Lack of association between insulin-like growth factor-I (IGF-I) gene polymorphism (rs35767) and power performance in professional, amateur athletes and non-athlete individuals in Iranian population

Mohammad Reza Batavani[1*], Sayyed Mohammad Marandi[2], Kamran Ghaedi[3], Fahime Esfarjani[2]

1. Department of Physiology, Faculty of Center of Physical Education, Isfahan University of Technology, Isfahan 84156-83111, Iran
2. Department of Physiology, Faculty of Physical Education and Sport Sciences, University of Isfahan, Isfahan, Iran
3. Department of Biology, Faculty of Sciences, University of Isfahan, Isfahan, Iran

***Corresponding author:**Tel: +98 9132398493 Fax: +98 3133912940-1
Address: Department of Physiology, Faculty of Center of Physical Education, Isfahan University of Technology, Isfahan 84156-83111, Iran
E-mail: batavani@cc.iut.ac.ir
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Abstract

Introduction: Genetic variation between individuals may include single nucleotide polymorphisms. These individual differences could change human phenotypes (such as skeletal structure, heart and lung size). Insulin-like growth factor-I (IGF-I) is a critical factor in growth and exercise-associated muscle development. In this study we analyzed the frequency of IGF-I rs35767 polymorphism in karate-Kas athletes to find whether there is a difference among professional, amateur athletes and non- Karate-Kas.
Materials and methods: Two hundred and fifty-four athletes (95 professional Karate-Kas, and 159 amateur Karate-Kas) and 159 non-athletic healthy individuals participated in the current study. Following provision of informed consent, 2 mL of blood samples were taken. After DNA isolation, the genotyping frequencies of the IGF-I gene C/T polymorphism was assessed by restriction fragment length polymorphism (RFLP) analysis. Statistical analyses carried out using SPSS software (version 20) and data with $P<0.05$ were considered to be significant.
Results: Frequency of T/T and C/T genotypes in comparison to C/C genotype was increased in professional Karate-Kas and amateur Karate-Kas versus to control volunteer (odd ratio > 1) however, was not significant. Odd ratios of T/T and T/C genotypes compared to C/C in professional group versus controls were 1.125 (95%CI: 0.556 - 2.275) and 1.034 (0.538 - 1.984), respectively. Also, odd ratio of T/T and T/C in amateur group versus controls were 1.002 (0.535 - 1.876) and 1.256 (0.715 - 2.205), respectively.
Conclusion: The IGF-I gene C/T polymorphism is not significantly associated with the physical performance´s levels of karate-kas athletes in Iranian population.

Keywords: IGF-I, Polymorphism, Professional Karate-Kas, Amateur Karate-Kas

Introduction

Physical performance is a multifaceted phenomenon; furthermore it is dependent to both intrinsic factors (e.g., genetics and psychological profile) and extrinsic factors (e.g., training and overall health conditions). One of the most complex

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human traits is athletic performance, since depends on several body systems such as: respiratory, nervous and musculoskeletal. Individual differences of body system, are origin of variety in strength, power, body morphology and endurance, which are key factors underlying physical performance (1).

Around 40% of body mass in human consists of skeletal muscle. Since this type of muscles have numerous critical functions such as locomotion, glucose and fat metabolism and thermogenesis, the mechanisms regulating muscle growth and development are widely studied(2).Muscle is an extremely plastic tissue, which, exercise, nutrition and hormonal factors are key regulators of muscle mass(3). According dramatic effects of Anabolic steroids on muscle bulk, these hormones have long been used by body builders. Anabolic activity of insulin-like growth factor-I (IGF-I) have been demonstrated and recent studies demonstrated that exercise-associated muscle growth and development depends on IGF-I expression level(2). IGF-1 participates in diverse biological process such as: skeletal growth, cell cycle progression, differentiation, and mediator for various hormones. So numerous SNPs of IGF-1 have been studied to be involved in cancer, myopia, muscle function, BMD regulation and affects age at menarche(4, 5). C-1245T (rs35767) is one of these genetic variation that is located in IGF-I gene's promoter. Previous studies have demonstrated that rs35767 is associated with the endurance performance and power in athletics (6).

Karate, the Japanese word for "empty hands", developed in Japan in the early 17th century. Four karate styles are categorized by the World Karate Federation: Goju, Shito, Shotokan and Wado. Chaabène et al. reviewed physical and physiological features of karate athletes in several aspect such as Body Composition, Bone Mineral Density, Aerobic and Anaerobic Profile, Muscle Power and Flexibility (7). It is reported that achieving top karate performances depends on muscle explosive power plays a major role in(8, 9). In this study, we aimed to investigate the frequency of rs35767, in three groups of professional karate-kas, amateur athletes and non-athletes.

Materials and methods

Subjects: 254 Karate-kas (95 professional Karate-kas, and 159 amateur Karate-kas) and 159 non-athletes participated in this case-control study. All subjects in professional group were member of Iran national teams who has earned a medal or worthy score in world, Asian, international or national championships. All members in amateur group succeeded to take the black belt in karate (at least four years' experience in karate), but they didn't succeeded to get any score or medal in aforementioned championships. The main condition for including subjects in the control group was the absence of previous experience in any sport on a regular basis. After signing an informed consent form for agreement to participation in this study by the persons, 2 mL of blood sample was taken from each individual and collected in EDTA contain tubes.

DNA isolation: In this process, 2mL blood samples were taken from each individual, after completing a consent form. Genomic DNA was extracted from peripheral blood samples using standard salting out procedure and for evaluate quantity and quality of extracted DNA, absorbance at 260/280 nm, measured by a NanoDrop spectrometer (Nanodrop 1000; Thermo Scientific, Wilmington, NC, USA), and by electrophoresis on 1% agarose gel. Extracted DNAs were stored at -20 °C till 2further usage for genotyping analysis.

Genotyping: Allele specific PCR (ASP) was utilized to genotyping rs35767. To perform ASP, three primers designed (Table 1) and purchased from MacrogenInc (Seoul, Korea), one common primer and two allele specific primers

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which used in separate set of PCR. Primers were purchased from MacrogenInc (Seoul, Korea). PCR reactions were performed by PCR kit (Fermentas, Thermo Scientific, Germany) in a total volume of 25 µL containing 50-100 ng of genomic DNA, 2.5 µL of 10X PCR buffer containing 50 mM MgCl2 (Sinaclon), 1 µL of 5 pmol of each primer, 0.5 µL of dNTPs mix (10 mM), 1 U/µL of taq DNA polymerase and deionized water. PCR products were analyzed by electrophoretic separation on 1% agarose gel followed by staining with ethidium bromide and visualization in transmitted ultraviolet light.

Statistical analysis

Statistical Package for the Social Sciences (SPSS) (Ver. 20) software used for statistical analysis. Calculation of Odds ratio (OR) with 95%, confidence intervals (95% CI) was used to assess correlations between SNPs and Professional, Amateur Athletes possibility.

Results

The electrophoresis of the PCR products were led to diagnosis of the types of the allele (C/C or T/T in case of 1 response [just response of C or T allele, respectively], and C/T in case of 2 response [both responses of C and T allele]) and genotype of all subjects (Figure 1). The distribution analyses of genotype frequency by the IGF1 gene (C/T polymorphism) in Professional, Amateur karate-kas and Non-athlete groups are shown in Figure 2.

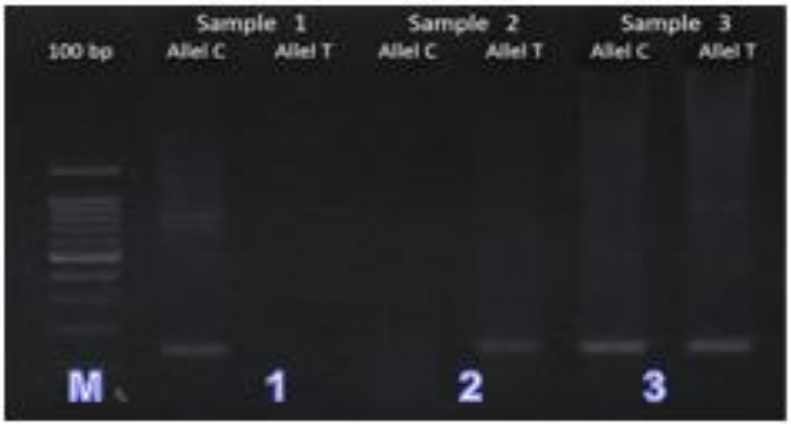


Figure1. Representative of an agaros gel electrophoresis of PCR partial products of IGF1 gene. 1; CC genotype (1 response to C allele), 2; TT genotype (1 response to T allele) and lanes 3; CT genotype (1 response to C allele and 1 response to T allele). M is DNA 100 bp ladder (Fermentas**).**

Statistical analysis performed by multinomial logistic regression for differentiating three groups to estimate OR of each genotype (Table 2). As shown in Table 2, there is no statistical significant association between IGF-I Gene Polymorphism (rs35767) and Professional, Amateur karate-kas and Non-athlete groups. Frequency of T/T and C/T genotype in comparison to C/C genotype increased in two groups of Karate-kas versus to Non-athlete group (OR > 1).

Discussion

As mentioned previously, rs35767 is located in IGF-I gene's promoter. Association between IGF1 and rs35767 polymorphism demonstrated by Palles and colleagues already (10).Since IGF1 have key role in variety of physiological process, lot of studies focused on rs35767 in diversity of condition such as age-related decrease in muscle mass and strength, physical ability, baseline free fat

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mass, body composition and muscle function(11-14).

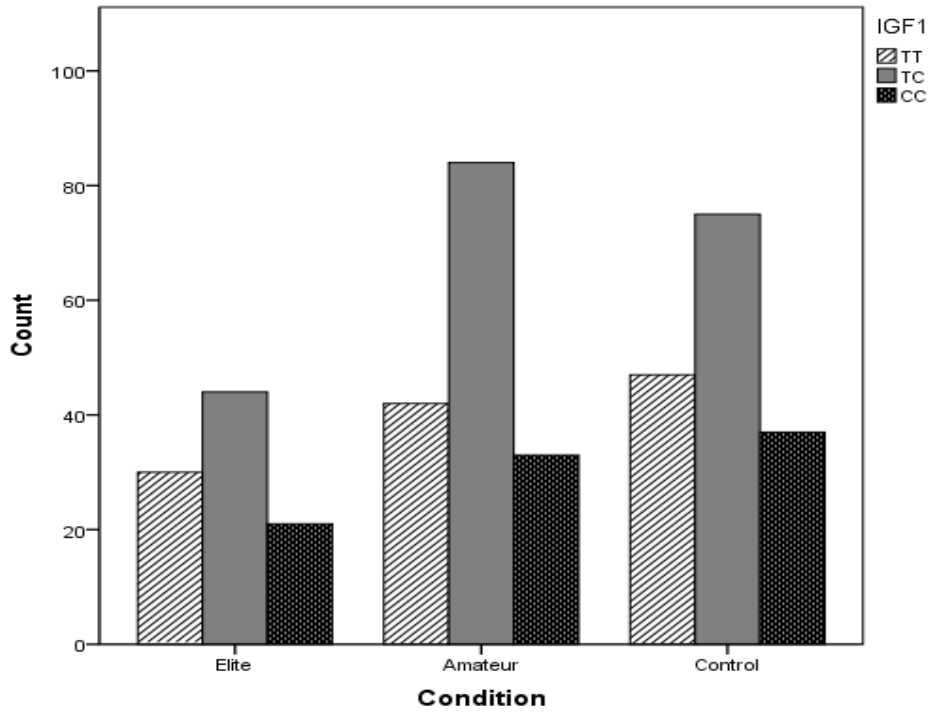


Figure 2. Frequency of TT, TC and CC genotype in three groups of study. Professional Karate-Kas (Elite), amateur Karate-Kas (Amateur) and nonathletic healthy individuals (Control).

Table 2. Multinomial logistic regressions for rs35767 in professional, amateur athletes and non-athlete groups.

	Professional Karate-Kas		Amateurs	
IGF-I Gene Polymorphism (rs35767)	Odds Ratio(95% CI)	P value	Odds Ratio(95%CI)	P value
T / T	1.12(0.56-2.27)	0.74	1.00(0.54-1.88)	0.99
T / C	1.03(0.54-1.98)	0.92	1.27(0.71-2.20)	0.43
C / C	1.000		1.000	

The reference category was nonathletic healthy individuals group. CI, confidence interval.

Although association of IGF1 polymorphism and professional athletes reported previously in Poland and Israel's population(6,15), but in Iranian population no significant association observed. Recently Miyamoto-Mikami and colleagues(16),21 genetic markers in Japanese sprint/power track and field athletes and assessed total genetic score (TGS), proposed by Williams and Folland (17) as a genetic algorithm to predict the genetic potential for endurance performance. But finally reported that the TGSs based on the selected genetic markers, were not able to predict the difference in Japanese population. Therefore based on this study and our result, there is differences in genotype–phenotype associations between Asian and Caucasian populations.

Conclusion

Taken together with our present case-control study indicate that rs35767, the IGF-I gene C/T polymorphism, is not significantly associated with the physical performance´s levels of karate-kas athletes in Iranian population. However, genetic analysis on a larger population is required.

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Conflict of interests

None of the authors declare competing financial interests.

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