Genetic Diversity Testing for Samoyed

Overview

The Veterinary Genetics Laboratory (VGL), in collaboration with Dr. Niels C. Pedersen and staff, has developed a panel of short tandem repeat (STR) markers that will determine genetic diversity within the genome and in the Dog Leukocyte Antigen (DLA) class I and II regions.

Price: $80
Price reduced to $70 when combined with a diagnostic test.

ORDER TEST KITS
Allow 5-10 business days for results.

Results reported as:
Short tandem repeat (STR) loci: A total of 33 STR loci from across 25 of 39 chromosomes were used to gauge genetic diversity within an individual and across the breed. The alleles inherited from each parent are displayed graphically to highlight heterozygosity.

DLA haplotypes: Seven additional STR loci linked to the DLA class I and II genes were used to identify genetic differences in regions regulating immune responses and self/non-self-recognition. Problems with self/non-self-recognition, along with non-genetic factors in the environment, are responsible for autoimmune disease, allergies and reduced resistance to certain infections.

Internal Relatedness: The IR value is a measure of genetic diversity within an individual that takes into consideration both heterozygosity of alleles at each STR loci and their relative frequency in the population. Therefore, IR values heterozygosity over homozygosity and uncommon alleles over common alleles. IR values are unique to each dog and cannot be compared between dogs. Two dogs may have identical IR values but with very different genetic makeups.

I. Introduction

A. Breed history

The Samoyed is considered one of the basal breeds developed in the late Victorian era from indigenous Laika used by the Nenets (Samoyede) peoples of Northwest Russia and Siberia. Like many pure breeds, Samoyed have a convoluted history with several versions [1].[a-f] Samoyed belong to what is known as the Arctic or Nordic group that include the Alaskan Malamute, Chow Chow, German Shepherd and several other Spitz-type dogs. The peoples of the arctic regions have kept dogs over millennia and selectively bred dogs that were most helpful to them for hunting, guarding camps and villages, pulling sledges and carrying packs, companionship and even sharing the hearth and bed. Dogs of this utilitarian type first appeared in Russian dog shows in the late 19th century.[g] Robert Peary was the first westerner to use dogs to pull sledges during his expeditions to Greenland in 1891-92.[h] Sledge dogs were first brought keenly to the attention

of the Western world by Fridtjof Nansen, a Norwegian explorer who used teams of "Samoyedes" as sledge dogs during his 1894 expedition to the North Pole [2]. Although the breed traces its ancestry to these early sled and sledge dogs of the North, the modern Samoyed is largely an idealized and stereotypic version of a variety of rugged and thick coated medium-sized dogs that evolved in the arctic regions. However, with proper conditioning Samoyed are still very capable of pulling sleds.

The rise of the Samoyed in America and other countries was preceded in the UK by the activities of Sir Ernest Kilburn-Scott and his wife Clara.[a-e, i] The first foundation dog for the breed in England was a brown and white dog named Sabarka that was purchased by Ernest in Archangel, Russia in 1889 as a gift for Clara.[i] However, it was the white dogs that he encountered in his travels that caught his fancy and led them to purchase a cream-colored bitch named Whitey Petchora [1]. The offspring of these two dogs were bred by the Kilburn-Scott's and others and helped popularize the breed. During their stay in Australia from 1907-1910, the Kilburn-Scott's became involved in the plight of former sledge dogs returning from Antarctic explorations. Returning dogs were sometimes returned to sledge-dog breeding kennels in South New Zealand, but most were subjected to strict, long-term, expensive and therefore fatal quarantines in New Zealand and some were exhibited in zoos in New Zealand and Australia. Dogs saved from such conditions and environments were a common source of breeding stock for evolving Arctic-type breeds during the late 19th and early 20th centuries. Ernest and Clara brought several of these dogs back with them from Australia in 1910, including a large white dog named Antarctic Buck, which they acquired from the Sydney zoo [1]. Antarctic Buck died of canine distemper after coming to the UK but at least five of his progeny survived and were integrated into the Samoyede in the UK. The Kilburn-Scott's showed their dogs in the foreign dog classes in the UK for several years and the breed was given conditional registration in 1902 and formal recognition under the original name Samoyede in 1912 by the UK kennel club. The name was later changed to Samoyed by both the AKC and UKC. The American connection was stimulated when a member of a famous European royal family, Rose de Mercy-Argenteau (Princess de Montglyon), was smitten with a large white champion Russian Samoyed named Moustan entered the 1902 St. Petersburg dog show by Grand Duke Michael of Russia.[g] Moustan was given to the countess as a gift and brought along with three other Samoyed to the USA in 1904. Moustan was shown extensively in America and the breed recognized by the AKC in 1906.

Changes in Samoyed population size over time

The Samoyed breed has retained an average popularity compared to other breeds over the century and registration numbers have been relatively steady in in both the UK and USA except for a brief decline during WWI and a sharp rise after 1980 to a peak of 1200 registrants in the UK in 1995 and decline to pre-peak levels of 300 by 2015.[j] The Samoyed currently ranks 64th in popularity in the USA with a steady 1500 new registrations each year per AKC records.

II. Baseline genetic diversity testing and what it tells about Samoyeds

A. Standard genetic assessments based on 33 STR loci on 25 chromosomes and allele frequencies

STR markers are highly polymorphic and have great power to determine genetic differences among individuals and breeds. The routine test panel contains 33 STRs, those recommended for universal parentage determination for domestic dogs by the International Society of Animal Genetics (ISAG) and additional markers developed by the VGL for forensic purposes. Each STR locus manifests several different genetic configurations known as alleles. Everyone inherits one of these alleles from the sire and the other from the dam. Table 1 lists the alleles recognized at each STR locus among 190 Samoyed tested to date, as well as listing the frequency of any given allele in the population.

Table 1: STR alleles from 33 genomic loci and their frequencies in Samoyeds

(link to table 1)

The distribution of alleles at each of the 33 autosomal STR loci is typical of most pure breeds of dogs. Only a portion of the alleles known to exist in all dogs occurs at each locus in Samoyed, and one allele was virtually fixed at locus INRA21 (94% of dogs) (Table 1). Alleles at two other loci (LE1004, VGL1828) were found in 75 and 80% of dogs tested. A relatively small number of other alleles were identified at each locus with two to four of them predominating. These dominant alleles have been strongly conserved throughout breed evolution indicating that these regions of the genome are very important in preserving certain breed-defining phenotypic traits.

1. Standard genetic assessment values for individual STR loci

The allele frequencies (Table 1) can be used to do a standard genetic assessment of heterozygosity at each STR locus (Table 2). The value Na is the number of alleles that are observed at each locus for a specific breed, while Ne is the number of effective alleles observed at each locus. Effective alleles are those alleles that contribute to the bulk of the genetic diversity. The Na values for individual STR loci for this population of 190 Samoyed ranged from a low of 3 to a high of 12, while the Ne ranged from 1.13 to 5.93.

Observed heterozygosity (Ho) is based on the actual allele frequencies at each STR locus and their distribution, while the expected heterozygosity (He) is the value that would be predicted if allele frequencies were in the equivalent of Hardy-Weinberg equilibrium (HWE). HWE is achieved when all alleles at a specific locus within a specific population or breed are segregating randomly. A Ho value of 1.0 would be observed when alleles at each locus are unique to everyone in the population. A Ho value of 0.00 would occur if there is no heterozygosity, e.g. every individual has the same alleles at a given locus. Ho ranged from 0.111 to 0.789, indicating a large range of genetic diversity from one locus to another. The He ranged from 0.115 to 0.825 across the 33 STR loci (Table 2). The Ho and He values were used to calculate the F value (1-Ho/He), a measure of deviation from HWE. F values ranged from -0.084 to +0.175. Twenty-five loci had FIS values greater than zero, while 6 loci had FIS scores less than zero, indicating an excess of inbred alleles in the population.

Table 2. Standard Genetic Assessment for Samoyed using 33 STR loci (Updated October 09, 2019)

#	Locus	N	Na	Ne	Ho	He	F
1	**AHT121**	192	9	4.574	0.745	0.781	0.047
2	**AHT137**	192	7	4.497	0.771	0.778	0.009
3	**AHTH130**	192	4	1.982	0.474	0.495	0.043
4	**AHTh171-A**	192	5	3.587	0.724	0.721	-0.004
5	**AHTh260**	192	7	4.263	0.714	0.765	0.068
6	**AHTk211**	192	3	2.071	0.521	0.517	-0.007
7	**AHTk253**	192	6	3.516	0.651	0.716	0.090
8	**C22.279**	192	7	3.734	0.693	0.732	0.054
9	**FH2001**	192	8	2.915	0.609	0.657	0.072
10	**FH2054**	192	8	2.222	0.516	0.550	0.062
11	**FH2848**	192	8	3.567	0.620	0.720	0.139
12	**INRA21**	192	3	1.129	0.109	0.114	0.041
13	**INU005**	192	5	3.093	0.615	0.677	0.092
14	**INU030**	192	4	2.455	0.646	0.593	-0.090
15	**INU055**	192	5	1.945	0.500	0.486	-0.029
16	**LEI004**	192	3	1.617	0.385	0.381	-0.010
17	**REN105L03**	192	10	3.044	0.698	0.672	-0.039
18	**REN162C04**	192	5	2.451	0.536	0.592	0.094
19	**REN169D01**	192	8	3.676	0.714	0.728	0.020
20	**REN169O18**	192	5	2.588	0.563	0.614	0.083
21	**REN247M23**	192	7	3.609	0.688	0.723	0.049
22	**REN54P11**	192	5	2.348	0.510	0.574	0.111
23	**REN64E19**	192	4	2.969	0.677	0.663	-0.021
24	**VGL0760**	192	9	5.912	0.792	0.831	0.047
25	**VGL0910**	192	8	4.812	0.776	0.792	0.020
26	**VGL1063**	192	7	2.280	0.552	0.561	0.017
27	**VGL1165**	192	9	4.573	0.646	0.781	0.173
28	**VGL1828**	192	7	1.493	0.276	0.330	0.164
29	**VGL2009**	192	6	3.844	0.714	0.740	0.036
30	**VGL2409**	192	7	4.944	0.807	0.798	-0.012
31	**VGL2918**	192	6	2.359	0.599	0.576	-0.040
32	**VGL3008**	192	12	5.764	0.776	0.827	0.061
33	**VGL3235**	192	6	3.394	0.688	0.705	0.025

2. Using allele frequency data to do standard genetic assessments on the entire population.

Allele frequencies across all 33 STR loci taken from Table 1 can also be used to calculate a mean observed heterozygosity (Ho) and expected heterozygosity (He) for the 190 Samoyed (Table 3). The population of Samoyed, when examined as a whole population, had a mean number of alleles (Na) of 6.455 across all 33 genomic STR loci. The mean effective alleles (Ne) per locus were 3.249. This means that about one half of available alleles accounted for most of the diversity. The Na and Ne values for Samoyed are lower than genetically diverse breeds such as Miniature and Toy Poodles, similar to Labrador and Golden Retrievers, somewhat more than Flat Coated Retrievers and much greater than for Swedish Vullhund.

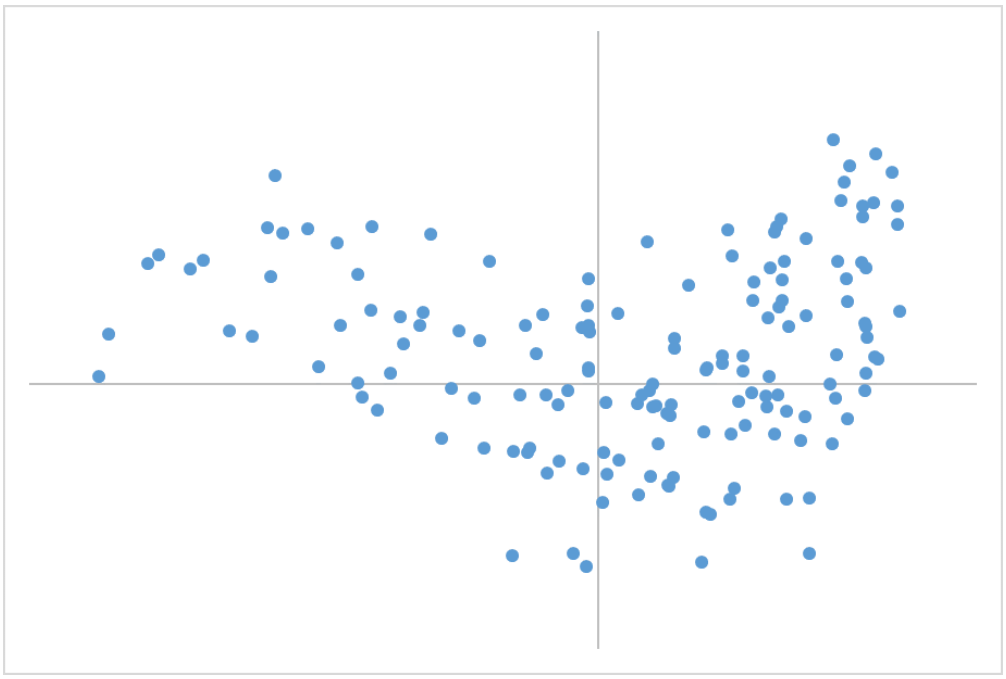
Table 3. Summary of Standard Genetic Assessment for Samoyed using 33 STR loci (Updated October 09, 2019)

	N	Na	Ne	Ho	He	F
Mean	192	6.455	3.249	0.615	0.642	0.041
SE		0.364	0.207	0.026	0.027	0.010

The mean observed heterozygosity (Ho) across the population was 0.619, which was lower than the expected heterozygosity (He) of 0.650. This resulted in an F value (0.047) that is slightly higher than zero, indicating that a small proportion of individuals were more inbred than the average Samoyed. The relatively high breed-wide heterozygosity and small excess of less heterozygous individuals is similar to many other pure breeds and suggests that breeders have done a reasonable job in selecting the least related parents. However, Ho and He are average scores for the population and may not accurately reflect individuals that are more outbred or inbred than the overall population. This is better reflected by the IR scores (see section on internal relatedness).

Principal coordinate analysis (PCoA) uses genetic distance based on allele sharing to demonstrate genetic differentiation between individuals in related or unrelated populations. The resulting data is multi-dimensional but can be accurately portrayed in a two-dimensional graph by selecting values from the two coordinates that represent the greatest proportions of individuals (coordinate 1 and 2 in this case). Figure 1 is a PCoA plot of 190 Samoyed. The plot shows that the 190 dogs cluster as a single breed, although the plot is somewhat diffuse with several more distant outliers. This suggests that there is still phenotypic, and therefore genotypic, variation in the breed.

Fig. 1: PCoA plot of 190 Samoyed from around the world. The relationship between various individuals varied with no distinct clustering in the center of the graph.



B. The use of genomic allele frequencies to determine internal relatedness

1. Internal relatedness of individuals and the population as a whole

Genetic assessments such as those presented in Table 4 are indicators of population-wide heterozygosity and do not reflect the genetic diversity of individuals within the population. The genetic diversity of an individual dog is largely determined by the diversity inherited from each of its parents. Internal Relatedness (IR) is a calculation that has been used to determine the relative genetic contributions of both parents to an individual. The IR calculation evaluates homozygosity and uses allele frequencies to give more importance to rare and uncommon alleles. IR scores of all individuals in a population can be graphed to form a curve ranging from -1.0 to +1.0. A dog with a value of -1.0 would have parents that were totally unrelated at all 33 STR loci), while a dog with an IR value of +1.0 has parents that were genetically identical at all loci. An IR value of +0.25 would equivalent to offspring of full sibling parents from a random breeding population. IR values >0.25 occur when the parents of the full sibling parents were themselves highly inbred.

The IR curve (red line Fig. 3) calculated from the 190 Samoyed ranged from around -0.276 for the most outbred dog to +0.498 for the most inbred, with a mean value for the population of +0.042 (Table 5). One half of the dogs had IR values over 0.042 and one quarter had values of 0.132-0.503. A value of 0.25 would be average for a litter of puppies born to parents that were full siblings in a random breeding population. An IR value of 0.498 would only occur if the full sibling parents were more highly related to each other that the average dog in the population. This highly inbred quarter of the population is balanced by the one quarter of outbred dogs with IR values of -0.276 to -0.056 to This balancing of highly outbred and inbred dogs is why the Ho, He and F values calculated for the whole population using allele frequencies of the 33 genomic STR loci gave the impression that the population of 190 dogs tested were being randomly bred.

Table 5: Statistical breakdown of IR and IRVD values used to create population curve shown in Figure 3. (n=190)

	IR	IRVD
Min	-0.276	-0.118
1st Qu	-0.056	0.192
Mean	0.042	0.276
Median	0.027	0.269
3rd Qu	0.124	0.356
Max	0.498	0.682

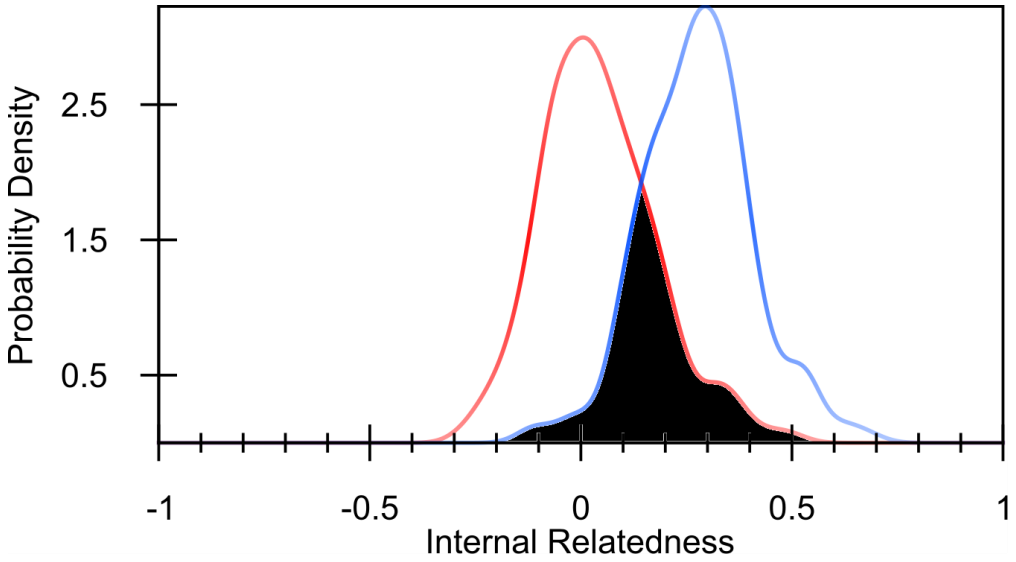


Fig. 3. Distribution of IR estimated in Samoyed (n=190) based on intra-breed diversity (red line), compared with IR adjusted for available genetic diversity that has been lost during breed development (blue line). The black area is an estimate of lost genetic diversity using randomly breeding village dogs as the standard.

The IR values can be adjusted in such a way as to provide an estimate of the amount of genetic diversity that has been during breed evolution. This is done by using allele frequencies obtained from DNA of present day village dogs from the Middle East, SE Asia and Island Pacific nations. This population retains most of the genetic diversity that still exists in dogdom. The adjusted IR value is known as IR-village dogs or IRVD. The IRVD curve (blue line) was shifted to the right, reflecting the loss of genetic diversity that occurred during breed development (Fig. 3). The dark area of overlap between the IR and IRVD curves is an estimate of the amount of genetic diversity in modern village dogs that is still present in Samoyed (i.e., 35%). This is lower than the 60% or so retained diversity seen in genetically diverse breeds such as the toy poodle or 54% in Labrador Retriever, but higher than the 7% observed in Swedish Vallhund, 15% in Doberman Pinschers, or 27% in Shiloh Shepherd.

C. DLA Class I and II Haplotype frequencies and genetic diversity

The DLA consists of four gene rich regions making up a small part of canine chromosome 12. Two of these regions contain genes that help regulate normal cell- (Class I) and antibody-mediated (Class II) immunity. Polymorphisms in these regions have also been associated with abnormal immune responses responsible for autoimmune diseases. The Class I region contains several genes, but only one, DLA-88, is highly polymorphic (with many allelic forms) and is therefore most important for immune regulation. Specific alleles at the four STR loci associated with the DLA88 are linked together in various combinations, forming specific haplotypes (Table 4). Groups of genes and their alleles inherited as a block, rather than singly, are called haplotypes. The class II region also contains several genes, three of which are highly polymorphic, DLA-DRB1, DLA-DQB1 and DLA-DQA1. Specific alleles at STR loci associated with each of the three Class II genes are strongly linked and inherited as a single block or haplotype (Table 6). One haplotype comes from each of the parents.

The STR-based haplotype nomenclature used in this breed diversity analysis is based on numerical ranking with the first haplotypes identified in Standard Poodles being named 1001, 1002, ... for class I haplotypes and 2001, 2002, ... for class II haplotypes. It is common for various dog breeds to share common and even rare haplotypes, depending on common ancestry.

Table 6. Samoyed DLA class I and Class II haplotypes and their frequencies

DLA Class I Haplotype Frequencies (Updated Oct 9, 2019)		
DLA1 #	**STR types**	**Samoyed (n=189)**
1006	387 375 293 180	0.005
1009	382 377 277 184	0.124
1011	376 365 281 180	0.272
1012	388 369 289 188	0.013
1014	375 373 287 178	0.003
1061	380 365 281 183	0.003
1068	380 373 287 181	0.042
1085	376 373 277 186	0.003
1152	390 373 281 180	0.495
1153	389 373 287 183	0.019
1157	386 373 281 180	0.003
1158	390 371 281 180	0.003
1160	386 369 289 176	0.016

DLA Class II Haplotype Frequencies (Updated Oct 9, 2019)		
DLA2 #	**STR types**	**Samoyed (n=189)**
2002	343 327 280	0.003
2003	343 324 282	0.013
2007	351 327 280	0.005
2015	339 327 280	0.016
2022	339 327 282	0.108
2024	343 323 280	0.016
2042	341 324 286	0.003
2050	341 327 284	0.003
2053	343 324 280	0.558
2095	355 322 280	0.159
2096	351 322 280	0.114
2099	345 324 276	0.003

We have identified 13 STR-associated DLA Class I and 12 DLA Class II haplotypes among the Samoyeds that we tested (Table 6). DLA class I haplotypes 1085, 1152, 1153 1157 and 1158 and class II haplotypes 2095, 2096 and 2099 have not been seen in other breeds studied to date. The 1152 and 2053 haplotypes were found in 50-56% of individuals, respectively. These two haplotypes were in strong linkage, forming a unique 1152/2053 haplotype. Other unique linked DLA class I/II haplotypes include 1011/2095, 1011/2096, 1068/2053, 1085/2002, 1152/2053, 1153/2053, 1157/2053 and 1158/2099. These unique haplotypes have been preserved in Samoyed because of their direct or indirect association with genotypes that determine the phenotypes that are highly desired by the breed standard.

A lack of genetic diversity in the DLA class I and II region and over-representation of certain haplotypes has been linked to a higher incidence of autoimmune disorders, allergies, and susceptibility to certain infections. Genetic diversity in the DLA class I and II regions of Samoyed is limited compared to breeds such as the Standard, Miniature and Toy Poodles, Havanese, Italian Greyhound, Golden and Labrador Retriever, and Akita. It is similar to the diversity seen in Doberman Pinscher, Giant Schlnauzer and Flat Coated Retriever, and more than breeds such as the Alaskan Klee Kai, Black Russian Terrier, Swedish Vallhund and Shiloh Shepherd.

The genes found in the DLA region are strongly linked in individual dogs and undergo limited recombination over the time which breeds such as the Samoyed evolved. These large regions of strong linkage are inherited as blocks from each parent and are therefore useful in estimating the founders that contributed most to the breed (see above) and the relationship between breeds (Table 7). The 2053 haplotype was found at high frequency in Swedish Vallhund (0.49), Flat Coat Retriever (0.126), and Shiloh Shepherd (0.317), but not in association with 1152 (Table 7). Several other class I and II haplotypes found in Samoyed were shared at low frequency with other breeds, Labrador Retriever, Golden Retriever, Havanese, Miniature Poodle, Standard

Poodle, and Swedish Vallhund. Unfortunately, no comparative data was available for Nordic breeds such as the Siberian Husky and Malamute.

Table 7. Comparison of DLA class I and II haplotypes between Samoyed and several other breeds.

DLA Class I Haplotype Frequencies (Updated Jul 9, 2018)

DLA1 #	STR types	Alaskan Klee Kai (n=461)	Black Russian Terrier (n=113)	Shiloh Shepherd (n=115)	Labrador Retriever (n=128)	American Akita (n=84)	Japanese Akita (n=311)	Blend Akita (n=53)	Golden Retriever (n=690)	Doberman Pinscher (n=448)	Flat Coated Retriever (n=386)	Havanese (n=379)	Samoyed (n=187)	Miniature Poodle (n=245)	Swedish Vallhund (n=142)	Poodle (n=2170)	Giant Schnauzer (n=165)	English Bulldog (n=163)	Biewer (n=103)	Biewer Yorshire Terrier (n=53)	Biewer Terrier (n=65)	Yorkshire Terrier (n=16)	Biro Biewer (n=3)	Italian Greyhound (n=691)	Toy Poodle (n=109)
1006	387 375 291 180	--	--	0.04	--	0.027	0.054	--	--	0.0145	--	--	0.045	0.005	0.004	0.254	0.0461	0.048	0.003	--	--	--	--	--	--	--
1009	382 377 277 184	--	--	--	--	--	--	--	--	--	--	--	--	0.123	0.082	0.056	0.018	--	--	--	--	--	--	--	--	0.083
1011	376 365 281 180	--	0.06	--	--	--	--	--	--	0.0007	--	--	--	0.273	0.004	--	0.0212	0.015	--	--	--	--	--	--	--	0.028
1012	388 369 289 188	--	--	--	--	--	--	--	--	0.0014	--	--	0.017	0.013	0.047	--	0.0101	--	0.414	0.175	0.151	0.262	0.28	0.2	0.0094	0.032
1014	375 373 287 178	--	0.166	--	0.04	--	--	--	--	0.0399	--	--	0.036	0.003	0.004	--	0.0078	0.245	--	0.019	0.009	0.062	0.09	--	--	0.037
1063	380 365 281 183	--	0.047	--	--	--	0.036	0.18	0.075	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--
1068	380 373 287 181	--	--	--	0.248	0.047	--	--	--	0.05	--	0.268	0.016	0.043	0.014	0.342	--	0.036	--	--	0.009	--	--	--	--	0.014
1085	376 373 277 186	--	--	--	--	--	--	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--
1152	390 373 281 180	--	--	--	--	--	--	--	--	--	--	--	--	0.495	--	--	--	--	--	--	--	--	--	--	--	--
1153	389 373 287 183	--	--	--	--	--	--	--	--	--	--	--	--	0.019	--	--	--	--	--	--	--	--	--	--	--	--
1157	386 373 281 180	--	--	--	--	--	--	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--
1158	390 371 281 180	--	--	--	--	--	--	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--
1160	386 369 289 176	--	--	0.031	--	0.006	--	--	--	--	--	--	--	0.016	--	--	--	--	--	--	--	--	--	--	--	--

DLA Class II Haplotype Frequencies (Updated Jul 9, 2018)

DLA2 #	STR types	Alaskan Klee Kai (n=461)	Black Russian Terrier (n=113)	Shiloh Shepherd (n=115)	Labrador Retriever (n=128)	American Akita (n=84)	Japanese Akita (n=311)	Blend Akita (n=53)	Golden Retriever (n=690)	Doberman Pinscher (n=448)	Flat Coated Retriever (n=386)	Havanese (n=379)	Samoyed (n=187)	Miniature Poodle (n=245)	Swedish Vallhund (n=142)	Poodle (n=2170)	Giant Schnauzer (n=165)	English Bulldog (n=163)	Biewer (n=103)	Biewer Yorshire Terrier (n=53)	Biewer Terrier (n=65)	Yorkshire Terrier (n=16)	Biro Biewer (n=3)	Italian Greyhound (n=691)	Toy Poodle (n=109)
2002	343 327 280	--	--	--	--	--	--	--	--	--	--	--	0.003	0.004	--	0.0906	--	--	--	--	--	--	--	--	--	--
2003	343 324 282	--	--	--	0.023	--	--	--	0.0225	--	0.13	0.223	0.013	0.508	--	0.0915	0.042	0.598	0.218	0.208	0.292	0.38	0.2	0.0072	0.445
2007	351 327 280	0.014	0.04	--	0.031	0.054	--	--	0.0145	--	--	0.049	0.005	0.002	0.261	0.0138	0.048	--	--	--	--	--	--	--	--	--
2015	339 327 280	--	--	--	--	--	--	--	--	--	--	--	0.016	0.051	--	0.0067	--	--	--	--	--	--	--	--	0.0087	0.046
2022	339 327 282	--	--	0.057	0.086	--	--	--	0.0007	0.002	0.126	0.116	0.107	0.002	0.056	0.0002	0.006	0.015	--	--	--	--	--	--	--	0.014
2024	343 323 280	--	--	--	0.066	--	--	--	--	0.001	--	0.004	0.016	0.01	--	0.0002	--	0.006	--	--	--	--	--	--	--	0.069
2042	341 324 286	0.047	--	--	--	--	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--	--
2050	341 327 284	--	--	--	--	--	--	--	0.0399	--	--	--	0.003	--	--	--	0.161	--	--	--	--	--	--	--	--	0.037
2053	343 324 280	--	--	0.317	0.043	--	--	--	0.029	--	0.14	0.038	0.559	0.014	0.493	--	0.039	--	--	--	0.009	--	--	--	--	0.014
2095	355 322 280	--	--	--	--	--	--	--	--	--	--	--	0.158	--	--	--	--	--	--	--	--	--	--	--	--	--
2096	351 322 280	--	--	--	--	--	--	--	--	--	--	--	0.115	--	--	--	--	--	--	--	--	--	--	--	--	--
2099	345 324 276	--	--	--	--	--	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--	--

D. Using standard genetic assessment parameters and DLA class I and II STR allele frequencies to gauge diversity in the entire DLA region.

It is important to maintain as much genetic diversity in the DLA region as possible and to select mates in a random manner to maintain that diversity. Genetic diversity in the DLA region can be assessed by studying the frequency of the DLA class I and II alleles of the four DLA class I and three DLA class II STR loci (Tables 8, 9), in the same manner as employed with the 33 genomic STR loci. Although these STRs are associated only with the DLA class I and II regions on chromosome 12, the numerous genes and their alleles that make the entire DLA are in strong linkage disequilibrium and inherited by descent with limited genetic recombination. Therefore, genetic diversity in the small DLA region should mirror the more genome-wide genetic diversity shown by the 33 genomic STR markers.

Table 8 shows the frequency of alleles each of the four DLA class I and three DLA class II STR loci. There are 4-9 alleles at each loci (Na), but like the 33 genomic STR loci, only 1.29-2.94 alleles are responsible the observed heterozygosity found in the 187 Samoyed tested. The observed heterozygosity (Ho) at each locus varied from 0.21 to 0.64 and the expected heterozygosity (He) 0.23 to 0.66, yielding F values ranging from -0.01 to 0.073. The inbreeding coefficient (F) was slightly positive for 6/7 alleles and less than zero for only one allele (Table 9). Therefore, a standard genetic assessment of alleles within the DLA region mirrored the findings for the 33 autosomal STR loci (compare Tables 3 and 8). The 187 dogs tested appeared to be in near HWE, but with a slight excess of more inbred individuals. The lower Na and Ne scores for alleles in the DLA class I and II regions may be due to the over-representation of founders with the 1152/2053 haplotype.

Table 8. Standard Genetic Assessment Samoyed using 7 STRs in the DLA region (Updated October 09, 2019)

	N	Na	Ne	Ho	He	F
Mean	192	6.143	2.100	0.472	0.486	0.027
SE		0.620	0.208	0.052	0.055	0.009

Table 9. Assessment of genetic diversity within DLA region using the frequencies of alleles for each of the 4 STR loci associated with DLA class I and the 3 STR loci associated with class II. (Updated October 09, 2019)

#	Locus	N	Na	Ne	Ho	He	F
1	**DLA I-3CCA**	192	9	2.941	0.639	0.660	0.032
2	**DLA I-4ACA**	192	6	2.442	0.571	0.590	0.032
3	**DLA I-4BCT**	192	5	1.619	0.387	0.382	-0.013
4	**DLA1131**	192	8	1.612	0.380	0.380	-0.001
5	**5ACA**	192	6	2.458	0.576	0.593	0.029
6	**5ACT**	192	4	2.334	0.534	0.571	0.066
7	**5BCA**	192	5	1.293	0.216	0.227	0.048

III. Known or presumed heritable diseases of Samoyed

Several diseases that appear to be heritable, but not genetically defined, have been observed at low frequency in the breed.[k,m] Many of these disorders have evolved with the domestic dog over time and inherited by descent as breeds have been created [3]. Except for hip dysplasia, which is considered one of the more serious heritable disorders of Samoyed, most heritable and potentially heritable disease traits of the breed have been of minor importance.[k] There are only four simple deleterious genetic disorders in Samoyed with defined causes: 1) X-linked glomerulopathy [4], 2) X-linked progressive retinal atrophy [5][m], 3) an incomplete dominant short-limbed defect with ocular abnormalities [6, 7], and 4) an autosomal recessive ameliogenesis imperfecta (enamel hypoplasia) has been recently reported in the breed [11]. Posterior cataracts have also been described in the breed and are probably of simple genetic origin.[n]

Autoimmune disorders are common among pure breeds of dogs and are polygenic in nature. They have been linked to the degree of inbreeding in breeds such as the Standard Poodle [12] and Italian Greyhound [13] and are likely similar in other breeds. Autoimmune disorders reported in Samoyed include endocrine alopecia, type 1 diabetes mellitus, uveoermatological syndrome (VKH), keratoconjunctivitis sicca, and nasal planum depigmentation.

IV. Interpretation of genetic diversity test data for Samoyed

A. Genetic diversity

The Samoyed breed has average genetic diversity among the breeds that we have studied based mainly on the Na and Ne values, IR/IRVD curve overlap and DLA class I and II haplotypes. The breed has less genetic diversity than Miniature, Toy and Standard Poodles, Italian Greyhounds, Golden Retriever, Labrador Retriever, Havanese, Akita and Alaskan Klee Kai; similar genetic diversity to Black Russian Terriers, Flat Coated Retriever, Doberman Pinscher and Giant Schnauzer; and greater genetic diversity than Shiloh Shepherds and Swedish Vallhunds.

A lack of genetic diversity is not serious, providing that the founders of the breed were genetically healthy and if this original diversity has been maintained and in balance through sound breeding practices. There is evidence that some genetic defects such as recessive X-linked PRA and posterior cataracts were present in founders common to other Nordic breeds and then passed down within the breed by descent. Such disorders will often stay at low incidence if genetic diversity is maintained, population size kept large, and artificial genetic bottlenecks avoided. X-linked glomerulopathy, incomplete dominant short-limb defect with ocular abnormalities, and autosomal recessive enamel hypoplasia are genetic disorders are genetic disorders that appear to have arisen after the breed was founded. The fact that several genetic disorders have appeared in Samoyed indicates that there have been periods of excessive positive artificial selection since its inception, and this coupled with relatively low genetic diversity, allowed these deleterious traits to be amplified and become clinically apparent [11].

Although most breeds have been successful in maintaining equality in observed (actual) and expected heterozygosity, there are always a small number of individuals in a breed that are products of excessive inbreeding, but fortunately these dogs are usually balanced by a small group of dogs that are highly outbred. The bulk of the population would be in between, resulting in a population that is in a state equivalent to HWE in a wild population. The most accurate way to identify highly inbred and outbred individuals would be through DNA testing and IR values.

B. Origin of breed

The origins of most breeds are open to differences of opinion, memory, and historical records. The Samoyed is held up as an example of the outstanding sled and sledge dogs used in the late nineteenth and early twentieth century by Arctic and Antarctic explorers. However, numerous photographs of these dogs showed them to be very heterogeneous in appearance, medium-sized with thick double coats, large feet, smaller ears, and fluffy curved tails. There is also anecdotal evidence that the lighter colored dogs fared better in Arctic and Antarctic conditions than dark coated dogs. Therefore, the modern Samoyed tends to be a conceptual model of the ideal sled/sledge dog as seen by the public of the time. However, well-conditioned Samoyed are still capable of working in snow and ice and pulling sleds.

There is genetic evidence that supports a common origin of Samoyed, Siberian Husky, and Malamute breeds. Samoyed and Siberian Husky suffer from the identical genetic late onset form of recessive X-linked progressive retinal atrophy. An inherited disorder causing bilateral

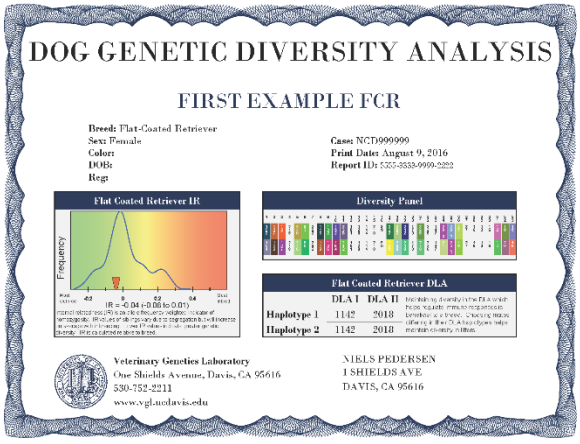
posterior cortical type cataracts in 1-3- year old Siberian Husky, Samoyed and Alaskan Malamute has also been reported. Where and when these mutations occurred is unknown, but it appears that they were present in founders of these breeds and then inherited by descent as the breeds were standardized and refined to their present states.

The DLA class I and II haplotypes are inherited by descent as relatively conserved blocks of genes that can be also used to investigate breed founders. The greatest sharing of currently known STR-associated DLA haplotypes is with the Labrador and Golden Retrievers, Miniature and Standard Poodles, Havanese, and Swedish Vallhund (Table 7). The Vallhund relationship is most interesting as the Vallhund, except for the chondrodystrophic short legs and coat color/pattern, appears much like common Nordic breeds. Unfortunately, we do not yet have DLA haplotype information on other Nordic breeds such as the Siberian Husky and Malamute with which to compare Samoyed.

The 1152/2053 haplotype is unique to the breed and is present in over one-half of contemporary Samoyeds. The origin of this haplotype was either from a single dog or a small number of closely related individuals, the genetics of which have been strongly conserved during breed standardization and refinements. It is likely, therefore, that physical traits of this dog(s) are strongly tied to the breed standard. Three dogs appear to play key roles in the foundation of the breed: 1) Sabarka was a white dog from Archangel, Russia that was gifted to Clara Kilburn-Scott by her husband Ernest, 2) Whitey Petchora was a cream-colored dog that caught the fancy of Ernest during his travels in Northern Europe, and 3) Antarctic Buck was a large white dog that had been part of an Antarctic expedition and subsequently rescued from an Australian zoo by the Kilburn-Scott's.

C. How will you be given the results of DNA-based genetic diversity testing on your dog?

After a sample is submitted for genetic testing, the identity of the dog and owner will be replaced by a laboratory barcode identifier. This identifier will be used for all subsequent activities and each owner will be provided with a certificate that reports the internal relatedness, genomic STR genotypes and DLA class I and II haplotypes for the dog(s) tested. The internal relatedness value for the dog compared to the entire population.


DOG GENETIC DIVERSITY ANALYSIS

FIRST EXAMPLE FCR

Breed: Flat-Coated Retriever
Sex: Female
Color:
DOB:
Reg:

Case: NCD999999
Print Date: August 9, 2016
Report ID: 5555-3553-9999-9999

Flat Coated Retriever IR

Frequency

Diversity Panel

Flat Coated Retriever DLA

	DLA I	DLA II
Haplotype 1	1142	2018
Haplotype 2	1142	2018

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D. What should you do with this information?

The goal for Samoyed breeders should be to continue to produce puppies with IR scores less than 0, and with time even lower scores. Although Samoyed appeared to be randomly breeding, there was a subpopulation of dogs that were much more inbred than the rest of the population. Fortunately, there is also a small population of dogs that were much more outbred (heterozygous) than the average Samoyed. Therefore, there is a possibility to better balance genetic diversity in the breed. Mates should be selected to avoid homozygosity at any genomic loci or DLA class I and II haplotype and encourage the use of dogs with less common genomic alleles or DLA haplotypes. Maintaining existing genomic diversity will require using IR values of potential mates based on the 33 STR loci to assure puppies of equal or greater overall diversity, like what is being done by many Standard Poodle breeders. However, IR values, because they reflect the unique genetics of each individual, cannot be used as the criteria for selecting ideal mates. Mates with identical IR values may produce puppies significantly more or less diverse than their parents. Conversely, a mating between dogs with high IR values, providing they are genetically different, may produce puppies having much lower IR scores than either parent. A mating between a dog with a high IR value and a low IR value, providing the latter has few alleles and DLA haplotypes in common, will produce puppies much more diverse than the highly inbred parent. Breeders should also realize that a litter of puppies may have a wide range of IR values, depending on the comparative contributions of each of the parents. The more genetically diverse and different the parents, the greater the range of IR values in their offspring.

The next step is to compare the DLA class I and II haplotypes. You want to avoid breeding pairs that will produce puppies that will be homozygous for the same haplotypes, and once again, less common haplotypes may offer more diversity than common ones.

Breeders who do not have access to computer programs to predict the outcome of matings based on IR values of sire and dam can also compare values by manual screening. Potential sires and dams should be first screened for genetic differences in alleles and allele frequencies for the 33 genomic STR loci. Some extra weight should be given to rare vs common alleles. This information is included on all certificates and on the breed-wide data on the VGL website.

Puppies, once born, should be tested for their actual IR values, which will reflect the actual genetic impact of each parent on internal diversity. Considerations of mate choices for genetic diversity should be balanced with other breeding goals but maintaining and/or improving genetic diversity in puppies should be paramount.

An additional goal of this study is to contribute this genetic information to a web repository, hopefully under the control of the registry. The best format for such a repository and testing has been provided by Standard Poodle breeders. This information could be incorporated into a mate selection service that will allow a breeder to identify, among all of the dogs tested, potential mates that would be most ideal for increasing genetic diversity in their litters- https://www.betterbred.com/.

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