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ScienceDirect
Metabolism Clinical and Experimental 56 (2007) 1649 – 1651
Metabolism
Clinical and Experimental
www.elsevier.com/locate/metabol

Association between a β_2-adrenergic receptor polymorphism and elite endurance performance

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Received 16 December 2006; accepted 3 July 2007

Abstract

The Arg16Gly single nucleotide polymorphism of the human β_2-adrenoceptor (*ADRB2*) gene was evaluated in a case-control study that included 313 white male elite endurance athletes and 297 white male sedentary controls (SCs) recruited in a multicenter project from North America, Finland, and Germany. The groups were matched by country of origin. The elite endurance athletes were required to have a maximum oxygen uptake $\geq$75 mL·kg^{-1}·min^{-1} (mean [SD], 79.0 [3.5]), whereas SC subjects had to be sedentary with a measured maximum oxygen uptake $\leq$50 mL·kg^{-1}·min^{-1} (40.1 [7.0]). Polymerase chain reaction technique was used to amplify the single nucleotide polymorphism–containing region in codon 16 of the *ADRB2* gene. *ADRB2* genotypes were in Hardy-Weinberg equilibrium in both groups. Genotypes did not differ between countries or sports of the athletes. The χ^2 analysis for the genotype distribution showed a significant difference between the 2 cohorts (P = .030), suggesting a positive association between the tested Arg16Gly polymorphism and endurance performance. Comparing carriers vs non-carriers for the 2 alleles, an excess of Gly allele carriers was seen in the SC group (P = .009), indicating an unfavorable effect of the Gly allele with respect to the performance status. In conclusion, we found suggestive evidence that the Arg16Gly polymorphism in the gene encoding for the β_2-adrenergic receptor may associate with endurance performance status in white men.

1. Introduction

Elite endurance performance is a multifactorial and oligogenic trait. However, not much is known about single genetic markers contributing to this complex phenotype. The adrenergic receptors are involved in several performance-related pathways and are therefore of particular interest as candidate genes for performance phenotypes. Especially the β_2-adrenergic receptor (*ADRB2*) gene is a possible candidate for the variation in endurance performance levels because of its contribution to the regulation of energy expenditure and lipid mobilization from human adipose tissue. In addition, *ADRB2* has a pivot role in the regulation of cardiovascular function. Based on these physiological implications, several studies have investigated the role of genetic polymorphisms known to alter the ADR β-receptor function [1,2]. Based on these findings, we summarize that even subtle changes in the function of ADR β-receptors may lead to changes in cardiovascular, respiratory, or metabolic control. Therefore, we hypothesize that the Arg16/Gly single nucleotide polymorphism (SNP) of the *ADRB2* could be associated with elite endurance performance.

2. Material and methods

A total of 610 male white subjects were included in the study: 313 male endurance athletes with a maximum oxygen uptake (VO_{2max}) of at least 75 mL·kg^{-1}·min^{-1} (mean, 79.0 mL·kg^{-1}·min^{-1}; SD, 3.46; range, 75.0-92.9) and 297

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0026-0495/$ – see front matter © 2007 Elsevier Inc. All rights reserved.
doi:10.1016/j.metabol.2007.07.006

sedentary male controls (mean VO_{2max}, 40.1 mL·kg^{-1}·min^{-1}; SD, 7.0; range, 17.2-50.0). The athletes were recruited from Germany (n = 186), North America (n = 76), and Finland (n = 52) and represented the following sports: cross-country skiing (n = 104), biathlon (n = 86), cycling (n = 71), running (n = 38), and other endurance sports such as triathlon and rowing (n = 14). All the athletes had been competing at the national or international level for several years. The control group comprised healthy sedentary subjects from the same geographical areas as the athletes. The VO_{2max} of the athletes was determined in the course of incremental exercise tests on cycle ergometers or motor-driven treadmills when the athletes were at their peak. The VO_{2max} values of the control subjects were measured in incremental exercise tests on cycle ergometers. The study was approved by the ethics committee of the participating institutions, and all subjects provided written informed consent for participation.

Genomic DNA was isolated from lymphoblastoid cell lines or white blood cells following a standard protocol. Polymerase chain reaction amplification was performed using a Biometra Thermocycler (Biometra, Goettingen, Germany). The 20-μL reaction volume contained 50 ng DNA, 10 pmol of each oligonucleotide, 0.2 mmol/L of each deoxyribonucleotide triphosphate (dNTP), 1.5 mmol/L $MgCl_2$, 0.5 U Taq polymerase, 16 mmol/L ammonium sulfate, 50 mmol/L Tris-HCl (pH 8.8), and 0.01% Tween 20. The following oligonucleotides were used to amplify the 200–base pair (bp) fragment: 5′-CTT CTT GCT GGC ACG CAA T-3′ and 5′-CCA GTG AAG TGA TGA AGT AGT TGG-3′. The polymerase chain reaction program was a modification of Large et al [2]. After an initial 3-minute denaturation, followed by 35 cycles of 1 minute with an annealing temperature at 62°C, a final elongation step was performed for 10 minutes at 72°C. The 200-bp fragment was digested with 1 U of *Bst*DI at 65°C for 2 hours. The obtained fragments showed up with 16, 56, and 128 bp for the Arg16 allele and 16, 20, 56, and 108 bp for the Gly16 allele. The fragments were visualized after electrophoresis on an ethidium bromide–stained 2% agarose gel using UV illumination.

All statistical analyses were performed with version 12.0 of the SPSS statistical software package (SPSS, Chicago, IL). A χ^2 test was used to compare allele and genotype frequencies between athletes and controls, as well as among different sports and places of origin of athletes.

Table 1
Allele and genotype frequencies for the *ADRB2* Arg16Gly polymorphism in EEAs and SCs

	EEAs	SCs	χ^2 statistics
Genotype			
Arg/Arg	52 (0.17)	28 (0.09)	
Arg/Gly	141 (0.45)	149 (0.50)	
Gly/Gly	120 (0.38)	120 (0.41)	$\chi^2 = 7.01/P = .030$
Allele			
Arg	245 (0.39)	205 (0.35)	
Gly	381 (0.61)	389 (0.65)	$\chi^2 = 2.801/P = .094$

Numbers for genotype and allele counts are total numbers; percentage values are given in parentheses.

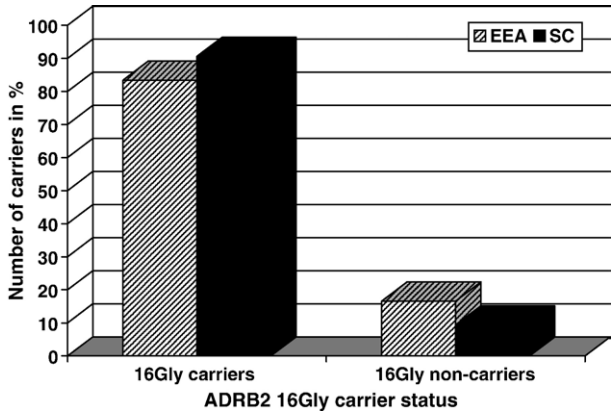

Fig. 1. Carriers and noncarriers of the 16Gly allele of the Arg16Gly polymorphism in the *ADRB2* gene ($\chi^2 = 6.91$, $df = 1$, $P = .009$).

3. Results

Genotype distributions for both groups were in Hardy-Weinberg equilibrium. Allele and genotype frequencies of the Arg16Gly marker for both groups are shown in Table 1. A χ^2 test revealed a significant difference ($\chi^2 = 7.01$, $df = 2$, $P = .030$) in the genotype distribution between athletes and controls. Comparing carriers vs non-carriers of the Arg16Gly alleles, we found a statistically significant excess of the 16Gly allele in the sedentary control (SC) group (Fig. 1; $\chi^2 = 6.91$, $df = 1$, $P = .009$). No difference in allele frequencies or genotype distributions was seen in the athlete group with regard to sports ($P > .05$) or places of origin ($P > .05$). In addition, we found no differences in allele frequencies with regard to the anthropometric data in the elite endurance athlete (EEA) group ($P > .05$) as well as in the SC group ($P > .05$).

4. Discussion

An increasing number of articles on physical performance and fitness phenotypes focus on genetic markers of human performance traits [3]. In the present study, we found evidence for a significant association between the Arg16Gly SNP in the *ADRB2* gene and endurance performance status.

Based on previous findings, 2 major physiological mechanisms appear to be relevant. The first has to do with the central role of catecholamines in the regulation of substrate mobilization. In an early study by Large and coworkers, the Arg16Gly SNP was shown to be of physiological relevance. The Gly16 carriers showed a 5-fold increase in agonist sensitivity without any change in *ADRB2* expression [2]. In a large sample of the Bogalusa Heart Study, the Gly16 carriers showed a significantly greater increase in body mass index over a long follow-up

period compared with Arg16 homozygotes [4]. In practice, body mass and composition play a major role in elite endurance performance. In our study, the frequency of the Gly16 allele carriers was greater in the SCs, whereas the Arg/Arg genotype was found to be more prevalent in the athlete group (17% vs 9%; Table 1). Thus, subjects carrying 2 copies of the Arg allele could, compared with subjects with 1 or 2 Gly alleles, benefit in terms of a lower body weight and a more favorable weight-to-strength ratio, which are important factors for all athletes competing in endurance sports.

The second potential explanation for the results of our study comes from the cardiovascular impact of the β-adrenergic receptors. *ADRB2* is highly expressed throughout the cardiovascular system, contributing to the increase in myocardial inotropy, chronotropy, and arterial vasodilatation. Several *ADRB2* polymorphisms were found to be associated with various cardiovascular phenotypes. Patients with heart failure showed significantly different exercise-capacity measures depending on the *ADRB2* genotypes [5]. In patients with the Arg16 allele, a higher peak VO_2 value compared with that in the Gly16 carriers was found. This was explained by a greater degree of down-regulation of the β2-ADR in the Gly16 carriers resulting in a lower peak VO_2. Therefore, it is reasonable to hypothesize that athletes could profit from the Arg16 genotype as opposed to the Gly16 variant, which was more prevalent in our control group.

The presence of other variants in the *ADRB2* gene may influence these findings as well. Arg16 homozygotes are often homozygous for the Glu27 allele at the Glu27Gln SNP [2]. Glu27 carriers in a cohort of 63 older women with different fitness levels were shown to have better endurance performance [6]. As the Glu27 allele is often inherited with the Gly16 allele of the Arg16Gly SNP, the results of both studies are generally consistent.

The quintessence of the *ADRB2* polymorphism research thus far is that genetic variants in this gene do not contribute to extreme phenotypes such as single-gene diseases. Sequence variants in *ADRB2* appear more likely to influence risk factors and modulate physiological phenotypes. From the present study, we conclude that the Arg16Gly SNP in the *ADRB2* gene plays a role in discriminating SC subjects from EEAs and that the Gly allele does not appear to favor a high aerobic capacity, known as an essential requirement for an extraordinary endurance performance.

Acknowledgments

Thanks are expressed to the colleagues who kindly provided some of the DNA samples. Furthermore, we would like to thank J. Fischer for her technical assistance. This work was supported by the German Institute for Sports Science Grant VF 0408/03/01/2002-2005. C. Bouchard is partially supported by the George A Bray Chair in Nutrition.

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