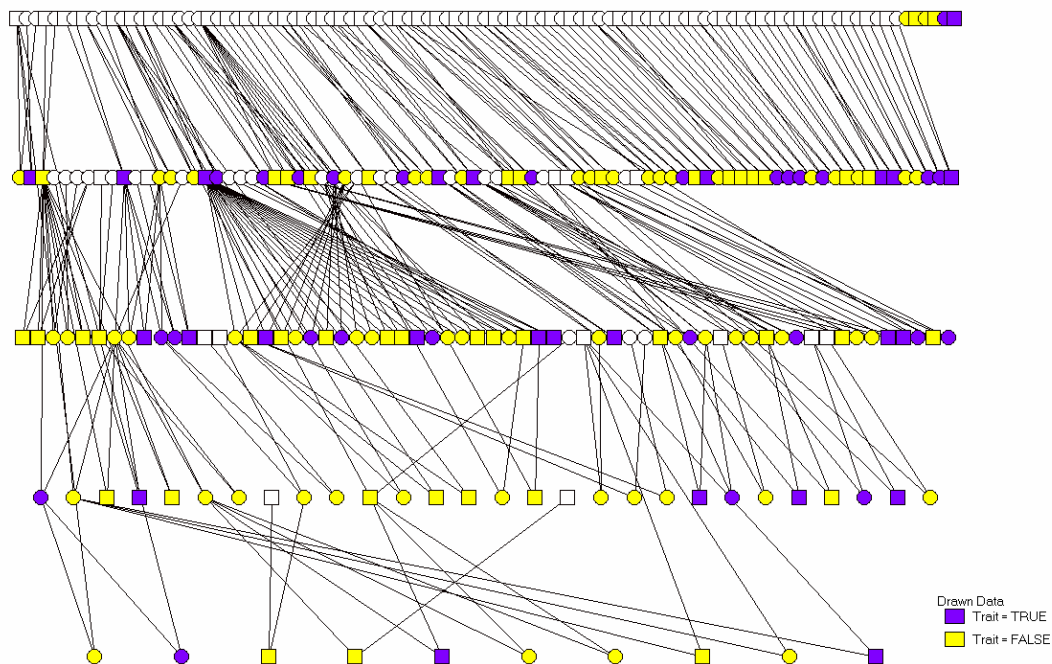


August 12, 2006

Progress Update: *Cairn Terrier DNA Genotyping for Determination of the Genetic Basis for Hepatic Vascular Malformations-Portosystemic Vascular Anomalies (PSVA) & Microvascular Dysplasia (MVD)*
SA Center, DVM, Professor, College of Veterinary Medicine, Cornell University

We have collected blood from 152 dogs including some dogs related over 4 generations and have constructed a pedigree involving 450 dogs. The large pedigree represents dogs in North America and we have identified a number of kindreds that have generated one or more dog with PSVA. We have measured total serum bile acids in each of 152 dogs from which we have collected blood and have mailed results to their owners. In most dogs, we also have conducted an independent study of urine bile acid concentrations. We have isolated DNA from each blood sample and have determined the concentration and quality of DNA in one-half of these (using spectrophotometric estimation and agarose gel electrophoresis).

Below is a simplified graphic demonstration of relationships among Cairn Terriers from which we have collected blood samples (dogs located in Canada, Missouri, California, Colorado, Texas, New Jersey, Pennsylvania, West Virginia, and New York).



Circles = females, Squares = males, purple color = high total serum bile acids, yellow color normal total serum bile acids, no color: blood unavailable for bile acid determinations.

Thirty-five of 152 dogs (23%) have high total serum bile acid concentrations (≥ 25 uM/L). Of 35 dogs with high total serum bile acids, 7/35 (20%) dogs have a PSVA and the remainder (80%) are classified having MVD. Using power prediction software we have determined that we have enough dogs to demonstrate linkage between the hepatic vascular malformation trait and genetic markers. It is important to include dogs with MVD in this data set as this trait is invariably linked with PSVA and otherwise, these dogs (if considered unaffected) would confuse the mathematical probability calculations determining linkage association. We have a few remaining dogs that we would like to

collect blood samples from to fill in data gaps in certain pedigrees to increase the informativeness of their kindred. It is important to have related dogs with and without high total serum bile acid values rather than a large number of dogs that are not directly related. Through the rich cooperation of the Cairn Terrier Breed Clubs this has been possible to achieve.

It is most likely that PSVA and MVD are genetically related representing variants of the same gene(s) based on their dual occurrence in the breeds and kindreds and their similar histological appearance. We still suspect a dominant trait with variable penetrance as the mode of transmission and have not ruled out the possibility of more than one gene. Over the past nine months we have progressed in our genome wide investigation of an informative large pedigree of Tibetan Spaniels (our foundation investigation for these hepatic vascular disorders). Linkage analysis has identified a probable chromosomal location for hepatic vascular malformation and we are currently further investigating this area using Tibetan Spaniel samples with an expanded microsatellite set and SNPs. As this progresses, we will begin to evaluate DNA from Cairn Terriers. The goal is to use the Tibetan Spaniel information to focus on particular loci. Over the next 6 months we plan to demonstrate whether similar linkage exists at the identified chromosomal site in Cairn Terriers. This is the most cost effective method of defining the genetic basis for this disorder, using a foundation investigation and layering evidence with demonstration in a second breed (in this case the Cairn Terrier). The Cairn Terrier and Tibetan Spaniel Breed Clubs have been the most progressive and cooperative with this investigation and have made this important discovery work possible

In addition, we have collected and isolated DNA from 50 small breed dogs with extrahepatic PSVA to demonstrate common involvement of the loci we are investigating. This association analyses will be conducted after we have further refine the genetic loci to conserve resources and focus our investigation.

I am currently on sabbotic leave working primarily on this project with a team of researchers that have focused expertise in the area of genetic investigation. Cornell has provided a post-doc for project support over the next year as well as veterinary student that assisted with pedigree documentation. We hope to have further information to share with your club in the next 6 months.