We have received the following report on the AHT Goniodysgenesis/Glaucoma research project in the Flatcoated Retriever from James Oliver Specialist Veterinary Ophthalmologist at the Animal Health Trust.
March 2016

The Project:

"Estimation of prevalence of pectinate ligament dysplasia and identification of genetic risk factors for glaucoma and pectinate ligament dysplasia in the Flatcoated Retriever and development of DNA tests to reduce disease prevalence"

Aims and objectives of the project

Objectives of the study

1. To provide robust and current prevalence data for pectinate ligament dysplasia (PLD) in the Flatcoated Retriever (FCR) in the UK
2. To collect DNA from i) PLD cases ii) PG cases and ii) controls and perform genome-wide association analyses for PLD and PG
3. To identify genetic variants that confer susceptibility to PLD and PG in the FCR

Summary of the scientific achievements to date.

Objective 1.

We have performed gonioscopy on and measured intraocular pressures (IOP) in 170 Flatcoated retrievers (FCRs). One hundred six of 170 (62.4%) FCR were affected by PLD (ordinal grades 1-3); 70 (41.2%) being mildly affected (grade 1), 36 (21.2%) moderately affected (grade 2) and 0 severely affected (grade 3) (Table 2). A significant positive correlation was observed between PLD and age (rho = 0.34, $P < 0.01$). No correlation was observed between PLD and IOP (rho = -0.02, $P = 0.85$). These findings were presented at the European College of Veterinary Ophthalmologist's annual congress in Helsinki, 2015. A manuscript detailing our findings has been published in *Canine Genetics and Epidemiology*. We continue to examine and collect DNA from further Flatcoated Retrievers.

Objective 2.

We have collected DNA from all 170 FCRs described above. We now have DNA from 63 FCRs with PLD, 91 controls > 5 years old with no evidence of PLD and 7 FCRs

that have been diagnosed with PG and all these will contribute directly to the study. Sample collection is continually ongoing.

We have performed a genome-wide association study with DNA from 148 FCRs (comprising i) 83 with normal eyes (controls) ii) 65 with PLD or PG (cases). We compared the results of controls and cases. Data were first represented as quantile-quantile plots (Q-Q plot) to compare the distribution of observed test statistics with the distribution expected under the null. There was inflation of the observed findings across the distribution, indicative of population stratification or cryptic relatedness without compelling evidence for an excess of disease associations. Genome-wide Manhattan plots were created to display the GWAS findings with respect to their genomic positions to highlight any signals of potential interest. None of the SNPs reached genome wide significance. Owing to the absence of discovery of any associations between the SNPs and disease (PLD and/or PG) in our FCR cohort we performed a combined GWAS analysis using GWAS data from Welsh Springer Spaniels; another breed we are also investigating as part of our wider studies into the genetics of PLD and PG (funded by a grant from the American Kennel Club Canine Health Foundation). We had already performed a GWAS in 92 Welsh Springer Spaniels comprising i) 24 with normal eyes (controls) ii) 68 with PLD or PG. GWAS data from the two breeds was combined using meta-analysis (143 cases and 97 controls). We combined summary statistics from each breed (log odds ratios and standard errors) using a fixed effects model and inverse-variance weighted averages of regression coefficients to obtain a combined estimate of the overall odds ratio for SNPs that passed quality control filters and were therefore informative in both breeds. This analysis revealed a single locus on chromosome 11 that reached a genome-wide significant level of association with disease (PLD and PG combined; P-value 5.53×10^{-7}).

Objective 3.

We are now beginning to analyse the variants within this locus for association with disease using whole-genome sequencing data from two affected FCR and one unaffected FCR (funded by the Flat Coated Retriever Society Rescue and Rehoming DNA Research Fund).

FCRS Health Sub Committee.