

EVALUATION OF POLYMORPHISM IN KLK1 GENE AFFECTING PERFORMANCE TRAITS IN CASPIAN HORSE BREED

Mohsen Goudarzi¹, Ghodrat Rahimi¹, Ardeshir Nejati-Javaremi²

¹*Department of Animal Sciences, Faculty of Animal and Fisheries, Sari Agricultural Sciences and Natural Resources University,*

²*Department of Animal Sciences, Faculty of Agronomy Sciences, College of Agriculture & Natural Resources, University of Tehran,*

Corresponding Author E-mail address: mohsengodarzi95@yahoo.com

ABSTRACT

Today, Caspian horse or that same Iran's miniature horse is more found in northern areas of Alborz mountain ranges. This horse is very similar to Arab horse and is one of oldest horses in the world. Low genetic illnesses and naturalness of their choice causes preserving their genetic sources during thousands of years. Kallikreins are a group serine proteases found in most diverse tissues and biological fluids. They were first specified and extracted as blood pressure regulator in 1930 (2). Most Kallikrein activities are related to the released products. Bradykinin and Kallidin are some of these products (1). Kallikrein gene contains 15 exons that has a role in regulation and control of blood pressure. Pancreas-renal Kallikrein gene or KLK1 is encoding for human Kallikrein 1 (hK1). This enzyme releases Lysyl-Bradykinin controlling blood pressure, electrolyte balance, inflammation and other physiologic reactions. The hK1 may affect some proteins, i.e. growth factor, some hormones, cytokines, and pleiotropic effect (2). The human KLK1 gene is located on chromosome 19q13.3 and its protein includes 262 amino acids. The most published reports are related to the polymorphism of Kallikrein gene in human and no any reports are available on KLK gene in horse. According to the importance of Kallikrein protein as an important plasma factor and its effect on performance and health traits, characterization of KLK gene based on molecular genetic markers is of priority in horse. In this study, blood samples were collected from 150 Caspian horses. The samples were kept in a cooling chain, transferred to the laboratory and stored at -20°C for further analysis. Genomic DNA was extracted using modified salting out method and the quantitative DNA specifications were specified by agarose gel electrophoresis. Polymerase chain reaction (PCR) were carried out in a total volume of 20 µl, containing 10 µl of master mix from Pars Tous Co. and added 3 µl of each primers (30 pmoles), 1.5 µl of genomic DNA (100 nano-gram), and 2.5 µl water. The thermo-cycling conditions included an initial denaturation at 95°C for 5 min, followed by 34 cycles of 30 seconds at 94°C, 30 seconds at 58°C for annealing temperature and 30 second at 72°C. A final elongation step was carried out at 72°C for 7 min. The presence of PCR product was tested on a 1% agarose gel and the polymorphism was detected by SSCP method on 14% polyacrylamide gel electrophoresis using silver staining. Genotype of each individual was specified based on band pattern on the gel. A fragment with the size of 155bp was amplified from exon 3 of Kallikeirn gene in all samples. Detection of two distinct SSCP patterns in all genotyped horses in the present study indicates the absence of polymorphism in Turkmen horse population. Further studies with the larger sample size and/or using other marker sites at this gene may increase the chance of finding polymorphism in Kallikeirn gene in horse population.

References

- 1- Hecquet C., Tan F., Marcic B.M., Erdos E.G. (2000). Human Bradykinin B2 receptor is activated by Kallikrein and other serine proteases. *Mol Pharmacol* 58: 828-36.
- 2- Yousef G.M., Diamandis E.P. (2001). The new human tissue Kallikrein gene family: structure, function, and association to disease. *Endocrine Reviews* 22(2): 184-204.