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Large Animal Models for Batten Disease: A Review

Krystal Weber, BS and David A. Pearce, PhD

Sanford Children's Health Research Center, Sanford Research/University of South Dakota, Sioux Falls, South Dakota (KW, DAP), and the Department of Pediatrics, University of South Dakota Sanford School of Medicine, Sioux Falls, South Dakota (DAP)

Abstract

The neuronal ceroid lipofuscinoses, collectively referred to as Batten disease, make up a group of inherited childhood disorders that result in blindness, motor and cognitive regression, brain atrophy, and seizures, ultimately leading to premature death. So far more than 10 genes have been implicated in different forms of the neuronal ceroid lipofuscinoses. Most related research has involved mouse models, but several naturally occurring large animal models have recently been discovered. In this review, we discuss the different large animal models and their significance in Batten disease research.

Keywords

Animal models; Batten disease; neuronal ceroid lipofuscinosis; ONCL

Introduction

Neuronal ceroid lipofuscinosis (NCL), also known as Batten disease, is a neurodegenerative disease characterized by lysosomal accumulation of autofluorescent storage material in all tissues.¹ Until recently, 10 different genes that cause various forms of neuronal ceroid lipofuscinosis have been identified. Recently, new genes associated with new and existing variants have been discovered (Table 1).² The incidence of neuronal ceroid lipofuscinosis worldwide is 1 to 8 in 100 000 live births, with the juvenile being the most common.¹ Currently there is no known cure, and treatments are limited to palliative care.

Much of the work in the field of neuronal ceroid lipofuscinosis is centered on the use of murine models. These murine models have been immensely useful in studying the disease development and the function of the neuronal ceroid lipofuscinosis-causing genes, leading to major advancements. Although murine models have significantly advanced our knowledge

Corresponding Author: Dr David A Pearce, Sanford Childrens Health Research Center, 2301 East 60th Street North, Sioux Falls, SD, 57104-0589; Phone: 605-312-6004; Fax: 605-312-6071; david.pearce@sanfordhealth.org.

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of neuronal ceroid lipofuscinosis, they do not completely recapitulate the human disease pathology.³⁻⁵ Neurodegeneration between human and murine models is not always directly comparable because of the pathophysiological differences in the murine model's lissencephalic brain. The limitations of the inability to completely replicate the disease phenotype can hinder development of therapeutic interventions. Advancements made in neuronal ceroid lipofuscinosis murine models cannot always directly translate to humans. Incidentally, there have been a number of naturally occurring large animals displaying neuronal ceroid lipofuscinosis pathology, such as dogs, sheep, and cattle. The study of large animal models has proved useful because of the close mimicry of disease pathology as well as similarities to human anatomy and physiology.^{3,4} The large animal models have displayed similar phenotypic features to that of human neuronal ceroid lipofuscinosis. The longer lifespan of large animal models to that of murine models also proves beneficial to long-term evaluations of disease pathology and treatments.

Three forms of neuronal ceroid lipofuscinosis have been linked to genetic mutations affecting proteins with known enzymatic function: palmitoyl protein thioesterase-1, tripeptidyl peptidase-1, and cathepsin D.¹ The functions of the remaining neuronal ceroid lipofuscinosis proteins have yet to be fully understood. The three major subtypes of neuronal ceroid lipofuscinosis are infantile, late-infantile, and juvenile, with having the greatest prevalence.^{1,6} Symptoms of all subtypes include loss of vision, physical delays and loss of motor coordination, cognitive deterioration, and seizures.

Neuronal ceroid lipofuscinosis has been found in many animal species; however, only a few have provided useful models.⁴ Canine neuronal ceroid lipofuscinosis is the most researched of the large animal models, with the most established model being the English setter dog.^{7,8}

English Setter – *Cln8*

Neuronal ceroid lipofuscinosis in the English setter was first described in the 1950s.⁷ Affected dogs appear normal at birth and do not exhibit symptoms until 1 year to 2 years of age. Affected dogs display rapid decline and die at approximately 2 years of age. Symptoms in the English setter include visual deterioration, cognitive impairment, motor decline, and seizures. The seizures tend to be the cause of death, as they become intractable in the end stage of the disease.

Because of the similar pathological, biochemical, and clinical feature of neuronal ceroid lipofuscinosis in the English setter, the breed was thought to be a good model for human juvenile neuronal ceroid lipofuscinosis.⁹ After the exclusion of *Cln2* and *Cln3* by Katz and colleagues⁷ and Shibuya and colleagues,⁹ respectively, Lignaas and colleagues were able to determine that mutations of *Cln8* caused English setter neuronal ceroid lipofuscinosis.⁸

Border Collie – *Cln5*

The age of onset and severity of symptoms seen in Border collie neuronal ceroid lipofuscinosis varies greatly between individual dogs. Owners, however, tend to observe symptoms as early as 15 months of age, and dogs typically die no later than 28 months of age.¹⁰ Owners report agitation, hallucinations, aggressive behavior, seizures, and

hyperactivity. Loss of coordination is shown through incontinence, ataxia, and dysphagia. Vision impairment is seen in most cases, with lipopigment accumulation in retinal tissues.

Melville and colleagues were able to identify a C>T polymorphism leading to a premature stop codon in the coding region of the *Cln5* gene as the cause of Border collie neuronal ceroid lipofuscinosis.¹⁰

American Bulldog – Cathepsin D

Neuronal ceroid lipofuscinosis was first described in the American bulldog by Evans and colleagues in 2005.¹¹ The onset of neuronal ceroid lipofuscinosis is generally seen around 2 years of age, with death occurring as late as 7 years due to slow progression of the disease. Loss of motor coordination is the most obvious symptom generally displayed through ataxia, hypermetria, and a wide-base stance. Accumulation of storage material is found in the ganglion cells of the retina; however, there have been no reports of vision changes. Autofluorescent material is present in the cerebral cortex and cerebellum.

In 2005, Awano and colleagues determined the cause of neuronal ceroid lipofuscinosis in the American bulldog to be a missense mutation in cathepsin D.¹²

American Staffordshire Terriers – Arylsulfatase G

A severe cerebellar cortical abiotrophic disease was identified as the cause of ataxia in adult American Staffordshire terriers and the disease was later found to be neuronal ceroid lipofuscinosis.^{13,14} Progressive ataxia is the most obvious symptom, with initial disease onset occurring around 3 years to 5 years of age. Visual impairments are not reported and upon examination there is an absence of lipofuscin in the retina. Affected dogs exhibit cerebellar atrophy, loss of Purkinje cells, and accumulation of ceroid lipofuscin in several areas of the brain. In 2009, Abitbol and colleagues were able to identify an arylsulfatase G mutation as the cause of American Staffordshire terrier neuronal ceroid lipofuscinosis.¹³

Tibetan Terrier – *ATP13A2*

Farias and colleagues and Wöhlke and colleagues determined the cause of Tibetan terrier neuronal ceroid lipofuscinosis to be a mutation causing a truncation of the *ATP13A2* gene.^{15,16} The onset of symptoms begin with mild neurodegeneration between 4 years and 6 years of age. As the disease progresses, behavioral symptoms develop, such as aggression, anxiety, and nervousness. Other symptoms include ataxia, cognitive deterioration, seizures, and visual impairment. Massive accumulation of autofluorescent storage material is seen in the cortex, cerebellum, and retina.^{15,16}

Dachshund – *Tpp1* and *Ppt1*

The first cases of suspected neuronal ceroid lipofuscinosis in the dachshund were described in 1977 and 1980 in wirehaired and long-haired varieties, respectively.^{17,18} The age of onset and clinical signs differed between the 2 cases, suggesting that in the dachshund, the disease results from different mutations.

Neuronal ceroid lipofuscinosis was described in 2 juvenile dachshund siblings by Awano and colleagues in 2006.¹⁹ Affected dogs begin to display symptoms at 9 months of age, with rapid progression and death around 12 months of age. Symptoms include vomiting, mental dullness, ataxia, myoclonus, seizures, and loss of vision. Cognitive impairment is also noted by unresponsiveness to previously learned commands. Behavioral changes develop near the end stage of the disease, including hyperactivity, aggression, and repetitive circling. Autofluorescent storage material is present in the central nervous system.

Awano and colleagues identified a single base-pair deletion leading to a premature stop codon in *Tpp1*, which was shown to be one cause of neuronal ceroid lipofuscinosis in the dachshund.¹⁹

Sanders and colleagues described neuronal ceroid lipofuscinosis in a 9-month-old miniature dachshund in 2010.²⁰ The affected dog displayed symptoms such as ataxia, weakness, visual impairment, and behavioral changes, as well as the presence of autofluorescent storage material in the retina and cerebral cortex. Brain tissue acquired showed a significant lack of palmitoyl-protein thioesterase 1 activity. An insertion mutation leading to premature stop codon in *Ppt1* was linked to the disease phenotype.

Recently, enzyme-replacement therapy has shown promise in a dachshund model for neuronal ceroid lipofuscinosis. Vuilleminot and colleagues performed intrathecal TPP1 enzyme replacement in a neuronal ceroid lipofuscinosis-affected dachshund, elevating TPP1 activity throughout most of the brain and attenuating degenerative changes.²¹ This was the first study achieving widespread delivery of biochemically active TPP1 to the brain in a large animal model of late-infantile neuronal ceroid lipofuscinosis.

Australian Shepherd – *Cln6*

O'Brien and Katz were able to identify neuronal ceroid lipofuscinosis in Australian shepherd littermates in 2008.²² Disease onset in the Australian shepherd tends to occur at approximately 18 months of age. Affected dogs display changes in behavior exhibited by extreme anxiety or nervousness. Loss of motor coordination is evident as the dogs display hypermetria, ataxia, and weakness. The dogs also show signs of hallucinations and vision loss. A massive amount of autofluorescent material is present in the cerebral cortex, cerebellum, and retinas, and diffuse brain atrophy occurs.^{22,23}

Katz and colleagues identified the cause of neuronal ceroid lipofuscinosis in Australian shepherds as a T>C transition resulting in a missense mutation in the *Cln6* gene.²³

Although dogs have provided remarkable advancement in the understanding of Batten disease, they are not the only beneficial large animal model. Livestock models are valuable due to their production animal status, making them economical to breed and manage.

South Hampshire Sheep – *Cln6*

The first ovine model for neuronal ceroid lipofuscinosis was segregated in 1976 by RD Jolly and colleagues using South Hampshire sheep and has been maintained since.^{4,24} Sheep

affected with ovine ceroid lipofuscinosis generally start showing symptoms at 9 months to 12 months of age, with death occurring around 24 months of age. Symptoms include brain atrophy, retinal degeneration, lipopigment accumulation, and tremors.

The strong synteny between South Hampshire ovine neuronal ceroid lipofuscinosis and *CLN6* chromosomal regions identified by Broom and colleagues strongly suggested that mutations in *Cln6* cause the ovine disease.⁵ The suggestion of mutant *Cln6* being the cause of South Hampshire ovine neuronal ceroid lipofuscinosis was confirmed through mRNA analysis by Tammen and colleagues in 2006.²⁶

Merino Sheep – *Cln6*

The clinical, pathological, and biochemical symptoms of Merino sheep ovine neuronal ceroid lipofuscinosis was first characterized in 2002 by Cook and colleagues, and an ovine neuronal ceroid lipofuscinosis flock has since been established.²⁷ Signs of neurological disease first develop around 8 months to 12 months and include decreased menace response and mild behavioral changes. Later in the disease pathology, blindness, motor deficits, and anorexia began to develop, with seizures occurring in the latest stages of the disease. Brain atrophy and storage bodies are also observed postmortem. Death generally occurs at 24 months to 26 months of age.²⁷

In 2006, Tammen and colleagues were able to isolate a mutation in exon 5 of *Cln6* as the cause of Merino ovine neuronal ceroid lipofuscinosis. The mutation is a frame-shift resulting in a premature stop codon in *Cln6*.²⁶

Borderdale Sheep – *Cln5*

Neuronal ceroid lipofuscinosis was first described in the Borderdale sheep by Jolly and colleagues in 2002, and later a colony was established.²⁸ Symptoms of ovine neuronal ceroid lipofuscinosis in Borderdale sheep were described as vision loss and apparent deafness. The sheep also display fearlessness and difficulty in moving with the rest of the flock. Frugier and colleagues discovered a G>A substitution resulting in a premature stop codon in *Cln5*.³

Swedish Landrace Sheep – Cathepsin D

In 1993, a flock of white Swedish Landrace sheep in Northern Sweden was observed to be affected by a congenital form of ovine neuronal ceroid lipofuscinosis.³⁰ The affected newborn lambs are weak and unable to support themselves when attempting to stand. Autopsy reveals severe brain atrophy, with the weight being about half of normal brains. The cerebrum and cerebellum are largely devoid of myelin. Autofluorescent material is seen in the cerebral cortex.

Tyynela and colleagues isolated the cause of congenital ovine neuronal ceroid lipofuscinosis to be a G>A missense mutation in the *cathepsin D* gene. Affected sheep show a significant deficiency in cathepsin D enzymatic activity.³¹

Devon Cattle – *Cln5*

A small group of cattle from a herd in Australia were found to display pathology associated with neuronal ceroid lipofuscinosis.³² Symptoms were observed in these cattle from 14 months of age. They showed loss of vision and tracking in a circular motion, often with mild torticollis. Histology findings showed retinal atrophy and accumulation of storage material. There was also accumulation of storage material in neurons.

In 2006, Houweling and colleagues were able to identify the cause of neuronal ceroid lipofuscinosis in Devon cattle.³³ A single base duplication in exon 4 of bovine *Cln5* causes a frame-shift mutation, resulting in a premature stop codon.

Conclusion

Large animal models may be highly instrumental for future Batten disease research. Dogs, sheep, cattle, and other large animal models share a close resemblance to the human pathology of neuronal ceroid lipofuscinosis due to patterns of neurodegeneration and motor skill deficiencies that are more similar than those seen in mouse models. Advances made in large animal models are likely to provide new avenues for discovering treatments for the human form of this disease.

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References

1. Jalanko A, Thomas B. Neuronal ceroid lipofuscinosis. *Biochim Biophys Acta*. 2009; 1793(4):697–709. [PubMed: 19084560]
2. Williams RE, Mole SE. New nomenclature and classification scheme for the neuronal ceroid lipofuscinoses. *Neurology*. 2012; 79(2):183–191. [PubMed: 22778232]
3. Frugier T, Mitchell NL, Tammen I, et al. A new large animal model of CLN5 neuronal ceroid lipofuscinosis in Borderdale sheep is caused by a nucleotide substitution at a consensus splice site (c.571+1G>A) leading to excision of exon 3. *Neurobiol Dis*. 2008; 29(2):306–315. [PubMed: 17988881]
4. Jolly RD, Martinus RD, Palmer DN. Sheep and other animals with ceroid-lipofuscinoses: their relevance to Batten disease. *Am J Med Genet*. 1992; 42(4):609–614. [PubMed: 1535180]
5. Getty AL, Pearce DA. Interactions of the proteins of neuronal ceroid lipofuscinosis: clues to function. *Cell Mol Life Sci*. 2011; 68(3):453–474. [PubMed: 20680390]
6. Weimer JM, Custer AW, Benedict JW. Visual deficits in a mouse model of Batten disease are the result of optic nerve degeneration and loss of dorsal lateral geniculate thalamic neurons. *Neurobiol Dis*. 2006; 22(2):284–293. [PubMed: 16412658]
7. Katz ML, Khan S, Awano T, et al. A mutation in the CLN8 gene in English Setter dogs with neuronal ceroid-lipofuscinosis. *Biochem Biophys Res Commun*. 2005; 327(2):541–547. [PubMed: 15629147]

8. Lingaas F, Aarskaug T, Sletten M, et al. Genetic markers linked to neuronal ceroid lipofuscinosis in English setter dogs. *Anim Genet.* 1998; 29(5):371–376. [PubMed: 9800325]
9. Shibuya H, Liu PC, Katz ML, et al. Coding sequence and exon/intron organization of the canine CLN3 (Batten disease) gene and its exclusion as the locus for ceroid-lipofuscinosis in English setter dogs. *J Neurosci Res.* 1998; 52(3):268–275. [PubMed: 9590435]
10. Melville SA, Wilson CL, Chiang CS, et al. A mutation in canine CLN5 causes neuronal ceroid lipofuscinosis in Border collie dogs. *Genomics.* 2008; 86(3):287–294. [PubMed: 16033706]
11. Evans J, Katz ML, Levesque D, et al. A variant form of neuronal ceroid lipofuscinosis in American bulldogs. *J Vet Intern Med.* 2005; 19(1):44–51. [PubMed: 15715047]
12. Awano T, Katz ML, O'Brien DP, et al. A mutation in the cathepsin D gene (CTSD) in American Bulldogs with neuronal ceroid lipofuscinosis. *Mol Genet Metab.* 2006; 87(4):341–348. [PubMed: 16386934]
13. Abitbol M, Thibaud JL, Olby NJ, et al. A canine Arylsulfatase G (ARSG) mutation leading to a sulfatase deficiency is associated with neuronal ceroid lipofuscinosis. *Proc Natl Acad Sci U S A.* 2010; 107(33):14775–14780. [PubMed: 20679209]
14. Olby N, Blot S, Thibaud JL, et al. Cerebellar cortical degeneration in adult American Staffordshire Terriers. *J Vet Intern Med.* 2004; 18(2):201–208. [PubMed: 15058771]
15. Farias FH, Zeng R, Johnson GS, et al. A truncating mutation in ATP13A2 is responsible for adult-onset neuronal ceroid lipofuscinosis in Tibetan terriers. *Neurobiol Disord.* 2011; 42(3):468–474.
16. Wöhlke A, Phillip U, Bock P, et al. A one base pair deletion in the canine ATP13A2 gene causes exon skipping and late-onset neuronal ceroid lipofuscinosis in the Tibetan terrier. *PLoS Genet.* 2011; 7(10):e1002304. [PubMed: 22022275]
17. Cummings JF, de Lahunta A. An adult case of canine neuronal ceroid-lipofuscinosis. *Acta Neuropathol.* 1977; 39(1):43–51. [PubMed: 197773]
18. Vandevelde M, Fatzer R. Neuronal ceroid-lipofuscinosis in older dachshunds. *Vet Pathol.* 1980; 17(6):686–692. [PubMed: 7423829]
19. Awano T, Katz ML, O'Brien DP, et al. A frame shift mutation in canine TPP1 (the ortholog of human CLN2) in a juvenile Dachshund with neuronal ceroid lipofuscinosis. *Mol Genet Metab.* 2006; 89(3):254–260. [PubMed: 16621647]
20. Sanders DN, Farias FH, Johnson GS, et al. A mutation in canine PPT1 causes early onset neuronal ceroid lipofuscinosis in a Dachshund. *Mol Genet Metab.* 2010; 100(4):349–356. [PubMed: 20494602]
21. Vuilleminot BR, Katz ML, Coates JR, et al. Intrathecal tripeptidyl-peptidase 1 reduces lysosomal storage in a canine model of late infantile neuronal ceroid lipofuscinosis. *Mol Genet Metab.* 2011; 104(3):325–337. [PubMed: 21784683]
22. O'Brien DP, Katz ML. Neuronal ceroid lipofuscinosis in 3 Australian shepherd littermates. *J Vet Intern Med.* 2008; 22(2):572–475.
23. Katz ML, Farias FH, Sanders DN, et al. A missense mutation in canine CLN6 in an Australian shepherd with neuronal ceroid lipofuscinosis. *J Biomed Biotech.* 2011; 2011:198042.
24. Jolly RD, West DM. Blindness in South Hampshire sheep: a neuronal ceroidlipofuscinosis. *N Z Vet J.* 1976; 24(6):123. [PubMed: 1067509]
25. Broom MF, Zhou C, Broom JE, et al. Ovine neuronal ceroid lipofuscinosis: a large animal model syntenic with the human neuronal ceroid lipofuscinosis variant CLN6. *J Med Genet.* 1998; 35(9):717–721. [PubMed: 9733028]
26. Tammen I, Houweling PJ, Frugier T, et al. A missense mutation (c.184C>T) in ovine CLN6 causes neuronal ceroid lipofuscinosis in Merino sheep whereas affected South Hampshire sheep have reduced levels of CLN6 mRNA. *Biochim Biophys Acta.* 2006; 1762(10):898–905. [PubMed: 17046213]
27. Cook RW, Jolly RD, Palmer DN, et al. Neuronal ceroid lipofuscinosis in Merino sheep. *Aust Vet J.* 2002; 80(5):292–297. [PubMed: 12074311]
28. Jolly RD, Arthur DG, Kay GW, Palmer DN. Neuronal ceroid-lipofuscinosis in Borderdale sheep. *N Z Vet J.* 2002; 50(5):199–202. [PubMed: 16032271]
29. Frugier T, Mitchell NL, Tammen I, et al. A new large animal model of CLN5 neuronal ceroid lipofuscinosis in Borderdale sheep is caused by a nucleotide substitution at a consensus splice site

- (c.571+1G>A) leading to excision of exon 3. *Neurobiol Dis.* 2008; 29(2):306–315. [PubMed: 17988881]
30. Tyynelä J, Sohar I, Sleat DE, et al. Congenital ovine neuronal ceroid lipofuscinosis--a cathepsin D deficiency with increased levels of the inactive enzyme. *Eur J Paediatr Neurol.* 2001; 5 (Suppl A): 43–45. [PubMed: 11589006]
31. Tyynelä J, Sohar I, Sleat DE, et al. A mutation in the ovine cathepsin D gene causes a congenital lysosomal storage disease with profound neurodegeneration. *EMBO J.* 2000; 19(12):2786–2792. [PubMed: 10856224]
32. Harper PA, Walker KH, Healy PJ, et al. Neurovisceral ceroid-lipofuscinosis in blind Devon cattle. *Acta Neuropathol.* 1988; 75(6):632–636. [PubMed: 3376766]
33. Houweling PJ, Cavanagh JA, Palmer DN, et al. Neuronal ceroid lipofuscinosis in Devon cattle is caused by a single base duplication (c.662dupG) in the bovine CLN5 gene. *Biochim Biophys Acta.* 2006; 1762(10):890–897. [PubMed: 16935476]

Table 1

Categorization of Known NCL Mutations

	Gene Symbol	Protein	NCL Diseases
Soluble lysosomal enzyme deficiencies	<i>CTSD</i>	Cathepsin D	Congenital, late-infantile, juvenile, adult
	<i>PPT1 (CLN1)</i>	Palmitoyl protein thioesterase-1	Infantile, late-infantile, juvenile, adult
	<i>TPP1 (CLN2)</i>	Tripeptidyl peptidase-1	Late-infantile, juvenile
	<i>CTSF</i>	Cathepsin F	Adult Kufs type B
Non-enzyme deficiencies, (functions of identified proteins generally poorly understood at present time)	<i>CLN3</i>	Transmembrane protein	Juvenile
	<i>CLN5</i>	Soluble lysosomal protein	Late-infantile, juvenile, adult
	<i>CLN6</i>	Transmembrane protein	Late-infantile, adult Kufs type A
	<i>MFSD8</i>	Major facilitator superfamily domain-containing protein 8transmembrane protein	Late-infantile
	<i>CLN8</i>	Transmembrane protein	Late-infantile, EPMR
	<i>DNAJC5</i>	Soluble cysteine string protein α	Adult autosomal dominant
	<i>GRN</i>	Progranulin	Adult; also causes frontotemporal lobar dementia in heterozygous mutants
	<i>ATP13A2</i>	P-type ATPase	Juvenile
	<i>KCTD7</i>	Potassium channel tetramerization domain-containing protein 7	Infantile; also causes progressive myoclonic epilepsy-3
Others	?	Mutations yet to be defined	Congenital, infantile
	?	Mutations yet to be defined	Late-infantile
	? <i>CLN9</i> ?	Mutations yet to be defined	Juvenile
	?	Mutations yet to be defined	Late-onset/Adult, including adult Kufs type B
	<i>CLCN6</i>	Mutations yet to be found on both alleles in human disease	Adult
	<i>SGSH</i>	Mutations usually cause Mucopolysaccharidoses type IIIA	Adult