multifocal Lafora bodies (incidental).

Contributor's Comment:
The acute neurologic presentation in this case was associated with lymphoma of the spinal cord, spinal nerve roots, and leptomeninges. Infiltrates of neoplastic lymphocytes with a mixed inflammatory response extended into the leptomeninges of the cerebrum, cerebellum, and brainstem (tissues not provided).

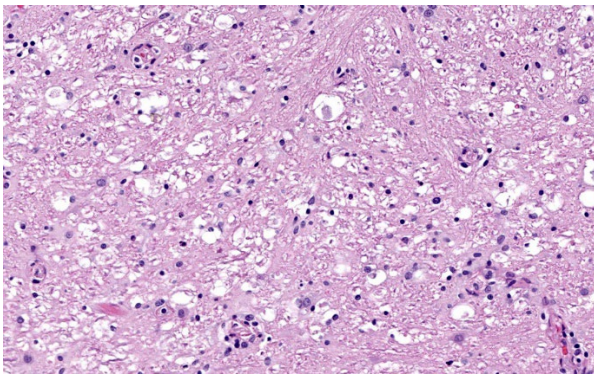

Figure 4-4. Spinal cord, cat. Adjacent to the neoplasm, white matter contains numerous dilated myelin sheaths and swollen axons. (HE, 255X)

Because some areas had more mixed inflammatory infiltrates, immunohistochemistry for FIP antigen was performed to rule out Feline Infectious Peritonitis and no immunoreactivity was noted. Immunohistochemistry for CD3 demonstrates strong immunoreactivity in the neoplastic cells infiltrating or expanding neural tissue, consistent with a T cell lymphoma. Small aggregates of CD20 expressing cells (B-cells) are attributed to secondary inflammation which does not infiltrate into neural tissue. Formalin-fixed paraffin embedded (FFPE) tissue was submitted for PARR, which identified a clonally rearranged T cell receptor gene, further supporting the diagnosis of T cell lymphoma in this case. A complete postmortem examination was performed with histologic review of select samples of all major organ systems, and no involvement of any other organ systems was noted in this case (tissues not provided).

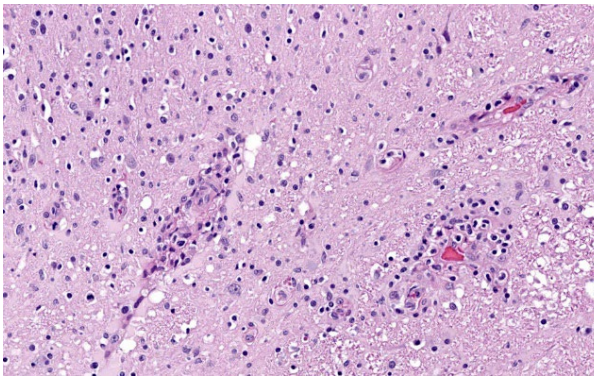

Figure 4-3. Spinal cord, cat. Neoplastic lymphocytes extend from the infiltrated meninges along Virchow-Robin spaces. (HE, 306X)

While lymphomas in cats are common and nervous system involvement may be part of a multicentric process, primary lymphomas of the nervous system are considered rare.[6] Primary lymphomas of the nervous system include both neuroinvasive and intravascular lymphomas. Our case is an example of the neuroinvasive subtype. Intravascular lymphoma is a rare subtype in which there is preferential accumulation of neoplastic lymphocytes within small blood vessels in the central nervous system (CNS) and extraneurally in some organs. This usually occurs without leukemia or systemic mass lesions. Complete vessel occlusion by neoplastic cells can lead to infarction in these cases.[6]

When there is diffuse infiltration of malignant lymphoid cells into peripheral nerves, the term neurolymphomatosis is used.[5,8] This is most common in humans due to non-Hodgkin's lymphoma and may encompass cases with or without CNS involvement. In

humans, it is almost exclusively B cell origin, though rare reports of T cell associated lymphomas have been reported.[4] Though a distinct mechanism for neurotropism has not been identified in either human or veterinary literature, CD56 (NCAM, neural cell adhesion molecule) has been implicated.[7,9]

There have been scattered case reports of neurotropic lymphoma or neurolymphomatosis of cats in the veterinary literature.[3,5,7,10-14,16,18,19] An older case series and more recent studies have also described primary central nervous system feline lymphoma with variable gross and clinical presentations and immunophenotypes.[1,11,14,15,19] There is roughly equal distribution of both T and B cell lineages re-

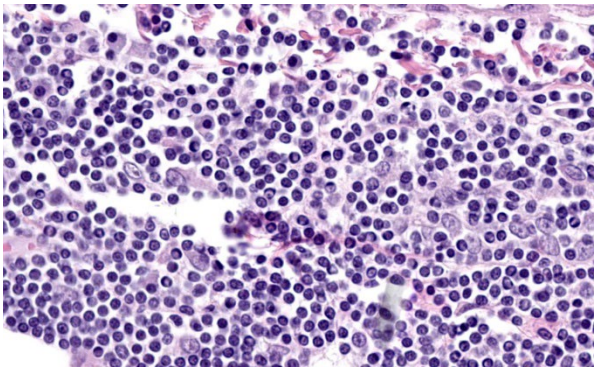

Figure 4-5. Spinal cord, cat. High magnification of lymphocytes within the meninges. (HE, 731X)

ported in both case reports and case series, which contrasts to the majority B cell immunophenotype reported in humans.[4] The most common location for primary CNS lymphoma in cats is the spinal cord. Similarly, peripheral nervous system (PNS) involvement can occur exclusively in the PNS or simultaneously with CNS lymphoma.

The clinical (antemortem) diagnosis of primary nervous system lymphoma may be challenging. Advanced imaging combined with cerebrospinal fluid (CSF) analysis may offer the best chance at antemortem diagnosis, but often results can vary and be nonspecific.[1] CSF analysis was not performed in this case. Gross findings may be minimal, as neoplastic infiltrates may not create grossly visible enlargement or a discrete mass effect. Histologic examination and immunohistochemistry remain the gold standard in both human and veterinary medicine.[17] Important clinical differential diagnoses for hindlimb paresis/paralysis include thromboembolic disease (such as saddle thrombus or fibrocartilaginous embolism), demyelinating diseases (i.e. peripheral neuropathies), other neoplasms of the spinal cord or spinal canal, infectious meningomyelitis/neuritis (e.g., Feline Infectious Peritonitis, toxoplasmosis, rabies), trauma, degenerative disk disease, neurotoxins (i.e. bromethalin), or metabolic abnormalities (such as hypoglycemia or electrolyte disturbances).

Contributing Institution:
Colorado State University
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Fort Collins, CO
https://vetmedbiosci.colostate.edu/vdl/

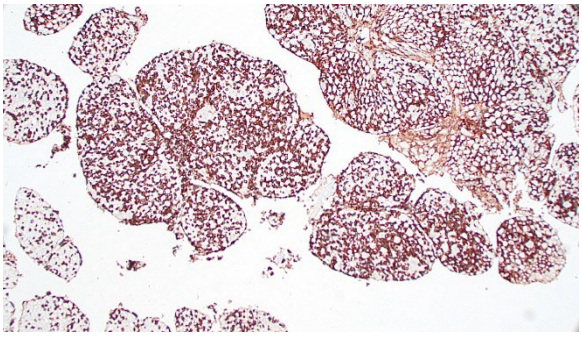

Figure 4-6. Spinal cord, cat. Neoplastic cells demonstrate strong cytomembranous staining for CD3. (anti-CD3, 100X)

JPC Diagnosis:
Spinal cord and spinal nerves: Lymphoma.

JPC Comment:
The contributor provides an excellent review of neurolymphomatosis and correctly notes

that this entity presents a particular diagnostic challenge.

In addition to cats, neurolymphomatosis has been reported in a smattering of dogs and horses, often with minimal perivascular cuffing of clonal lymphocytes throughout the CNS.[6] Clinical signs in affected animals are variable, typically non-specific, and stem from Wallerian degeneration of axons due to heavy lymphocytic infiltration of the peripheral nerves. The most common presenting complaints are single or multi-limb lameness, muscle weakness, monoparesis or plegia, and cauda equine syndrome, depending on the particular nerves affected.[2]

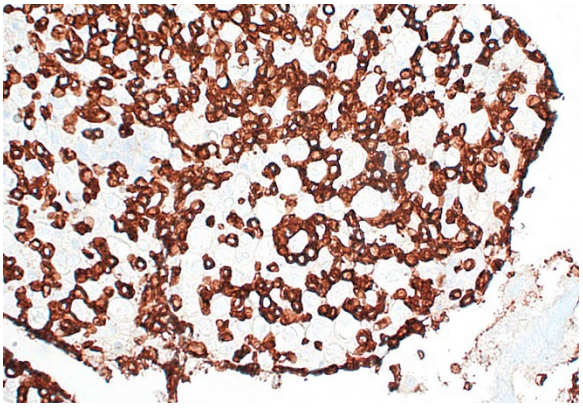

Figure 4-7. Spinal cord, cat. Neoplastic cells demonstrate strong cytomembranous staining for CD3. (anti-CD3, 400X)

Antemortem diagnosis in humans is typically made on the basis of enlargement or enhancement of nerves and nerve roots on MRI and PET/CT images and the examination of peripheral nerve biopsy speciments.[2] Even so, in one case study series, 45% of cases were diagnosed as neurolymphomatosis only at autopsy.[17] In veterinary medicine, MRI exam is typically cannot provide definitive diagnosis due to the lack of historically consistent MRI findings among histologically confirmed cases of neurolymphomatosis.[6]

Postmortem gross findings can include bilateral asymmetrical or symmetrical thickening of the spinal or cranial nerves, though it is more common to find no gross lesions, as illustrated in this case. Definitive diagnosis requires histologic evidence of infiltration of nerve fascicles of B- or T-cell lymphocytes in rows between peripheral nerve axons and within intraneural and epineural spaces.[6]

Conference participants discussed two histologic features of this case that do not strictly fit the case definition of neurolymphomatosis: the abundant mixed inflammatory infiltrate composed of B lymphocytes and plasma cells within the leptomeninges and the presence of abundant neoplastic T lymphocytes within the spinal cord parenchyma. Prior to reviewing IHC results, conference participants assumed meningeal lymphocytes were an extension of the neoplastic process; however, IHC results convincingly indicate that the meningeal lymphocytes are B lymphocytes admixed with fewer plasma cells and few histiocytes, indicating that these aggregates most likely represent chronic inflammation. Conference participants felt the chronic inflammation could be plausibly explained by the presence of long-standing neoplastic infiltration of the spinal nerves, though this feature has not been consistently described in this entity. More confounding was the presence of neoplastic lymphocytes within the spinal cord. Neurolymphomatosis is, by definition, neoplastic infiltration of the peripheral nervous system without infiltration of other organs. The presence of lymphocytes within the spinal cord parenchyma, along with the inability to conclusively rule out other lymphomatous foci elsewhere in the animal, led to a conservative JPC diagnosis of lymphoma.

References:

1. Durand A, Keenihan E, Schweizer D, et al. Clinical and magnetic resonance im-

aging features of lymphoma involving the nervous system in cats. *J Vet Intern Med*. 2022;36(2):679-693.
2. Fantaconi N, Walker JJA, Ives EJ. What is your neurologic diagnosis? *JAVMA.* 2020;257(10):1013-1016.
3. Fondevila D, Vilafranca M, Pumarola M. Primary central nervous system T-cell lymphoma in a cat. *Vet Pathol*. 1998;35(6):550-553.
4. Gan HK, Azad A, Cher L, Mitchell PL. Neurolymphomatosis: diagnosis, management, and outcomes in patients treated with rituximab. *Neuro Oncol*. 2010; 12(2):212-215.
5. Higgins MA, Rossmeisl JH Jr, Saunders GK, Hayes S, Kiupel M. B-cell lymphoma in the peripheral nerves of a cat. *Vet Pathol.* 2008;45(1):54-57.
6. Higgins RJ, Bollen AW, Dickinson PJ, Siso-Llonch S. Tumors of the Nervous System. In: Meuten DJ, ed. *Tumors in Domestic Species*. 5th ed. Wiley Blackwell; 2020:872-877.
7. Hsueh CS, Tsai CY, Lee JC, et al. $CD56^+$ B-cell neurolymphomatosis in a cat. *J Comp Pathol*. 2019;169:25-29.
8. Kelly JJ, Karcher DS. Lymphoma and peripheral neuropathy: a clinical review. *Muscle Nerve*. 2005;31(3):301-313.
9. Kern WF, Spier CM, Hanneman EH, Miller TP, Matzner M, Grogan TM. Neural cell adhesion molecule-positive peripheral T-cell lymphoma: a rare variant with a propensity for unusual sites of involvement. *Blood*. 1992;79(9):2432-2437.
10. Linzmann H, Brunnberg L, Gruber AD, Klopfleisch R. A neurotropic lymphoma in the brachial plexus of a cat. *J Feline Med Surg.* 2009;11(6):522-524.
11. Mandara MT, Domini A, Giglia G. Feline lymphoma of the nervous system: immunophenotype and anatomical patterns in 24 cases. *Front Vet Sci*. 2022; 9:959466.
12. Mandrioli L, Morini M, Biserni R, Gentilini F, Turba ME. A case of feline neurolymphomatosis: pathological and molecular investigations. *J Vet Diagn Invest*. 2012;24(6):1083-1086.
13. Mellanby RJ, Jeffery ND, Baines EA, Woodger N, Herrtage ME. Magnetic resonance imaging in the diagnosis of lymphoma involving the brachial plexus in a cat. *Vet Radiol Ultrasound*. 2003;44 (5):522-525.
14. Mello LS, Leite-Filho RV, Panziera W, et al. Feline lymphoma in the nervous system: pathological, immunohistochemical, and etiological aspects in 16 cats. *Pesq Vet Bras.* 2019;39(06):393-401.
15. Rissi DR, McHale BJ, Miller AD. Primary nervous system lymphoma in cats. *J Vet Diagn Invest*. 2022;34(4):712-717.
16. Sakurai M, Azuma K, Nagai A, et al. Neurolymphomatosis in a cat. *J Vet Med Sci*. 2016;78(6):1063-1066.
17. Shree R, Goyal MK, Modi M, et al. The diagnostic dilemma of neurolymphomatosis. *J Clin Neurol*. 2016;12(3):274-281.
18. van Koulil Q, Santifort KM, Beukers M, Ioannidis M, Van Soens I. Neurolymphomatosis in a cat with diffuse neuromuscular signs including cranial nerve involvement. *Vet Rec Case* Rep. 2022; 10:e482.
19. Zaki FA, Hurvitz AI. Spontaneous neoplasms of the central nervous system of the cat. *J Small Anim Pract*. 1976;17 (12):773-782.