

Distribution of *GABPB1* gene rs7181866 polymorphism in rugby players

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Nuclear respiration factor 2 is an important respiratory factor for cellular metabolism formed by polypeptides produced by the GA binding protein transcription factor subunit alpha (*GABPA*) and GA binding protein transcription factor subunit beta 1 (*GAPPB1*) genes. A functional polymorphism on the *GABPB1* gene (rs7181866, G/A) alters the cellular respiration metabolism by affecting the protein dynamics. In this context, here, we examined the distribution of *GABPB1* rs7181866 polymorphism in professional rugby players to see its relationship with their stamina. Twelve rugby players participated in the study. Genotyping was performed by Real-Time PCR after DNAs were isolated from blood cells using commercially available isolation kits. In our cohort, the AA genotype and the A allele were superior to the AG and GG genotypes and the G allele. There was no statistically significant difference in terms of Genotypes ($p=0.9027$) and allelic groups ($p=0.6948$) in the athlete and control groups. Studies have revealed the relationship between endurance characteristics of athletes and rs7181866 AA genotype and A allele. These studies will be an important data source for future studies in sports genetics and *GAPPB1* analysis.

Keywords: Athletic performance, Endurance, Nuclear respiration factor, Sport genetics

Sport genetics is relatively a new field of science that examines the genetic regulation and functioning of athletes. One of the main objectives of sport genetics is to identify the genes that affect athletic performance and to determine their susceptibility to athletic performance^{1,2}. Genetic factors play an important role in athletic performance. Athletic performance is a common combination of an individual's inherent genetic abilities and subsequent environmental factors. It is reported that individual traits such as endurance, muscle coordination, strength, motivation, etc. have genetic background³⁻⁵. Existing research on athletic performance contributes to improving the performance of individuals⁶. Environmental factors such as training and nutrition are important for the development of elite athletes. However, these factors alone are not enough. Elite athletic performance is a complex sports phenotype that is largely determined by genetic potential⁷.

Rugby is a high-intensity sport that combines a variety of physical abilities. Rugby players must have a high level of strength, especially those involved in the

forward attack. Rugby players other than strikers should have the ability to sprint on a higher surface⁸⁻¹⁰.

Nuclear respiration factor 2 (also known as GA binding protein or *NRF2*) consists of subunits synthesized by the *GABPA* (GA binding protein transcription factor subunit alpha) and *GAPPB1* (GA binding protein transcription factor subunit beta 1) genes¹¹. During exercise, NRF-2 protein is thought to increase cellular respiration capacity and ATP production rate. This is due to its role in inducing mitochondrial biogenesis^{12,13}. The NRF-2 protein consists of five subunits (α , β 1, β 2, γ 1, γ 2) encoded by two genes. The α subunit is encoded by the α gene located at 21q21. The β subunit is encoded by the β gene located at 15q21.29¹⁴. NRF-2 protein associated with maximum oxygen consumption (VO_{2max}) in response to physical exercise has been reported. The polymorphism most associated with VO_{2max} is the rs7181866 polymorphism^{3,15}. Studies to date suggest that specific polymorphisms may be associated with endurance performance.

Although the *GAPPB1* rs7181866 polymorphism has considerable impact, there are not enough studies in the literature about this polymorphism. Here, we studied the distribution of *GABPB1* rs7181866 polymorphism in professional rugby players and

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compared the results with sedentary individuals. Additionally, we examined the advantage of the dominant “A” allele of rs7181866 polymorphism on athletic performance is also.

Materials and Methods

Study groups

Twelve rugby players participated in our study. As a control group, 110 individuals who did not exercise regularly participated in our study. Our study protocol, which was prepared in accordance with the Helsinki Declaration 2 (2015) guideline, was also approved by the Üsküdar University Non-Interventional Ethics Committee. Before the study, all participants signed consent forms containing all the information such as the study protocol, results, and evaluation of the results.

Genotyping

DNA isolations from the blood cells of the athletes participating in the study were performed with a commercially obtained PureLink DNA isolation kit (Invitrogen, Thermo Fisher Scientific, Inc.). Genotyping of the *GABPB1* rs7181866 polymorphism was performed using Real-Time PCR on a StepOnePlus (Thermo Fisher Scientific, Inc.) device and Taqman SNP Genotyping Assays genotyping kits according to the manufacturers’ protocols (cat. no. 4362691, Thermo Fisher Scientific, Inc.). A and G alleles were determined using FAM and VIC primers, respectively (Fig. 1). For a total volume of 10 µL reaction, 5 µL of Genotyping Master Mix (Applied Biosystems, Foster City, CA), 3.5 µL of nuclease-free H₂O (ThermoFisher, USA), 0.5 µL of genotyping test (Applied Biosystems), and 1 µL of DNA were used¹⁶. The sequences of the TaqMan Probe used for genotyping are listed in Fig. 2A.

Results and Discussion

All the rugby players had the AA genotype. No GG and AG genotypes were detected for *GABPB1* rs7181866 polymorphism. When allele distributions were examined, the ‘A’ allele was counted as 100%. No G allele was detected in our player cohort. In the control group (n = 110), 100 individuals had AA and 10 individuals had AG genotypes. No GG genotype was detected for *GABPB1* rs7181866 polymorphism. A allele was counted as 210 (95%) and G allele as 10 (5%). The genotype and allele number distributions of the athletes are summarized in Fig. 2 A & B.

Studies conducted so far show the formation and development of athletic performance through the interaction of different gene groups and environmental factors¹⁷. There are more than 200 polymorphisms associated with elite endurance performance in athletic performance¹⁸. The *GABPB1* gene plays an important role in most of the nuclear genes encoding various mitochondrial enzymes¹⁹. Therefore, *GABPB1* gene contributes to increased respiration capacity and ATP production rate during exercise. In this way, it is considered as a genetic marker of endurance²⁰.

GABPB1 gene is associated with high maximum oxygen consumption (VO_{2max}). There are 2 important

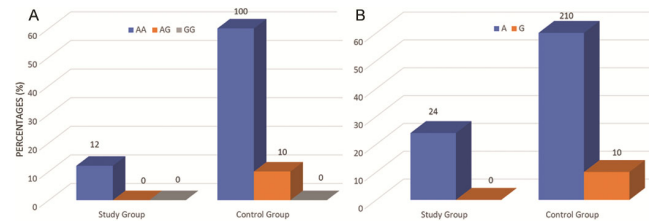


Fig. 2 — (A) Genotypic; and (B) Allelic distributions of *GABPB1* rs7181866 polymorphism in our cohort

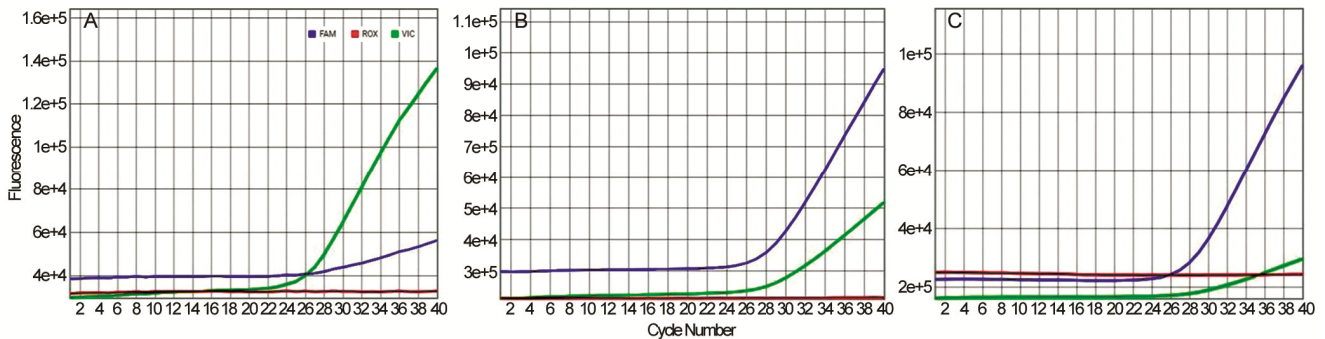


Fig. 1 — Quantitative Real-Time PCR amplification of the AA, AG and GG genotype of *GABPB1* rs7181866 polymorphism. (VIC (green curve) indicates A allele, FAM (blue curve) indicates G allele. (A) Single green curve indicates homozygous genotype of AA; (B) green and blue curves indicate heterozygous genotype of AG; and (C) single blue curve shows homozygous genotype of GG.

polymorphisms related to this relationship: rs8031031 (intron4; C/T) and rs7181866 (intron3; A/G). The most relevant polymorphism is the A/G transition polymorphism in intron 3 (rs7181866). In the studies, the polymorphism of *GABPB1* rs7181866 was examined and it has been shown that the AA genotype and the A allele are more common in rugby players^{15,21-23}.

GABPB1, known as nuclear respiratory factor 2 (NRF2), activates mitochondrial genes that are responsible for oxidative phosphorylation. Earlier studies on *GABPB1* reported that two single nucleotide polymorphisms (SNPs), such as rs7181866 and rs8031031, are associated with increased endurance in athletes^{24,25}.

In the literature, there are not enough studies on *GABPB1* rs7181866 polymorphism. *GABPB1* AA and AG genotypes stimulate higher NRF2 protein expression, an advantage for endurance in athletes¹³. Further, *GABPB1* AA polymorphism has been shown to be associated with success in endurance-type sports¹⁴. Studies have also established the relationship between *GABPB1* AG genotype and G allele and elite endurance athletes. In addition, the *GABPB1* AG genotype and the *GABPB1* G allele can provide elite endurance athletes an advantage^{26,27}.

Conclusion

In the present study, where we examined the *GABPB1* rs7181866 polymorphisms in endurance athletes, we observed that the AA genotype and the A allele were more dominant than the other genotypes and alleles. Considering that individuals from different populations were included in the studies, we can say that the *GABPB1* rs7181866 polymorphism provides a general predisposition to athletic performance, especially in endurance athletes. In order to make clearer interpretations, analysis should be made in larger groups of athletes with the same ethnicity and similar characteristics.

Conflict of interest

Authors declare no competing interests.

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