Is there an association between *ACTN3* R577X polymorphism and muscle power phenotypes in young, non-athletic adults?

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We investigated the association between *ACTN3* R577X polymorphism and jumping (vertical squat and counter-movement jump tests) and sprint ability (30 m dash) in non-athletic, healthy young adults [$N = 284$ (217 male), mean (SD) age: 21 (2) years]. We analyzed the differences in the study phenotypes among *ACTN3* R577X genotypes by one-way analysis of covariance before and after adjusting for sex, age, weight and height (confounders). We also compared the genotype and allele frequencies between those with the best and worst results in the aforementioned tests ($\geq$ 90th vs <90th of the sex-specific percentile, respectively). We used logistic regression to calculate the odds ratio (OR) for having the best performance. We did not observe a significant association between *ACTN3* R577X genotypes and the study phenotypes before and after adjusting for potential confounders, nor after analyzing males and females separately. We did not observe significant differences in genotype frequencies between those with the best or the worst performance. The OR for an individual with the RR genotype to be in the top 10 percentile was <1.00 for jump tests and <1.015 for sprint tests (all $P > 0.05$). In summary, α-actinin-3 deficiency does not negatively influence the ability to generate explosive leg muscle power in a young non-athletic population.

The expression of α-actinin-3 is almost exclusively restricted to fast twitch (type II) muscle fibers (Mills et al., 2001). In this fiber subtype, it constitutes the predominant component of the Z-disc, where it acts as a lattice structure that anchors actin-containing thin filaments; it stabilizes the muscle contractile apparatus, thereby conferring a higher capacity for force absorption/transmission compared with slow (type I) fibers (Squire, 1997; Mills et al., 2001). Through an interaction with the signalling protein calcineurin, α-actinin-3 might also promote the formation of fast twitch fibers (Yang et al., 2003).

A premature stop codon polymorphism (R577X, rs1815739) in *ACTN3*, the gene encoding for the synthesis of α-actinin-3 in skeletal muscle fibers, was first described by North et al. (1999). Although this genetic variation is not associated with any known disease phenotype, with few notable exceptions (Lucia et al., 2007), the α-actinin-3-deficient XX genotype is believed to preclude top-level athletic performance in "pure" power and sprint sports (sprinting, jumping, weightlifting and throwing events), especially in women (Yang et al., 2003).

More controversy exists on the putative role of the *ACTN3* R577X polymorphism in muscle phenotypes (particularly, the ability to produce peak power) in a non-athletic population. Walsh et al. (2008) reported that women, but not men, deficient in α-actinin-3 (i.e. *ACTN3* XX genotype) had lower knee extension shortening and lengthening peak isokinetic torque compared with their RR/RX referents, across the adult age span (22–40 years). The high-velocity torque of knee extensor muscles was unaffected by *ACTN3* genotypes in Caucasian adult men (McCauley et al., 2009). In another report, however, healthy young men with the RR genotype showed significantly higher relative dynamic quadricep torques at 300°/s and a greater percentage of type IIX fibers than those with the XX genotype (Vincent et al., 2007). Additional controversy stems from the fact that in older adults (mean age ∼ 65 years), the XX genotype was associated with a higher knee extensor concentric peak power compared with the RR and RX genotypes (Delmonico et al., 2007).

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Discrepancies between studies might be related to the differences in age, gender, ethnic background or fitness level of the selected subjects. A source of disparity can also arise from the different testing batteries used to characterize muscle power phenotypes. Naturally occurring jumping, throwing or sprinting actions are multi-joint movements that involve the coordinated participation of the majority of lower limb muscles (Ashley & Weiss, 1994; Brown & Weir, 2001). During actual high muscle power actions (e.g. in team sport games), angular velocities at the hip or knee joints in the aforementioned actions can approach 800–1000°/s (Bosco et al., 1983). However, the previously mentioned studies on non-athletes used predominantly isokinetic tests involving single-joint movements (e.g. knee extension) at relatively low angular velocities ($\leq 300°/s$).

The main purpose of our study was to examine the association between *ACTN3* R577X polymorphism and the ability to produce peak power in non-athletic, young adults of both genders. Peak power was evaluated using jumping and sprint tests of practical applicability and involving naturally occurring multi-joint movements. Based on the fact that previous findings on elite athletics [as those by Yang et al., (2003)] cannot be extrapolated to the general population and that muscle phenotypes (e.g. "explosive" power) are complex traits not reducible to a single polymorphism, we hypothesized that the ability to produce peak power is not associated with *ACTN3* R577X genotypes in non-athletic, physically active young adults.

Methods
Subjects

Written consent was obtained from each subject. The study protocol was approved by the institutional ethics committee [Universidad Europea de Madrid (UEM), Spain] and was in accordance with the Declaration of Helsinki for Human Research. The sample was comprised of 284 healthy young adults (university students) [mean (SD) age: 21 (2) years (range: 21, 32)] of both genders (217 men, 67 women). Inclusion criteria were to be free of any diagnosed cardiorespiratory disease, and not to be engaged in competitive sports nor in (i) formal, supervised "power" (weight lifting, alpine skiing) or jumping-oriented type of training (plyometrics, volleyball and basketball) or in (ii) endurance training (running, swimming and bicycling), that is, performing less than one (power) or three (endurance) structured weekly training sessions within the previous year. All participants were of the same Spanish (Caucasian) ancestry for at least three generations.

Genotype assessment

Our study was designed and performed in accordance with the recommendations for the human genotype–phenotype association studies recently published by the NCI-NHGRI Working Group on Replication in Association Studies (Chanock et al., 2007). These recommendations include, among others, the

following items: indicating the time period and location of subject recruitment, success rate for DNA acquisition, sample tracking methods or genotyping with a second technology in a second, well-reputed laboratory.

During winter–spring 2008, we extracted genomic DNA from saliva samples of students in two different universities of the same city (Madrid, Spain): Universidad Politécnica and UEM ($N = 200$ and 84 subjects, respectively).

"Reference" genotyping

Genotyping was originally performed during fall 2008 in the genetics laboratory of UEM (Madrid). The polymerase chain reaction (PCR) was performed in order to amplify the sequence containing the mutation. A fragment of 303 bp was amplified with the following primers: forward CTGTTGCC TGTGGTAAGTGGG, with 5′; VIC labelling and reverse TGGTCACAGTATGCAGGAGGG (Lucia et al., 2006). The PCR conditions were as follows: initial denaturing at 95 °C for 5 min; 35 cycles at 95 °C for 30 s, 60 °C for 30 s, 72 °C 30 s and a final extension at 72 °C for 10 min.

ACTN3 genotypes were established by enzymatic digestion of amplicons with *Dde*I (Lucia et al., 2006). The R577X change creates a restriction site resulting in fragments of 108, 97 and 86 bp. Digestion of the R577 allele yields fragments of 205 and 86 bp. Digestion products (108 bp for 577X and 205 bp for R577) were detected by capillary electrophoresis in an ABI Prism 310 genetic analyzer (Applied Biosystems, Foster City, California, USA).

Reliability assessment of genotype analysis in a second laboratory

Following recent recommendations (Chanock et al., 2007), genotype results in 100 samples were corroborated in a different laboratory (Progenika Biopharma, Parque Tecnológico de Zamudio, Derio-Vizcaya, Spain) during fall 2008 using a different technology, i.e. 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 elsewhere (Tejedor et al., 2005). In brief, the PCR products were fluorescently labelled and hybridized to the DNA microarray in an automated platform (Ventana Medical Systems Inc., Tucson, Arizona, USA). The microarrays were scanned (Innopsys SA, Carbonne, France) and we determined variants using a developed software that converts the intensity of the spots into the genotype of each variant.

Phenotype assessment

Vertical jump tests

Squat (SJ) and counter-movement jump (CMJ) tests were performed using an infrared contact timing platform (Globus Ergo Tester, Codognè, Italy) to evaluate leg muscles' ability to produce "explosive" power (Young et al., 2001).

SJ tests. The SJ tests were performed without rebound or previous counter movement. Subjects kept both hands on the hips and trunk straight before and during jumps. Before the jumps, they reached 90° of knee flexion angle for ~ 1 s and during jumps they could not perform hip or knee flexions (Bosco et al., 1982, 1983).

CMJ tests. Subjects started from a standing position, with the trunk straight, legs extended and both hands on hips, and

performed a vertical jump with a prior fast counter movement allowing 90° knee flexion (Bosco et al., 1982, 1983). During jumps, they kept both hands on the hips, the trunk straight and they could not perform hip or knee flexions.

Both tests were performed three times (each separated by a 2-min rest period) and the best score was retained.

Sprint test

Subjects performed a 30-m sprint test in an indoor rubberized track under two conditions: (i) starting from the stationary (standing) position (with one foot in front of the other) (Young et al., 2001) and (ii) starting with a previous 15 m run (running), thereby allowing achieving higher speeds in the first meters of the test (Alcaraz et al., 2009). The difference in performance time between both tests (at 15 and 30 m, respectively) was used as an index of a subject's ability to produce acceleration, i.e. lesser difference implies higher acceleration capacity. We used photoelectric gates at 0, 15 and 30 m to start and stop a digital timer.

The participants were encouraged to do their best when performing the tests and were advised not to perform strenuous physical activity within the previous 48 h. Because the subjects were students of the School of Sports Sciences, they were familiarized with the tests. Nevertheless, 1 week before the tests, all the participants received comprehensive instructions on the tests, after which a familiarization session took place. All the tests were performed during spring 2008 in the same location (UEM) and under the supervision of the same researchers.

Reliability assessment of phenotype measurements

A subgroup of subjects [age: 21 (2) years (range: 21, 28) of both genders (nine men, five women)] were asked to participate in a reliability study. They performed the same tests under the same conditions 7 days later. Realistically, a certain amount of error is always present when collecting data. The main components of measurement error are systematic bias (e.g. general learning on the tests) and random error due to biological or mechanical variation. To avoid systematic bias, all subjects received a familiarization session 1 week before the tests.

Statistical analysis

Hardy–Weinberg equilibrium within the study groups was tested using a chi-square test. We compared the genotype frequencies between males and females with the chi-square test.

A required sample size of 144 subjects was determined to be large enough to detect a medium effect size of 0.30 with a power of 90% and an α of 5%.

Reliability assessment of phenotype measurements

Differences between scores obtained on day 1 and day 2 (intertrial difference D1–D2) were analyzed by one-way analysis of covariance (ANCOVA) for repeated measures, where D1 and D2 were entered as factors and sex, age, weight and height as covariates. Because no interaction was found between sex × test, all the analyses were performed for both men and women together.

Association between the ACTN3 R577X polymorphism and leg muscle power

We analyzed the differences in the study phenotypes among variants of the ACTN3 R577X polymorphism by one-way analysis of variance (Model 1), where the ACTN3 R577X polymorphism was entered as a fixed factor and the phenotype was entered as a dependent variable. Because age, weight and height might be potential confounders, we performed one-way ANCOVA (Model 2) where the ACTN3 R577X polymorphism was entered as a fixed factor, the phenotype was entered as a dependent variable and age, weight and height were entered as covariates. We did not observe an interaction effect between sex × ACTN3 R577X polymorphism and phenotypes (all $P > 0.2$); therefore, all the analyses were performed with men and women together and sex was included as a covariate in Model 2. We used the Bonferroni and Holm method to correct for multiple testing (Holm, 1979; Shaffer, 1995).

Genotype and allele frequency comparisons between those with the best and the worst performance in the study tests

In order to examine whether subjects with the best performance had a different genotype and allele frequency compared with those with lower performance, we classified the population into two groups based on an arbitrary cut-off point: ≥ 90th of the sex-specific percentile and < 90th of the sex-specific percentile. The rationale for choosing 90th percentile was based on the fact that subjects in the top 10% might have a favorable genetic endowment to perform better. To investigate the influence of these cut-offs on the findings, we performed sensitivity analyses after varying those cut-offs (≥ 75th and ≥ 95th). We compared the genotype and allele frequencies between groups (<90th and ≥ 90th sex-specific percentile) with the chi-square test. In order to estimate the effect size, we used logistic regression analysis to calculate the odds ratio (OR) for having the best performance in the jump and sprint tests (≥ 90th of the sex-specific percentile) after adjusting for sex, age, weight and height, and using the dominant model.

Finally, we compared the mean level of the study phenotypes between groups by one-way ANCOVA, where group was entered as a fixed factor, the phenotype was entered as a dependent variable and sex, age, weight and height were entered as covariates.

All the analyses were performed with the Statistical Package for Social Sciences (SPSS, v. 16.0 for Windows; SPSS Inc., Chicago, Illinois, USA), and the level of significance was set at $\alpha \leq 0.05$.

Results

Genotype distributions met Hardy–Weinberg equilibrium ($\chi^2 = 0.061$, $P = 0.803$). We did not observe differences ($\chi^2 = 2.109$, $P = 0.348$) in the genotype distributions in men [70 (32.4%), 103 (47.7%) and 43 (19.9%) for RR, RX and XX, respectively] and women [20 (29.9%), 38 (56.7%) and 9 (13.4%) for RR, RX and XX, respectively]. Genotype and allele frequencies were within the range previously reported for Caucasians of European ancestry, with ~ 18% of individuals carrying the α-actinin-3-deficient (XX) genotype (Yang et al., 2003; Lucia et al., 2006).

Reliability of genotype analysis

No failures occurred in sample collection and DNA acquisition. Genotyping success rate was > 99.6%

(only one missing data). Parallel genotyping results of the *ACTN3* R577X polymorphism showed 100% concordance between the two laboratories.

Reliability of phenotype measurements

The mean values and SD for the two trials and the mean intertrial difference for the study phenotypes are shown in Table 1. We did not observe an intertrial difference in any of the study tests.

Association between the *ACTN3* R577X polymorphism and leg muscle power

The association between the *ACTN3* R577X polymorphism and study phenotypes is presented in Table 2. We did not observe any effect of the *ACTN3* R577X polymorphism on the study phenotypes before (Model 1) and after adjusting for sex, age, weight and height (Model 2). We repeated the analyses separately in men and women and the results did not materially change.

Genotype and allele frequency comparisons between those with the best and the worst performance in the phenotype tests

To examine whether those with the best performance (≥ 90th of the sex-specific percentile) had different genotype frequencies from those with lower performance (<90th of the sex-specific percentile), we compared the genotype frequencies between groups for the jump and sprint tests. We did not observe differences in genotype (Table 3) or allele (Table 4) frequencies between groups. The OR [95% confidence interval (CI)] for an individual with the RR variant to be in the top 10 percentile (≥ 90th of the sex-specific percentile) was 0.907 (95% CI: 0.473–1.740, $P = 0.777$) and 0.769 (95% CI: 0.392–1.511, $P = 0.446$) for SJ and CMJ, respectively. The OR for being in the top 10 percentile in the 15 and 30 m (30 m running start spring test) was 1.022 (95% CI: 0.515–

2.029, $P = 0.950$) and 0.699 (95% CI: 0.324–1.377, $P = 0.275$), respectively. The OR for the 15 and 30 s (30 m standing start spring test) was 1.014 (95% CI: 0.531–1.934, $P = 0.967$) and 0.875 (95% CI: 0.450–1.703, $P = 0.695$). The results remained the same after changing the cut-off point to ≥ 75th percentile or to ≥ 95th percentile (data not shown).

Mean levels of the study phenotypes were significantly higher in the group classified in the ≥ 90th percentile compared with the group <90th percentile (Fig. 1).

Discussion

The main finding of our study was that the *ACTN3* R577X polymorphism does not seem to influence the ability to produce peak (explosive) power in non-athletic, young adults of both genders. Leg muscles' peak power was evaluated using jumping and sprint tests of practical applicability. Subjects' performance in these tests did not show considerable interindividual variability and was considerably lower (e.g. 20% lower performance in CMJ tests) than that reported previously for power-trained young adults (Baker et al., 1994). Thus, we can discard the existence of a major confounding effect arising from an eventual strong power training background of the subjects or a large individual variability in training levels. We did not find any interaction effect between gender, genotypes and the phenotypes studied. Although previous research showed a stronger association between R577X genotypes and exercise capacity phenotypes in women than in men (Yang et al., 2003; Clarkson et al., 2005; Walsh et al., 2008), our findings are in overall agreement with others that did not report the R577X effect on muscle phenotypes to be gender specific (Delmonico et al., 2007).

ACTN3 is the first structural skeletal-muscle gene for which a genotype–elite sports performance phenotype association has been clearly demonstrated, especially in women (MacArthur & North, 2004,

Table 1. Mean values (standard deviation) for the day 1 (D1) and day 2 (D2) trials and the mean intertrial difference for the tests

	D1	D2	Mean difference	95% CI	*P*-value
SJ					
Flight time (s)	0.525 (0.052)	0.527 (0.056)	− 0.004	− 0.006, 0.001	0.562
Vertical displacement of CG (cm)	0.341 (0.066)	0.345 (0.072)	− 0.003	− 0.008, 0.002	0.439
CMJ					
Flight time (s)	0.543 (0.052)	0.545 (0.052)	− 0.002	− 0.005, 0.001	0.997
Vertical displacement of CG (cm)	0.364 (0.068)	0.367 (0.069)	− 0.003	− 0.007, 0.000	0.967
30 m running start					
Time at 15 m (s)	2.058 (0.122)	2.045 (0.142)	0.013	− 0.008, 0.035	0.983
Time at 30 m (s)	4.006 (0.273)	3.985 (0.308)	0.024	− 0.012, 0.052	0.790
30 m standing start					
Time at 15 m (s)	2.630 (0.129)	2.616 (0.124)	0.013	− 0.006, 0.032	0.479
Time at 30 m (s)	4.611 (0.250)	4.591 (0.256)	0.019	− 0.009, 0.048	0.724

SJ, squat jump; CMJ, counter-movement jump; CG, center of gravity.

Table 2. Mean (standard error) estimates of study phenotypes by genotypes of *ACTN3* R577X (rs1815739) polymorphism

	Model 1						Model 2					
	RR (n = 90)	RX (n = 141)	XX (n = 52)	P overall	P dom.	P recess.	RR (n = 90)	RX (n = 141)	XX (n = 52)	P overall	P dom.	P recess.
Vertical jump tests												
SJ												
Flight time (s)	546.6 (5.1)	545.0 (4.1)	540.3 (6.6)	>0.1	>0.1	>0.1	548.6 (4.4)	547.5 (3.3)	536.2 (5.5)	>0.1	>0.1	>0.05
Vertical displacement of CG (cm)	36.9 (0.7)	36.7 (0.5)	36.1 (0.9)	>0.1	>0.1	>0.1	37.2 (0.6)	37.1 (0.5)	35.6 (0.7)	>0.1	>0.1	>0.05
CMJ												
Flight time (s)	555.4 (5.1)	553.7 (4.3)	555.9 (6.9)	>0.1	>0.1	>0.1	559.1 (4.6)	556.2 (3.5)	550.6 (5.7)	>0.1	>0.1	>0.1
Vertical displacement of CG (cm)	38.1 (0.7)	37.9 (0.6)	38.2 (0.9)	>0.1	>0.1	>0.1	38.6 (0.6)	38.2 (0.5)	37.5 (0.8)	>0.1	>0.1	>0.1
Sprint tests												
30 m running start												
Time at 15 m (s) (A)	2.00 (0.02)	2.01 (0.19)	1.98 (0.02)	>0.1	>0.1	>0.1	1.97 (0.02)	2.00 (0.01)	2.01 (0.02)	>0.1	>0.1	>0.1
Time at 30 m (s) (B)	3.91 (0.04)	3.93 (0.03)	3.89 (0.05)	>0.1	>0.1	>0.1	3.88 (0.03)	3.91 (0.02)	3.94 (0.03)	>0.1	>0.1	>0.1
30 m standing start												
Time at 15 m (s) (C)	2.61 (0.02)	2.61 (0.02)	2.61 (0.02)	>0.1	>0.1	>0.1	3.60 (0.02)	2.60 (0.01)	2.63 (0.02)	>0.1	>0.1	>0.1
Time at 30 m (s) (D)	4.54 (0.04)	4.56 (0.03)	4.57 (0.04)	>0.1	>0.1	>0.1	4.53 (0.03)	4.55 (0.02)	4.61 (0.03)	>0.1	>0.1	>0.05
Acceleration index												
C–A (s)	0.61 (0.01)	0.60 (0.01)	0.62 (0.01)	>0.1	>0.1	>0.1	0.62 (0.02)	0.60 (0.01)	0.62 (0.02)	>0.1	>0.1	>0.1
D–B (s)	0.63 (0.02)	0.64 (0.02)	0.67 (0.01)	>0.1	>0.1	>0.1	0.65 (0.02)	0.64 (0.02)	0.67 (0.03)	>0.1	>0.1	>0.1

Model 1: unadjusted.

Model 2: adjusted for sex, weight, height and age.

dom., dominant; recess, recessive; SJ, squat jump; CMJ, counter-movement jump; CG, center of gravity; RR, major allele; XX, minor allele.

Table 3. Genotype frequencies (*n*) in subjects with the best performance ($\geq$ 90th of the sex-specific percentile) vs those with lower performance (<90th of the sex-specific percentile)

	<90th			$\geq$ 90th			χ^2, *P*-value
	RR	RX	XX	RR	RX	XX	
Jump tests							
SJ	32.7 (74)	48.7 (110)	18.6 (42)	28.1 (16)	54.4 (31)	17.5 (10)	0.638, 0.727
CMJ	33.0 (75)	47.6 (108)	19.4 (44)	26.8 (15)	58.9 (33)	14.3 (8)	2.349, 0.309
30 m running start							
15 m	30.4 (69)	51.1 (116)	18.5 (42)	37.5 (21)	44.6 (25)	17.9 (10)	1.099, 0.577
30 m	31.7 (72)	49.8 (113)	18.5 (42)	32.1 (18)	50.0 (28)	17.9 (10)	0.013, 0.993
30 m standing start							
15 m	31.4 (71)	49.6 (112)	19.0 (43)	33.3 (19)	50.9 (29)	15.8 (9)	0.328, 0.849
30 m	31.4 (71)	51.8 (117)	16.8 (38)	33.3 (19)	42.1 (24)	24.6 (14)	2.393, 0.302

RR: major allele, and XX: minor allele of *ACTN3* R577X (rs1815739) polymorphism.
SJ, squat jump; CMJ, counter-movement jump.

Table 4. Allele frequencies in subjects with the best performance ($\geq$ 90th of the sex-specific percentile) vs those with lower performance (<90th of the sex-specific percentile)

	<90th		$\geq$ 90th		χ^2, *P*-value
	p(R)	*q*(X)	*p*(R)	*q*(X)	
Jump tests					
SJ	0.327	0.673	0.281	0.719	0.458, 0.498
CMJ	0.330	0.670	0.268	0.732	0.810, 0.368
30 m running start					
15 m	0.304	0.696	0.318	0.682	1.045, 0.307
30 m	0.317	0.683	0.321	0.679	0.004, 0.951
30 m standing start					
15 m	0.314	0.686	0.333	0.667	0.077, 0.781
30 m	0.314	0.686	0.333	0.667	0.077, 0.781

RR: major allele, and XX: minor allele of *ACTN3* R577X (rs1815739) polymorphism.
SJ, squat jump; CMJ, counter-movement jump, VO_{2max}, maximum oxygen uptake.

2007). This would suggest the existence of a "trade-off" between power/sprint and endurance phenotypic traits such that an individual could be inherently predisposed toward athletic performance in either sprint/power or endurance events (Garland et al., 1990). However, the putative role of the *ACTN3* R577X polymorphism on muscle phenotypes in the Caucasian non-athletic population, particularly in men, is not well established. Others have reported that the absence of α-actinin-3 protein influences negatively knee extension peak isokinetic torque in middle-aged women but not in men (Walsh et al., 2008). Similarly, McCauley et al. (2009) reported no association between *ACTN3* R577X genotypes and high-velocity torque of knee extensor muscles in adult men. However, Vincent et al. (2007) showed that healthy young men with the RR genotype had significantly higher relative dynamic quadriceps torques at 300°/s and a greater percentage of type IIX fibers than those with the XX genotype. A source of

disparity between studies can arise from the different testing batteries used to characterize muscle power phenotypes. Most jumping or sprinting actions that are naturally performed by young humans are multi-joint movements involving high angular velocities that involve the coordinated participation of the majority of lower limb muscles (Ashley & Weiss, 1994; Brown & Weir, 2001), whereas most previous studies on *ACTN3* R577X in non-athletes used predominantly isokinetic tests involving single-joint movements (e.g. knee extension) at relatively low angular velocities ($\leq 300°/s$). While there is a high likelihood that α-actinin-3 is important to produce high levels of skeletal muscle power, other factors may be at least as important as the intrinsic characteristics of muscle proteins; these include the ability to coordinate and sequence complex muscle actions (e.g. take-off in the case of jumping) and properties of the skeletal muscle tissue (muscle mass, the ratio of muscle fiber to tendon cross-sectional area or the relative length of fibers and tendons) (Lucia et al., 2007). In fact, we recently reported α-actinin-3 deficiency in a former Olympic-class long jumper (Lucia et al., 2007). Long jump involves complex muscle actions (i.e. a combination of sprint, take-off and landing abilities) that are not only influenced by muscle protein characteristics.

In summary, we found no overall evidence that α-actinin-3 deficiency negatively influences the ability to generate explosive leg muscle power (jumping, sprinting) in a young non-athletic population, irrespective of gender.

Perspectives

ACTN3, also known as "the gene for speed," is the first structural skeletal muscle gene for which a genotype–phenotype association has been clearly documented in elite athletes, especially in women. The results obtained in athletes are, however, difficult to extra-
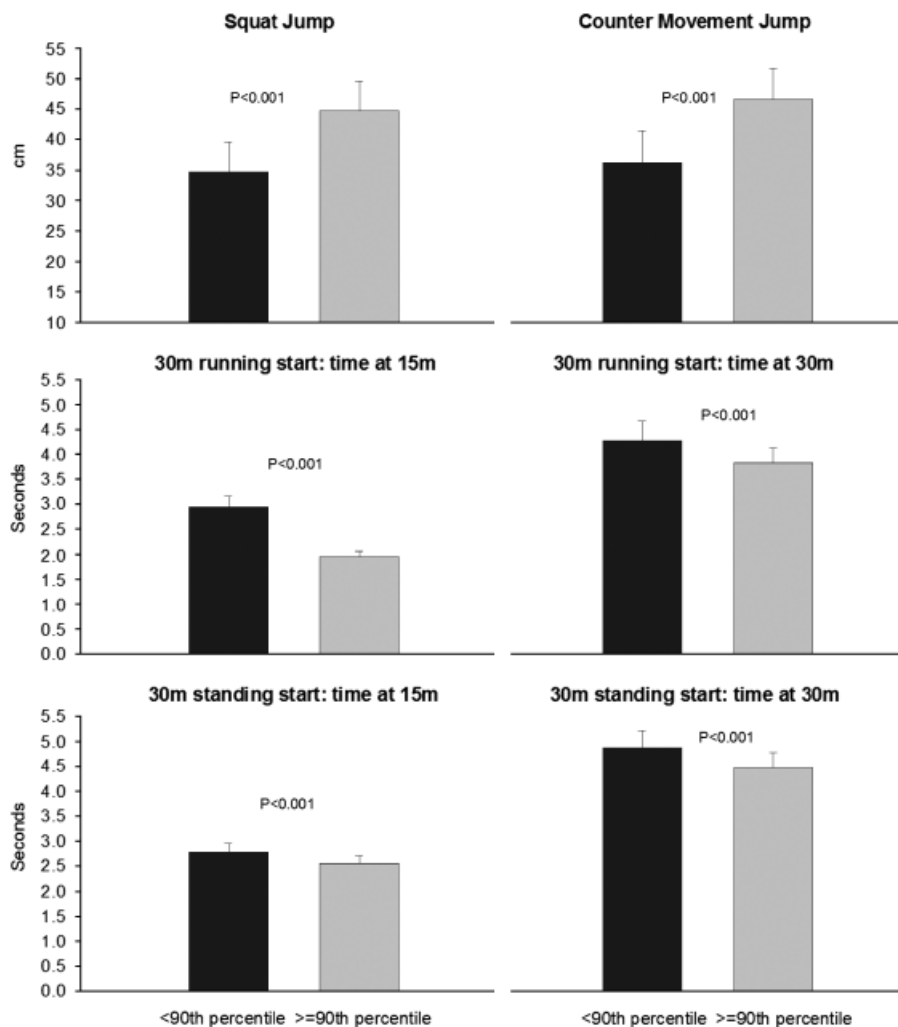
Fig. 1. Mean levels of the study phenotypes by groups classified by percentiles ($\geq$ 90th vs < 90th percentile)

polate to the general population. Advances in the field of genetics and exercise capacity seem to parallel the growth of complexity and disparity between studies. Differences in the age, baseline physical capacity and ethnic origin of subjects and particularly in the different tests used to characterize muscle phenotypes make a comparison between studies difficult. Further, most exercise phenotype traits are complex and are thus not likely to be reducible to a single polymorphism, e.g. *ACTN3* R577X. The effects of epigenetic mechanisms on gene expression are probably more important than genetic polymorphisms per se. Further, there might be other genetic variants, still to be determined, that might not influence muscle phenotype individually, but could exert complex interactions with candidate genes as the one studied here. Finally, beyond geno-type:phenotype interactions, the effect of MicroRNAs on human muscle phenotypes remains to be determined. These short, non-coding RNA molecules regulate skeletal muscle post-transcriptional gene expression and thus modulate important muscle aspects of muscle function, including contractility (van Roiij et al., 2008).

Key words: genotype, allele, sports, endurance.

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