Genetic Diversity Testing for the Rat Terrier

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 assess genetic diversity across the genome and in the Dog Leukocyte Antigen (DLA) class I and II regions. This test panel will be useful to breeders who wish to manage genetic diversity of their breed as a long-term goal.

Genetic diversity testing of Rat Terrier is now in the preliminary results phase. During this phase, we continue to test more registered dogs to build genetic data necessary to provide breeders with an accurate assessment of genetic diversity. This report is based on testing of 34 specifically selected Rat Terriers from across the USA. This number of dogs is sufficient for a reasonable genetic assessment of the breed given their heterogeneity. Allele and DLA haplotype frequencies will be updated as more Rat Terriers are tested. It is anticipated that new alleles at the 33 STR loci and additional DLA class I and II haplotypes will be identified in the future, but these will tend to be of much lower incidence than those detected in the present population.

ORDER TEST KITS

Results reported as:
Short tandem repeat (STR) loci: A total of 33 STR loci from across the genome 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, and breed-wide allele frequency is provided.

DLA haplotypes: 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. DLA haplotypes are also useful in studying founder lineages.

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. History [1-4]

The Rat Terrier originated in America and the breed name was said to be coined by Teddy Roosevelt [1]. However, small fast and agile dogs have been used for centuries in many parts of the world for vermin control [2]. A good "ratter" was a standard feature on old-time farms, where a rodent infestation could mean the difference between having enough grain to last the winter and going hungry [1]. However, farmers expected their dogs to be more than control vermin, so 'ratters' also became hunting partners, watchdogs, henhouse guardians, and children's playmates.

The ancestors of the breed included small mixed-breed hunting dogs known as "feists" [3]. Feists were mongrels involving crosses between the Smooth Fox Terrier, the Manchester Terrier, and the now extinct English White Terrier, and brought to America by English working-class immigrants. The modern Rat Terrier evolved later in the 19th century with further outcrossing to breeds such as the Beagle, Italian Greyhounds and Manchester Terrier. Rat Terriers were one of the most popular dog types from the 1920s to the 1940s, only losing popularity with the advent of modern rodent control methods.

Recognition of the Rat Terrier as a breed was relatively recent [2]. The United Kennel Club (UKC) officially recognized the breed on January 1, 1999 and Rat terriers have been competing in UKC events for the last 15 years. The American Rat Terrier Association is the national parent breed club for Rat Terriers in the UKC. The American Kennel Club (AKC) first allowed Rat Terriers in sanctioned AKC Companion events (Obedience, Agility, Rally) beginning January 1, 2006. However, the breed was not formally recognized by the AKC until July 1, 2010 and accepted into the AKC's terrier group in 2012. The Rat Terrier Club of America is the official parent club within the AKC. The National Rat Terrier Association is the largest independent registry for the breed and lineage records have been maintained for decades. It is the most prominent of clubs and associations opposing closed-registry breeding rules and over-emphasis on conformation traits and showing.

The contemporary Rat Terrier is becoming increasingly popular as both a family pet and a working dog in several fields and ranks 86 of 195 breeds recognized by the AKC [3]. Their size and temperament have also made them popular as service dogs in hospice, treatment for depression, in Assisted Living, and other human-care jobs. Police departments have also found them useful as search dogs for contraband. Their small size allows search of cars, homes, and prison cells without causing damage.

B. Appearance [1-4]

The Rat Terrier ranges from about 10 to 25 pounds and stands 10 to 18 inches at the shoulder. The miniature size, 13 inches and under as defined by the UKC, is becoming increasingly popular as a house pet and companion dog. A larger variety, often more than 25 pounds, is called the Decker and created as a larger hunting companion [3]. The

Decker is recognized by the National Rat Terrier Association. UKC and AKC recognize the Decker as a standard variety and not a breed. The NRTA recognizes a Toy variety weighing 10 pounds or less and continues to classify the Teddy Roosevelt Terrier as the Type B Rat Terrier. In the 1970s, a hairless mutation appeared in a single Rat Terrier and was propagated into a line of the Rat Terrier. After a period of development this lineage resulted in the American Hairless Terrier and is recognized as a separate breed by several registries [4].

The Rat Terrier comes in a variety of coat colors. The classic coloring is black tan point with piebald spotting (black tricolor), although chocolate, tan varying in shade from pale gold to dark mahogany, blue, isabella (pearl), lemon and apricot are also common. The coat can be tricolor or bicolor, always with some amount of white present. Sable may overlay any of these colors. Creeping tan (Calico), is also acceptable. Brindle, currently disallowed, is considered a traditional Rat Terrier pattern, and there is a movement to have this pattern accepted. However, merle is widely considered to be the result of recent outcrosses, and because of associated health problems, it is rejected by most Rat Terrier breeders.

Rat Terriers usually have naturally erect ears and an alert expression. Ear carriage is erect, but can also be tipped, or button. The tail has been traditionally docked to about 2–3 inches, but the bobtail gene is common in Rat Terriers and can result in a variety of tail lengths. Some breeders prefer a natural, undocked tail, which is accepted in the breed standards.

C. Temperament [1-4]

Rat Terriers tend to be both intelligent and stubborn and are considered good family pets because of their energy and compatibility with kids. They are playful and require much exercise and enjoy outdoor activities. Their closeness to people makes them very trainable but socialization from an early age is critical. Proper socialization includes exposing puppies to a wide variety of people and places. Like most active and intelligent breeds, Rat Terriers are happier when they receive a lot of mental stimulation and exercise. Rat terriers are loyal and respectful to their owners if they receive the proper training at a young age.

II. Genetic diversity studies of the contemporary Rat terrier

A. Population genetics based on 33 STR loci on 25 chromosomes

STR markers are highly polymorphic and have great power to determine genetic differences among individuals and breeds. The routine test panel contains 33 STRs consisting of those that are 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 of these STR loci is known to contain from 7 to 27 different alleles (avg. 15.4 alleles/locus) when tested across many breeds of dogs. Each breed, having evolved from a small number of founders and having

been exposed to artificial genetic bottlenecks will end up with only a portion of the total available diversity. Artificial genetic bottlenecks include such things as popular sire effects, geographic isolation, catastrophes, outbreaks of disease, and ups and downs in popularity and resulting increases and decreases in population size. The alleles identified at each of the 33 STR loci and their relative frequencies were determined for 34 Rat Terriers and are listed in Table 1.

Table 1. Allele frequencies for 33 STR markers in Rat Terrier (n=34)

AHT121	AHT137	AHTH130	AHTh171-A	AHTh260	AHTk211
92 (0.10)	131 (0.29)	119 (0.16)	219 (0.32)	234 (0.01)	87 (0.40)
96 (0.04)	133 (0.04)	121 (0.18)	221 (0.12)	238 (0.19)	89 (0.25)
98 (0.19)	137 (0.12)	123 (0.01)	225 (0.26)	240 (0.01)	91 (0.24)
100 (0.28)	139 (0.04)	125 (0.07)	227 (0.03)	242 (0.06)	95 (0.04)
102 (0.01)	141 (0.06)	127 (0.38)	233 (0.15)	244 (0.12)	97 (0.07)
104 (0.15)	145 (0.09)	129 (0.13)	235 (0.03)	246 (0.47)	
106 (0.15)	147 (0.16)	131 (0.04)	237 (0.09)	248 (0.07)	
108 (0.04)	149 (0.01)	133 (0.01)		250 (0.04)	
110 (0.03)	151 (0.18)			252 (0.01)	

AHTk253	C22.279	FH2001	FH2054	FH2848	INRA21
282 (0.03)	114 (0.03)	128 (0.06)	144 (0.09)	228 (0.06)	95 (0.24)
284 (0.01)	116 (0.16)	132 (0.12)	148 (0.04)	230 (0.01)	97 (0.22)
286 (0.28)	118 (0.10)	136 (0.03)	152 (0.03)	234 (0.07)	99 (0.12)
288 (0.24)	120 (0.24)	140 (0.06)	156 (0.40)	236 (0.21)	101 (0.19)
290 (0.32)	122 (0.04)	144 (0.19)	164 (0.19)	238 (0.18)	103 (0.07)
292 (0.09)	124 (0.24)	148 (0.31)	168 (0.07)	240 (0.32)	105 (0.16)
294 (0.03)	126 (0.19)	152 (0.21)	172 (0.13)	242 (0.07)	
		158 (0.03)	176 (0.03)	244 (0.07)	
			180 (0.01)		

INU005	INU030	INU055	LEI004	REN105L03	REN162C04
106 (0.06)	144 (0.35)	204 (0.06)	85 (0.13)	227 (0.06)	200 (0.01)
110 (0.07)	148 (0.13)	208 (0.04)	95 (0.71)	229 (0.16)	202 (0.15)
122 (0.01)	150 (0.43)	210 (0.47)	97 (0.10)	231 (0.16)	204 (0.24)
124 (0.44)	152 (0.06)	212 (0.19)	101 (0.01)	233 (0.03)	206 (0.46)
126 (0.18)	156 (0.03)	214 (0.01)	107 (0.04)	235 (0.25)	208 (0.04)
130 (0.10)		216 (0.01)		237 (0.06)	210 (0.10)
132 (0.13)		218 (0.16)		239 (0.13)	
		220 (0.04)		241 (0.15)	

REN169D01	REN169O18	REN247M23	REN54P11	REN64E19	VGL0760
202 (0.04)	162 (0.10)	268 (0.63)	222 (0.18)	139 (0.07)	12 (0.04)

210 (0.07)	164 (0.06)	270 (0.03)	226 (0.40)	143 (0.01)	18.2 (0.01)
212 (0.31)	166 (0.32)	272 (0.21)	228 (0.01)	145 (0.35)	19.2 (0.12)
214 (0.04)	168 (0.21)	274 (0.04)	230 (0.01)	147 (0.28)	20.2 (0.13)
216 (0.43)	170 (0.31)	276 (0.09)	232 (0.15)	151 (0.06)	21.2 (0.21)
218 (0.03)			234 (0.13)	153 (0.19)	22.2 (0.22)
220 (0.07)			236 (0.07)	155 (0.03)	23.2 (0.12)
			238 (0.04)		24.2 (0.10)
					25.2 (0.03)
					26.2 (0.01)

VGL0910	**VGL1063**	**VGL1165**	**VGL1828**	**VGL2009**	**VGL2409**
13 (0.04)	8 (0.04)	17 (0.03)	14 (0.04)	9 (0.07)	13 (0.01)
15.1 (0.01)	9 (0.07)	18 (0.09)	16 (0.12)	10 (0.13)	14 (0.09)
16 (0.04)	12 (0.15)	20 (0.06)	17 (0.18)	11 (0.04)	15 (0.24)
17 (0.01)	13 (0.07)	21 (0.24)	19 (0.04)	13 (0.26)	16 (0.16)
17.1 (0.09)	14 (0.22)	22 (0.16)	20 (0.15)	14 (0.29)	17 (0.28)
18.1 (0.06)	15 (0.09)	24 (0.01)	21 (0.07)	15 (0.19)	18 (0.16)
19.1 (0.47)	17 (0.01)	25 (0.09)	22 (0.29)		19 (0.06)
20.1 (0.19)	18 (0.01)	26 (0.06)	23 (0.10)		
21.1 (0.01)	19 (0.12)	27 (0.03)			
22.1 (0.01)	20 (0.18)	28 (0.10)			
26.1 (0.04)	22 (0.03)	30 (0.01)			
		31 (0.12)			

VGL2918	**VGL3008**	**VGL3235**
12 (0.06)	12 (0.01)	9 (0.01)
13 (0.26)	15 (0.38)	11 (0.01)
14 (0.28)	17 (0.10)	12 (0.03)
15 (0.06)	18 (0.07)	13 (0.21)
17.3 (0.22)	19 (0.22)	14 (0.37)
18.3 (0.07)	20 (0.10)	15 (0.07)
19.3 (0.03)	21 (0.09)	16 (0.03)
21.3 (0.01)	23 (0.01)	17 (0.10)
		18 (0.16)

B. Assessment of population diversity using standard genetic parameters

Allele and allele frequencies at each of the 33 STR loci are listed in Table 1 and used to determine basic genetic parameters (Table 2) such as the number of alleles found at each STR locus (Na); the number of effective alleles (Ne) per locus (i.e., the number of alleles that contribute to heterozygosity in the population); the observed or actual heterozygosity (Ho) that was found in the population; and the heterozygosity that would be expected

(He) if the existing population was being randomly bred. The value F is a coefficient of inbreeding derived from the Ho and He values. A value of +1.0 would occur only if every individual were genetically indistinguishable at each of the 33 STR loci, while a value of -1.0 would be seen when all the dogs were completely different at each of the 33 loci. A value of 0.00 would be seen if the selection of sires and dams were entirely random within the existing gene pool.

The allele frequency data obtained from the 33 STR panel can be used to assess heterozygosity within a population (Table 2). Using the 33-marker panel, the 34 Rat Terrier had an average of 7.67 alleles/locus (Na). This is much higher than for the Shiloh Shepherd (Na=4.0), Lakeland Terrier (Na=4.24), Swedish Vallhund (Na=4.91) and Irish Red and White Setter (Na=5.09), Llewellin Setter (Na=5.94) and Flat-coated Retriever (Na=5.94), slightly higher than the Labrador Retriever (Na=7.33), and somewhat lower than the Golden Retriever (Na=8.39) and Miniature Poodle (Na=8.91). However, the average number of alleles is less important than the number of alleles that have the greatest genetic influence on heterozygosity, a figure known as average effective alleles/loci or Ne. Ne in this group of dogs averaged 4.56 effective alleles per locus. Therefore, an average of 4.56 alleles at each locus contribute to most of the heterozygosity within the breed. The observed (actual) heterozygosity of this group of 34 dogs was 0.75, while the expected heterozygosity (He) for a population in a state resembling Hardy-Weinberg equilibrium (HWE) was 0.76, yielding a coefficient of inbreeding (F) of 0.015 (i.e., only 1.5% more inbred than predicted for HWE). The average observed heterozygosity score for Rat Terriers is among the highest for any breed tested to date. The fact that the expected and observed heterozygosity scores were nearly identical, leading to a near zero inbreeding coefficient, is an indication that this group of dogs is both genetically diverse and their parents selected in an almost totally random manner.

Ne is also a measure of the number of individuals that are necessary to keep a free-breeding population in a state of HWE from one generation to the next. This number is also known as the effective population size. Pure breeds of dogs have Ne values as low as 2.3-2.6 (Doberman pinscher, Japanese Akita, Swedish Vallhund, Flat coated retriever), 3.3-3.8 (Havanese, Labrador and Golden Retriever, Border Colle, Samoyed, Magyar agar), and a previous high of 4.2 (Toy Poodle). An Ne is 4.56 for the Rat Terrier is the highest for any dog breed tested to date and suggests that the breed has been built on many genetically diverse founder lines and is in no danger of population decline/loss of vigor (Table 2).

Table 2: Standard genetic assessment of 34 Rat Terriers based on allele frequencies at 33 genomic STR loci on 25 chromosomes. Values are expressed as means (averages) with one standard error (SE).

	N	Na	Ne	Ho	He	F
Mean	34	7.67	4.56	0.75	0.76	0.015
SE		0.31	0.22	0.02	0.01	0.015

B. Standard genetic assessment values for individual STR loci

The allele frequencies can be also used to do a standard genetic assessment of heterozygosity at each STR locus (Table 3). This provides an estimate of genetic similarities in the specific regions of the genome that are associated with each STR marker. Phenotypic differences equate to genotypic differences. Therefore, alleles that are widely shared across the population are indicators that positive selection is occurring for certain desired traits. The Na values for an individual STR locus for this population of 34 Rat Terrier ranged from a low of 5 to a high of 12 alleles per locus, while the Ne ranged from 2.21 to 7.76 alleles per locus. It is important to remember that each STR locus can have from 7-27 different alleles (avg. 15.4 alleles/locus) when testing across all dogs. The observed heterozygosity (Ho) for an individual STR locus ranged from 0.53 to 0.91, while He ranged from 0.47 to 0.87 (Table 3). Twenty loci had positive F values and 13 were negative. Six of the positive loci had F values of 0.10 or greater, indicating linkage with regions of the genome that were under greater than expected positive selection. However, the influences of these various inbred and outbred regions of the genome defined by these 33 STR loci have been kept in balance by breeders as evidenced by a very low F value for the overall population (Table 2). However, this assessment is for the entire group of dogs. The degree of inbreeding or outbreeding for individual dogs in this group of 34 will be made apparent from internal relatedness (IR) scores.

Table 3. Standard Genetic Assessment for Rat Terrier using 33 STR loci

#	Locus	N	Na	Ne	Ho	He	F
1	**AHT121**	34	9	5.77	0.77	0.83	0.08
2	**AHT137**	34	9	5.78	0.68	0.83	0.18
3	**AHTH130**	34	8	4.37	0.85	0.77	-0.11
4	**AHTh171-A**	34	7	4.55	0.77	0.78	0.02
5	**AHTh260**	34	9	3.53	0.71	0.72	0.02
6	**AHTk211**	34	5	3.54	0.65	0.72	0.10
7	**AHTk253**	34	7	4.04	0.79	0.75	-0.06
8	**C22.279**	34	7	5.35	0.91	0.81	-0.12
9	**FH2001**	34	8	5.08	0.82	0.80	-0.03
10	**FH2054**	34	9	4.37	0.79	0.77	-0.03
11	**FH2848**	34	8	5.05	0.79	0.80	0.01
12	**INRA21**	34	6	5.38	0.88	0.81	-0.08
13	**INU005**	34	7	3.80	0.77	0.74	-0.04
14	**INU030**	34	5	3.05	0.68	0.67	-0.01
15	**INU055**	34	8	3.43	0.62	0.71	0.13
16	**LEI004**	34	5	1.89	0.53	0.47	-0.12
17	**REN105L03**	34	8	6.18	0.74	0.84	0.12
18	**REN162C04**	34	6	3.36	0.79	0.70	-0.13
19	**REN169D01**	34	7	3.42	0.71	0.71	0.002
20	**REN169O18**	34	5	3.90	0.59	0.74	0.21
21	**REN247M23**	34	5	2.21	0.53	0.55	0.03
22	**REN54P11**	34	8	4.24	0.71	0.76	0.08
23	**REN64E19**	34	7	4.01	0.74	0.75	0.02
24	**VGL0760**	34	10	6.66	0.85	0.85	0.00
25	**VGL0910**	34	11	3.62	0.77	0.72	-0.06

26	**VGL1063**	34	11	7.29	0.91	0.86	-0.06
27	**VGL1165**	34	12	7.66	0.82	0.87	0.05
28	**VGL1828**	34	8	5.78	0.82	0.83	0.004
29	**VGL2009**	34	6	4.59	0.74	0.78	0.06
30	**VGL2409**	34	7	5.07	0.77	0.80	0.05
31	**VGL2918**	34	8	4.76	0.74	0.79	0.07
32	**VGL3008**	34	8	4.35	0.68	0.77	0.12
33	**VGL3235**	34	9	4.51	0.79	0.78	-0.02

On average, the alleles identified in this group of 34 dogs represented 7.67/15.4=49.8% of alleles known to exist in all canids tested at the VGL. This is higher than the Swedish Vallhund (31.9%) and Irish Red and White Setter (34.8%); similar to the Flat-coated Retriever (38.6%), Magyar Agar (40.4%) and Borzoi (40.9%); but lower than breeds such as the Golden Retriever (54.5%), Toy Poodle (55.6%) and Standard Poodle (58%).

D. Differences in population structure as determined by principal coordinate analysis (PCoA)

PCoA measures the genetic relatedness of individuals in a population. The data is computed in a spherical form, but it is often presented in the two dimensions that most closely represent its three-dimensional form (usually coordinates 1 and 2). The more closely individuals cluster together around the XY axis, the more related they are to each other.

The 34 Rat Terriers formed a single population (i.e., breed) by PCoA (Fig. 1). Individual dogs were clearly segregated genetically from each other and widely dispersed across all quadrants of the graph. Only two dogs near the XY axis appeared to be closely related to each other. This was another indication that this group of 34 dogs was representative of the breed.

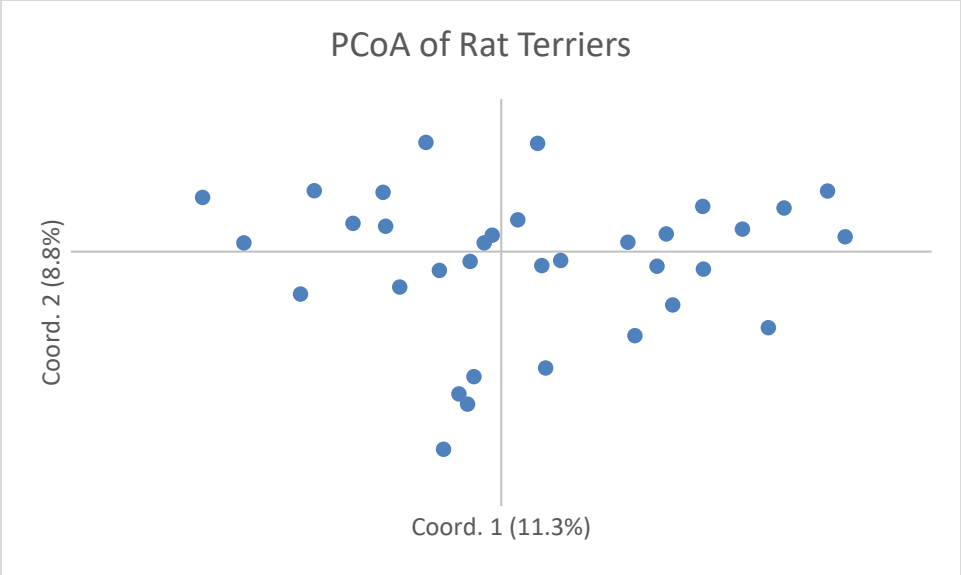


Figure 1. PCoA graph portraying the genetic relatedness of 34 Rat Terriers.

The degree of relatedness of individuals within a breed can be further emphasized by comparing small dog breeds, such as the Havanese, Italian greyhound, and Rat Terrier (Fig. 2). Comparing related breeds with a very unrelated breed will enhance the degree of relatedness between related breeds (or varieties/bloodlines). The three breeds are genetically distinct in this plot, although some individual Havanese appear more closely related to the Rat Terrier than to Italian Greyhound.

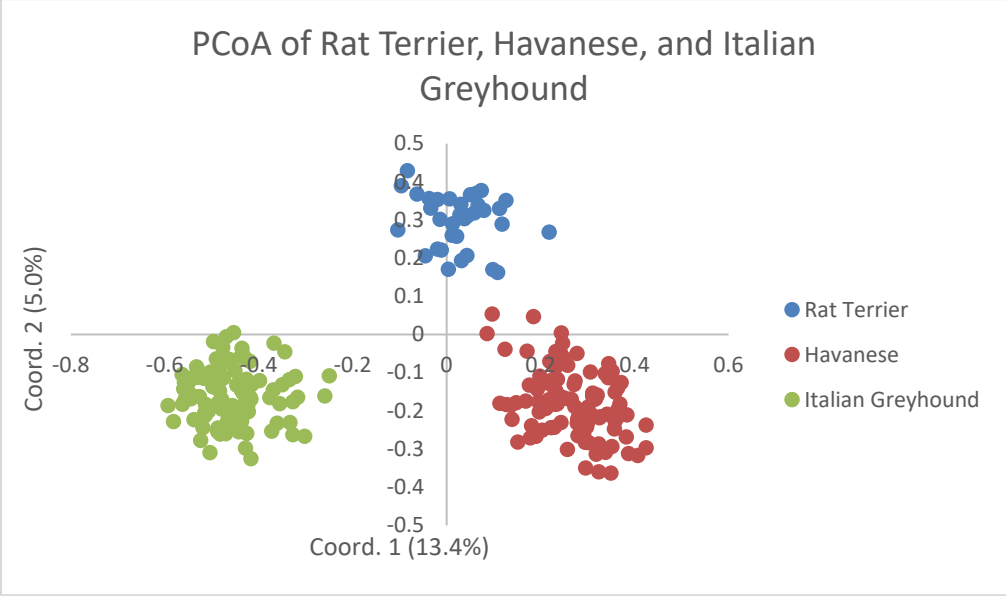


Figure 2. PCoA comparing 3 small dog breeds, Rat Terrier, Havanese, and Italian Greyhound.

D. Internal relatedness (IR) of individuals and the population as a whole

1. IR testing

Genetic assessments such as those presented in Tables 1-3 are indicators of population-wide heterozygosity and do not reflect the genetic diversity being provided to individuals by their parents. Internal Relatedness (IR) is a calculation that has been used to determine the degree to which the two parents of an individual dog were related. The IR calculation takes into consideration homozygosity at each locus and gives more importance to rare and uncommon alleles. Rare and uncommon alleles would presumably be present in less related individuals. 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 be found among 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 higher the IR value above 0.25 the more closely related were the parents and grandparents of the siblings.

Table 4 lists the IR values for the 34 Rat Terriers that were initially tested. The 25% of most outbred dog in the population had an IR scores of -0.087 to -0.200, while the 25% of most inbred dog in the group had an IR score of 0.104 to 0.399, while the mean (average) IR score for the group was 0.017. Therefore, IR values give a different picture than seen with the population average scores from the standard genetic assessment (Table 2). While the standard genetic assessments indicated a population in HWE, the IR scores showed a population of individuals that ranged from very outbred to very inbred. The most inbred dogs in the group was interrelated to a greater degree than offspring of full cousins or half sibling parents from within the breed. The more inbred dogs are balanced by outbred dogs, making it appear that the overall population was in a state of HWE. This is a common feature of all pure breeds of dogs.

Table 4: Internal relatedness (IR) values calculated using allele numbers and frequencies for 34 Rat Terriers. The IR values can be adjusted to reflect how these same dogs would score if they were to exist in a large population of village dogs (IRVD).

	IR	IRVD
Min	-0.200	-0.130
1st Qu	-0.087	-0.018
Mean	0.017	0.100
Median	-0.044	0.028
3rd Qu	0.104	0.256
Max	0.399	0.447

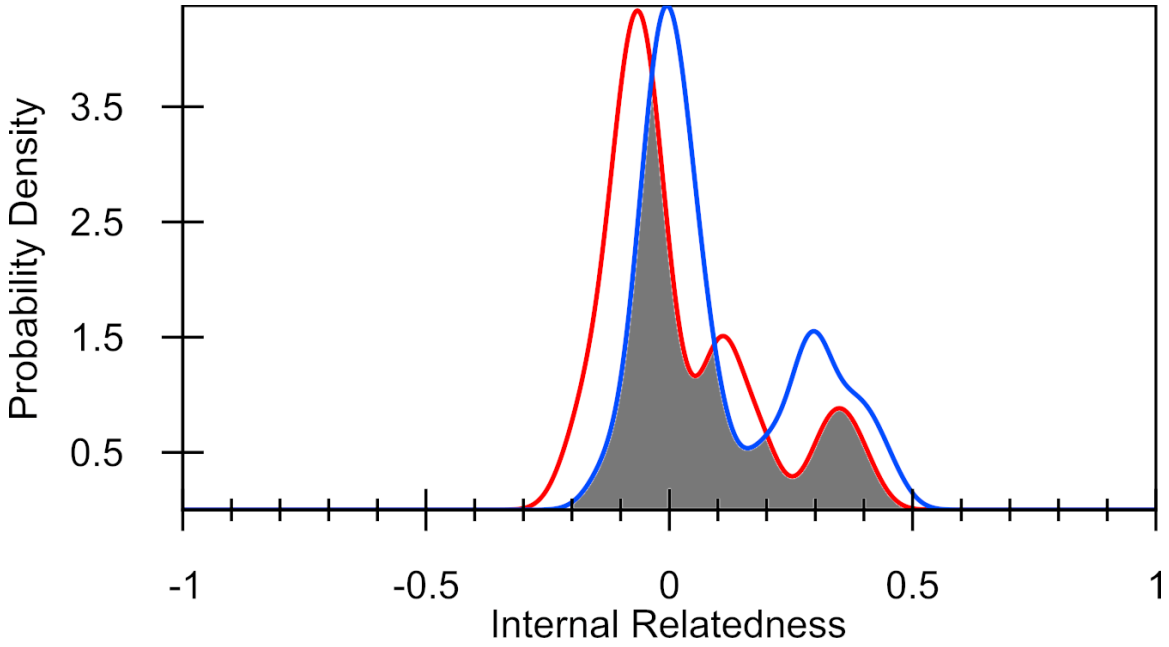


Figure 3. Distribution of IR (red line) and IR-village dog (IRVD) (blue line) values for the Rat Terrier (n=34). The area under the curve (black) represents the degree of allele sharing (64.7%) between Rat Terriers and village dogs.

2. Adjusted IR values (IRVD) as a measure of genetic diversity lost during breed evolution from time of origin to the present time.

It is possible to determine the amount of canid genetic diversity a breed has retained as it evolved to present day. This is done by assuming that individual Rat Terriers were members of the current village dog population found in the Middle East, SE Asia, and the Island Pacific nations. The IR values and IR values adjusted to village dogs (IRVD) (Table 4) can then be graphed and the graphs overlaid (Fig. 3). One quarter of the dogs have IRVD scores from 0.256 to 0.447. Therefore, if this group of dogs were found among modern village dogs, three-fourths of them would be considered equally or more inbred than offspring of full sibling village dog parents. This degree of inbreeding in individuals is seen in many breeds of dogs.

The IRVD curve for the Rat Terriers was nearly superimposed on the IR curve (Fig. 3). This figure is close to the 64.7% of retained canid genetic diversity calculated from comparison with a somewhat different population, i.e., all canids ever tested at the VGL (Tables 1, 2). This level of retained village dog genetics was even higher than the about 60% retained diversity observed in the Miniature/toy poodle or 54% in Labrador Retriever, and much higher than the 23% for Irish wolfhound, 15% in Doberman Pinchers and 7% in Swedish Vallhund. This would place the Rat Terrier as the breed most simlar to village dogs as tested by the VGL to date.

E. 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, allergies, and resistance/susceptibility to infectious 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 5). 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 the three STR loci associated with the three Class II genes are strongly linked and inherited as a single block or haplotype (Table 5). One haplotype comes from each of the parents.

Specific class I and II haplotypes are often linked to each other and inherited as a genetic block. However, there is enough distance between these two regions to allow for a degree of recombination resulting in many class I/II combinations. 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.

1. DLA class I and II haplotypes existing in Rat Terriers

The 34 Rat Terrier in this study possessed 26 DLA class I and 22 DLA class II haplotypes (Table 5). Five class I (1252-1258) were unique to the breed and the rest shared with several other breeds (Table 5). All class II haplotypes have been recognized in other breeds (Table 6). No class I or II haplotype was dominant in the breed, different from many other breeds. The large number of DLA haplotypes indicates that the breed has evolved from a relatively large and diverse founder population and no single founder line dominated the breed.

The number of DLA class I and II haplotypes found in these 34 Rat Terriers was above average compared to many other breeds studied to date. The numbers of DLA class I and II haplotypes (26, 22) haplotypes found in Rat Terriers were higher than the Swedish Vallhund (6, 4), Shiloh Shepherd (7, 6), Giant Schnauzer (14, 15), Samoyed (13, 12) and Shiba Inu (16, 15); similar to the Golden Retriever (26, 23), and lower than the Miniature Poodle (33, 23). It is possible that more haplotypes will be identified as more dogs are tested, but the numbers and incidences are likely to be low.

Table 6: DLA class I and Class II haplotypes in Rat Terriers and their frequencies

DLA1 #	STR types	Frequency
1002	380 365 281 181	0.01
1008	386 373 289 182	0.03
1009	382 377 277 184	0.06
1011	376 365 281 180	0.06
1012	388 369 289 188	0.03
1016	382 371 277 178	0.01
1030	380 373 293 178	0.03
1040	380 371 277 186	0.18
1046	376 379 291 180	0.03
1054	382 379 277 184	0.01
1062	382 371 277 183	0.04
1074	386 383 289 186	0.03
1087	380 371 277 178	0.12
1092	376 379 277 181	0.06
1105	382 379 277 178	0.03
1128	384 376 287 182	0.01
1200	394 367 273 178	0.01
1211	386 369 277 183	0.04
1234	380 371 281 181	0.01
1252	389 371 277 182	0.03
1253	389 365 289 182	0.03
1254	388 371 291 180	0.01
1255	388 371 277 186	0.01
1256	386 365 281 180	0.01
1257	384 376 289 180	0.06
1258	384 373 287 180	0.01
DLA Class II Haplotype Frequencies		
2001	343 324 284	0.01
2002	343 327 280	0.01
2003	343 324 282	0.07
2005	339 322 280	0.07
2009	351 324 280	0.01
2011	345 322 284	0.06
2012	345 322 280	0.01
2014	339 322 284	0.01
2015	339 327 280	0.01
2017	343 322 280	0.03
2021	339 324 268	0.03
2022	339 327 282	0.18

2023	341 323 282	0.03
2025	351 321 280	0.09
2028	345 327 288	0.09
2031	339 322 282	0.12
2035	341 323 280	0.01
2037	341 327 280	0.06
2040	345 327 280	0.01
2043	343 324 296	0.01
2079	343 323 278	0.01
2081	343 322 282	0.03

2. DLA haplotype sharing with other dog breeds

DLA haplotypes are much more conserved than most other regions of the genome and each DLA region inherited as a block of linked genes from each parent and passed on by descent. Therefore, the number and incidence of DLA haplotypes found in a breed can be used to estimate the founder/founder lines that were used to create a breed and the importance of the various lines in subsequent breed evolution. The DLA class I and II regions are frequently shared between breeds, reflecting common distant ancestry and inheritance by descent (Table 6). The Rat Terrier shared class I and II DLA haplotypes with 41 other breeds tested previously by the VGL. This degree of haplotype sharing was expected given the high genetic diversity present in the breed and its strong relationship to village dogs and to all canids tested to date by the VGL. As might be expected, the greatest sharing of DLA haplotypes was with European breeds, particularly the Poodles and Golden Retrievers (Table 6). As has been observed with almost all other breeds, the Rat Terrier shares many of the same haplotypes as the Havanese. The lowest degree of haplotype sharing was with Asian breeds such as the Akita and Shikoku.

Table 6. Sharing of specific DLA class I and II haplotypes between the Rat Terrier and various breeds.

DLA1 #	STR types	Rat Terrier (n=34)	Black Russian Terrier (n=140)	Lakeland Terrier (n=92)	Labrador Retriever (n=191)	Irish Red and White Setter (n=61)	Doberman Pinscher (n=710)	Flat Coated Retriever (n=575)	Havanese (n=505)	Samoyed (n=191)	Saint Bernard (n=61)	Shiba Inu (n=118)	Giant Schnauzer (n=237)	Polish Lowland Sheepdog (n=25)	Borzoi (n=127)	English Bulldog (n=163)	Whippet (n=59)	Biewer (n=121)	Biewer Yorshire Terrier (n=53)	Biewer Terrier (n=132)	Yorkshire Terrier (n=16)	Italian Greyhound (n=968)	Shikoku (n=47)	Alaskan Klee Kai (n=171)	Shiloh Shepherd , ISSA (n=207)	Magyar Agar (n=77)	English Mastiff (n=21)	Irish Setter (n=49)	Llewellin Setter (n=94)	American Akita (n=141)	Japanese Akita (n=416)	Blend Akita (n=59)	Berger Picard (n=89)	Golden Retriever (n=734)	Collie (n=49)	Irish Wolfhound (n=55)	Border Collie (n=54)	Miniature Poodle (n=301)	Scottish Collie (n=83)	Barbet (n=68)	Swedish Vallhund (n=271)	Poodle (n=1197)	Toy Poodle (n=158)
1002	380 365 281 181	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.042	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.0007	--	--	--	0.002	--	0.066	--	0.1711	0.006
1008	386 373 289 182	0.03	--	--	--	--	--	--	--	--	--	--	--	--	--	0.006	0.076	0.008	--	--	--	0.06	0.1255	--	0.0595	--	--	--	0.11	0.069	--	--	--	--	0.0014	--	--	0.009	--	0.006	--	--	0.0152	0.016
1009	382 377 277 184	0.06	--	--	--	--	--	--	--	0.123	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.076	--	--	--	0.046	0.0166	0.073
1011	376 365 281 180	0.06	--	--	--	0.033	--	--	0.002	0.272	0.066	--	0.011	--	--	--	--	--	--	--	--	--	--	--	0.0587	--	--	--	0.1	--	--	--	--	--	0.0007	--	--	0.074	0.003	--	--	--	0.0188	0.012
1012	388 369 289 188	0.03	--	0.75	--	--	0.0007	--	0.0139	0.013	--	--	--	--	--	0.414	0.051	0.19	0.151	0.235	0.28	0.0072	--	--	--	--	--	--	--	--	--	--	--	0.0014	--	--	0.046	0.055	0.006	--	--	0.0092	0.025
1016	382 371 277 178	0.01	0.011	0.011	0.01	--	0.0162	--	0.2089	--	--	--	0.04	0.02	--	0.095	0.093	0.025	0.019	0.015	0.03	0.0594	--	--	--	--	0.12	--	0.021	--	--	--	0.002	0.04	--	--	0.027	0.084	--	--	--	0.0195	0.028
1030	380 373 283 178	0.03	--	--	0.039	--	0.0058	--	0.003	--	--	--	--	--	--	--	--	0.459	0.349	0.485	0.31	0.0274	--	--	--	--	0.12	--	0.027	--	--	--	0.0007	--	--	--	0.009	--	--	0.029	--	0.0025	--
1040	380 371 277 186	0.18	--	0.179	--	--	0.0113	--	0.0198	--	--	0.016	--	0.62	--	0.04	0.059	0.107	0.208	0.076	--	0.0857	--	--	0.2102	--	--	--	--	0.007	--	--	--	0.0007	--	--	--	0.007	--	--	--	0.0005	0.003
1046	376 379 291 180	0.03	--	--	0.003	--	--	--	--	--	--	--	--	--	--	0.003	--	0.033	--	0.038	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.029	--	0.0003	--
1054	382 379 277 184	0.01	--	--	0.076	--	--	0.1148	0.1203	--	--	--	0.368	0.004	--	--	0.008	--	--	--	--	0.015	0.01	--	--	0.141	--	0.18	0.09	--	--	--	--	--	--	--	--	--	0.002	--	--	--	--	--
1062	382 371 277 183	0.04	--	--	0.018	--	--	--	--	--	0.066	--	--	--	--	0.187	--	--	--	--	--	--	--	--	0.2137	--	--	--	--	--	--	--	--	0.0892	--	--	--	--	--	--	--	--	--	--
1074	386 383 289 186	0.03	--	--	--	--	--	--	--	--	--	--	--	--	--	0.037	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1087	380 371 277 178	0.12	--	--	--	--	--	--	0.001	--	--	--	--	--	--	--	--	0.004	--	--	--	--	--	--	--	--	--	--	--	0.014	--	--	--	--	--	--	--	--	--	0.018	--	--	--	--
1092	376 379 277 181	0.06	0.243	--	--	--	--	--	0.0911	--	--	--	0.255	--	--	0.006	--	--	0.018	--	0.06	--	--	--	--	--	--	--	--	--	0.306	0.008	--	--	--	--	0.009	--	--	0.037	--	0.0002	--
1105	382 379 277 179	0.03	--	--	0.052	--	0.0007	--	0.001	--	--	--	--	--	--	--	--	0.004	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.318	--	--	0.056	--	--	--	--	0.0025	0.073
1128	384 376 287 182	0.01	--	--	--	--	--	--	0.002	--	0.008	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.0007	--	--	--	--	--	--	--	--	--	--
1200	394 367 273 178	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.11	--	--	--
1211	386 369 277 183	0.04	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.07	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1234	380 371 281 181	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.0003	--
1252	389 371 277 182	0.03	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1253	389 365 289 182	0.03	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1254	388 371 291 180	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1255	388 371 277 186	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1256	386 365 281 180	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1257	384 376 289 180	0.06	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1258	384 373 287 180	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--

DLA Class II Haplotype Frequencies (Updated Jun 19, 2020)

DLA2 #	STR types	Rat Terrier (n=34)	Black Russian Terrier (n=140)	Lakeland Terrier (n=92)	Labrador Retriever (n=191)	Irish Red and White Setter (n=61)	Doberman Pinscher (n=710)	Flat Coated Retriever (n=575)	Havanese (n=505)	Samoyed (n=191)	Saint Bernard (n=61)	Shiba Inu (n=118)	Giant Schnauzer (n=237)	Polish Lowland Sheepdog (n=25)	Borzoi (n=127)	English Bulldog (n=163)	Whippet (n=59)	Biewer (n=121)	Biewer Yorshire Terrier (n=53)	Biewer Terrier (n=132)	Yorkshire Terrier (n=16)	Italian Greyhound (n=968)	Shikoku (n=47)	Alaskan Klee Kai (n=171)	Shiloh Shepherd , ISSA (n=207)	Magyar Agar (n=77)	English Mastiff (n=21)	Irish Setter (n=49)	Llewellin Setter (n=94)	American Akita (n=141)	Japanese Akita (n=416)	Blend Akita (n=59)	Berger Picard (n=89)	Golden Retriever (n=734)	Collie (n=49)	Irish Wolfhound (n=55)	Border Collie (n=54)	Miniature Poodle (n=301)	Scottish Collie (n=83)	Barbet (n=68)	Swedish Vallhund (n=271)	Poodle (n=1197)	Toy Poodle (n=158)
2001	343 324 284	0.01	--	--	0.003	--	--	--	0.0485	--	--	0.016	0.005	0.008	--	--	0.068	--	--	--	--	--	--	--	0.005	0.071	--	--	--	0.021	--	--	--	--	0.1403	--	--	0.213	0.015	--	0.199	--	0.639	0.006
2002	343 327 280	0.01	--	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.021	--	--	--	--	--	--	0.13	0.007	0.006	--	--	--	0.0885	--
2003	343 324 282	0.07	--	0.75	0.016	--	0.0007	0.1452	0.2158	0.013	--	--	--	0.014	--	0.598	0.136	0.231	0.208	0.258	0.38	0.0067	--	--	0.007	--	--	--	--	--	--	--	--	0.0225	--	0.046	0.522	0.012	0.132	--	0.0916	0.424
2005	339 322 280	0.07	0.014	0.005	0.052	0.115	--	0.4217	0.002	--	0.074	--	--	0.008	--	0.025	--	0.045	0.009	0.038	--	--	--	--	--	--	--	0.04	0.793	--	--	--	--	0.0157	--	0.227	0.009	--	--	--	--	--	0.0185	0.003
2009	351 324 280	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.01	--	0.029	--	0.0117	0.032
2011	345 322 284	0.06	--	--	--	0.0007	--	--	0.001	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.003	--	--	--	0.0189	0.032
2012	345 322 280	0.01	--	--	0.189	--	--	--	0.0059	--	--	--	0.011	--	--	--	--	--	--	--	--	--	0.022	0.0622	--	0.078	--	--	0.06	0.037	--	0.001	--	--	0.0007	--	--	--	0.06	--	--	0.025	0.054
2014	339 322 284	0.01	--	0.011	0.003	--	--	0.0296	0.0069	--	0.09	--	--	0.002	0.64	--	0.092	--	--	--	--	--	0.0015	--	0.0701	--	--	0.12	--	--	--	--	--	--	--	--	--	--	0.027	--	--	--	0.0183	0.025
2015	339 327 280	0.01	--	0.011	--	--	--	--	--	0.016	--	0.059	--	--	--	--	--	--	--	--	--	--	0.0098	--	--	--	0.12	--	--	--	--	--	--	--	--	--	0.045	--	--	--	--	--	0.0056	0.041
2017	343 322 280	0.03	--	0.038	--	--	--	0.0009	0.0069	--	--	--	0.14	0.008	0.215	0.237	--	--	--	--	--	--	0.2185	--	0.362	0.455	0.17	--	--	0.011	0.005	0.017	0.079	0.0409	--	--	0.204	--	--	--	0.029	--	0.0025	0.003
2021	339 324 268	0.03	--	--	0.018	--	--	--	0.002	--	0.066	--	--	--	--	--	--	0.004	--	--	--	--	0.2119	--	--	--	--	--	--	--	--	--	--	--	0.0892	--	0.282	--	0.056	--	--	--	0.0028	0.073
2022	339 327 282	0.18	--	0.168	0.076	--	0.0014	0.12	0.1158	0.107	--	--	0.004	--	0.598	0.025	--	--	--	--	--	--	--	--	0.056	--	--	0.18	0.09	--	--	--	--	0.0007	--	0.382	0.009	0.002	--	--	0.046	0.0002	0.009
2023	341 323 282	0.03	0.004	--	0.039	--	0.0058	--	0.003	--	0.385	--	--	--	--	--	--	0.459	0.349	0.489	0.31	0.0274	--	--	--	--	0.12	--	0.016	--	--	--	0.0007	--	--	0.009	--	--	0.029	--	0.0025	--
2025	351 321 280	0.09	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.051	--	--	--	0.0022	0.016
2028	345 327 288	0.09	--	--	--	--	--	--	--	--	--	0.016	--	--	--	0.037	0.059	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.007	--	--	--	0.0025	--
2031	339 322 282	0.12	0.036	--	0.008	--	--	--	--	--	--	--	0.04	--	--	--	0.093	--	--	--	--	--	0.0579	--	--	--	--	--	--	--	--	--	--	--	--	--	0.018	0.009	--	--	--	--	--	--
2035	341 323 280	0.01	--	--	--	0.082	--	--	0.001	--	0.008	--	--	--	--	--	--	--	--	--	--	--	0.0857	--	--	--	--	--	0.005	--	0.306	0.008	--	--	--	--	--	--	--	--	0.051	--	0.0002	--
2037	341 327 280	0.06	0.275	--	--	--	--	--	0.0287	--	--	--	0.352	--	--	--	--	0.029	0.009	0.068	0.09	0.0077	--	--	0.3792	--	0.013	--	0.37	--	0.177	0.257	0.153	--	--	--	0.083	0.007	0.006	--	--	--	0.009
2040	345 327 280	0.01	--	--	--	--	--	--	0.003	--	--	--	--	--	--	--	--	0.07	0.085	0.027	0.03	0.0015	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.005	--	--	--	--	0.025
2043	343 324 296	0.01	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.218	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.103	--	--	--
2079	343 323 278	0.01	--	--	--	--	--	--	0.002	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.0007	--	--	--	--	--	--	--	--	--
2081	343 322 282	0.03	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.012	0.019	0.004	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	0.006	--	--	--	--	--

3. Heterozygosity in the DLA region

The 7 loci that define the DLA class I and II haplotypes are in stronger linkage disequilibrium that other parts of the genome that are measured by the 33 autosomal STR markers. However, the expectation is that these loci have achieved an equilibrium with other loci in the genome with random mating and over enough time. This can be tested by doing a standard genetic assessment of each locus (Table 7), as well as all the loci taken together (Table 8). Standard genetic assessment of each of the 7 loci demonstrates values for F (inbreeding coefficient) that range from slightly negative to slightly positive (Table 7). The F values for each of the 7 DLA class I and II loci are all negative with values ranging from 5-20% excess of heterozygosity over HWE within the DLA region in this group of 34 dogs. The average F value for all DLA loci was 0.1 or 10% excess heterozygosity (Table 8). However, the F value for the 33 genomic STR loci was nearly zero (Table 2), suggesting that this outcrossing was intentionally or accidentally related to lines with more unique DLA haplotypes.

Table 7. Standard Genetic Assessment for Rat Terrier using 7 STRs in the DLA region

#	Locus	N	Na	Ne	Ho	He	F
1	**DLA I-3CCA**	34	8	4.97	0.88	0.8	-0.1
2	**DLA I-4ACA**	34	9	4.27	0.88	0.77	-0.15
3	**DLA I-4BCT**	34	7	2.44	0.71	0.59	-0.2
4	**DLA1131**	34	8	6.25	0.91	0.84	-0.09
5	**5ACA**	34	5	3.69	0.77	0.73	-0.05
6	**5ACT**	34	5	3.54	0.77	0.72	-0.07
7	**5BCA**	34	7	3.19	0.74	0.69	-0.07

Table 8. Summary of Standard Genetic Assessment for Rat Terrier using 7 STRs in the DLA region

	N	Na	Ne	Ho	He	F
Mean	34	7	4.05	0.81	0.73	-0.1
SE		0.54	0.44	0.03	0.03	0.02

III. Health

A. Lifespan

The Rat Terrier is considered a hardy breed with a life expectancy of 12-18 years according to the AKC [1] and 16-19 years by the Canine Health Information Center (CHIC) [6].

B. General health problems

Due to regular outcrossing throughout the breed history, complex genetic disorders have been limited mainly to diseases that existed in small dogs prior to breed formation.

However, the increase in popularity in recent years has tended to bring some of these disorders into focus. CHIC recommends that Rat Terriers be tested for patellar luxation, cardiac abnormalities, pancreatic issues, hip dysplasia, and Legg–Calvé–Perthes syndrome [6].

The Rat Terrier, like many small breeds, is more likely to have problems with their teeth, starting with tartar build-up that often leads to gingivitis, periodontitis, infection of the tooth roots, and tooth loss [7]. This is most likely due to the tendency of smaller dogs to avoid the heavy chewing activities of larger dogs. Tarter build up and gum disease can be prevented by encouraging chewing exercise and routine teeth exams and cleaning.

C. Heritable disease problems

The high level of genetic diversity and overall randomness of parent selection has greatly limited the expression of the simple Mendelian disease traits that have plagued many other breeds. The one exception is a recessive genetic mutation that induced hairlessness in a single Rat Terrier. This mutation was then used to crate the American Hairless Terrier [5].

Hairlessness in the dog has evolved independently at least twice [8, 9]. One form of hairlessness present in several breeds (Peruvian Inca Orchid, Chinese Crested, Mexican Xoloitzcuintle) is inherited as a dominant trait and is lethal in the homozygous state (two copies of the mutation). A second recessive form occurred initially in the Rat Terrier. Contrary to the dominant form, there are no adverse effects on dentition or fecundity associated with the recessive Terrier hairlessness.

The recessive Terrier hairlessness trait is caused by a frameshift deletion in the serum/glucocorticoid regulated kinase family member 3 gene (*SGK3*) [8, 9]. A 4 bp deletion (TTAG) in exon 4 of *SGK3* disrupts the protein coding sequence and is predicted to knock out the function of the gene. *SGK3* has been shown to affect postnatal hair follicle development in mice and appears to have a similar function in dogs: American Hairless Terriers are born with a thin coat of hair that is lost within first months of life. A commercial test is available for both mutations [9].

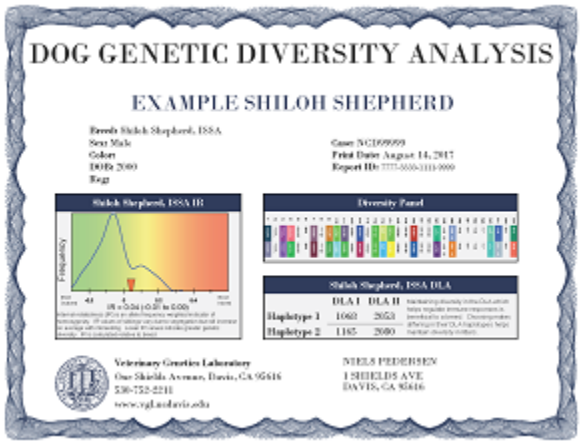
IV. What does this assessment of genetic diversity tell us about the contemporary Rat Terrier

The genetic diversity of the Rat Terrier has contributed to the overall soundness of the breed [6]. Whereas most modern breeds have evolved from a few founding dogs and then propagated from a closed gene pool, the Rat Terrier has benefited from a long history of refinement with regular outcrosses to bring in useful qualities and genetic variability.

V. Results of Diversity Testing

A. 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 being tested is related to the entire population. The alleles at each of the 33 STR loci are presented as numbers that correspond to those found in Table 1. Each locus will have two alleles, which are either different (heterozygous) or the same (homozygous). Each allele is inherited from each of the parents. More of the alleles at each locus will be homozygous in dogs from closely related parents or that in regions of the genome that are under strong positive selection for some favored phenotypic trait or traits. Dogs with a predominance of rarer (i.e., low incidence) alleles will be more distantly related to the bulk of the population than dogs that have a predominance of common (i.e., high incidence) alleles.


DOG GENETIC DIVERSITY ANALYSIS

EXAMPLE SHILOH SHEPHERD

Breed: Shiloh Shepherd, ISSA
Sex: Male
Color:
DOB: 2010
Reg:

Case: NG109999
Print Date: August 14, 2017
Report ID: ????-????-????-????

Shiloh Shepherd, ISSA IR

Diversity Panel

Shiloh Shepherd, ISSA DLA

	DLA I	DLA II
Haplotype 1	1060	2053
Haplotype 2	1165	2000

Veterinary Genetics Laboratory
One Shields Avenue, Davis, CA 95616
530-752-2211
www.vgl.ucdavis.edu

NIELS PEDERSEN
1 SHIELDS AVE
DAVIS, CA 95616

B. What should you do with this information?

The Rat Terrier possesses a great deal of genetic diversity similar to other small dog breeds such as the Jack Russell Terrier, Chihuahua, Miniature Poodle and Havanese. Except for the recessive hairless mutation and a low incidence of orthopedic and immune disorders inherited by descent from ancestral dogs, the breed is amazingly free of genetic problems. However, the increasing popularity of the breed and shifting emphasis towards conformation and showing puts the breed at risk for problems in the future. Therefore, it is timely for Rat Terrier breeders to come to agreement on performance and show breeding strategies and to continue to breed as randomly as possible and maintain their present heterogeneity and effective population size. This can best be done by using routine DNA testing to augment pedigree-based parent selection. In this scheme, pedigrees become most important for breed history and genealogy and DNA testing to maintain genetic diversity.

Although the breed is genetically diverse and average heterogeneity high, there was a small subpopulation of dogs that were much more inbred than the breed average.. Therefore, the goal for breeders should be to continue to produce puppies with IR scores less than 0, and with time, even lower scores. Although most of the individuals tested were randomly bred, 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 individuals, 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 pairings 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 v*s* 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.

A more effective use of this study is to contribute the genetic information to a web repository. 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 the dogs tested, potential mates that would be most ideal for increasing genetic diversity in their litters- https://www.betterbred.com/.

References

1. American Kennel Club. Rat Terrier. https://www.akc.org/dog-breeds/rat-terrier/

2. Wikipedia. Rat Terrier. https://en.wikipedia.org/wiki/Rat_Terrier

3. Wikipedia. Fiest (dog). https://en.wikipedia.org/wiki/Feist_(dog).

4. A Kennel Club Book (Google Books). Rat Terrier: A Comprehensive Owner's Guide. https://books.google.com/books?id=ZL8JBgAAQBAJ&pg=PT12&lpg=PT12

5. Wikipedia. American Hairless Terrier. https://en.wikipedia.org/wiki/American_Hairless_Terrier

6. OFA-CHIC health testing requirement. Rat Terrier. https://www.ofa.org/recommended-tests?breed=RAT

7. Animal Health Center. Rat Terrier. https://animalhealthcenternh.com/client-resources/breed-info/rat-terrier/.

8. Veterinary Genetics Laboratory, UC Davis, Hairlessness in Terriers. https://vgl.ucdavis.edu/test/hairlessness-terriers.

9. Parker, H. G., Harris, A., Dreger, D.L., Davis, B.W., & Ostrander, E.A. (2017). The Bald and the Beautiful: Hairlessness in Domestic Dog Breeds. Philosophical Transactions of the Royal Society B, 372(1713). doi: 10.1098/rstb.2015.0488.