NEVPC Case

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SIGNALMENT: Approximately 1 year old, male, mixed breed, *Canis familiaris*

HISTORY:
Initially presented with progressive stumbling, weakness, and regurgitation. The dog weighed 6.5 kg, had a body condition score of 2/5 (i.e. thin), was approximately 10% dehydrated, and walked with difficulty. Bilateral sublingual masses were present resulting in difficulty in food prehension and chewing, hypersalivation, halitosis, pain, and regurgitation were noted. Supportive care was initiated. The CBC revealed a mild anemia (hematocrit 31%, normal reference interval [RI] 37–55) and vacuolated lymphocytes. The serum biochemical profile was normal. An infectious disease screen was positive for antibodies against *Ehrlichia canis*. Survey radiographs of the thorax and abdomen were unremarkable. On the following day, the dog was anesthetized, and the sublingual masses were resected. The dog was discharged with antibiotics and anti-inflammatory medications for post-operative management and observation. Histopathology of the resected sublingual masses revealed pyogranulomatous and fibrinous sialoadenitis and stomatitis with multifocal ulceration of oral mucosa. Macrophages infiltrating the lesions were often heavily distended with foamy, vacuolated cytoplasm.
During the following week the foster owner noted frequent regurgitation after eating, weakness, occasional falling, episodes of aerophagia and incoordination. A neurological examination did not identify any localizing lesions. The dog responded to visual and auditory stimuli, but audition was not specifically examined. An ophthalmic examination was normal and otoscopic examination showed normal ear canals. The dog was euthanized one month after initial presentation.

GROSS FINDINGS:
Absence of subcutaneous and inner adipose tissue and severe muscle atrophy. Severe enlargements of the dorsal and ventral vagal branches, vagosympathetic trunk, phrenic nerve, cervical nerves and dorsal root ganglia were noted. The aboral esophagus was mildly distended (megaesophagus, Abnormalities of the central nervous system (CNS) included a mild internal bilateral symmetric hydrocephalus (lateral ventricles) and white matter atrophy. The kidneys were mildly enlarged and pale. The liver was enlarged, and pale brown, with left lateral lobe liver rupture and fibrosis. Both thyroid glands, spleen, and multiple lymph nodes were mildly enlarged. Multifocal small gastric pyloric ulcers were noted. Other findings included bone callus of left ribs 7, 8 and 9 that was assumed to be post-traumatic.

HISTOPATHOLOGIC FINDINGS:
The slide has sections of 2 tissues: esophagus and esophagogastric junction.
Esophagus: there is marked enlargement of the submucosal (Meissner's) and myenteric plexus (Auerbach's plexus) with extensive vacuolation of neuronal cells. Neurons sometime display nuclear peripheralization or remain centrally located.
Stomach: similar changes affecting the submucosal and myenteric plexus. Withing the mucosa, parietal cells are diffusely enlarged, with vacuolated cytoplasm. Similar cytoplasmic vacuolation is seen in some endothelial cells and interstitial macrophages.

A nerve around a large artery in the serosa of the stomach is markedly enlarged due to edema as well as endo-, peri-, and epineurial infiltration of vacuolated macrophages with mild demyelination and Schwann cell vacuolation. Some Schwann cell nuclei are hypertrophic. There is also depletion of myelin sheaths, which was interpreted as hypomyelination and demyelination.

MORPHOLOGIC/ETIOLOGIC DIAGNOSIS:
MDx: Esophagus and stomach, submucosal and myenteric plexus (Meissner's and Auerbach's) neuronal cells, cytoplasmic vacuolation diffuse, moderate to severe.
Endothelial cells and interstitial macrophages: cytoplasmic vacuolation diffuse, moderate.
Stomach: parietal cells cytoplasmic vacuolation diffuse, severe
Peripheral nerves: edema, fibroplasia with macrophage infiltration, Schwann cell and macrophage vacuolation, axonal degeneration and demyelination, severe, diffuse.

CONDITION:
Canine β-mannosidosis (lysosomal storage disorder, oligosaccharide storage disease).

DISCUSSION:
The clinical signs in this dog were consistent with a central and peripheral neuropathy. The histological findings were consistent with a systemic storage disease. Lesions in the submucosal and myenteric plexus extended throughout the length of the gastrointestinal tract. In humans with β-mannosidosis, behavioral changes, deafness, and mental disabilities are noted as early as a few months of age with other vague neurological signs. In contrast, ruminants with β-mannosidosis exhibit a severe CNS disorder with facial and skeletal dysmorphia and neonatal death.
Other findings in this dog included a mild megaesophagus, enlarged thyroid glands, and mild hydrocephalus which have also been observed in some human patients. One human patient had hypotonia and feeding difficulty in the first months of life, with swallowing abnormalities, decreased esophageal motility and achalasia around the age of 2 years, but whether it was associated with her β-mannosidosis was not determined. Another case in humans presented with ventricular dilatation (hydrocephalus) that developed around the age of 2 years. A report in one cat with α-mannosidosis described megaesophagus likely associated with peripheral nerve involvement.
The marked thickening of nerves and the dorsal root ganglia in this dog have not been previously reported in other species with β-mannosidosis but have been reported in English Springer Spaniels with fucosidosis.
Plasma concentration of the β-mannosidosis specific oligosaccharide was approximately 75 fold that of controls. The plasma beta-mannosidase activity was severely reduced to ~5% of controls; five other lysosomal acid hydrolase activities were increased or within their normal reference interval. Genomic sequencing of this dog's MANBA gene identified a homozygous exonic five bp tandem duplication in the penultimate exon of the MANBA gene (c.2377_2381dupTATCA) which results in a reading frame shift, altering the subsequent amino acid sequence and creating a premature stop codon. The truncated beta-mannosidase enzyme is expected to be dysfunctional. This enzyme deficiency causes the accumulation of un-degraded oligosaccharides in cells, which affect the myelination of the peripheral and central nervous systems.

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