Genome-Wide Association & Whole Genome Sequencing of an Inherited Progressive Retinal Atrophy in the Bengal Cat

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1. Introduction

- Inherited retinal diseases of humans are a leading cause of blindness throughout the world. These include retinitis pigmentosa, Leber's congenital amaurosis and some forms of macular degeneration (1).
- Progressive retinal atrophy (PRA) is a term describing a group of inherited retinal diseases in animals, similar to those of humans (1).
- PRA is characterized by degeneration of the retinal photoreceptors of the eye, which eventually leads to blindness. It is an age-dependent disease with varying rates of progression (2).
- Animal models of human inherited retinal degenerations are useful for development of genetic testing methods and therapies to benefit both animals and humans (2).
- Four types of PRA have been found in cats—two in Abyssinian cats and one in Persians (1). **The fourth is a novel, autosomal recessive PRA that has been observed in the Bengal cat breed.**

2. Objectives

- Characterize the phenotype of a novel feline PRA using state-of-the-art imaging methods
- Confirm the chromosomal location of the PRA with GWAS and haplotype analysis
- Determine the causal mutation of the PRA in Bengal cats

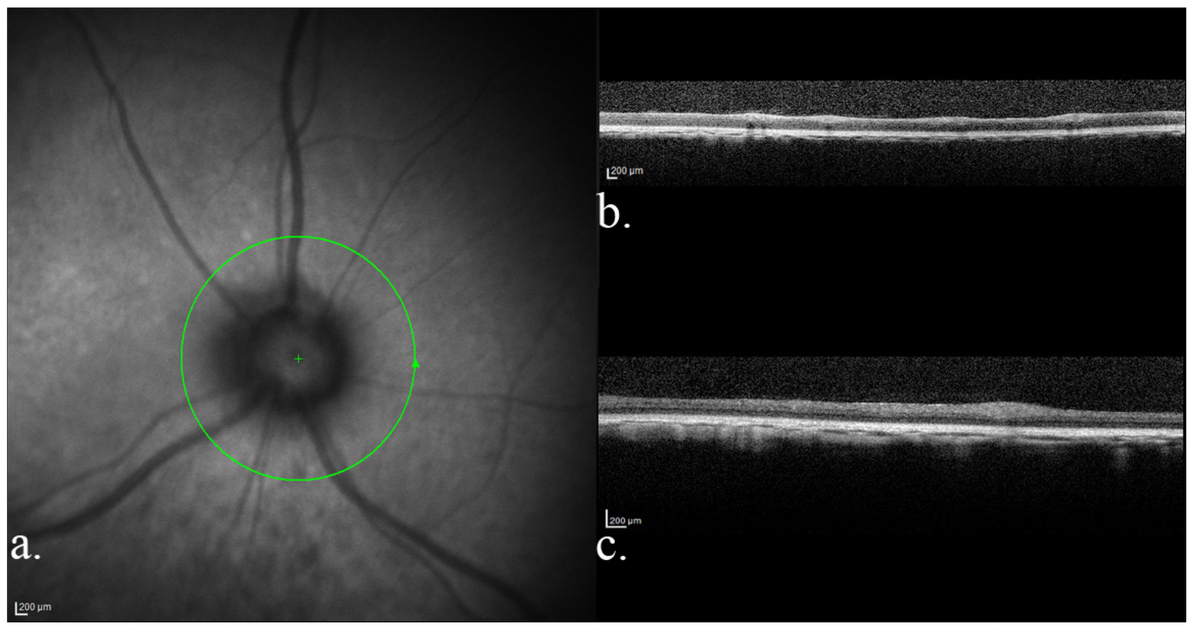

Figure 1: Optical Coherence Tomography scans. Cross-sectional images were taken of the area of the retina outlined in green (**a**). Retinal thickness is decreased in the 6 month old affected cat (**c**) compared to the non-affected cat (**b**).

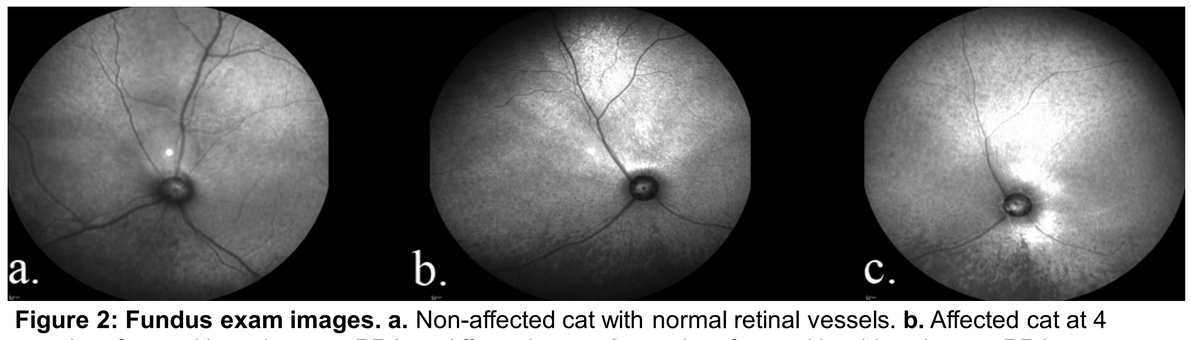

Figure 2: Fundus exam images. a. Non-affected cat with normal retinal vessels. **b.** Affected cat at 4 months of age with early stage PRA. **c.** Affected cat at 6 months of age with mid-end stage PRA.

3. Methods

- Complete ophthalmic exams including biomicroscopy and indirect ophthalmoscopy were performed every 4 weeks on 4 affected Bengal kittens in an established breeding colony.
- Optical Coherence Tomography (OCT) scans were performed on all kittens every 4 weeks and within 1 week of complete ophthalmic examination (**Figures 1 & 2**).
- A genome-wide association study (GWAS) was previously performed using the Illumina Infinium Feline 63K iSelect DNA array data from 45 affected cases and 53 unaffected controls.
- A trio of cats with the known PRA genotype from the Bengal colony (**Figure 3**) was whole genome sequenced. Sequencing data was screened and polymorphisms in the associated region were evaluated.
- Five missense mutations in a gene known to be involved with vision in humans and one splice site variant in a gene involved in energy production were chosen to be verified with Sanger Sequencing.

Figure 3: A trio of cats with the known PRA genotype from the Bengal colony was whole genome sequenced. These included 1 carrier cat (left) and 2 affected cats (center & right).

4. Results

- Complete ophthalmic examinations of affected kittens showed the earliest signs of retinal photoreceptor degeneration to be around 15 weeks of age. Degeneration was determined to be complete by 8 months of age.
- The GWAS suggested a 6 Mb region between base pairs 20,000,000 and 26,000,000 on chromosome A3 to be strongly associated with PRA (**Figure 4**).
- Whole genome sequencing revealed 569 polymorphisms in 90 genes located in the associated region provided by the GWAS.
- Of the SNPs that were Sanger Sequenced, only 1 was still concordant after population screening.

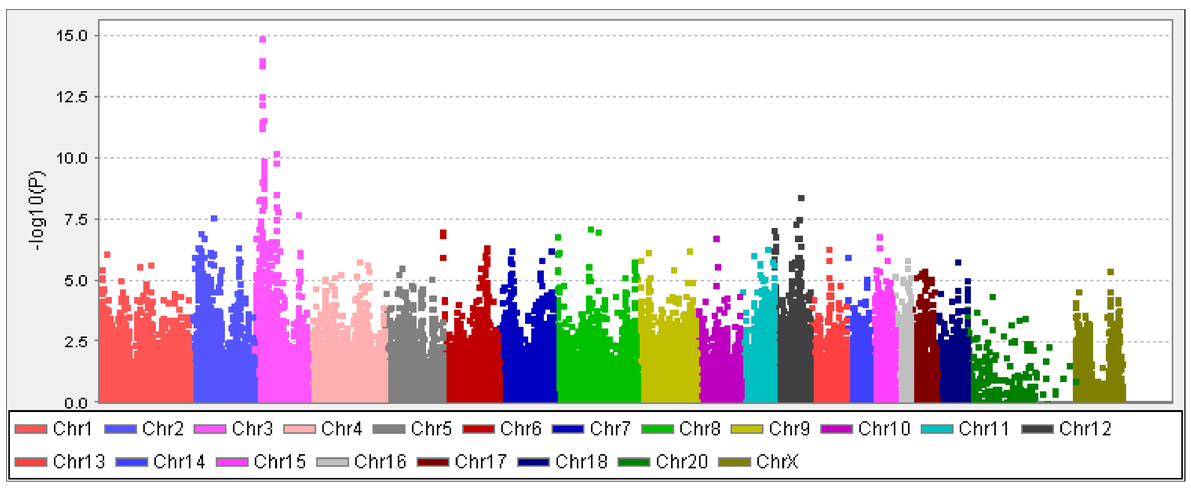

Figure 4: Manhattan plot summarizing the case–control GWAS for Bengal cats with PRA. A significant association with the disease was obtained with SNPs making up a 6 Mb region on chromosome A3 (chromosome 3 here).

5. Conclusion

- Complete ophthalmic examinations confirmed the phenotype of a mid-onset form of PRA in the Bengal cat breed.
- OCT scans further characterized the phenotype and retinal thickness changes associated with PRA in affected cats. Retinal thickness data will be quantified at a later time.
- The concordant SNP is currently being evaluated with additional Sanger Sequencing.
- If the mutation is confirmed, a genetic test can be produced to eliminate the disease in the breed.
- A feline model could identify a new gene associated with retinal degeneration in humans and other species. Animal models can be used further to develop gene and drug therapies.

6. Acknowledgements

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