Neuronal Ceroid-lipofuscinosis in a Cat

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Abstract. Neuronal ceroid-lipofuscinosis was diagnosed in a young adult domestic short-haired cat euthanatized because of severe progressive neurologic disease. Clinical signs included blindness, seizures, and decreased mentation. An autofluorescent pigment, identified as ceroid-lipofuscin by electron microscopy and staining properties, was found within neurons of the central and peripheral nervous systems. A diffuse reactive astrocytosis accompanied by multifocal microgliosis was visible in all areas of the brain. Retinal atrophy with intraneuronal lipopigment accumulation was also identified. Contrary to the human neuronal ceroid-lipofuscinoses, pigment deposition appeared to be restricted to neural tissues.

Key words: Cat; central nervous system; ceroid-lipofuscinosis; peripheral nervous system; retinal atrophy; storage disease.

Descriptions of several feline lysosomal storage diseases can be found in standard veterinary texts. These diseases include GM₁ and GM₂ gangliosidoses, sphingomyelin lipidosis, alpha-mannosidosis, mucopolysaccharidoses types I and VI, globoid cell leukodystrophy, and ceroid-lipofuscinosis. A literature search revealed only two reports of feline ceroid-lipofuscinosis: a neuronal form identified in two Siamese cats and a multisystemic form found in a Japanese domestic cat. This report describes a case of neuronal ceroid-lipofuscinosis in a domestic short-haired cat (DSH).

A male DSH with a clinical history of lethargy and altered mentation was examined at the Veterinary Teaching Hospital of the Atlantic Veterinary College. The animal was estimated to be 1.5 years of age and had been a stray (barn cat) until adopted 4 months previously by one of the authors (P. Ward). Following adoption, the animal was housed indoors and fed a balanced diet of commercial cat food. The cat appeared healthy for 3 months but had been less active and less alert during the 3 weeks prior to presentation.

A neurologic examination identified the following clinical abnormalities: pelvic limb hyperreflexia, generalized hyperesthesia, bilateral absence of an ocular menace response, and poor visual perception. Pupillary light reflexes were intact, and no other cranial nerve deficits were identified. An ophthalmologic exam revealed mild hyperreflectivity of the tapetal fundus. The cat had a normal gait and no difficulty with locomotory tests (wheelbarrowing, hopping, etc.). During the physical examination, the animal had a seizure characterized by whole-body clonic convulsions. A hemogram and serum chemistry profile revealed no abnormalities. Tests for feline leukemia virus and feline immunodeficiency virus were negative, and further clinical work-up was declined. The cat was euthanatized 2 months later, as the clinical signs had progressed to include complete blindness, mental dullness, and seizures of increasing frequency and severity.

No gross abnormalities were noted at necropsy. The presence of a well-developed thymus confirmed the cat to be a young adult. In addition to the entire brain and spinal cord, samples of lung, heart, liver, spleen, thymus, skeletal muscle, kidney, adrenal gland, pancreas, small intestine, and sciatic nerve were fixed by immersion in neutral-buffered 10% formalin. The eyes were placed in Bouin’s solution for 24 hours and then transferred into 70% alcohol for 24 hours. Paraffin-embedded tissues were sectioned at 6 μm and stained with hematoxylin and eosin (HE) and with Luxol fast blue (LFB). Sections of the brain and spinal cord were also stained with periodic acid–Schiff (PAS), Ziehl-Neelsen acid fast (AF), and Sudan Black B (SB) stains. Control tissues for light microscopy were obtained from a 1-year-old cat (all tissues) and a 10-year-old cat (eye only). Following several months of immersion in formalin, the brain of the affected cat was also sampled for ultrastructural examination. Thalamic tissue was minced into 1-mm³ pieces, placed in 2% glutaraldehyde with 0.1 M phosphate buffer (pH 7.4) for 1 hour, and then post-fixed for 1 hour in 1% osmium tetroxide using the same buffer. Following dehydration in alcohols, samples were embedded in Epon resin (J.B. EM Services, Pointe-Claire, Quebec, Canada). Ultrathin sections were stained with uranyl acetate and Sa-
Fig. 1. Lateral thalamic nucleus; cat with ceroid-lipofuscinosis. Most neurons contain granular pigment (arrowheads). LFB. Bar = 30 μm.

to's stain\(^1\) and examined with a Hitachi 600 transmission electron microscope. Control tissue was not examined ultrastructurally.

Microscopically, a mild degree of cerebrocortical atrophy was characterized by diffuse thinning of the gray matter and a slight decrease in the number of cortical neurons. Neurons throughout the brain and spinal cord contained various amounts of a granular cytoplasmic pigment. These irregularly shaped, 1-6-μm-diameter, slightly refractile granules varied from light brown to bright red with HE stain. The pigment granules stained pink with PAS, dark blue with LFB (Fig. 1), and black with SB but were negative with AF. These staining properties were consistent with previous reports describing ceroid-lipofuscin at the light microscopic level.\(^6-8\) The pigment reacted most strongly with LFB, and application of this stain revealed fine cytoplasmic granules in many neurons of the central and peripheral nervous systems that had appeared unaffected in HE sections. Cytoplasmic granules of LFB-positive material were detected in the exocrine pancreas, but these were also present in the control tissue. Pigment granules and other significant lesions were not observed in the other tissues examined, including the components of the mononuclear-phagocyte system within the spleen and liver.

The intraneuronal pigment seldom resulted in cell swelling or distortion. A reactive astrocytosis was present throughout the brain but was most prominent in the thalamus and cerebral cortex (Fig. 2). Microgliosis was prominent in many areas, with some of these cells being markedly swollen by an intracytoplasmic pigment. In comparison to the intraneuronal pigment, these granules tended to be larger and more eosinophilic with HE; their staining properties with LFB, SB, PAS, and AF were unaltered. In the spinal cord, pigment deposition was most severe in the neurons of the ventral horn and the spinal ganglia. A few blood vessels in the gray matter of cerebral cortex and the meninges of sacral spinal cord were cuffed by lymphocytes.

Diffuse retinal degeneration was characterized by thinning of the outer nuclear and photoreceptor layers plus loss of ganglion cells. Staining with LFB revealed small numbers of granules within the cytoplasm of many cells in all retinal layers (Fig. 3). Application of
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LFB also revealed granules within the neurons of the sinoatrial node, the coeliaco-mesenteric ganglion, and Meissner’s and Auerbach’s ganglia of the small intestine.

When examined under ultraviolet light, unstained paraffin sections of cerebral cortex emitted a yellow-green fluorescence typical of ceroid-lipofuscin pigment.7 Electron microscopic examination of affected thalamic neurons demonstrated membrane-bound intracytoplasmic bodies composed of a variety of osmiophilic deposits, which included multilamellar arrays (Fig. 4) characteristic of ceroid-lipofuscin.8

The human neuronal ceroid-lipofuscinoses (NCLs) are a heterogeneous group of diseases that include several common neurodegenerative conditions in children.1-5 Symptoms for some of these conditions include progressive visual deficits, seizures, hyperesthesia, and altered mentation;1-5 these clinical signs were observed in this cat. NCL in humans is characterized by pigment accumulation in neurons, but a variety of other cell types may also be affected, including the mononuclear phagocytes of the liver, spleen, and skin.1-5 The conspicuous neurologic effects may be partly due to the inability of neurons to decrease the intracellular accumulation of abnormal metabolites through cell division or exocytosis.3

Multisystemic involvement has been reported in feline NCL10 and in other domestic species including
sheep,7 various dog breeds,8 and cattle.9 Lesions restricted to the nervous system have been recorded in NCL of Nubian goats,4 Rambouillet sheep,2 and two Siamese cats.6 The Siamese cats became clinically affected at 1.5–2 years of age. The clinical presentation in one cat included hyperesthesia and convulsions while in the other cat decreased vision was noted. Both the age at onset and the clinical signs of our case correlated well with these prior feline NCL forms. Histopathologic findings in these cats were also generally comparable to those of the present case although pigment deposition in autonomic or retinal neurons was not reported. Retinal atrophy with loss of photoreceptors and lipopigment accumulation has been described in dogs,6,9 sheep,2,7 and humans1,5 suffering from some forms of NCL but, until this case, had not been observed in cats.

Neuronal storage diseases can be caused by the ingestion of certain toxins.9 A toxic etiology is unlikely in this case because the clinical signs arose and steadily progressed several months after the cat was placed on a commercial cat food. During this period, the cat was housed indoors, and no other cat in this multiple-cat household was affected.

Most lysosomal storage diseases are inherited, often in an autosomal recessive pattern.3 This pattern has been established for some variants of human NCL, and specific chromosomes have been implicated in the infantile and juvenile forms of this disease.3 The exact nature of the genetic defects that result in NCL remain unknown. It is probable that a specific substrate accumulates due to an alteration in enzymatic function, as is true of most storage diseases.3 The bulk of the stored material in some forms of NCL has been identified as subunit c of mitochondrial adenosine triphosphate synthetase,7,8 suggesting a defect in the catabolism of this enzyme.

Although this was not a purebred cat, inbreeding of barn cat populations is likely very common, and an inherited origin cannot be ruled out. Development of a feline animal model of NCL is desirable. Unfortunately, the parentage of this cat could not be established.

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References