ACE ID Genotype and the Muscle Strength and Size Response to Unilateral Resistance Training

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ABSTRACT

PESCATELLO, L. S., M. A. KOSTEK, H. GORDISH-DRESSMAN, P. D. THOMPSON, R. L. SEIP, T. B. PRICE, T. J. ANGELOPOULOS, P. M. CLARKSON, P. M. GORDON, N. M. MOYNA, P. S. VISICH, R. F. ZOELLER, J. M. DEVANEY, and E. P. HOFFMAN. ACE ID Genotype and the Muscle Strength and Size Response to Unilateral Resistance Training. *Med. Sci. Sports Exerc.*, Vol. 38, No. 6, pp. 1074–1081, 2006. **Purpose:** To examine associations among the angiotensin I–converting enzyme (ACE) insertion (I)/deletion (D) polymorphism and the response to a 12-wk (2 d·wk^{-1}) unilateral, upper-arm resistance training (RT) program in the trained (T, nondominant) and untrained (UT, dominant) arms. **Methods:** Subjects were 631 (mean ± SEM, 24.2 ± 0.2 yr) white (80%) men (42%) and women (58%). The ACE ID genotype was in Hardy–Weinberg equilibrium with frequencies of 23.1, 46.1, and 30.8% for ACE II, ID, and DD, respectively ($\chi^2 = 1.688$, $P = 0.430$). Maximum voluntary contraction (MVC) and one-repetition maximum (1RM) assessed peak elbow flexor muscle strength. Magnetic resonance imaging measured biceps muscle cross-sectional area (CSA). Multiple variable and repeated-measures ANCOVA tested whether muscle strength and size differed at baseline and pre- to post-RT among T and UT and ACE ID genotype. **Results:** Baseline muscle strength and size were greater in UT than T ($P < 0.001$) and did not differ among ACE ID genotype in either arm ($P \geq 0.05$). In T, MVC increases were greater for ACE II/ID (22%) than DD (17%) ($P < 0.05$), whereas 1RM (51%) and CSA (19%) gains were not different among ACE ID genotype pre- to post-RT ($P \geq 0.05$). In UT, MVC increased among ACE II/ID (7%) ($P < 0.001$) but was similar among ACE DD (2%) pre- to post-RT ($P \geq 0.05$). In UT, 1RM (11%) and CSA (2%) increases were greater for ACE DD/ID than ACE II (1RM, 7%; CSA, −0.1%) ($P < 0.05$). ACE ID genotype explained approximately 1% of the MVC response to RT in T and approximately 2% of MVC, 2% of 1RM, and 4% of CSA response in UT ($P < 0.05$). **Conclusion:** ACE ID genotype is associated with the contralateral effects of unilateral RT, perhaps more so than with the muscle strength and size adaptations that result from RT. **Key Words:** EXERCISE, GENETICS, STRENGTH TRAINING, QUANTITATIVE TRAIT LOCI, RENIN–ANGIOTENSIN SYSTEM

The renin–angiotensin system (RAS) is a hormonal cascade that regulates cardiovascular function (2). The pathway begins with the production of renin that acts on angiotensinogen to form angiotensin I. Angiotensin I–converting enzyme (ACE) transforms biologically inactive angiotensin I into angiotensin II, a potent vasopressor whose actions include vasoconstriction, renal sodium reabsorption, and aldosterone production. ACE also degrades bradykinin, a vasodilator that has favorable effects on endothelial function and substrate utilization. A functional polymorphism of the ACE gene is the insertion (I) and deletion (D) polymorphism, depending on the presence or absence of a 287 amino acid base pair Alu repeat sequence within intron 16 on chromosome 17. Hence, three ACE ID genotypes exist: II, ID, and DD, the distributions of which within a white population are approximately 25, 50, and 25%, respectively (1).

The D allele of the ACE ID polymorphism is associated with higher serum and tissue ACE activity, resulting in greater production of angiotensin II and aldosterone, increased vascular smooth muscle growth, and a decreased half-life of bradykinin compared with the ACE I allele (2,26,27). Although serum ACE levels are generally stable within an individual, there is considerable variability in

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Submitted for publication November 2005.
Accepted for publication January 2006.

0195-9131/06/3806-1074/0
MEDICINE & SCIENCE IN SPORTS & EXERCISE®

DOI: 10.1249/01.mss.0000222835.28273.80

serum ACE levels in the general population (11,17,23,26), largely attributed to the presence of the ACE I or D allele. The differential actions of the ACE I and D alleles on the RAS have been associated with the response of the cardiovascular and musculoskeletal systems to environmental perturbations such as exercise (4,16).

The ACE I allele is more common in elite male endurance athletes including distance runners (15,27), rowers (7), triathletes (4), and mountaineers (13,29) and is associated with an enhanced cardiovascular response to endurance exercise training than the ACE D allele (8,12,13,27–30). In contrast, the ACE D allele is reported to be more common in muscle strength and power athletes (15,28) and to be associated with a superior muscle size and strength response to exercise training (6,12,13). However, only three investigative teams (6,21,26) have used a resistance training (RT) intervention to examine these associations, and they have arrived at different conclusions. Folland et al. (6) reported greater maximum voluntary contraction (MVC) and similar one-repetition maximum (1RM) gains after RT among subjects with the ACE D allele compared with those with the ACE II genotype. Williams et al. (26) found no associations among the ACE ID polymorphism and the muscle strength response to RT. Thomis et al. (21) reported borderline significance for greater concentric flexion torque gains for ACE I allele carriers but found no associations among the ACE ID genotype and the muscle size response to RT of the elbow flexors.

Because evidence is limited, we examined the influence of the ACE ID genotype on the muscle strength and size adaptations to a standardized 12-wk unilateral, upper-arm RT intervention in a large sample of apparently healthy young adults. Consistent with the conclusions of previous reports (6,12,13,15,28), we hypothesized that the muscle strength and size gains from RT would be greater among ACE DD homozygotes compared to carriers of the ACE I allele.

METHODS

Study overview. This study was part of the Functional Single Nucleotide Polymorphisms Associated with Human Muscle Size and Strength (FAMuSS) multicenter trial conducted by the Exercise and Genetics Collaborative Research Group (22). FAMuSS was designed to identify nonsynonymous single nucleotide polymorphisms (SNP) that associate with various phenotypes of human muscle size and strength. The institutional review boards from the 10 institutions involved with FAMuSS approved the study protocol.

The experimental design of FAMuSS has been described (3,9,22). Potential study volunteers were recruited from the eight RT sites via strategic flyer placement and inhouse, listserv, and radio announcements. The mean (± SEM) number of subjects trained at the eight sites was 75 ± 9, with a range of 49 to 114 individuals per site. After obtaining informed consent from all participants, isometric (MVC) and dynamic strength (1RM) and cross-sectional area (CSA) of the upper arms by magnetic resonance imaging (MRI) were measured before and after RT. The standardized, progressive RT intervention consisted of 12 wk of elbow flexor and extensor training of the nondominant arm (i.e., the hand with which the subject did not write; trained, T) in 264 men and 367 women who had not performed RT in the previous year. The dominant arm (untrained, UT) served as the nonexercise comparison in response to RT. Biannual investigator training was performed to ensure standardization of study protocols among sites.

Study population. Study volunteers who used medications known to affect skeletal muscle function such as corticosteroids, antihypertensive or hyperlipidemic medications, anabolic steroids, diuretics, arthritis medications (Vioxx, Celebrex), Depo-Provera Contraceptive Injection, Clenbuterol, Rhinocort nasal inhaler, lithium, and chronic use of nonsteroidal antiinflammatory drugs were excluded. In addition, those who had chronic medical conditions such as diabetes; had metal implants in arms, eyes, head, brain, neck, or heart; had performed any resistance training and/or regular activity that required repetitive use of the arms beyond normal daily activities within the prior year; consumed on average more than two alcoholic drinks daily; or used dietary supplements reported to build muscle size/strength or to cause weight gain such as protein supplements, creatine, or androgenic precursors were also excluded. Due to the potential confounding effects of weight fluctuation in the muscle strength and size response to RT, subjects that reported trying to lose weight, beginning a special diet purported to bring about a weight change, or having a weight change of more than 5 lb within 3 months of study participation were excluded. All subjects gave written informed consent prior to participation.

Anthropometric measurements. Pre- and post-RT assessments included body weight (in pounds using a standard balance beam scale) and height (in inches) that were used to calculate the body mass index (BMI) ($kg \cdot m^{-2}$). Subjects were instructed to follow their usual diet throughout the study. To ensure weight stability defined as ± 5.0 lb of pre-RT weight, body weight was also measured every 3 wk during study participation.

Isometric strength testing. MVC of the elbow flexor muscles of the T and UT arms was assessed using a custom-made preacher curl bench and strain gauge (model 32628CTL, Lafayette Instrument Company, Lafayette, IN). Baseline MVC was assessed on three separate days 24–48 h apart, and post-RT measurements were assessed on two separate days 24–48 h apart and within 48 h of the final RT session. Baseline values were recorded as the average of the second and third pre-RT testing days, with the first day serving as a familiarization session. Each MVC attempt began with a verbal cue from the tester, with subjects gradually increasing to a maximal effort that was sustained for 3 s with 1 min allowed between contractions. The same investigator measured MVC for a subject pre- and post-RT after calibrating the strain gauge and using a fixed seat height and a goniometer to maintain the arm angle at 90°. To minimize compensatory movement, the investigator ensured that the arm not being tested was relaxed and placed in neutral position

with the hand pronated and arm extended on the lap. The testing session ended once three measurements were within 5 feet per pound of each other or a maximum of six attempts had been made. The closest three measurements were averaged and recorded in kilograms. The intersite within-subject coefficient of variation based on baseline measurements made on days 2 and 3 was 7.4%, indicating adequate reliability among sites.

1RM maximum strength testing. The dynamic strength of the elbow flexor muscles of T and UT arms was assessed by 1RM on a standard preacher curl bench (Yukon International Inc., Cleveland, OH) holding a Powerblock (Powerblocks, Intellbell, Inc., Owatonna, MN) before and 48 h after the last RT session. Subjects were familiarized with the 1RM procedures on another occasion prior to testing. Powerblocks are handheld weights resembling dumbbells in increments of 2.50 and 5.00 lb. If needed, additional weight could also be added in 1.25-lb increments using Platemates® (Benoit Built Inc., Boothbay Harbor, ME). The same investigator measured 1RM for a subject and ensured that the arm not being tested was relaxed and placed in neutral position. Each subject performed two warm-up sets with increasing weight. Study investigators then verbally instructed subjects to perform one full range of motion repetition, extending the elbow to 180° and curling the weight back up to the shoulder with the weight at 100% of estimated maximum. If the lift was unsuccessful, a 3-min rest was taken and the weight decreased slightly. If the lift was successful, a 3-min rest was taken and the weight increased. The procedure was repeated until subjects failed to complete a full range of motion lift. Weights were chosen so that the 1RM could be determined in three to five attempts. Maximum weight lifted was recorded in kilograms as the greatest amount of weight successfully lifted one time. As with the MVC testing, the same study investigator measured 1RM and provided verbal encouragement during each 1RM attempt for a given subject pre- and post-RT.

MRI. CSA of the T and UT biceps brachii was measured using MRI with 1.5-T systems. MRI were performed prior to the first RT session and within 48–96 h after the final RT session. Prior to entering the MR magnet, a radiographic bead (Beekley Spots, Beekley Corp., Bristol, CT) was placed at the maximum circumference or the point of measure (POM) of the upper arms of each subject. The POM was determined with subject's arm abducted 90° at the shoulder, flexed 90° at the elbow, hand open and the biceps maximally contracted. The same investigator visually located the POM on the T and UT arm for a subject pre- and post-RT.

The MRI consisted of 15 axial slices comprising 24 cm of the upper arm. Subjects laid supine on the scanning bed in an anatomical position, with the arm aligned to the isocenter of the magnet. The hand was supinated and taped in place on the scanner bed, and the POM was centered to the alignment light of the MRI. Coronal and sagittal scout images were produced to locate the long axis of the humerus and to align the eighth to ninth axial slices with the POM, respectively. Fifteen spoiled gradient images were generated (time to echo (TE) = 1.9 s, time to repeat (TR) = 200 ms, flow artifact suppression, 30° flip angle), with POM as the center point. Axial imaging began at the superior portion of the upper arm and proceeded distally toward the elbow joint. Each individual image slice was 16 mm thick with a 0-mm interslice gap, 256 × 192 matrix resolution, 22 × 22-cm field of view, and number of experiments (NEX) = 6.

Muscle CSA measurements. MR images from each investigative site were saved via magnetic optical disk or CD-ROM in a DICOM format and sent to the central imaging facility for analysis. The same investigator analyzed MR images using a custom-designed program created to function within Matlab (The Math Works, Inc., Natick, MA). This software enables the user to assign regions of interest in an image set by tracing region borders with a mouse. Because muscle is easily identifiable on MR images, CSA could be measured using this computerized planimetry technique. Once the region of interest was defined, the program reported the number of pixels contained in the selected region of interest. CSA was then determined by multiplying the number of pixels within the defined area by a preset CSA value of 0.01 cm^2, determined from the MRI matrix and field of view. MRI standardization between sites was accomplished by comparing the radiographic bead's measured CSA with the MRI determined CSA.

To assign the slice to be assessed, the analyst identified the slice immediately following the axilla and then counted down slice by slice to the slice showing the POM in arm. In the rare instance that the number of slices between the axilla and POM differed pre- and post-RT POM, readily apparent discernible irregularities in the contour of the muscle and shape of the arm pre-RT were compared with slices adjacent to the post-RT POM until an identical anatomical match was found. The ninth axial slice was measured for maximum biceps CSA for most subjects. The pre-CSA was subtracted from the post-CSA, yielding the RT response for that arm. Interobserver reliability was ± 3.5%, and the intraobserver reliability for the entire process of image acquisition and analysis was calculated to be a correlation coefficient of 0.99. To further validate the CSA calculation, a subset of data from 70 subjects was analyzed by volumetric analysis in the T and UT arms. The CSA (cm^2) of 11 successive slices was determined over 17.6 cm of the scanned length of the upper arm. Each CSA was multiplied by the known slice thickness (1.6 cm) to yield a slice volume (cm^3). Slice volumes were then summed over the length of the biceps. Comparison of the relative percentage from baseline training-induced change in volume versus CSA within the subgroup of 70 subjects revealed no significant differences between the two methods ($P = 0.315$).

RT program. A unilateral upper-arm RT program was chosen as the exercise intervention to minimize the confounding influence of activities of daily living on the muscle strength and size response to RT (22). In addition, this study design allowed the UT arm to serve as a nonexercise comparison. Subjects underwent 12 wk of

gradually progressive, supervised RT of their nondominant arm, twice weekly, separated by a minimum of 48 h. The exercises consisted of the biceps preacher curl, biceps concentration curl, standing biceps curl, overhead triceps extension, and triceps kickback. The primary purpose was to train the elbow flexors, but the elbow extensors were also trained to balance muscle strength across the elbow. Each RT session began with a warm-up consisting of two sets of 12 repetitions of the biceps preacher curl and the overhead triceps extension. A 3-min rest followed each warm-up set. Following the warm-up series, subjects performed three sets of 12 repetitions at 65–75% of their 1RM for each of the five above-mentioned exercises. The speed of each repetition was 4 s: 2 s for the concentric and 2 s for the eccentric phase. A 2-min rest followed each set. At week 5, the number of repetitions was decreased to eight and then to six at week 10. Thus, the exercise intensity at weeks 5 and 10 increased to 75–82% 1RM and 83–90% 1RM, respectively. All exercises were performed with Powerblocks, and some exercises used the preacher curl bench. All training sessions were supervised and lasted approximately 45–60 min.

Genotyping methods. Blood samples were obtained from all subjects in vacutainer tubes containing ethylenediamine tetraacetic acid and sent to the coordinating site in Washington, DC, for DNA isolation using Puregene kits (Gentra Systems, Inc., Minneapolis, MN). For the ACE ID genotype, we used a method (23) that has been validated using three additional primers that produce unambiguous results for the ACE ID genotype. The primers (F: CTGGAGACCACTCCCAATCCTTTC and R: GATGTGGCCATCACATTCGTC) were used to amplify the region of *Alu* insertion, with the *Alu* element's presence or absence detected by running the product on an agarose gel. Polymerase chain reaction amplification was performed in a 10-μL reaction containing 20 ng of DNA, 0.10 mmol·L^{-1} of each primer, 0.125 mmol·L^{-1} of each dNTP, 2 mmol·L^{-1} $MgCl_2$, and 0.1 U of Ampli*Taq* Gold™ (Applied Biosystems). The cycling conditions were 95°C for 10 min, then 35 cycles of 94°C for 1 min, 58°C for 45 s, 72°C for 1.5 min, and, finally, 72°C for 10 min. All gels were called by two investigators, and if any disagreement in genotyping was found, the genotyping was repeated.

Data administration. Data from all investigative sites were compiled in a master database maintained by a statistical consultant at the coordinating site in Washington, DC. Each investigative site was able to access the database and manually enter data via a secure intranet using a confidential username and password.

Quality control. To ensure standardization between sites, the following procedures were followed: tester training and protocol and methodology reviews were done biannually; there were annual site visits to ensure protocol adherence; common standard operating procedure manual was used at each site; and there was frequent contact among the investigators through regular conference calls. Additionally, identical testing and workout equipment were used at each site.

Statistical analysis. The attrition rate from the RT program was 9% with no difference among the ACE ID genotypes. Consequently, all analyses included only subjects who completed the study. Descriptive statistics and frequencies were calculated for study variables. The χ^2 test was used to determine whether the ACE ID genotype was in Hardy–Weinberg equilibrium for white populations. Dependent variables included baseline and change in muscle strength (post-RT − pre-RT) for MVC and 1RM and muscle size (CSA) in the T and UT arms, which are presented in absolute (no correction; MVC, 1RM, and CSA), relative percentage (post-RT − pre-RT/pre-RT × 100; MVC, 1RM, and CSA), and allometric (strength (kg) × body weight (kg)$^{-0.67}$; MVC and 1RM) terms (24).

Multiple-variable and repeated-measures ANCOVA, with age and BMI as covariates and ACE ID genotype and gender as fixed factors, tested whether muscle strength and size differed in the T and UT arms among the total sample and ACE ID genotype at baseline and pre- to post-RT, respectively. *Multiple variable regression* tested whether the relative percentage of change from baseline differed from 0 in the T and UT arms pre- to post-RT among the total sample and ACE genotype. The resulting P values from these linear tests were adjusted for multiple comparisons using the Sidak *post hoc* multiple comparison test. No gender × ACE ID genotype interactions were found for any of the muscle phenotypes examined. Thus, gender was found not to alter the influence of ACE ID genotype on the muscle strength and size response to RT. In addition, the ACE genotype distribution was in Hardy–Weinberg equilibrium for both men and women. For these reasons, and because our findings on the influence of gender on the muscle strength and size response to RT have been published (9), results for this study are presented for the T and UT arms among the total sample and ACE ID genotype only.

Because of the large sample size, the proportion of all variance in the muscle strength and size response that could be attributable to ACE genotype was calculated for all study variables in the T and UT arms with the likelihood test ratio. The likelihood test ratio compares the full statistical model containing ACE ID genotype, gender, age, and BMI to the constrained model without ACE ID genotype. Statistical significance was set at $P < 0.05$, with all data reported as mean ± SEM. All calculations were made using SPSS 13.0 for Windows.

RESULTS

Subjects. The study sample ($N = 631$) had a mean age of 24.2 ± 0.2 yr (± SEM) and was composed of 367 (58.2%) women and 264 (41.8%) men. The sample was predominately white (79.5%), with 8.2% of the subjects Asian, 4.6% Hispanic, 4.1% African American, and 3.3% other. The ACE ID genotype distribution of the study sample was in Hardy–Weinberg equilibrium for white populations, with frequencies of 23.1, 46.1, and 30.8% for

TABLE 1. Mean (± SEM) physical characteristics of the total sample and by ACE ID genotype.

Variable	Total Sample (N = 631)	Genotype II (N = 146)	ID (N = 291)	DD (N = 194)
Age (yr)	24.2 ± 0.2	24.4 ± 0.5	24.2 ± 0.3	24.1 ± 0.4
Weight (kg)	70.6 ± 0.6	69.6 ± 1.2	71.6 ± 1.0	69.9 ± 1.1
Height (cm)	170.0 ± 0.3	170.3 ± 0.7	169.8 ± 0.5	169.9 ± 0.6
BMI ($kg \cdot m^{-2}$)	24.5 ± 0.2	24.1 ± 0.3	24.9 ± 0.3	24.5 ± 0.2

There were no significant differences in physical characteristics among the ACE ID genotypes.
ACE, angiotensin I–converting enzyme; I, insertion allele; D, deletion allele; BMI, body mass index.

the ACE II, ID, and DD genotypes, respectively (χ^2 = 1.688; P = 0.430) and frequencies of the I and D alleles of 46.2 and 53.8%, respectively. Descriptive characteristics did not differ by ACE genotype (Table 1) ($P \geq 0.05$).

Muscle strength and size by ACE genotype. The muscle strength and size measures in the T and UT arms at baseline and in response to RT are displayed in Table 2. Baseline muscle strength (MVC and 1RM) and size (CSA) were greater in the UT (dominant) than T (nondominant) arm in the total sample and ACE ID genotype groups (P < 0.001). However, baseline muscle strength and size did not differ among the ACE ID genotype groups in either the T or UT arm ($P \geq 0.05$).

T arm. All muscle strength and size measures increased in the T arm pre- to post-RT in the total sample and ACE ID genotype groups (P < 0.001) (Table 2). MVC gains in the T arm pre- to post-RT were greater for subjects with the ACE I allele (ACE II/ID) than those with the ACE DD genotype (Table 2), whether these gains were expressed in absolute terms, adjusted for baseline values, or allometrically scaled for body mass (P < 0.05). In contrast, unadjusted (absolute) and adjusted (relative percentage of baseline and allometric) 1RM increases in the T arm pre- to post-RT were not different among ACE ID genotype groups ($P \geq 0.05$) (Table 2). Absolute and relative percentage of muscle size increases in the T arm also were not different among ACE ID genotype groups ($P \geq 0.05$) (Table 2).

UT arm. All muscle strength and size measures increased in the UT arm pre- to post-RT among the total sample (P < 0.05); however, there were variable muscle strength and size responses to RT in the UT arm among ACE ID genotype groups (Table 2). MVC increased in the UT arm pre- to post-RT among those with the ACE I allele (ACE II/ID) (P < 0.001), whereas MVC was not different in the UT arm pre- to post-RT among those with the ACE DD genotype ($P \geq 0.05$), whether the responses were expressed in absolute terms, adjusted for baseline values, or allometrically scaled for body mass (Table 2). Unadjusted (absolute) and adjusted (relative percentage of baseline and allometric) 1 RM increased in the UT arm pre- to post-RT among ACE ID genotype groups (P < 0.05), and these gains were greater for subjects with the ACE D allele (ACE ID/DD) than those with the ACE II genotype (P < 0.05) (Table 2). Unadjusted (absolute) and adjusted (relative percentage of baseline) CSA increased in the UT arm pre- to post-RT among subjects with the ACE D allele (ACE DD/ID), whereas unadjusted (absolute) CSA decreased (P = 0.049) and adjusted (relative percentage of baseline) CSA was similar ($P \geq 0.05$) pre- to post-RT among those with the ACE II genotype (Table 2).

Proportion of variance attributable to ACE genotype. Our sample was large, enabling us to calculate the proportion of variance in the muscle

TABLE 2. Mean (± SEM) muscle strength and size measures in the trained and untrained arms at baseline and pre- to post–resistance training in the total sample and by ACE ID genotype.[a,b]

Variable	Arm	Total Sample (N = 631)	Genotype II (N = 146)	ID (N = 291)	DD (N = 194)
Baseline MVC (kg)	Trained	44.8 ± 0.6	43.1 ± 1.2	46.0 ± 0.9	45.4 ± 1.1
	Untrained	46.9 ± 0.7	45.6 ± 1.3	48.0 ± 1.0	47.1 ± 1.2
MVC absolute	Trained	8.0 ± 0.3	8.7 ± 0.6	8.6 ± 0.5	6.8 ± 0.6†
Change (kg)	Untrained	1.8 ± 0.3	2.3 ± 0.6	2.4 ± 0.4	0.6 ± 0.5^^^, ‡
MVC relative	Trained	20.5 ± 0.9	22.8 ± 1.7	22.0 ± 1.3	16.8 ± 1.6†
Change (%)	Untrained	5.2 ± 0.8	6.6 ± 1.4	7.0 ± 1.0	2.0 ± 1.3^^^, ‡
MVC allometric	Trained	0.45 ± 0.43	0.50 ± 0.04	0.50 ± 0.03	0.40 ± 0.03†
Change ($kg \cdot kg^{-0.67}$)	Untrained	0.10 ± 0.02	0.14 ± 0.03	0.14 ± 0.02	0.04 ± 0.03 ^^^, ‡
Baseline 1RM (kg)	Trained	8.6 ± 0.2	8.8 ± 0.2	9.2 ± 0.1	9.2 ± 0.2
	Untrained	9.8 ± 0.1	9.6 ± 0.2	9.9 ± 0.1	10.0 ± 0.2
1RM Absolute	Trained	3.9 ± 0.1	3.8 ± 0.2	4.1 ± 0.1	4.0 ± 0.1
Change (kg)	Untrained	0.8 ± 0.1	0.5 ± 0.1**	0.9 ± 0.1	1.0 ± 0.1
1RM relative	Trained	52.8 ± 1.3	51.6 ± 2.5	50.9 ± 1.8	51.2 ± 2.3
Change (%)	Untrained	9.9 ± 0.8	7.1 ± 1.5*	10.7 ± 1.1	11.9 ± 1.3
1RM allometric	Trained	0.23 ± 0.00	0.24 ± 0.01	0.24 ± 0.01	0.23 ± 0.01
Change ($kg \cdot kg^{-0.67}$)	Untrained	0.05 ± 0.00	0.03 ± 0.00**	0.05 ± 0.00	0.06 ± 0.01
Baseline CSA (cm^2)	Trained	17.0 ± 0.3	17.6 ± 0.4	18.0 ± 0.3	17.2 ± 0.4
	Untrained	18.3 ± 0.2	18.5 ± 0.4	18.5 ± 0.3	17.9 ± 0.4
CSA absolute	Trained	3.2 ± 0.1	3.1 ± 0.2	3.4 ± 0.1	3.4 ± 0.2
Change (cm^2)	Untrained	0.1 ± 0.5	−0.2 ± 0.1*$	0.3 ± 0.1	0.2 ± 0.1
CSA relative	Trained	18.7 ± 0.0	18.1 ± 0.9	19.1 ± 0.6	19.9 ± 0.8
Change (%)	Untrained	0.8 ± 0.3	−0.9 ± 0.5^^^,*$	1.7 ± 0.4	1.5 ± 0.5

[a] Covariates included age and body mass index with gender and ACE ID genotype as fixed factors.
[b] All muscle and sizes changes pre- to post-RT were significant among the total sample and the ACE ID genotype groups (P < 0.001); except ^^^ $P \geq 0.05$.
† P < 0.05, ‡ P < 0.01, ACE DD vs ACE ID/ID.
* P < 0.05, ** P < 0.01, $ P < 0.001, ACE II vs ACE ID/DD.
ACE, angiotensin I–converting enzyme; I, insertion allele; D, deletion allele; MVC, maximal voluntary contraction; 1RM, one-repetition maximum; CSA, cross-sectional area.

TABLE 3. Variability attributable to ACE genotype in muscle strength and size at baseline and pre- to post–resistance training in the trained and untrained arms.

Variable	Arm	R^2 Full Model	R^2 Constrained Model	Variation due to Genotype (%)	Likelihood Ratio[a] *P* Value
Baseline MVC (kg)	Trained	0.5471	0.5452	0.19	0.290
	Untrained	0.5531	0.5517	0.14	0.391
MVC absolute change (kg)	Trained	0.1042	0.0948	0.94	0.050
	Untrained	0.0271	0.0122	1.49	0.014
MVC relative change (%)	Trained	0.0253	0.0107	1.46	0.014
	Untrained	0.0282	0.0103	1.79	0.006
MVC allometric change ($kg \cdot kg^{-0.67}$)	Trained	0.0823	0.0724	0.99	0.046
	Untrained	0.0255	0.0113	1.42	0.017
Baseline 1RM (kg)	Trained	0.6243	0.6222	0.21	0.182
	Untrained	0.6484	0.6470	0.14	0.302
1RM absolute change (kg)	Trained	0.1009	0.0982	0.27	0.387
	Untrained	0.0448	0.0281	1.67	0.005
1RM relative change (%)	Trained	0.1635	0.1633	0.02	0.920
	Untrained	0.0720	0.0618	1.02	0.034
1RM allometric change ($kg \cdot kg^{-0.67}$)	Trained	0.1198	0.1176	0.22	0.460
	Untrained	0.0485	0.0337	1.48	0.008
Baseline CSA (cm^2)	Trained	0.5065	0.5042	0.20	0.322
	Untrained	0.5361	0.5344	0.17	0.417
CSA absolute change (cm^2)	Trained	0.2439	0.2393	0.46	0.231
	Untrained	0.0353	0.0017	3.36	< 0.001
CSA relative change (%)	Trained	0.0282	0.0234	0.48	0.297
	Untrained	0.0414	0.0049	3.65	< 0.001

[a] Likelihood ratio test comparing full model (with ACE genotype, gender, age, and body mass index) to constrained model (gender, age, and body mass index).
ACE, angiotensin I–converting enzyme; MVC, maximal voluntary contraction; 1RM, one-repetition maximum; CSA, cross-sectional area.

strength and size response to RT attributable to ACE ID genotype in the T and UT arms (Table 3).

T arm. As shown in Table 3, 0.9% of the variability in the absolute MVC increase ($P = 0.050$), 1.5% of the variability in the MVC increase adjusted for baseline values ($P < 0.01$), and 1.0% of the allometrically scaled MVC increase in the T arm were attributable to ACE ID genotype. The likelihood ratio tests of all other muscle strength (1RM) and size (CSA) measures at baseline and pre- to post-RT in the T arm did not achieve statistical significance ($P \geq 0.05$).

UT arm. ACE ID genotype accounted for a small proportion of the variance in the muscle strength (MVC, 1.0–1.5%; 1RM, 1–1.7%) and size (CSA, 3.4–3.6%) response to RT in the UT arm ($P < 0.05$), whereas the likelihood ratio tests for contributions of ACE ID genotype to baseline muscle strength and size did not achieve statistical significance ($P \geq 0.05$).

DISCUSSION

We tested whether the ACE DD genotype was associated with superior muscle strength and size response to a standardized, unilateral RT intervention in a large population of healthy young men and women who were mostly white. The 11 muscle strength and size measures examined in the T and UT arm were baseline MVC and 1RM and muscle size, absolute change in MVC and 1RM and muscle size, relative percentage of change in MVC and 1RM and muscle size, and allometric change in MVC and 1RM.

The major findings of this study were 1) subjects with the ACE I allele (ACE II/ID) had greater unadjusted (absolute) and adjusted (relative percentage of baseline and allometric) MVC gains compared with ACE DD homozygotes in the T and UT arms (Table 2); 2) the ACE ID genotype was not associated with unadjusted and adjusted gains in 1RM and muscle size in the T arm (Table 2); 3) subjects with the ACE D allele (ACE DD/ID) had greater unadjusted and adjusted gains in 1RM and muscle size than ACE II homozygotes in the UT arm (Table 2); and 4) ACE genotype was not associated with baseline muscle strength and size in either arm (Table 2). Thus, the ACE ID genotype was associated with the contralateral muscle strength and size effects of unilateral RT, and these associations were no longer apparent after dynamic RT when the T arm was tested dynamically. These unexpected results are contrary to our hypothesis and to reports of others (6,15,28) that the ACE D allele is associated with a superior muscle strength and size response to RT compared with the ACE I allele. Furthermore, they suggest the ACE ID genotype appears to be associated with the cross-education effects of unilateral RT (5,14,18), more so than with the inherent muscle strength and size adaptations that result from RT.

The assertion that the ACE D allele is associated with enhanced muscle strength and power-oriented performance is based primarily on observational evidence. An excess of the ACE D allele has been found among elite sprint runners (15) and swimmers (28). Folland et al. (6) is one of the few investigators to use an RT intervention to examine the associations of the ACE ID genotype and the muscle strength response to RT. In this study, 33 young men performed 9 wk of quadriceps muscle RT in which one leg was trained isometrically and the other leg dynamically. Folland et al. (6) observed greater MVC and similar 1RM strength gains among subjects with the ACE D allele (ACE DD/ID) than the ACE II genotype. Thomis et al. (21) studied 57 young male twins who underwent 10 wk of intense, dynamic RT of the elbow flexors. They found no associations of the ACE ID genotype with 1RM gains and muscle size assessed by computed tomography, but did find borderline significance for larger concentric flexion torque

gains in ACE II homozygotes than those with the ACE D allele. In contrast, Williams et al. (26) found no difference in the isokinetic muscle strength response to an 8-wk dynamic RT program of the legs among 44 men 23 yr of age.

The reasons for the discrepancies between the findings of Folland et al. (6), Thomis et al. (21), Williams et al. (26), and ours are not readily apparent. They could be related to differences in the RT intervention and assessments employed, that is, lower- versus upper-body and/or dynamic, isokinetic, or isometric training and/or testing. The large sample size of this study ($N = 631$) enabled us to statistically account for important confounding factors in the muscle strength and size response to RT (i.e., age, gender, and BMI) and provided us with sufficient statistical power to detect the differences we found in the muscle strength and size response to RT according to ACE ID genotype.

We observed similar (MVC) and even stronger (1RM and CSA) associations among the ACE genotype and the muscle strength and size response to RT in the UT compared with the T arm (Tables 2 and 3). These findings suggest the ACE ID genotype is associated more with the contralateral effects of unilateral RT than with the intrinsic muscle strength and size adaptations that result from RT. The proposed mechanisms for the contralateral effects of unilateral training, also termed cross-education, cross-training, and/or the cross-transfer effect, include improved voluntary activation via motor unit recruitment and increased motor unit firing rate resulting from bilateral activation of the central nervous system during a unilateral task (5,14,18). The recommended modality for evaluating cross-education effects when training the muscle dynamically is MVC in order to avoid learning effects (18,20). It is interesting to note that Folland et al. (6), Thomis et al. (21), and we have found associations among the ACE ID genotype and the muscle strength response to dynamic RT in the T limb when measured with isometric or isokinetic testing. Collectively, these findings support the notion that the ACE ID genotype associates with the cross-education and learning effects of RT that are primarily neurologically mediated and that these associations are no longer evident when the limb is trained *and* tested dynamically.

Mechanisms to explain our findings that the 1RM and muscle size gains were greater in carriers of the ACE D allele (ACE DD/ID) than those with the ACE II genotype in the UT arm, while the MVC gains were greater in carriers of the ACE I allele (II/ID) than those with the ACE DD genotype in the T and UT arms, are not readily apparent. However, these results suggest that the associations among the ACE genotype and the muscle strength and size response to RT are mediated by neurological mechanisms due to cross-education and learning effects. Wali (25) and Jonsson et al. (10) have shown that angiotensin II facilitates neuromuscular transmission, and that this response is exaggerated in spontaneously hypertensive rats. The ACE D allele is associated with higher ACE activity and angiotensin II production than the ACE I allele (2,11,23), lending support for a neurological explanation for our findings. Alternatively, the ACE I allele is associated with enhanced contractile efficiency by the skeletal muscles compared with the D allele (27,30) and could account for the greater MVC gains in carriers of the ACE I allele than subjects with the ACE DD genotype. Bradykinin is involved in the exercise pressor reflex, being released by high-intensity, static muscle contractions in direct proportion to lactate production and inversely related to pH (19). It seems plausible that the exercise-induced ACE signaling effects are greater with the ACE I allele due to its associations with increased bradykinin activity compared with the ACE D allele (23), resulting in enhanced contractile efficiency and a superior MVC response to RT.

Another explanation for our findings is that the ACE ID polymorphism is one of many genetic variants contributing to the observed variance in the muscle strength and size response to RT (3,11,23). Epistatic interactions between the ACE ID genotype and another gene variant in the kinin β_2 receptor associated with the regulation of skeletal muscle exercise performance have recently been observed (27). We found that approximately 1% of the variability in MVC strength in the T arm and 2–4% of the variability in the muscle strength and size response in the UT arm was attributable to the ACE genotype (Table 3). Such small contributions that were detected in a large sample suggest the ACE ID genotype may be a marker for some other functional genetic variant or be in linkage disequilibrium with another gene or genes more directly responsible for the effects that we observed. Clearly, more work is needed to better clarify the pathways through which the ACE ID genotype and other genetic variants may associate with the muscle strength and size response to RT.

In conclusion, we found the ACE ID genotype associated with the contralateral muscle strength and size effects of unilateral RT, and these associations remained after dynamic RT only when the T arm was tested isometrically. Contrary to our hypothesis, subjects with the ACE I allele (ACE II/ID) had greater MVC strength gains compared with ACE DD homozygotes in the T and UT arms. Thus, it appears that the ACE ID genotype is primarily associated with the cross-education and learning effects rather than the inherent muscle strength and size adaptations that result from RT. The variability of the muscle strength and size gains as a result of RT attributable to ACE ID genotype was approximately 1–4%. Such small contributions found in a large study sample indicate that ACE ID polymorphism may merely be a functional marker and/or is one of many genes that contribute to the muscle strength and size response to RT. The challenge remains to identify other genetic variants and their interactions that explain a larger proportion of the muscle strength and size response to RT.

We thank the students and technicians at the participating institutions for their time and effort, the subjects for their participation and commitment to the project, and Monica Hubal, M.S., and Stephen Bilbie, M.S., R.D., for the expert input they gave to the manuscript.

Supported by a grant from the NIH (NINDS/NIA/NIAMS NS40606), and the Parsons Family Foundation.

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