Phenotype and Gene Frequencies of Red Blood Cell Groups in Dogs of Various Breeds Reared in Japan

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ABSTRACT. The occurrence of various red blood cell groups and corresponding genes was studied with dogs of various breeds reared in Japan. Thirteen antisera were used to determine the red blood cell groups. The frequency of the red blood cell groups of the DEA 1, DEA 3, DEA 6, D, and J1 systems had distinct tendency towards certain canine breeds. In particular, there was a pronounced difference in the frequency of D gene system between indigenous Japanese and European and American breeds. The Japanese breeds, Akita and Shiba, had a high frequency in the D¹ gene, while the European and American breeds showed a high frequency in the D² gene. There was no other deviations in the frequency of genes in any other system of red blood cell groups that might characterize the indigenous Japanese breeds. There was no relationship between sex and the frequency of genes in any system of the red blood cell groups.—**KEY WORDS:** antibody, antigen, dog, red cell.

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Seven systems of red blood cell groups (RGs) have been standardized in dogs [9]. They include DEA 1, 3, 4, 5, 6, 7 and 8. Efforts were made in various laboratories [7–9, 11] including this one [1] to produce antisera that specifically detect each of RGs. Consequently, 13 antisera have been established for the determination of RGs [1, 3]. Ten systems of RGs can be detected by them; DEA 1, DEA 3, DEA 5, DEA 6, D, J1, J2, J3, J4, and J5. The DEA 1 system was examined with beagles and mongrels. In addition, a genetic pedigree survey was conducted with these breeds [4]. The RGs mentioned above have been used as indicators giving information on histocompatibility for blood transfusion and transplantation or as markers for the identification of individuals [2, 3, 8, 10].

The present study was carried out to examine the occurrence of RGs and their genes in dogs in Japan with the use of the 13 antisera.

MATERIALS AND METHODS

Procedures for determining the red blood cell groups: The specificity of the antisera was established in reference to the previous results in preliminary experiment [1]. Of these antisera, those against DEA-1·1, 2, –1·1, –1·2, –3, –5, –6, J3, J4, or J5 were prepared by homologous immunization, those against D1 and D2 by heterologous immunization in rabbits [1]. One against J1 was naturally occurring antisera [5]. One against J2 were extracted from the seeds of the *Cloerodendron* [5]. Previously described methods were used for the preparation of red blood cells and for the determination of RGs [1].

Breeds: The red blood cell groups were determined with 15 breeds, including three indigenous Japanese breeds (Akita, Shiba, and Mongrel) and 12 European and American breeds (Afghan Hound, Beagle, Boxer, Col-

Table 1. Phenotype and gene frequency of the red blood cell groups of dogs

Red blood group system	Phenotype	No. examined	No. positive	Phenotype frequency(%)	Gene frequency
DEA 1	1.1	545	242	44	DEA 1 ⁺ : 0.2406
	1.2		120	22	DEA 2 ⁺ : 0.1763
	1(-)		183	34	DEA 1 ⁻ : 0.5831
DEA 3	3(+)	448	109	24	DEA 3 ⁺ : 0.1282
	3(-)		339	76	DEA 3 ⁻ : 0.8718
DEA 5	5(+)	448	46	10	DEA 5 ⁺ : 0.0513
	5(-)		402	90	DEA 5 ⁻ : 0.9487
DEA 6	6(+)	455	227	50	DEA 6 ⁺ : 0.2929
	6(-)		228	50	DEA 6 ⁻ : 0.7071
D	D1	1318	63	5	D ¹ : 0.1450
	D2		1003	76	D ² : 0.8550
	D1D2		252	19	
J1	J1(+)	448	179	40	J1 ⁺ : 0.2254
	J1(-)		269	60	J1 ⁻ : 0.7746
J2	J1(+)	420	136	32	J2 ⁺ : 0.1754
	J2(-)		284	68	J2 ⁻ : 0.8246
J3	J3(+)	447	88	20	J3 ⁺ : 0.1056
	J3(-)		359	80	J3 ⁻ : 0.8944
J4	J4(+)	447	169	38	J4 ⁺ : 0.2126
	J4(-)		278	62	J4 ⁻ : 0.7874
J5	J5(+)	448	25	6	J5 ⁺ : 0.0305
	J5(-)		423	94	J5 ⁻ : 0.9695

lie, English Pointer, English Setter, German Shepherd, Keeshond, Maltese, Shetland Sheepdog, Shih Tsu, and Yorkshire Terrier). The numbers of dogs examined for each RGs were within ranged between 420 to 1318. A large portion of samples were collected from dogs submitted to the Veterinary Hospital of the Nippon Veterinary and Zootechnical College, Tokyo. Among the 15 breeds, the dogs of some breeds (7 Collies and 5 Keeshond) were of the same or closely related family.

RESULTS

Table 1 shows the over all results in terms of 10 RGs systems. Table 2 to 6 show the results comparing the frequency of each system of the RGs and genes depending on the breed. There were remarkable differences in

the frequency of the DEA 1, DEA 3, DEA 6, D, and J1 systems depending on the breed examined.

In the DEA 1 system, the frequency of the DEA 1(+) type was high in five breeds, Keeshond, Shiba, Mongrel, Akita and Yorkshire Terrier; all were born in Japan. The dogs of the Collie and Keeshond breeds had close family relations, but the DEA 1(+) type was not found in the Collie, while the DEA 1.2 type was found in 80% of the Keeshond. These two breeds were different from the other breeds concerning the frequency of the DEA 1 system (Table 2).

In the DEA 3 system, the frequency of the DEA 3(+) type was high in three indigenous Japanese breeds; Akita, Mongrel and Shiba. On the other hand, in European and American breeds, the frequency of the DEA 3(+) type

Table 2. Phenotype and gene frequency of the DEA 1 system of various canine breeds

Breed	No. examined	Phenotype frequency(%)			Gene frequency		
		1.1	1.2	1(-)	1 ⁺	2 ⁺	1-,
Keeshond	5	20	80	0	0.1056	0.8944	0.0000
Shiba	19	79	16	5	0.4578	0.3186	0.2236
Mongrel	135	55	34	11	0.2553	0.4131	0.3316
Akita	7	57	28	15	0.2610	0.3517	0.3873
Yorkshire Terrier	5	80	0	20	0.5528	0.0000	0.4472
Maltese	9	33	44	23	0.1270	0.3934	0.4796
Afghan Hound	4	50	25	25	0.2113	0.2887	0.5000
Shih Tsu	7	57	14	29	0.2566	0.2049	0.5385
English Pointer	13	62	8	30	0.2994	0.1529	0.5477
English Setter	21	43	10	47	0.1779	0.1366	0.6855
Beagle	188	36	13	51	0.1386	0.1472	0.7142
German Shepherd	5	0	40	60	0.0000	0.2254	0.7746
Boxer	5	0	20	80	0.0000	0.1056	0.8944
Shetland Sheepdog	9	0	11	89	0.0000	0.0566	0.9434
Collie	7	0	0	100	0.0000	0.0000	1.0000

Table 3. Phenotype and gene frequency of the DEA 3 system of various canine breeds

Breed	No. examined	Phenotype frequency(%)		Gene frequency	
		3(+)	3(-)	3 ⁺	3 ⁻
Akita	7	100	0	1.0000	0.0000
Mongrel	125	50	50	0.2929	0.7071
Shiba	19	42	58	0.2384	0.7616
Afghan Hound	4	25	75	0.1340	0.8660
Yorkshire Terrier	5	20	80	0.1056	0.8944
Maltese	9	11	89	0.0566	0.9434
Beagle	89	0	100	0.0000	1.0000
Boxer	5	0	100	0.0000	1.0000
Collie	7	0	100	0.0000	1.0000
English Pointer	15	0	100	0.0000	1.0000
English Setter	22	0	100	0.0000	1.0000
Keeshond	5	0	100	0.0000	1.0000
German Shepherd	6	0	100	0.0000	1.0000
Shetland Sheepdog	9	0	100	0.0000	1.0000
Shih Tsu	7	0	100	0.0000	1.0000

was low. in particular, the Beagle, Boxer, Collie, English Pointer, English Setter, German Shepherd, Keeshond, Shetland Sheepdog and Shih Tsu were 100% positive with the DEA 3(-) type (Table 3).

The DEA 6 system had a high frequency in Beagles and a low frequency in four breeds; Afghan Hound, Shetland Sheepdog, Kees-

hond and Collie (Table 4).

The D system is divided into three phenotypes; D1, D2 and D1D2, based on the responses to two antisera; one against D1 and the other against D2. The frequency of type D1 was the highest in the Akita breed and type D1D2 was the highest in Shiba breed. Both the Akita and the Shiba breed are indig-

Table 4. Phenotype and gene frequency of the DEA 6 system of various canine breeds

Breed	No. examined	Phenotype frequency(%)		Gene frequency	
		6(+)	6(-)	6 ⁺	6 ⁻
Beagle	89	76	24	0.5101	0.4899
English Pointer	15	60	40	0.3675	0.6325
Akita	14	57	43	0.3443	0.6557
Shih Tsu	7	57	43	0.3443	0.6557
Maltese	9	56	44	0.3367	0.6633
German Shepherd	6	50	50	0.2929	0.7071
Mongrel	125	42	58	0.2384	0.7616
Boxer	5	40	60	0.2254	0.7746
Yorkshire Terrier	5	40	60	0.2254	0.7746
English Setter	22	32	68	0.1754	0.8246
Shiba	19	32	68	0.1754	0.8246
Afghan Hound	4	25	75	0.1340	0.8660
Shetland Sheepdog	9	22	78	0.1168	0.8832
Keeshond	5	20	80	0.1056	0.8944
Collie	7	0	100	0.0000	1.0000

Table 5. Phenotype and gene frequency of the D system of various canine breeds

Breed	No. examined	Phenotype frequency(%)			Gene frequency	
		D1	D2	D1D2	D ¹	D ²
Akita	14	50	14	36	0.6800	0.3200
Shiba	32	13	41	46	0.3600	0.6400
Mongrel	657	7	62	31	0.2250	0.7750
Afghan Hound	4	25	75	0	0.2500	0.7500
Yorkshire Terrier	5	0	80	20	0.1000	0.9000
Maltese	9	0	89	11	0.0550	0.9450
Beagle	374	0	99	1	0.0500	0.9500
German Shepherd	45	0	96	4	0.0200	0.9800
Boxer	5	0	100	0	0.0000	1.0000
Collie	7	0	100	0	0.0000	1.0000
English Pointer	15	0	100	0	0.0000	1.0000
English Setter	22	0	100	0	0.0000	1.0000
Keeshond	5	0	100	0	0.0000	1.0000
Shetland Sheepdog	9	0	100	0	0.0000	1.0000
Shih Tsu	7	0	100	0	0.0000	1.0000

enous breeds of Japan. There was a tendency for the D2 type to appear predominantly in the other breeds, and there were small differences within these breeds (Table 5).

In the J1 system, 100% of the Boxers had J1(+) type but only a few of the Keeshond and Shih Tsu had this type (Table 6).

Low frequencies were observed in some

breeds as to other systems of RGs, such as DEA 5 and J2 to J5. However, only a small number of subjects were examined in these breeds. The J3 and J4 systems had high frequencies in the Collie compared to the other breeds. Studies were also done to see whether there was any relationship between any RGs and sex, but no such relationship

Table 6. Phenotype and gene frequency of the J1 system of various canine breeds

Breed	No. examined	Phenotype frequency(%)		Gene frequency	
		1(+)	1(-)	1 ⁺	1 ⁻
Boxer	5	100	0	1.0000	0.0000
Collie	7	57	43	0.3443	0.6557
Afghan Hound	4	50	50	0.2929	0.7071
Mongrel	125	48	52	0.2789	0.7211
Shetland Sheepdog	9	44	56	0.2517	0.7483
Shiba	19	42	58	0.2384	0.7616
Yorkshire Terrier	5	40	60	0.2254	0.7746
Beagle	89	38	62	0.2126	0.7174
English Setter	22	36	64	0.2000	0.8000
Maltese	9	33	67	0.1815	0.8185
German Shepherd	6	33	67	0.1815	0.8185
Akita	7	29	71	0.1574	0.8426
English Pointer	15	20	80	0.1056	0.8944
Shih Tsu	7	14	86	0.0726	0.9274
Keeshond	5	0	100	0.0000	1.0000

was observed.

DISCUSSION

Attempts have been made in the present study to classify the membranous antigens of canine red cells by using naturally-occurring antisera [5], heterologous immune antisera [4-6], and lectin [5]. Internationally, antisera and antigens are being identified and standardized for the determination of RGs [9, 11].

The present results suggest a positive correlation between certain breeds and particular RG systems. For example, the frequency of DEA 1(-) type was high in three breeds, Boxer, Shetland Sheepdog and Collie. All the seven Collies examined showed DEA 1(-) type, but this probably is due to the fact that these dogs are closely related. Another example is the fact that Keeshond predominantly showed DEA 1·2 type. Keeshond have been bred to produce a type of cardiac deformity, conotruncal anomaly, for experimental purposes. Although the relationship between DEA 1·2 type and the cardiac is not clear, such genetic selection apparently pro-

duced the homogeneity in the RG system. These characteristics in blood cell antigens must be evaluated with caution, because the numbers of dogs examined in many breeds were very small.

It has been mentioned that DEA 1 is an important antigen concerning histocompatibility [8, 9]. When the RGs of a recipient dog is unknown in blood transfusions, it is necessary to use a dog of the DEA 1(-) type as a donor [3, 8]. The average frequency of all the types of the DEA 1 system was close to that reported by Vriesendorp *et al.* [9, 11] and Mears *et al.* [7]. The frequencies of the DEA 1(-) type were high in four breeds; German Shepherd, Boxer, Shetland Sheepdog, and Collie. It is, however, premature to conclude that dogs of these breeds are fit as blood donors as the numbers of dogs examined were small. It seems, though, that the Beagle is a reasonable choice for a donor from the results of this experiment.

Previous results from this laboratory suggested that type D1 of D system and DEA 3(+) type share specificity [1]. The present results support the suggestion; two breeds indigenous to Japan, Akita and Shiba, dem-

onstrated high frequencies for D1 and DEA 3(+). Dogs belonging to another Japanese breed, Shikoku, also exhibited D1D2 type and DEA 3(+) specificity in high frequency (data not shown). These results are consistent with the hypothesis that D1 and DEA 3(+) are closely related and that the specificity occurs in high frequencies in indigenous Japanese breeds. Mongrels in Japan had intermediate frequencies for the specificity between Japanese and Western breeds.

The frequency of the J1 system was examined in only five boxers. The J1(+) type appeared in 100% of these dogs. In Collies examined, the frequency of the J3(+) and J4(+) types was high. The reason may be that Collies, in general, maintain a close genetic relationship.

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要 約

日本におけるイヌの品種別の赤血球型出現頻度と遺伝子頻度について：江島博康・黒川和雄・池本卯典¹⁾（日本獣医畜産大学獣医第一外科学教室，¹⁾自治医科大学人間生物科学研究室）——日本におけるイヌ15品種の赤血球型出現頻度と遺伝子頻度について，13種類の赤血球型判定用抗体を用いて調査した。DEA 1, DEA 3, DEA6, Dおよび J1 システムはその赤血球型の頻度に品種間の明らかな差を認めた。それらの中で，とくに，Dシステムは日本在来種と欧米産種との間に明らかな偏在性を認めた。すなわち，日本在来種の秋田種，柴種の D¹ 遺伝子頻度は高く，一方，欧米産の品種では逆に D² 遺伝子頻度が高い値を示し，日本産雑種はその中間に位置する値を示した。他のシステムにおいては，日本在来種を特徴づけるような偏在性は認められなかった。なお，いずれの赤血球システムもそれらの頻度と性差との関連性はみられなかった。