Generalized Progressive Retinal Atrophy in Two Akita Dogs

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Abstract. Two distantly related, two- to three-year-old male Akita dogs developed partial or complete night blindness which progressed to day blindness. Clinical features were attenuated retinal blood vessels, diffuse exaggerated tapetal reflex, and hyperreflective horizontal lines in the tapetal fundus. The principal histologic change was regional photoreceptor cell degeneration. Areas of moderately atrophic sensory retina alternated, concentrically to the papilla, with areas of severely atrophic retina. Ultrastructurally, there was disorganization of photoreceptor outer segments, retinal gliosis, focal loss with reorganization of the outer limiting membrane, and retinal invasion by pigment epithelium-derived macrophages. Clinical observations on five additional Akitas with generalized retinal atrophy suggest that the condition is heritable.

Early and late onset hereditary retinal degenerations have been described in the dog. These include rod-cone dysplasia type 1 in the Irish setter, rod-cone dysplasia type 2 in the collie, central progressive retinal atrophy in several breeds, and cone dystrophy in the Alaskan malamute. It is probable that most if not all dog breeds are affected by progressive retinal atrophy. These heritable forms of retinal atrophy are potentially important models for those human retinal degenerations known collectively as retinitis pigmentosa.

This report describes the features of a distinctive type of retinal atrophy in two Akita dogs. The clinical features of blindness in a larger group of Akita dogs will be reported separately [Roberts and Severin, manuscript in preparation].

Materials and Methods

The right eye of dog 1 was removed at 11:00 A.M. under closed circuit general anesthesia. It was measured, sectioned frontally, and the vitreous body was gently removed. The posterior segment was fixed in 4% Sörensen's phosphate-buffered glutaraldehyde at 4°C for 30 minutes. The fixative was brought to room temperature for an additional 30 minutes. The posterior segment was cut sagittally through the optic papilla. The temporal half was fixed in Bouin's solution and embedded in paraffin. Interrupted serial sections were cut at 5 μm and stained with hematoxylin-phloxine-safran. The nasal half was washed in Sörensen's phosphate buffer and examined and drawn with the aid of a dissecting microscope. Small strips of retina were osmicated, dehydrated, and embedded flat in epon.

Both globes from dog 2 were removed immediately following a lethal intravenous dose of pentobarbital sodium. They were measured, fixed in Zenker's acetic acid solution, and cut in the sagittal plane. The atrophic retina was examined grossly with the aid of a dissecting microscope. Following paraflin embedding, the globes were cut at 5 μm and stained with hematoxylin-phloxine-safran.

Results

Two distantly related male Akita dogs developed poor night vision. Deficits first were noted by the owner at 2.5 years of age in dog 1. When examined at three years of age, the dog had difficulty avoiding objects in dim light. Direct and indirect pupillary responses were slow. Retinal and optic disc blood vessels were attenuated 30%. The tapetal retina was mottled and hyperreflective. A horizontal area of intense hyperreflectivity was in the superior peripapillary area (figs. 1a, b).

Night vision deficits developed at two years of age in dog 2. A linear horizontal hyperreflective band, identical to that in dog 1, was present bilaterally. Over the following two years, both night and day vision gradually deteriorated. When re-examined at four years of age, the retina was mottled and hyperreflective (figs. 1c, d). Retinal vessels were attenuated 50 to 75%. A horizontal linear band of reflectivity was still identifiable in one of the two eyes. No electroretinography response was elicited following light or dark adaptation.

The three enucleated eyes were of normal size, and the posterior ciliary vessels were unremarkable. Gross changes were restricted to the posterior segment, where alternating light and dark curved bands, 0.1 cm wide and up to 1 cm long, were in the retina. These alter-
Fig. 1: Fundus photographs; left and right eyes. 
a, b. Dog 1; three years of age. Retinal vessels slightly attenuated. Hyperreflective line (arrow) in tapetal retina of both eyes. 
c, d. Dog 2; four years of age. Marked attenuation of retinal vessels. Tapetal retina is diffusely mottled and hyperreflective. Remnant of hyperreflective line, present at two years of age, is still evident in one eye (arrow).
Fig. 2: Drawing of fixed nasal fundus; right eye, dog 1. Dark bands of severe retinal atrophy separated by light bands of less severe atrophy. Hyperreflective bar (arrows) still evident following glutaraldehyde fixation.

Narating bands generally were concentric to the optic papilla and were most pronounced in peripheral retina (fig. 2). Each light band was a ridge of moderately thinned retina. Dark bands were zones of marked retinal thinning. Some, particularly those in peripheral retina, contained black foci. Other gross changes were attenuation of most retinal vessels, perivascular pigment cuffs, and microcystoid degeneration at the ora ciliaris retinae. The hyperreflective horizontal line in the tapetum still was grossly evident following glutaraldehyde fixation. The altered tapetal color did not appear to result from local variations in retinal thickness.

Histologic examination confirmed that the atrophic retinal change was zonal and that retina covering the tapetum lucidum was affected least severely. Rod and cone outer segments were short, widely separated, and randomly oriented (fig. 3a). Histiocytes, some in the process of phagocytosis, were in the interphotoreceptor space. Inner segments were better preserved than outer segments, but many were short and irregular. The outer nuclear layer thickness was reduced variably (figs. 3a, b). Some photoreceptor nuclei were pyknotic. Changes in other layers were present only where photoreceptor cell degeneration was marked. The pigment epithelium was hypertrophied and occasionally extended over the vitreal retinal surface. Pigment-laden macrophages were present in the retina. Most severe change was in peripheral and, to a lesser extent, midzonal retina and resulted in hillocks of moderately atrophic retina which

Fig. 3: Dog 1. a. Tapetal peripapillary retina. Marked thinning with disorganization of outer segments. Thinned photoreceptor nuclear layer contains two pyknotic nuclei (arrows). Lipopigment granules in pigment epithelium. Bar = 20 μm. b. Tapetal peripapillary retina. Absence of outer and inner segments with reduction of photoreceptor nuclear layer to a bilayer. Bar = 20 μm. c. Nontapetal midzonal retina. Sensory retina absent and flattened, slightly disorganized pigment epithelium appears to be in direct contact with vitreous body. This area corresponds to one of the black foci noted grossly. Bar = 20 μm.
Alternated with completely atrophic sensory retina (figs. 3c, 4).

Degenerative rod and cone photoreceptors was the predominant ultrastructural feature. Outer segment lamellae were short and disorganized (fig. 5a). Packets of disordered shed lamellae indented the apical surface of the pigment epithelium where they were intimately associated with and enveloped by long pigmented epithelium villi. Vesicle formation was in many shed lamellae (fig. 5a). The precise location of all shed lamellae was difficult to determine because of the close apposition of lamellae to pigment epithelium microvilli. Most lamellae were in the interphoteceptor space. Unequivocal intracytoplasmic lamellae were not present in pigment epithelium cells. In areas of gliosis, atrophic retina rested directly upon the pigment epithelium without any intervening photoreceptors or outer limiting membrane. The apical processes of Müller cells were interconnected by zonulae adherens and formed rosette-like structures. Numerous processes of other glial cells were oriented parallel to the inner limiting lamina. These processes formed excrescences which indented but did not penetrate the inner limiting lamina. Macrophages, which in light microscopic sections appeared to lie in the vitreous on the retinal surface, were usually interpolated between the inner limiting lamina and Müller cell end-feet.

Pigment epithelium changes were mild. Many cells contained lipopigment granules, some of which surrounded melanin granules (fig. 5b). Apical microvilli occasionally were more numerous. Where atrophy was most advanced, all that remained of the retina was a flattened monolayer of pigment epithelium upon which lay the inner limiting lamina. Tapetal cells in cross-section were unremarkable in all areas.

A clinically normal littermate of dog 1 was bred on two occasions to a normal Akita bitch. Four of the 12 offspring had ophthalmoscopic evidence of retinal degeneration and horizontal hyperreflective bands at 23 months of age. A fifth offspring had horizontal hyperreflective bands without retinal degeneration. A littermate of dog 2 developed identical clinical signs and ophthalmoscopic retinal changes.

Discussion

The visual deficits in these two dogs was the result of severe retinal degeneration which chiefly involved the photoreceptor cell layer. Both rod and cone cells were affected, although the clinical history suggests that rods underwent degeneration first. Vesicle formation in rod lamellae, seen in dog 1, also occurs in Norwegian elkhound rod dysplasia.2 Changes in other retinal layers were probably secondary to photoreceptor cell loss. Retinal gliosis, increased pigment epithelium lipopigment, pigment epithelium-derived macrophages and the formation of junctions between Müller cells are nonspecific changes which occur in other types of retinal atrophy.7,9,10

The pattern of atrophy in these dogs’ eyes was distinctive. Concentric alternating bands of retinal atrophy have not to our knowledge been seen in other types of retinal degeneration. Local variations in severity occur randomly in other types of retinal atrophy.2,5

The basis for concentric atrophy patterns in the Akita is unknown. They did not correspond to the distribution of retinal vessels11 or of cones.13 They may have resulted from different classes of photoreceptors undergoing degeneration at different rates. Concentric patterns of atrophy may occur in other types of retinal atrophy but may not have been recognized because of the difficulty in ophthalmoscopically evaluating the peripheral retina where they are most pronounced.

Shed outer segment lamellae were not identified in pigment epithelium cytoplasm. Phagocytosis with digestion of shed lamellae is an important function of the pigment epithelium and in one model of retinal atrophy, the RCS rat, this process is defective.8 We are unable to conclusively impute pigment epithelium phagocytosis as the cause of retinal degeneration in either dog. Many lipofuscin and compound lipofuscin–melanin granules were in the pigment epithelium of both dogs. Their presence suggests that a competent pigment epithelium phagolysosomal system existed in both dogs for a time. The lipid component of such granules is considered to originate primarily in photoreceptor outer segment disc phospholipid.12 The absence of phagosomes in dog 1 may also reflect the chronicity of the disease and the time of day at which the eye was enucleated. Rod and cone membrane shedding is rhythmical and in some species follows a diurnal pattern.6,20 Little is known about the rhythmical shedding of photoreceptor membranes, and therefore the expected number of pigment epithelium phagosomes, in the normal dog.

A major ophthalmoscopic feature in both dogs was a hyperreflective horizontal line superior to the optic papilla (fig. 1). The morphologic basis for this line was not determined in spite of the use of transmission electron microscopic sections and serial light microscopic sections through the line. Similar lines have been seen in beagles where they were not associated with vision deficits.16,18 Clinical observations indicate that
Fig. 4: Dog 2; nontapetal peripheral retina. Areas of marked retinal atrophy alternate with areas of moderate atrophy. Moderate choroidal atrophy. Bar = 200 μm.

Fig. 5: Dog 1; a. Tapetal peripapillary retina. Marked reduction in length and number of outer segments. Packets of shed outer segment lamellae indent apical surface of pigment epithelium. Some lamellae exhibit vesicle formation (arrow). Bar = 5 μm. b. Nontapetal peripapillary retina. Lipopigment granules, some with melanin core, are common (arrow). Bar = 1 μm.
these lines frequently are associated with retinal atrophy in the Akita, although they also may be seen in the absence of vision deficits [Roberts and Severin, manuscript in preparation]. The lines may be an optical effect due to local indentation of the tapetum by the long posterior ciliary arteries. The course of these arteries in these two Akitas was unremarkable.

It is probable that heritable factors were involved in these two cases. Both dogs shared ancestors at the sixth and seventh generation and littermates have either developed similar clinical signs or have given rise to litters with affected dogs.

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References

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