Published in final edited form as:

Hum Biol. 2008 February ; 80(1): 1–9.

Genetic Architecture of Knee Radiographic Joint Space in Healthy Young Adults

DANA L. DUREN^{1,2}, RICHARD J. SHERWOOD^{1,3}, STEFAN A. CZERWINSKI¹, WILLIAM CAMERON CHUMLEA¹, MIRYOUNG LEE¹, ELLEN W. DEMERATH⁴, SHUMEI S. SUN⁵, ROGER M. SIERVOGEL¹, and BRADFORD TOWNE¹

¹Department of Community Health, Lifespan Health Research Center, Boonshoft School of Medicine, Wright State University, 3171 Research Boulevard, Dayton, OH 45420-4014

²Department of Orthopaedic Surgery, Boonshoft School of Medicine, Wright State University, Dayton, OH

³Department of Neuroscience, Cell Biology and Physiology, Wright State University, Dayton, OH

⁴Division of Epidemiology and Community Health, School of Public Health, University of Minnesota, Minneapolis, MN

⁵Department of Biostatistics, Virginia Commonwealth University, Richmond, VA

Abstract

Evidence of a significant genetic component to the age-related degenerative joint disease osteoarthritis has been established, but the nature of genetic influences on normal joint morphology in healthy individuals remains unclear. Following up on our previous findings on the influence of body habitus on phenotypic variation in knee joint space [Duren et al., *Human Biology* 78:353–364 (2006)], the objective of the current study was to estimate the heritability of radiographic joint space in the knees of healthy young adults from a community-based sample of families. A sample of 253 subjects (mean age = 18.02 years) from 87 randomly ascertained nuclear and extended families was examined. Joint width (JW) and minimum joint space in the medial (MJS) and lateral (LJS) knee compartments were measured. A maximum-likelihood variance components method was used to estimate the heritability of MJS, LJS, and JW. Covariate effects of age, sex, age-by-sex interactions, stature, weight, and BMI were simultaneously estimated. Genetic correlation analyses were then conducted to examine relationships between trait pairs. MJS, LJS, and JW were each significantly heritable ($p < 0.001$), with heritabilities of 0.52, 0.53, and 0.63, respectively. The genetic correlation between MJS and LJS was not significantly different from 1. Genetic correlations between each joint space measure and JW were not significantly different from 0. This study demonstrates a significant genetic component to radiographic knee joint space during young adulthood in healthy subjects. This suggests that there are specific but as yet unidentified genes that influence the morphology of healthy articular cartilage, the target tissue of osteoarthritis. Genetic correlation analyses indicate complete pleiotropy between MJS and LJS but genetic independence of joint space and JW.

Keywords

KNEE JOINT SPACE; CARTILAGE; OSTEOARTHRITIS; HERITABILITY OF OSTEOARTHRITIS; X-RAY; FELS LONGITUDINAL STUDY

Idiopathic osteoarthritis (OA) is generally considered a disease of aging. Like many other aging conditions (e.g., osteoporosis), however, predisposition to OA can be recognized earlier in life, before overt degenerative changes become apparent in the joint, and may be oligogenic in nature. That is, the traits underlying the condition are influenced by a discrete set or sets of genes that together are responsible for variation in those traits. The genetic nature of predisposition to common complex diseases such as OA, however, has yet to be fully elucidated. In addition to their oligogenic nature, myriad potential epigenetic influences can significantly contribute, either individually or in combination, to variation in the expressed disease or disease traits. OA is particularly challenging for genetic epidemiologic study because of its multifactorial nature and the variation in the expression of subphenotypes that make up the disease (e.g., osteophytes, sclerosis, cysts, and joint space narrowing).

The first genetic study of OA was published by Stecher (1941), who noted that siblings of arthritic patients were more likely than unrelated individuals to demonstrate clinical signs of OA, particularly the presence of Heberden's nodes at the finger joints. More recent genetic studies have found that OA in many regions of the body is highly heritable (reviewed by Spector and MacGregor 2004). For example, twin studies have shown that hip, hand, and knee OA is 39–65% heritable (MacGregor et al. 2000; Spector et al. 1996). Heritability estimates for generalized OA and OA of the hand and spine based on sib-pair studies range from 56% to 78% (Bijkerk et al. 1999).

Recent evidence also suggests that two individual characteristic features of OA, osteophytes and joint space narrowing, may each have a unique genetic etiology. Both of these traits are heritable, but different genes that are associated with the two OA subphenotypes have been identified. The gene coding for COL2A1, a structural protein found in articular cartilage, has been shown to be associated with joint space narrowing in a population-based sample of older adults (Uitterlinden et al. 2000). In the same study sample the vitamin D receptor gene was identified as being involved in osteophytosis (Uitterlinden et al. 2000).

Genetic contributions to variation in articular cartilage thickness in particular are potentially the most important in terms of OA treatment and prevention. The nature of the genetic underpinnings of disease and nondisease states (i.e., normal healthy variation) is important for ultimately understanding complex traits and diseases. As researchers close in on specific genes involved in OA and look toward gene therapies for cartilage defects (Cucchiari and Madry 2005; Evans et al. 2006), it is important that genetic influences on variation in normal healthy joint cartilage and adjacent bone also be considered. In a previous study we showed that body habitus has minimal influence on radiographic knee joint space (Duren et al. 2006). The objective of the current study is to estimate heritabilities of radiographic knee joint space and joint width and the genetic correlations between them in this same sample of healthy, young, skeletally mature individuals.

Subjects and Methods

Subjects

The study sample consisted of 253 individuals (132 men, 121 women) participating in the Fels Longitudinal Study. Initiated in 1929, the Fels Longitudinal Study is the world's largest and longest running study of human growth, development, and body composition change over the life span (Roche 1992). Fels Longitudinal Study participants are primarily white and live in or near southwest Ohio (Roche 1992). The 253 subjects in this particular study sample are from 87 nuclear and extended families. Among these 253 subjects there are 641 relative pairings represented, ranging from first-degree to sixth-degree relationships (Table 1).

Knee radiographs of the study subjects were taken between 1946 and 1987. Inclusion in the study sample was restricted to individuals who (1) were between 16.0 and 22.4 years of age at the time of radiography, (2) had attained skeletal maturity at the knee [complete medial and lateral epiphyseal fusion in the femur and tibia (Roche et al. 1975)], and (3) exhibited cartilage that had not suffered any age-related degenerative changes. The unique nature of the Fels Longitudinal Study enabled us to examine radiographs of individuals from families spanning up to three generations. All subjects were healthy and free of orthopedic conditions at the time of radiography, and they provided informed written consent to procedures before participation.

Measurements

Radiographs were taken of the left knee in standard weight-bearing anteroposterior views with the x-ray beam set on the joint center. Measurements obtained from the knee radiographs for this study were minimum medial joint space (MJS), minimum lateral joint space (LJS), and joint width (JW). Minimum joint space in each compartment was measured to approximate the measurement technique used when assessing clinical severity of OA or other joint diseases. All joint measurements were made to the nearest 0.01 mm using digital calipers with direct computer entry.

Stature and body weight were taken using standard methods (Lohman et al. 1988). Height was measured to the nearest 0.1 cm using a stadiometer with subjects barefoot; body weight, measured to the nearest 0.1 kg, was obtained using a scale with subjects barefoot and wearing light clothing. The body mass index (BMI) was calculated as kilograms per meters squared.

Reliability of Study Data

Reliability of measurements from knee radiographs in the standing anteroposterior view taken at our research facility was tested in a separate sample of knee radiographs of 49 adult subjects (mean age 53.5 years). Each of these subjects had two radiographs taken at least 1 year apart, with no signs of OA present in either radiograph (Kellgren-Lawrence score of 0). MJS, LJS, and JW were measured on each radiograph by the same observer. Reliability of knee measures was assessed using intraclass correlation (ICC) procedures implemented in SAS, version 9.1.

ICC coefficients for radiographic view reliability were 0.99, 0.86, and 0.85 for JW, MJS, and LJS, respectively. In addition, analyses of repeated measures demonstrated that inter- and intra-observer reliability was high (see Duren et al. 2006).

Statistical and Genetic Analyses

Quantitative genetic methods were used to estimate the heritability of each radiographic knee joint trait. Narrow-sense heritability, the proportion of phenotypic variance attributable to additive genetic effects, was estimated using a maximum-likelihood-based variance components method that uses all familial information (i.e., all relative pairs) for parameter estimation (Lange and Boehnke 1983), as implemented in SOLAR, version 2.1.2 (Almasy and Blangero 1998; available at <http://www.sfbr.org/solar/index.html>). In this approach the covariance matrix for a pedigree is given by

$$\Omega = 2\Phi\sigma_a^2 + I_n\sigma_e^2, \quad (1)$$

where Φ is the kinship matrix for the pedigree, σ_a^2 is the trait variance resulting from additive genetic effects, I_n is the $n \times n$ identity matrix, and σ_e^2 is the trait variance resulting from random, individual-specific environmental effects. The basic parameters estimated include the trait mean, standard deviation, and h^2 , where

$$h^2 = \frac{\sigma_a^2}{\sigma_a^2 + \sigma_e^2}. \quad (2)$$

The null hypothesis is that σ_a^2 equals 0. The significance of heritability estimates was tested by comparing the likelihoods of nested models using the likelihood ratio test. Variables were examined for covariate effects of age, sex, age², age by sex interaction, age² by sex interaction, weight, stature, body mass index (BMI), and JW. In addition, covariate effects of age and its interactions with sex were tested to examine potential age effects that might possibly exist, even in this highly age restricted sample. To estimate the effects of these covariates on the study traits, we screened each covariate in SOLAR by comparing the likelihood of a model that included the covariate to the likelihood of a model that excluded the covariate. Only covariates significant at $p < 0.10$ were retained in the final model for each trait.

Bivariate quantitative genetic methods were used to determine the nature of shared genetic effects on MJS, LJS, and JW. Using bivariate methods in the SOLAR analytic platform, we partitioned phenotypic correlations between traits into additive genetic and random environmental components (Lange and Boehnke 1983) as

$$\rho_P = \rho_G (h_1^2)^{1/2} (h_2^2)^{1/2} + \rho_E (1-h_1^2)^{1/2} (1-h_2^2)^{1/2}, \quad (3)$$

where ρ_P is the phenotypic correlation, ρ_G is the genetic correlation, and ρ_E is the environmental correlation between trait pairs, and h_1^2 and h_2^2 are the heritabilities for trait 1

and trait 2, respectively. Additive genetic correlations (ρ_G) range from -1.0 to 1.0 , where a value of 1.0 indicates complete positive pleiotropy (i.e., the same genes are affecting the two traits in the same direction), a value of 0 between traits indicates that different genes control them, and a value of -1.0 indicates complete negative pleiotropy (i.e., genes that act to increase the value of one trait act to decrease the value of the other). A genetic correlation significantly different from both 0 and 1 (or -1.0) indicates incomplete pleiotropy, meaning that the two traits are influenced to some extent by the same genes but that each trait also has a genetic basis unique from the other. Because of multiple testing issues and increased chance of committing type I errors, our α is adjusted for the genetic correlation analyses using a Bonferroni correction, which sets an α of 0.0167 for determination of significance.

Results

Descriptive statistics for characteristics of the study sample are presented in Table 2. Results of quantitative genetic analyses, including maximum-likelihood estimates of trait means, heritabilities, standard errors, p values, and covariate effects, are presented in Table 3. The residual heritabilities (i.e., the heritability after accounting for all covariate effects) of MJS and LJS were significant ($p < 0.001$) and of high magnitude ($h^2 = 0.52$ and 0.53 , respectively). Significant covariates in the analysis of MJS included sex, BMI, and JW. The proportion of the total phenotypic variance attributable to these three covariates was 24% (20.9% due to sex alone). Sex was the only significant covariate in the analysis of LJS, accounting for 20% of the total phenotypic variance. The residual heritability of JW was 0.63 ($p < 0.001$), with significant covariate effects of age, sex, age², age² by sex, and weight, accounting for 76% of the total phenotypic variance in JW.

Each genetic correlation (ρ_G) between trait pairs was tested for significance from both 1 (or -1) and 0 . Covariates used in this analysis incorporated only those significant for the included trait in the univariate analysis. For the bivariate analyses involving JW, JW was not included as a covariate even though it was significant in the univariate analysis of MJS. The genetic correlation between MJS and LJS was 0.67 ($SE = 0.20$) and was not significantly different from 1 ($p > 0.0167$). Genetic correlations between each joint space measure and JW were not significantly different from 0 [$\rho_G = 0.14$ ($SE = 0.26$) between MJS and JW, and $\rho_G = 0.04$ ($SE = 0.22$) between LJS and JW].

Discussion

Based on studies of osteoarthritis, it is clear that genes play a significant role in cartilage health during older adulthood. What remains unclear is how much of this genetic effect is targeting cartilage integrity and how much is targeting cartilage destruction. A recent examination of 228 monozygotic twins revealed a significant genetic influence on progression of OA once established (Zhai et al. 2007). Most genetic studies of OA are designed to search for genes that are causally related to OA. However, in using this approach, one might overlook the role of genes acting before disease onset. A set of genes controlling cartilage building and/or maintenance is one example. Such a set of genes may be activated early in life and may control the attainment of peak cartilage thickness, which may then significantly affect the risk of OA later in life.

In the current study we have demonstrated a significant genetic effect (high heritability estimates) on knee joint space variability in a randomly ascertained community-based sample of healthy young adults. In our sample there is a significant sex effect on knee joint space that accounts for approximately 20% of the trait variance. We have previously shown that body size accounts for only a small proportion of the variation in joint space (Duren et al. 2006). This finding was further supported here by the covariate effects of BMI and JW, together accounting for only 3.1% of the phenotypic variance in MJS after controlling for sex and 0% of the phenotypic variance in LJS. The identification of a sex effect on joint space at this young age may help to explain the sex disparity of OA in later life.

Our study is the first to examine the genetic architecture of joint space phenotypes in a sample of healthy young adults. Compared with studies of older adults and subjects with OA, our residual heritability results are congruent with, and in some cases higher than, those reported previously. A sibling-pair study that examined knee cartilage using MRI showed that medial and lateral cartilage volumes are 65% and 77% heritable, respectively (Zhai et al. 2004). Similar results were found by Hunter et al. (2003), where femoral and tibial cartilage volumes were analyzed separately and found to be 61–76% heritable. Similarly, Spector et al. (1996) found that joint space narrowing was 41% heritable in a sample of female twins, age 48 to 70 years.

Heritabilities reported in these studies are based on populations that either have OA or are at risk for OA (e.g., menopausal or perimenopausal women). As posed earlier, it remains unclear whether the genetic architecture of OA-related traits represents a change in gene expression that maintains cartilage integrity or a change in genetic pathways that lead to degeneration, or both. The examination of populations with, or at risk of, OA obscures distinctions between these two potential genetically mediated pathways. In the current study we used only healthy young adults to elucidate the genetic effects on knee joint morphology. In so doing, we are examining the genetic underpinnings of variation in joint space before any pathway, genetic or otherwise, is deleterious to cartilage health.

In addition to a significant heritability of each joint space measure, MJS and LJS show complete pleiotropy. That is, the heritable component of joint space in both knee compartments is due to the same set of genes. In contrast, both measures of joint space show genetic independence from JW. Although these results are not unexpected, this is the first time these relationships have been empirically demonstrated. Genetic independence of joint space and joint width reinforces our previous assertion that variation in knee joint space is largely unrelated to body size.

A potential limitation of the current study is that the knee radiographs taken in the Fels Longitudinal Study, including the subset of 253 radiographs analyzed for the purpose of this study, were all taken in the traditional anteroposterior view. This technique is widely used in both clinical and research populations for the diagnosis of OA (Hart et al. 2002; Hunter et al. 2005; Manek et al. 2003), but it has been suggested that the traditional standing anteroposterior radiographic view of the knee increases variability in the appearance of joint space because of variation in knee positioning, specifically tibial plateau alignment (Vignon et al. 2003). Such an increase in stochastic variation would reduce the heritability estimate

by adding nongenetic variation. Thus we consider our heritability estimates presented here to be conservative. In addition, our demonstrated high reliability and reproducibility (ICC 0.85) of joint measurements taken from the anteroposterior view indicates that findings of the current study represent the basic genetic architecture of normal variation in knee joint space.

Elucidation of the role of genes in normal healthy cartilage thickness (or its surrogate, radiographic joint space) is a timely subject. Genes coding for key components of cartilage matrix and proteins are among the prominent candidate genes identified by linkage studies of OA. These include genes such as collagen genes (*COL2A1* and *COL5A2*), bone morphogenetic protein 2 (*BMP2*), prostaglandin-endoperoxide synthase 2 (*COX2*), frizzled-related protein (*FR2B*), and the inter-leukin 2 gene cluster (Min et al. 2005; Uitterlinden et al. 2000; Valdes et al. 2004). Previous studies have identified these as OA-related genes, but each of these genes may play a key role in the development and maintenance of articular cartilage thickness. And, much like peak bone mass has been identified as the best predictor of later osteoporosis risk (Hui et al. 1999), so may we find that the attainment of peak cartilage thickness during early adulthood predicts later risk of OA. Our current findings support the role of genetics in healthy cartilage integrity and represent a step toward identifying genes involved in the maintenance and integrity of healthy cartilage tissue and risk of later OA.

Acknowledgments

This work was supported by the Arthritis National Research Foundation and by the National Institutes of Health through grants HD12252 and HD36342. We thank two anonymous reviewers for their comments on the original manuscript. We also thank Kimberly Ritter for measurement assistance and respectfully acknowledge the Fels Longitudinal Study participants.

Literature Cited

- Almasy L, Blangero J. Multipoint quantitative-trait linkage analysis in general pedigrees. *Am J Hum Genet.* 1998; 62:1198–1211. [PubMed: 9545414]
- Bijkerk C, Houwing-Duistermaat JJ, Valkenburg HA, et al. Heritabilities of radiologic osteoarthritis in peripheral joints and of disc degeneration of the spine. *Arthritis Rheum.* 1999; 42:1729–1735. [PubMed: 10446874]
- Cucchiari M, Madry H. Gene therapy for cartilage defects. *J Gene Med.* 2005; 7:1495–1509. [PubMed: 16142829]
- Duren DL, Sherwood RJ, Chumlea WC, et al. Radiographic joint space of the knee in healthy young adults. *Hum Biol.* 2006; 78:353–364. [PubMed: 17216807]
- Evans CH, Ghivizzani SC, Robbins PD. Gene therapy for arthritis: What next? *Arthritis Rheum.* 2006; 54:1714–1729. [PubMed: 16729280]
- Hart DJ, Cronin C, Daniels M, et al. The relationship of bone density and fracture to incident and progressive radiographic osteoarthritis of the knee: The Chingford Study. *Arthritis Rheum.* 2002; 46:92–99. [PubMed: 11817613]
- Hui SL, Zhou L, Evans R, et al. Rates of growth and loss of bone mineral in the spine and femoral neck in white females. *Osteoporos Int.* 1999; 9:200–205. [PubMed: 10450407]
- Hunter DJ, Niu J, Zhang Y, et al. Knee height, knee pain, and knee osteoarthritis: The Beijing Osteoarthritis Study. *Arthritis Rheum.* 2005; 52:1418–1423. [PubMed: 15880346]
- Hunter DJ, Snieder H, March L, et al. Genetic contribution to cartilage volume in women: A classical twin study. *Rheumatology.* 2003; 42:1495–1500. [PubMed: 12832711]

- Lange K, Boehnke M. Extensions to pedigree analysis. VI. Covariance components models for multivariate traits. *Am J Med Genet.* 1983; 14:513–524. [PubMed: 6859102]
- Lohman, T.; Roche, A.; Martorell, R. *Anthropometric Standardization Reference Manual.* Champaign, IL: Human Kinetics Books; 1988.
- MacGregor AJ, Antoniades L, Matson M, et al. The genetic contribution to radiographic hip osteoarthritis in women: Results of a classic twin study. *Arthritis Rheum.* 2000; 43:2410–2416. [PubMed: 11083262]
- Manek NJ, Hart D, Spector TD, et al. The association of body mass index and osteoarthritis of the knee joint: An examination of genetic and environmental influences. *Arthritis Rheum.* 2003; 48:1024–1029. [PubMed: 12687544]
- Min JL, Meulenbelt I, Riyazi N, et al. Association of the Frizzled-related protein gene with symptomatic osteoarthritis at multiple sites. *Arthritis Rheum.* 2005; 52:1077–1080. [PubMed: 15818669]
- Roche, AF. *Growth, Maturation, and Body Composition: The Fels Longitudinal Study 1929–1991.* Cambridge, U.K: Cambridge University Press; 1992.
- Roche, AF.; Wainer, H.; Thissen, D. *Skeletal Maturity: The Knee Joint as a Biological Indicator.* New York: Plenum; 1975.
- Spector TD, Cicuttini F, Baker J, et al. Genetic influences on osteoarthritis in women: A twin study. *Br Med J.* 1996; 312:940–943. [PubMed: 8616305]
- Spector TD, MacGregor AJ. Risk factors for osteoarthritis: Genetics. *Osteoarthritis Cartilage.* 2004; 12(suppl A):S39–S44. [PubMed: 14698640]
- Stecher R. Heberden's nodes: Heredity in hypertrophic arthritis of finger joints. *Am J Med Sci.* 1941; 201:801–809.
- Uitterlinden AG, Burger H, van Duijn CM, et al. Adjacent genes, for *COL2A1* and the vitamin D receptor, are associated with separate features of radiographic osteoarthritis of the knee. *Arthritis Rheum.* 2000; 43:1456–1464. [PubMed: 10902746]
- Valdes AM, Hart DJ, Jones KA, et al. Association study of candidate genes for the prevalence and progression of knee osteoarthritis. *Arthritis Rheum.* 2004; 50:2497–2507. [PubMed: 15334463]
- Vignon E, Piperno M, Le Graverand MP, et al. Measurement of radiographic joint space width in the tibiofemoral compartment of the osteoarthritic knee: Comparison of standing anteroposterior and Lyon schuss views. *Arthritis Rheum.* 2003; 48:378–384. [PubMed: 12571846]
- Zhai G, Hart DJ, Kato BS, et al. Genetic influence on the progression of radiographic knee osteoarthritis: A longitudinal twin study. *Osteoarthritis Cartilage.* 2007; 15:222–225. [PubMed: 17045816]
- Zhai G, Stankovich J, Ding C, et al. The genetic contribution to muscle strength, knee pain, cartilage volume, bone size, and radiographic osteoarthritis: A sib pair study. *Arthritis Rheum.* 2004; 50:805–810. [PubMed: 15022322]

Table 1

Relative Pairs Represented Among the 253 Study Subjects in 87 Families

<i>Relationship</i>	<i>Degree</i>	<i>N</i>
Identical siblings		3
Parent-offspring	First	54
Siblings	First	138
Avuncular	Second	68
Half-siblings	Second	36
Grand avuncular	Third	1
Half avuncular	Third	9
First cousins	Third	155
First cousins, once removed	Fourth	46
Half first cousins	Fourth	5
Second cousins	Fifth	124
Second cousins, once removed	Sixth	2
Total		641

Table 2

Descriptive Statistics of Study Data: Mean and SD

	Male (n = 132)		Female (n = 121)	
	<i>Mean (SD)</i>	Range	<i>Mean (SD)</i>	Range
Age (yr)	18.1 (0.62)	16.0–22.4	17.9 (0.61)	16.0–22.4
Stature (cm)	178.1 (6.68)	158.7–193.0	164.4 (5.31)	152.1–177.5
Weight (kg)	70.6 (11.15)	48.1–115.3	57.1 (8.15)	40.7–83.5
BMI (kg/m ²)	22.2 (2.93)	16.8–36.4	21.1 (2.79)	15.9–32.2
Medial joint space (mm)	5.64 (0.93)	3.2–8.2	4.71 (0.88)	2.5–7.6
Lateral joint space (mm)	5.74 (1.35)	2.5–9.5	4.53 (1.14)	1.9–8.8
Joint width (mm)	83.36 (4.46)	73.2–94.6	72.51 (2.71)	66.3–78.3

Table 3

Maximum-Likelihood Estimates of Parameters from the Best-Fit Univariate Models of Radiographic Joint Space Measures (Estimate $\pm$ SE)

Parameter	Radiographic Measure		
	Medial Joint Space	Lateral Joint Space	Joint Width
h^2	0.52 ± 0.16	0.53 ± 0.13	0.63 ± 0.15
μ	5.43 ± 0.11	5.73 ± 0.12	82.00 ± 0.33
σ	0.88 ± 0.04	1.23 ± 0.06	3.19 ± 0.15
β_{age}	ns ^a	ns	-1.13 ± 0.76
β_{sex}	-0.56 ± 0.19	-1.23 ± 0.15	-8.36 ± 0.47
$\beta_{\text{age} \times \text{sex}}$	ns	ns	1.25 ± 0.93
β_{age^2}	ns	ns	0.64 ± 0.22
$\beta_{\text{age}^2 \times \text{sex}}$	ns	ns	-0.49 ± 0.29
$\beta_{\text{joint width}}$	0.03 ± 0.02	ns	–
β_{BMI}	0.04 ± 0.02	ns	ns
β_{weight}	ns	ns	0.18 ± 0.02
β_{stature}	ns	ns	ns
Percentage of total trait variance due to covariates	24.0%	19.9%	76.5%

μ = Trait mean.

σ = Trait standard deviation.

β = Regression coefficient of covariate.

^a Not significant ($p > 0.10$).