Trp64Arg polymorphism in ADRB3 gene is associated with elite endurance performance

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ABSTRACT

In this study, allele and genotype frequencies of the ADRB1 Arg389Gly (rs1801253), ADRB2 Gly16Arg (rs1042713) and Gln27Glu (rs1042714), and ADRB3 Trp64Arg (rs4994) variations were compared in the following three groups of Spanish (Caucasian) men: (1) world-class endurance athletes (E; runners and cyclists, n=100), (2) elite power athletes (P; sprinters, jumpers and throwers, n=53) and (3) non-athletic controls (C; n=100). No significant differences were observed in genotype and allele distributions among the study groups except for the ADRB3 Trp64Arg polymorphism in E versus C (27% vs 8% of carriers of the Arg allele in E and C, p<0.001; frequency of the minor Arg (C) allele of 14% vs 4% in E and C, p=0.001). Heterozygosity for the ADRB3 Trp64Arg polymorphism seems to be associated with elite endurance performance, while other variants of the β-adrenergic receptors’ genes do not seem to significantly influence top-level sports performance, at least in athletes of Spanish origin.

β-Adrenergic receptors (βARs) are members of a family of G protein-coupled receptors that are stimulated by naturally occurring catecholamines. In humans, three βARs are known: β1AR, β2AR and β3AR. The genes that encode these receptors are associated with metabolic and cardiovascular phenotypes (see below); as such, they are candidates to influence human exercise capacity.

The Arg389Gly variation (rs1801253) in the β1AR gene (ADRB1) influences exercise capacity phenotypes such as peak oxygen uptake in patients with cardiac disease, yet the results are so far contradictory.1 2 The Gly16Arg (rs1042713) polymorphism in the human β2AR gene (ADRB2) is associated with the metabolic syndrome3 and with cardiovascular phenotypes during exercise in healthy non-athletes4 5 and patients with cardiac disease.6 It seems that it can also influence elite endurance performance status, with the Gly allele exerting an unfavourable effect.5 Likewise, the ADRB2 Gln27Glu polymorphism (rs1042714) is associated with cardiovascular function during exertion7 and with endurance sports performance, at least in postmenopausal women.8

Less is known about the potential role of the β3AR gene (ADRB3) in exercise performance. The β3AR couples to Gs to activate adenylyl cyclase in adipose tissue, thereby stimulating lipolysis and thermogenesis.9 10 The Arg allele of the ADRB3 Trp64Arg polymorphism (rs4994), which is associated with reduced agonist-stimulated adenylyl cyclase activity in vitro,12 was originally linked with an increased risk of weight gain in obese people.13 Subsequent research failed, however, to show an association between the Trp64Arg variation and body composition phenotypes, at least in Caucasians.14 15 Beyond its metabolic functions, the β3AR is present and functional in the human heart.16 Stimulation of β3ARs can have a negative inotropic effect.17 18 The β3ARs also regulate angiogenesis and endothelium-dependent vasorelaxation in the coronary microvasculature.11

Whether the ADRB3 Trp64Arg polymorphism is associated with sports performance is unknown. The purpose of this study was to compare allele and genotype frequencies of the ADRB3 Trp64Arg variants in male Spanish (Caucasian) non-athletes (controls) and in elite male athletes who are at the two end points of the performance continuum, that is, endurance (professional road cyclists and endurance runners) and power (sprinters, jumpers and throwers). We hypothesized that owing to the putative role of β3ARs on some important phenotype traits involved in endurance exercise capacity (energy metabolism and particularly, cardiovascular function), the Trp64Arg polymorphism in ADRB3 gene could be associated with elite endurance performance. We also compared the allele and genotype frequencies of the ADRB1 Arg389Gly, and ADRB2 Gly16Arg and Gln27Glu polymorphisms.

METHODS

The studied population comprised (1) 100 healthy male non-athletic controls (age, 18–30 years, see below); (2) 100 male endurance athletes who competed within the last 10 years (age, 20–39 years; 50 world-class endurance runners, including European champions and Olympic finalists, and 50 professional road cyclists who were all Tour de France finishers, including top 3 finishers); and (3) 53 male power athletes (40 top national level and 13 Olympic level) who also competed within the last 10 years (age, 20–33 years; jumpers, throwers and sprinters). All participants were of the same Caucasian (Spanish) descent for at least three generations. All participants in the control group were undergraduate Physical Education students from the same university (Universidad Europea de Madrid, Spain). Inclusion and exclusion criteria for this group were to be free of any diagnosed cardiorespiratory disease and not to be engaged in formal, supervised endurance training (ie, performing less than three structured weekly sessions of strenuous endurance exercise as running,
swimming and bicycling), respectively. The athlete sample size is limited because we wanted to ensure that the athletes group were at the “world-class” level, that is, they were in the upper end of the human endurance and power performance continuum. The mean (SD) VO$_{2}$max of endurance athletes was 73.7 (5.7) ml/kg/min, and that of power athletes was 60.3 (5.5) ml/kg/min.

Written consent was obtained from each participant. The study protocol was approved by the institutional ethics committee (Universidad Europea de Madrid, Spain) and was in accordance with the Declaration of Helsinki for Human Research of 1974 (last modified in 2000). Our study was in accordance with stringent recommendations for replicating human genotype–phenotype association studies.

We extracted DNA from saliva or blood samples over the years 2004–2008. All genotyping was performed in the same laboratory (Progenika Biopharma, Parque Tecnológico de Zamudio, Derio-Vizcaya, Spain) during the year 2008 using a newly developed low-density DNA microarray based on allele-specific probes. The design, fabrication, validation and analysis of the arrays were performed following the procedure described in Tejedor et al with minor modifications. We compared the genotype and allele frequency for each variant between the three groups with the χ$^2$ test with a set at 0.05 and adjusted for multiple comparisons. Because of the low number of participants with the CC variant of the ADRB3 Trp64Arg polymorphism, the CT and TT genotypes were combined.

## RESULTS

All genotype distributions in the study participants were in Hardy–Weinberg equilibrium, except the ADRB2 Gln27Glu variant in the endurance athletes group. Table 1 shows the genotype and allele frequencies of the studied polymorphisms in Spanish controls and in endurance and power athletes. We did not observe significant differences in genotype and allele frequencies among the study groups except for the ADRB3 Trp64Arg polymorphism in the controls versus the endurance athletes.

### DISCUSSION

While keeping in mind the relatively limited sample of our cohorts (especially of control participants), the main finding of our study was that heterozygosity for the ADRB3 Trp64Arg polymorphism is associated with elite endurance performance, with the frequency of the minor Arg (C) allele and of C allele carriers being higher in world-class endurance athletes than in non-athletic controls. It is noteworthy that a similar trend was found when comparing the frequency of C allele carriers in endurance and power athletes, yet without reaching statistical significance. Only two participants (power athletes) had the rare CC genotype. Other variants of the βARs genes (ADRB1 Arg589Gly, and ADRB2 Gly16Arg and Gln27Glu variants) were not associated with sports performance in our cohort.

The ADRB2 Gly16Arg polymorphism was previously associated with elite endurance performance status, with the Gly (G) allele exerting an unfavourable effect. In our study, the G allele frequency did not differ significantly in controls and endurance athletes (table 1). Discrepancy in our results and those from previous research might lie on the different ethnic origins of the participants (North-American, Finnish and German Caucasians vs Spanish Caucasians) or in the endurance sports speciality (which was more homogeneous in our study, that is, composed of only world-class runners and cyclists). Though the ADRB2 Gln27Glu polymorphism was associated with endurance sports performance in post-menopausal Caucasian women, similar results remained to be corroborated in world-class athletes, that is, those who are at the “endurance” end point of the human sports performance continuum.

The minor ADRB3 Arg (C) allele in codon 64 was clearly over-represented in world-class runners and cyclists compared with non-athletic controls. Some caution is needed.

<table>
<thead>
<tr>
<th>Polymorphism</th>
<th>Genotype/allele</th>
<th>Controls (C)</th>
<th>Endurance (E)</th>
<th>Power (P)</th>
<th>Overall p value</th>
<th>p Value (C vs E)</th>
<th>p Value (C vs P)</th>
<th>p Value (E vs P)</th>
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<tbody>
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<td>ADRB1 Arg389Gly (rs1801253)</td>
<td>CC</td>
<td>43</td>
<td>48</td>
<td>40</td>
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<tr>
<td></td>
<td>CQ</td>
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<td>47</td>
<td>47</td>
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</tr>
<tr>
<td></td>
<td>GG</td>
<td>10</td>
<td>5</td>
<td>13</td>
<td></td>
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<td></td>
<td>ρ(C)</td>
<td>0.77</td>
<td>0.72</td>
<td>0.63</td>
<td>&gt;0.1</td>
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<tr>
<td></td>
<td>q(G)</td>
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<td>0.28</td>
<td>0.36</td>
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<tr>
<td>ADRB2 Gly16Arg (rs1042713)</td>
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<td></td>
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<td>0.43</td>
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<td>ADRB3 Trp64Arg (rs4994)</td>
<td>TT</td>
<td>92</td>
<td>73</td>
<td>81</td>
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<td>0.90</td>
<td>0.004</td>
<td>0.001</td>
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<td>&gt;0.1</td>
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<tr>
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<td>0.14</td>
<td>0.10</td>
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</table>

*p Values in bold indicate significance.*
when interpreting this result, as we found no significant differences between the endurance and power athletes. The association we reported for the ADRB3 Trp64Arg variation and endurance performance status can nevertheless be explained by the numerous effects that the β3AR exerts in human body tissues, namely, adipose tissue and especially the heart.17 The β3AR stimulates lipolysis and thermogenesis.9–11 Besides regulating angiogenesis and vasorelaxation in the coronary microvasculature,21 β3ARs can have a negative inotropic effect.18–20 Thus, the ADRB3 Trp64Arg polymorphism, which is associated with reduced agonist-stimulated adenylyl cyclase activity in vitro,12 could favour myocardial contractility, that is, by stimulating cardiac inotropism. Though more research is needed in the field, this could explain, at least partly, a certain positive effect of the minor C allele on endurance performance.

Besides the unique competition level of our athlete cohort, a potential strength of our study stems from the fact that we analysed all the main variations in the three genes that encode for the βARs family with a documented effect on clinically relevant phenotypes. Cross-sectional studies as the present one are, however, limited by several factors. Mainly, the present type of design does not allow to determine the changes that the “unique” environmental factors of sportsmen can induce for the gene expression during critical periods of prenatal and postnatal development, that is, through epigenetic mechanisms.23 The effects of epigenetic mechanisms on gene expression are probably more important than genetic polymorphisms per se. Finally, there might be other genetic variants yet to be determined that might not influence sports performance individually but could exert complex interactions with candidate genes as those studied here.

Acknowledgements This study was funded by Progenika Biopharma and Sabiobi S.L. (Spain) and partially supported by Consejo Superior de Deportes (CSD, ref # 2010/085). FAS received a PhD grant from the Spanish Ministry of Education (EX-2007-1124), Karolinska Institutet, and the Swedish Council for Working Life and Social Research (FAS).

Competing interests None.

Patient consent Obtained.

Ethics approval This study was conducted with the approval of the Universidade Europea de Madrid.

What is already known on the topic

► Variants in the genes (ADRB1 and ADRB2) that encode βARs are associated with metabolic and cardiovascular phenotypes, especially in non-athletes.

What this study adds

► Heterozygosity for the ADRB3 Trp64Arg polymorphism is associated with elite endurance performance.

Provenance and peer review Not commissioned; externally peer reviewed.

REFERENCES

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*Br J Sports Med* 2011 45: 147-149 originally published online June 23, 2009
doi: 10.1136/bjsm.2009.061366

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