

Allele Frequencies and Linkage Disequilibrium of Eight FVIII Gene SNPs in Dogs

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Introduction: Haemophilia A is an X-linked recessive bleeding disorder caused by a deficiency in factor VIII, which plays an essential role in blood coagulation.¹ The FVIII gene spans 144 kb dispersed across 26 exon in the dog. Single nucleotide polymorphisms (SNPs) are the available markers of choice for identifying individuals at risk of disease, measuring the variability between populations. In this study, the allele frequencies of eight FVIII gene SNPs were determined and linkage among these SNPs was analyzed in dogs.

Materials and Methods: Blood samples were obtained from 368 dogs (Sapsaree, n=170; German Shepherd, n=92; Jindo, n=74; small breed dog, n=24) and genomic DNA isolated from whole blood. The canine FVIII gene fragments including SNPs were amplified using seven primer sets and PCR. For the genotyping of SNP, we used SNaPshotTM multiplex kit (Applied Biosystems, Foster City, CA, USA) based on the addition of a specific fluorescently labeled ddNTP to extension primers.

Results: We have investigated 8 SNPs of FVIII gene in 368 dogs, found polymorphism in E14nt687 and E14nt1513 in exon 14, while not found in E8nt100, E10nt15, E12nt40, E12nt74, E14nt2991 and E25nt167. The heterozygotes of E14nt687 and E14nt1513 were seen in the all breed in this study. The allele frequencies of E14nt687 was A (n=38) A/G (n=33) G (n=67) and E14nt1513 was C (=42) C/T (=36) T (=68) in the Sapsaree. All of the E14nt687 heterozygotes were seen in the Sapsaree had heterozygotes for E14nt1513 SNP site. The E14nt687 A allele in 97% of the case (37/38 of the all observed A allele) cosegregated with the E14nt1513 C allele and the E14nt687 G allele in 99% (66/67) cosegregated with the E14nt1513 T allele. Linkage analysis showed strong linkage disequilibrium among these two loci in the Sapsaree.

Clinical relevance: We have examined allele frequencies and analyzed linkage of eight FVIII gene SNPs in the Sapsaree, the German Shepherd, the Jindo and the small breed dog. We found polymorphisms in E14nt687 and E14nt1513 in all breeds and strong linkage disequilibrium of these two loci in the only Sapsaree.

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