

ADAMTS17 mutation associated with primary lens luxation is widespread among breeds

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Abstract

Primary lens luxation (PLL) is a well-recognized, painful and potentially blinding inherited ocular condition in dogs. We screened PLL-affected dogs of 30 different breeds, to identify those which carried a previously described c.1473+1 G>A mutation in *ADAMTS17* that is associated with PLL in Miniature Bull terriers, Lancashire Heelers, and Jack Russell terriers. This *ADAMTS17* mutation was identified in PLL-affected dogs from 14 additional breeds. PLL-affected dogs from some breeds (most notably the Shar pei and the Brittany spaniel) did not carry the G1473+1A *ADAMTS17* mutation, indicating they must suffer from a genetically distinct form of the condition. We also estimated the frequency of this *ADAMTS17* mutation in some of the breeds. Our findings indicate the mutation segregates in a large number of different breeds of dog, many of which are terriers or breeds with terrier co-ancestry, but some of which have more diverse origins. Our results also indicate that the mutation is present at high frequency within most of the breeds in which it segregates. In the miniature bull terrier breed estimates of mutation frequency ranged from 0.27 to 0.39, corresponding to 7.3–15.2% PLL-affected dogs in this breed. We also identified an increased risk of PLL associated with heterozygosity at *ADAMTS17*, suggesting that carriers carry a low risk of developing PLL.

Key Words: *ADAMTS17*, canine, inherited, mutation, primary lens luxation

INTRODUCTION

Primary lens luxation (PLL) is a painful and potentially blinding inherited canine ocular condition that has been recognized for at least 100 years.¹ PLL describes the spontaneous displacement of the crystalline lens from its normal position within the patellar fossa of the eye as a result of rupture of the lens zonules and with no antecedent ocular disease. The lens is usually displaced to the anterior chamber where it can cause damage to the anterior chamber structures and acute glaucoma through obstruction of the pupil or the filtration angle.² PLL is invariably bilateral, although a period of several weeks or months might separate luxation of the two lenses. The condition is well documented to affect many breeds of dog, particularly terrier and terrier-type breeds.^{3,4} In breeds that have been studied in depth the condition is believed to be inherited as an autosomal recessive

trait with a variable age-of-onset of between 3 and 8 years.^{4,5} The existence of clinically unaffected carriers and the fact that many dogs do not develop the disease until after they have reproduced mean the condition has been difficult for breeders to control and in some breeds there is a high incidence of the disease. In the UK, PLL is listed on Schedule A of the British Veterinary Association/Kennel Club/International Sheepdog Society Eye Scheme, which lists the known inherited eye diseases in the breeds where it is considered that there is enough scientific information to show that the condition is inherited in that breed and often what the mode of inheritance is (<http://www.thekennelclub.org.uk/item/310>). One breed that is certified for PLL under the Scheme is the Miniature Bull terrier. Between 2000 and 2009, 366 Miniature Bull terriers were ophthalmoscopically examined under the Scheme and 19 (5.19%) were diagnosed as affected with PLL, which corresponds to a mutation

frequency of approximately 0.23, assuming the dogs that were examined were representative of the breed as a whole (data obtained by the Animal Health Trust from records held by the Kennel Club [<http://www.thekennelclub.org.uk/>]). However, it is probable that this incidence is under-representative, because a significant proportion of clinically affected dogs would be unlikely to be presented for routine eye testing under the Scheme due to the severe phenotype of the disease; most dogs presented for examination under the Scheme are breeding animals requiring a clear eye test prior to mating, so older dogs that have been retired from breeding are unlikely to be presented for testing, and owners are also unlikely to present for examination dogs with signs of acute blindness and ocular pain, which are common sequelae of PLL.

Another PLL-certified breed is the Lancashire Heeler. Data from eye examinations performed at Lancashire Heeler club shows between 2003 and 2006 identified signs of PLL in 26/162 (16%) animals examined (D. Gould, data not shown), which corresponds to a mutation frequency of 0.4. This may also be an under-representation for the same reason(s) as cited above. Furthermore, 49/162 (30%) of the Lancashire Heelers examined were ≤ 3 years of age, whereas the mean age of onset of PLL is reported to be 4.5 years (range: 3–7 years),⁴ so it is also possible that some pre-clinical PLL cases were not identified in this survey.

Recently a mutation associated with PLL in Miniature bull terriers, Lancashire Heelers and Jack Russell terriers has been described.⁶ The mutation is a single nucleotide substitution at the 5' end of intron 10 of *ADAMTS17* (*ADAMTS17c.1473+1 G>A*). The nucleotide that is altered is at a position that is highly conserved between eukaryotic splice donor sites and the substitution causes exon-skipping of exon 10 and the resulting frameshift introduces a premature stop codon that is predicted to result in a truncated *ADAMTS17* protein. PLL has been reported to affect many breeds in addition to the Miniature bull terrier, Lancashire Heeler, and Jack Russell terrier so the purpose of this study was:

- (1) to screen PLL-affected dogs of breeds other than the Miniature bull terrier, Lancashire Heeler, and Jack Russell terrier to identify which share the mutation, and
- (2) to estimate the frequency of the mutation in breeds in which it segregates.

MATERIALS AND METHODS

Sample collection

Screening PLL-affected dogs of different breeds Blood and buccal cheek samples from dogs diagnosed with PLL were volunteered by veterinary ophthalmologists, owners, and collaborators. All samples were accompanied by documentation from that examining veterinary ophthalmologist to

confirm a diagnosis of PLL. The samples from PLL-affected dogs were collected from a number of European countries, North America, and Australia. These samples were not included in the mutation frequency estimation.

Estimating the frequency of the mutation in PLL-affected breeds We estimated the frequency of the *ADAMTS17* mutation in three independent population samples of dogs from PLL-affected breeds. All dogs used to estimate the frequency of the PLL mutation were included in one sample only; there was no overlap between the three samples of dogs used. The three populations were:

- (1) dogs living in the UK whose DNA had been submitted to the Animal Health Trust (http://www.aht.org.uk/genetics_tests.html) between 19th October 2009 and 30th April 2010 for the commercially available PLL DNA test. These dogs were referred to as 'AHT tested' dogs.
- (2) dogs whose DNA has been submitted to the Orthopedic Foundation for Animals (<http://www.offa.org/dnatesting/>) between 1st September 2009 and 21st May 2010 for the commercially available PLL DNA test. These dogs are referred to as 'OFA tested' dogs.
- (3) Miniature bull terriers, Lancashire Heelers, and Tibetan terriers living in the UK that were obtained by inviting owners of to submit buccal cheek swabs from their dogs. This invitation was made via Breed Club nominated Health Co-coordinators and was intended to be made to a representative subset of each breed. No clinical information was requested and we selected those dogs that had a unique sire and dam within the cohort for the final analysis. These dogs are referred to as 'unrelated' dogs.

Genomic DNA isolation

For samples that had been submitted as buccal swabs DNA was extracted from two swabs (Medical Packaging Corporation, Camarillo, CA, USA) using QIAamp® DNA Mini Kit (Order number 51304; Qiagen, West Sussex, UK) according to manufacturer's instructions.

For samples that had been submitted as blood, preserved in EDTA, DNA was extracted from 5 mL using the nucleon genomic DNA extraction kit (Tepnel, Manchester, UK) according to manufacturer's instructions. Samples collected in Australia were extracted using a routine salt preparation.

Genotyping

Primary lens luxation-affected dogs and 'AHT tested' dogs were genotyped using the following method:

DNA was amplified using three primers, one forward primer labeled with fluorescent dye 6-FAM (6-FAM-AG-CTACCGGGCATGCACTACA) and two reverse primers, one specific to the normal allele (CCTCCGGTGGCTGCTTACC) and one specific to the PLL allele (TCCTCCGGTGGCTGCTTATC). The reverse primer specific to the

PLL allele had one additional base at the 5' end making the amplification product 1 bp longer than the wildtype amplification product. Two microliters of genomic DNA was added to 10 µL PCR mix (0.2 mM deoxynucleotides, 2.5 mM magnesium chloride, 1× GC buffer [Finnzyme, Espoo, Finland], 0.1 µM primers and 0.2 U/µL Phusion HS DNA polymerase [Finnzyme]). The DNA was amplified using the following conditions: 30 s at 98 °C followed by 35 cycles of 10 s at 98 °C, 5 s at 68 °C, 5 s at 67 °C, 5 s at 66 °C, 5 s at 65 °C, 10 s at 72 °C, and a final period of 60 s at 72 °C. This genotyping method, which is different from that reported previously,⁶ was validated by comparing the results obtained for 96 dogs using this method with those obtained by determining the nucleotide at the 5' end of intron 10 of *ADAMTS17* by direct sequencing. For all 96 dogs the genotyping method described above and direct sequencing gave identical results.

One microliter of PCR reaction was combined with 10 µL Hi-Di formamide on a 96-well PCR plate (ABgene; Abgene Limited, Epsom, UK), heated to 95 °C for 1 min and cooled on ice for 2 min before being loaded onto an ABI 3100 genetic analyzer (Applied Biosystems, Carlsbad, CA) for electrophoresis.

'Orthopedic Foundation for Animals (OFA) tested' dogs were genotyped using the method described previously.⁶

Genotype relative risk

Genotype relative risk and 95% confidence intervals were calculated for heterozygous and homozygous mutant genotypes relative to the homozygous nonmutant genotype using JAVA STAT [<http://statpages.org/ctab2x2.html>], and probabilities relative to the null hypothesis (relative risk of both genotypes is the same) were calculated using chi squared with Yates correction for continuity.

RESULTS

Screening PLL-affected dogs of different breeds

We screened 121 dogs of 30 different breeds that were clinically affected with PLL for the previously described *ADAMTS17* mutation.⁶ These dogs had been collected from a number of European countries, from North America, and Australia. In addition to the three previously reported breeds we identified at least one dog of each of 14 different breeds that was homozygous for the *ADAMTS17* mutation and clinically affected (Australian Cattle dog, Chinese crested dog, Jagdterrier [also known as the German hunt terrier], Parson Russell terrier, Patterdale terrier, Rat terrier, Sealyham terrier, Tenterfield terrier, Tibetan terrier, Toy fox terrier, Volpino Italiano, Welsh terrier, Wire-haired fox terrier, Yorkshire terrier). The results are shown in Table 1.

Estimating the frequency of the mutation in PLL-affected breeds

We estimated the frequency of the *ADAMTS17* mutation in various subsets of breeds known to carry the mutation. The

Table 1. Genotypes of primary lens luxation (PLL)-affected dogs screened for *ADAMTS17* mutation

Breed	No. PLL-affected dogs screened	No.		
		-/-	+/-	+/+
American Cocker Spaniel	1	0	0	1
Australian Cattle Dog	1	1	0	0
Basset Griffon Vendéen	4	0	0	4
Border Collie	1	0	0	1
Brazilian Terrier	1	0	0	1
Brittany Spaniel006C	5	0	0	5
Bull Terrier	2	0	0	2
Cavalier King Charles Spaniel	1	0	0	1
Chinese Crested	8	7	1	0
Czechoslovakian Wolfdog	3	0	0	3
Griffon bruxellois	2	0	0	2
Jagdterrier	11	7	0	4
Kromfohrländer	1	0	0	1
Lhasa Apso	2	0	0	2
Norfolk Terrier	1	0	0	1
Parson Russell terrier	10	9	1	0
Patterdale terrier	2	2	0	0
Rat Terrier	3	2	0	1
Sealyham terrier	1	1	0	0
Shar Pei	4	0	0	4
Shih Tzu	1	0	0	1
Spanish Water Dog	1	0	0	1
Standard Poodle	1	0	0	1
Tenterfield Terrier	9	7	1	1
Tibetan Terrier	27	27	0	0
Toy Fox Terrier	5	5	0	0
Volpino Italiano	2	2	0	0
Welsh terrier	1	1	0	0
Wire-haired fox terrier	10	6	0	4
Yorkshire terrier	1	1	0	0
Total	121			

The genotypes are defined as follows: +/+ denotes the homozygous wildtype genotype, -/- denotes homozygous for the mutation, and +/- denotes a carrier that is heterozygous for the wildtype and mutant alleles.

PLL, primary lens luxation.

reasons for screening different population samples were twofold. Firstly, we wanted to estimate the frequency of the mutation in dogs living in different countries. This would enable breeding advice regarding mutation elimination to be customized if the mutation frequency was found to differ dramatically between populations. Secondly, if the frequency was very different between populations of dogs collected by different methods it would indicate that ascertainment bias was indeed a factor whereas similar frequency estimates across population samples would indicate less of a need to sample and screen different populations when an estimate of mutation frequency was required. The first population sample to be investigated consisted of dogs living in the UK belonging to breeds for which The Animal Health Trust had performed a commercially available DNA test (http://www.aht.org.uk/genetics_pll.html) between 19th October 2009 and 30th April 2010. Data were analyzed for the five breeds for which at least 50 dogs had been screened. The results are shown in Table 2.

Table 2. Summary of *ADAMTS17* genotyping data from Animal Health Trust (AHT) tested dogs from UK

Breed	No. dogs screened with each genotype			Total no. dogs tested	Carriers (%)	Frequency of <i>ADAMTS17</i> mutation	Frequency of wildtype <i>ADAMTS17</i> allele
	-/-	-/+	+/+				
Lancashire Heeler	6	45	69	120	37.50	0.24	0.76
Miniature Bull terrier	9	74	75	158	46.84	0.29	0.71
Parson Russell terrier	2	39	118	159	24.53	0.14	0.86
Sealyham terrier	5	25	21	51	49.02	0.34	0.66
Tibetan terrier	10	156	360	526	29.66	0.17	0.83
				1014			

Table 3. Summary of *ADAMTS17* genotyping data from Orthopedic Foundation for Animals (OFA) tested dogs

Breed	No. dogs screened with each genotype			Total no. dogs tested	Carriers (%)	Frequency of <i>ADAMTS17</i> mutation	Frequency of wildtype <i>ADAMTS17</i> allele
	-/-	-/+	+/+				
American Hairless (Rat) Terrier	0	3	72	75	4.00	0.02	0.98
Chinese Crested	16	221	601	838	26.37	0.15	0.85
Jack Russell Terrier	10	107	192	309	34.63	0.21	0.79
Miniature Bull Terrier	151	550	385	1086	50.64	0.39	0.61
Parson Russell Terrier	6	84	285	375	22.40	0.13	0.87
Rat Terrier	13	125	194	332	37.65	0.23	0.77
Sealyham Terrier	10	72	118	200	36.00	0.23	0.77
Tibetan Terrier	7	126	313	446	28.25	0.16	0.84
Toy Fox Terrier	2	37	87	126	29.37	0.16	0.84
Welsh Terrier	0	23	41	64	35.94	0.18	0.82
				3926			

Table 4. Summary of *ADAMTS17* genotyping data from unrelated dogs

Breed	No. dogs screened with each genotype			Total no. dogs tested	Carriers (%)	Frequency of <i>ADAMTS17</i> mutation	Frequency of wildtype <i>ADAMTS17</i> allele
	-/-	-/+	+/+				
Lancashire Heeler	1	18	27	46	39.13	0.22	0.78
Miniature bull terrier	2	29	31	62	46.77	0.27	0.73
Tibetan terriers	0	11	21	32	34.38	0.17	0.83

The second population sample were dogs belonging to breeds for which a commercially available DNA test had been performed through the Orthopedic Foundation for Animals (<http://www.offa.org/dnatesting/>) between 1st September 2009 and 21st May 2010. The majority of these dogs were living in North America. Data were analyzed for the 10 breeds for which at least 50 dogs had been screened. The results are shown in Table 3.

The third populations to be investigated were Miniature Bull terriers, Tibetan terriers, and Lancashire Heelers living in the UK that had been recruited specifically for the purpose of estimating the *ADAMTS17* mutation frequency. Each dog included in the analysis had a unique sire and dam within the cohort. The results are shown in Table 4.

The frequency of the PLL mutation was calculated for 13 different breeds in at least one of the subsets described.

Three breeds (Sealyham terriers, Lancashire Heelers, and Parson Russell terriers) were included in two of the subsets and two breeds (Miniature bull terriers and Tibetan terriers) were included in all three subsets. Table 5 summarizes the findings for these breeds in the three different sample populations.

The frequency of the *ADAMTS17* mutation ranged from 0.02 (American Hairless terrier) to 0.39 (Miniature bull terriers tested by OFA).

DISCUSSION

Screening PLL-affected dogs of different breeds

A mutation in *ADAMTS17* associated with PLL has previously been described in three different breeds, the Miniature bull terrier, the Lancashire heeler and the Jack Russell

Table 5. Comparison of *ADAMTS17* mutation frequency between different subsets of dogs screened

	AHT tested dogs		OFA tested dogs		Unrelated dogs	
	Frequency of <i>ADAMTS17</i> mutation	Frequency of wildtype <i>ADAMTS17</i> allele	Frequency of <i>ADAMTS17</i> mutation	Frequency of wildtype <i>ADAMTS17</i> allele	Frequency of <i>ADAMTS17</i> mutation	Frequency of wildtype <i>ADAMTS17</i> allele
Lancashire Heeler	0.24	0.76	NA	NA	0.22	0.78
Miniature bull terrier	0.29	0.71	0.39	0.61	0.27	0.73
Parson Russell terrier	0.14	0.86	0.13	0.87	NA	NA
Sealyham terrier	0.34	0.66	0.23	0.77	NA	NA
Tibetan terrier	0.17	0.83	0.16	0.84	0.17	0.83

AHT, Animal Health Trust; OFA, Orthopedic Foundation for Animals.

terrier.⁶ PLL is well documented to affect many breeds of dog^{3,7} so we screened clinically affected dogs of 30 additional breeds to investigate if any others shared the mutation. We identified the identical mutation in 14 additional breeds as detailed above. For four breeds (Australian Cattle dog, Sealyham terrier, Welsh terrier, and Yorkshire terrier) we only screened a single affected dog, all four of which were homozygous for the mutation. The Yorkshire terrier has not been described, to our knowledge, as being predisposed to PLL and the possibility that the Yorkshire terrier in our study was a cross-breed should be considered. In view of the small numbers of individuals analyzed for these breeds it is not formally possible to exclude the PLL mutation from involvement in the development of the condition in these breeds; analysis of greater numbers of dogs would be necessary to draw that conclusion with more confidence. For all other breeds we identified at least two clinically affected homozygous dogs so we are confident that these breeds do in fact segregate the mutation. Most of the breeds are terriers, or breeds with presumed terrier co-ancestry, a group of breeds that are themselves fairly divergent⁸ but some of the breeds carrying the PLL mutation appear even more distantly related. For example, the Tibetan terrier is more closely related to breeds of ancient Asian origin than to the majority of modern terrier breeds, and unsurprisingly the Australian cattle dog is more closely related to herding/sight hound breeds than to the majority of terriers.⁸ The Lancashire Heeler was developed by crossing a terrier with a herding dog and shows a close relationship to the Border Collie⁸ although interestingly the single Border Collie with PLL that we screened did not carry the *ADAMTS17* mutation. Studies to investigate the relatedness of various dog breeds consistently indicate the Miniature Bull terrier is more closely related to mastiff-type breeds than to the majority of the small terriers.^{8,9} The presence of the mutation in a large number of breeds with diverse origins and phenotypic characteristics suggests it is an ancient mutation that arose in a dog that served as a common founder for a large number of modern breeds, prior to the rigorous breed isolation that exists today. A comparable origin has been speculated for the mutation that causes progressive rod-cone degeneration which has

been identified in upward of 20 different breeds, many of which also have diverse ancestries.^{10,11}

Sixteen of the breeds we screened did not carry the *ADAMTS17* mutation. Several of these breeds, such as the Shar Pei and the Brittany spaniel, are at increased risk of PLL¹² indicating they must suffer from a genetically distinct form of the condition caused either by a mutation in a different gene or a different mutation in *ADAMTS17*. Currently we cannot distinguish between these two possibilities because we have relied on a simple test for a single mutation and have not resequenced *ADAMTS17* in these breeds.

The majority of affected dogs that carried the PLL mutation were homozygous but a single clinically affected Parson Russell terrier, Chinese Crested dog, and Tenterfield terrier were heterozygous for the mutation. This observation is consistent with our previous observation that heterozygotes might be at increased risk of PLL,⁶ compared to dogs that are homozygous for the wildtype allele of *ADAMTS17* (discussed further below) although it is also formally possible that additional mutations, either in *ADAMTS17* or elsewhere in the genome, are contributing to the phenotype of these dogs.

We identified Rat terriers, Jagdterriers, and Wire-haired fox terriers that were homozygous for the PLL mutation, but also dogs of the same three breeds that were reported to be clinically affected but that were homozygous for the wildtype allele. PLL may be genetically heterogeneous in these three breeds, although we cannot formally exclude the possibility that the dogs that did not carry the mutation had been mis-diagnosed.

Estimating the frequency of the mutation in PLL-affected breeds

The frequency of the *ADAMTS17* mutation ranged from 0.02 (American Hairless terrier tested by OFA) to 0.39 (Miniature bull terriers tested by OFA). The low frequency in the American Hairless terrier breed was an exception, however, with the mutation frequency being at least 0.13 in all other breeds analyzed. None of the subsets of dogs analyzed are likely to be truly representative of their respective breeds. Breeders may be motivated either to refrain from testing or to intensively test particular lines or pedigrees that

have been known to produce affected dogs, so estimating the frequency of any mutation in such a subset of dogs may result in under or over estimation of the true frequency in the overall population. It is possible, therefore, that the mutation frequencies we have calculated in the 'AHT tested' and 'OFA tested' groups of dogs differ from those in the general UK and North American populations. Furthermore, PLL-affected dogs showing the common secondary signs of acute blindness and ocular pain will be unlikely to have been included in these two groups.

In an attempt to overcome these obvious biases we recruited additional Miniature bull terriers, Lancashire Heelers, and Tibetan terriers, living in the UK, that had neither been submitted for DNA testing, nor had their eyes examined by a veterinary ophthalmologist, so were not selected on the basis of any phenotypic criteria. Once again, this group of dogs, although unrelated at parental level, cannot be considered truly random, because they are likely to belong to a subset of owners who for example, may be more aware of breed-associated inherited conditions; may be more likely to be members of Breed Clubs; and may be familiar with the dog press and websites.

However, while acknowledging the ascertainment bias associated with all three of our subsets of dogs, we observe remarkable consistency between subsets in the frequency of the PLL mutation, especially for the two subsets of dogs from the UK ('AHT tested' and 'Unrelated' dogs) (Table 5). The frequency of the mutation is high in most of the breeds analyzed (Tables 2–4). In Miniature Bull terriers it ranges from 0.27 ('unrelated' dogs) to 0.39 (OFA tested dogs); corresponding in a fully recessive model to between 7.3% and 15.2% affected PLL dogs in these populations. The mutation frequency of 0.23 estimated previously from the clinical examination of Miniature Bull terriers from the UK, although lower, is not dramatically different from the estimates of 0.27 and 0.29 obtained from Miniature Bull terriers living in the UK in this study. The frequency of the mutation in Tibetan terriers was very consistent between all three subsets of dogs analyzed (between 0.16 and 0.17). This frequency suggests approximately 2.5% of this breed will be clinically affected with PLL during their lives. During this study two very similar estimates were obtained for the frequency of the *ADAMTS17* mutation in Lancashire Heelers in the UK (0.24 in AHT tested dogs and 0.22 in unrelated dogs). Both estimates are lower than that obtained through clinical examination of dogs between 2003 and 2006, as discussed in the Introduction. The difference might reflect a genuine reduction in mutation frequency over the last few years, or might result from an over representation of affected dogs at eye testing events. All populations analyzed were in Hardy–Weinberg equilibrium with respect to this mutation (data not shown). The fact that the PLL mutation is at such high frequency in many breeds could mean that dog breeders are well advised to consider breeding from heterozygote dogs, at least in the short-term, provided they are mated to dogs homozygous for the wildtype allele. This

practice would enable the mutation to be eliminated from a breed without a significant loss of genetic diversity.

Heterozygous risk

Some affected dogs were heterozygous, so it is important to know whether there is increased risk of PLL associated with heterozygosity at *ADAMTS17*. In our previous study 15.5% of miniature bull terriers affected with PLL (16/103), collected from these populations, but independently of the population sample groups presented here, were heterozygous for the *ADAMTS17* mutation.⁶ Assuming that ascertainment of PLL status is equally efficient for all affected genotypes, the population sample genotypes here can be used to estimate the relative risk of heterozygous and homozygous mutation causing PLL. The risks of a clinically assigned PLL phenotype conferred by a given mutant genotype relative to the risk of the homozygous wildtype genotype are: for heterozygotes – 5.89 (CI₉₅ = 1.36–25.5; *P* [Yates] = 0.013); and for homozygotes – 122.2 (CI₉₅ = 30.4–490.8; *P* [Yates] < 0.0001). The second figure is typical of many presumed monogenic diseases. In the previous study,⁶ 161/176 dogs of all breeds with the A/A (homozygous mutant) genotype had suffered lens luxation at the time of sample collection. All but one of the A/A dogs with normal phenotype were under 6 years old, and the average age of this group was 2.4 years. It seems likely that all homozygous mutant dogs develop PLL if they live long enough. These figures also suggest that nearly 5% of heterozygotes in this breed will develop PLL. Overall lifetime incidence of clinically recognizable PLL across the whole breed will be around 12%, with the great majority of this associated with the *ADAMTS17* mutation, but background incidence from trauma, other environmental factors or phenocopies located elsewhere in the genome of around 0.4%. The number of heterozygotes observed with PLL in the miniature bull terrier was higher than that seen in other breeds, where we have too few heterozygous affected animals to prove significant heterozygote risk of PLL. However, as it is likely that carriers are at a low risk of developing PLL it would seem prudent for owners to have such dogs examined by a veterinary ophthalmologist on a regular basis throughout their lives, so clinical signs can be detected at the earliest opportunity.

It is interesting to speculate why the *ADAMTS17* mutation frequency is high in so many breeds in which it segregates (Table 3), when it is associated with such a serious and debilitating condition as PLL. Although heterozygous individuals have an increased risk of the developing PLL compared to dogs that do not carry the mutation, the large majority of carriers remain clinically unaffected during their lives. A reservoir of healthy dogs that carry a disease-associated mutation presents a challenge for dog breeders wishing to eliminate the mutation from a breed. For a relatively late-onset disease, such as PLL, the problem is confounded as many dogs have reproduced prior to developing clinical signs of disease. These difficulties notwithstanding, it is

reasonable to assume selection pressure against the mutation will have operated in most breeds. In human patients mutations in *ADAMTS17* have been associated with short stature, as well as with ectopia lentis and other ocular anomalies.¹³ Although PLL affected dogs do not apparently suffer ocular anomalies apart from secondary glaucoma³ it is interesting to observe that most of the breeds carrying the mutation are small breeds. If the mutation is associated with shortness in dogs we may speculate (in the absence of data) that it has been inadvertently selected for, especially in breeds such as the Miniature Bull terrier, where the breed standard dictates a maximum desirable height.

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