Clinical Research Article

Insulin-like Growth Factor-1, Bone Mineral Density, and Fracture: A Mendelian Randomization Study

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Abbreviations: eBMD, estimated bone mineral density; GWAS, genome-wide association study; IGF-1, insulin-like growth factor-1; MR, mendelian randomization.

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Abstract

Context: The associations of circulating insulin-like growth factor-1 (IGF-1) levels with bone mineral density and fracture risk are inconclusive in observational studies.

Objective: We conducted a mendelian randomization study to assess the associations of serum IGF-1 levels with estimated bone mineral density (eBMD) and fracture.

Methods: Genetic instruments for IGF-1 were selected at the genome-wide significance level ($P < 5 \times 10^{-8}$) from a genome-wide association study including 358 072 individuals of European ancestry. Summary-level data for eBMD (426 824 individuals) and fracture (53 184 fracture cases and 373 611 noncases) were obtained from the UK Biobank study. Univariable and multivariable mendelian randomization analyses methods were used to estimate the associations of IGF-1 with eBMD and fracture. The main outcome measure included the change of eBMD and odds ratio of fracture per genetically predicted 1-SD increase of serum IGF-1 levels.

Results: For 1-SD increase in IGF-1, the change of eBMD levels was 0.04 g/cm² (95% CI, 0.01-0.07; $P = .011$) and the odds ratio of fracture was 0.94 (95% CI, 0.91-0.98; $P = .003$). The associations persisted with similar magnitude after adjustment for height. The association was consistent for fracture but not for eBMD after excluding genetic instruments that might directly influence these outcomes. The association between IGF-1 and fracture was somewhat attenuated after adjustment for eBMD (odds ratio 0.96; 95% CI, 0.92-0.99; $P = .012$).
Insulin-like growth factor-1 (IGF-1) may exert anabolic effects on bone, specifically on the acquisition of bone mass during adolescence and the maintenance of skeletal architecture during adult life, which affect the risk of subsequent fracture (1, 2). Observational studies assessing the role of IGF-1 for bone health have been conflicting, with a reduced risk of bone mineral density loss and fracture with higher circulating IGF-1 levels found in some studies (3-6) but not in others (7, 8). Results from observational studies may be affected by residual confounding and cannot with certainty infer causality. Increased serum IGF-1 levels were shown to attenuate proximal femur bone loss and to shorten hospital stays among patients with recent hip fracture in a randomized controlled trial (9). However, in another randomized controlled trial with 16 individuals, 1 year of IGF-1 treatment showed no effects on bone density (10). The baseline difference in bone mineral density, unequal group sizes, high dropout rates, and inadequate power may question this null finding. Thus, the causal role of increased IGF-1 levels in bone mineral density and fracture remains unknown.

Mendelian randomization (MR) analysis is an epidemiological method using genetic variants as instrumental variables for an exposure (eg, serum IGF-1 levels) (11, 12). This method diminishes residual confounding and minimizes reverse causation bias, and therefore, can strengthen the causal inference in an exposure-outcome association. Here, we conducted an MR study to determine the associations of genetically proxied higher serum IGF-1 levels with bone mineral density and fracture.

Materials and Methods

Study design

The present study was based on summary-level data from published genome-wide association studies (GWASs) and the UK Biobank study. We first assessed the associations of genetically proxied high levels of IGF-1 with bone mineral density and fracture. We further conducted multivariable MR analysis to estimate the possible mediation of bone mineral density on the association between IGF-1 and fracture. Given that sex-specific associations of IGF-1 with bone mineral density and fracture have been reported in observational studies (13), we additionally conducted sex-specific MR analyses.

Genetic instrument selection

Genetic instrument selection for serum IGF-1 levels was based on a GWAS including 358 072 individuals of European ancestry from the UK Biobank study (14). A total of 416 independent (defined by linkage disequilibrium, r² < 0.01) single-nucleotide variations (SNVs, formerly single-nucleotide polymorphisms [SNPs]) were selected at the genome-wide significant threshold (P < 5 × 10⁻⁸) as genetic instruments for IGF-1. These SNVs explained approximately 9.4% of the phenotypic variance. The average (range) baseline age of included participants in the UK Biobank was 56 years (37-73 years) and the mean ± SD of IGF-1 concentration is 21.4 nmol/L (5.7 nmol/L). The IGF-1 concentration in the first and ninth decile is 14.2 nmol/L and 28.4 nmol/L, respectively. The genetic instrument for IGF-1 has been used in previous MR studies (15, 16).

Outcome data sources

Summary-level data for estimated bone mineral density (eBMD) and fracture were derived from the GWAS in the UK Biobank (17). This eBMD, derived from heel quantitative ultrasound, was used to measure bone mineral density and the GWAS on this outcome was based on 426 824 individuals of European descent (17). Heel quantitative ultrasound method is mobile, inexpensive, easy to perform, and radiation free, and can measure bone mineral density to the same extent as dual-energy x-ray absorptiometry (18). The mean (± SD) levels of eBMD were 0.51 ± 0.11 g/cm² in women and 0.56 ± 0.12 g/cm² in men. The GWAS on fracture included 416 795 individuals of European ancestry (33 184 fracture cases and 373 611 noncases) (17). Fracture was defined by 2 mutually nonexclusive methods, which were hospital episodes statistics and questionnaire-based self-reported fracture. Certain fracture cases were excluded, including fractures of the skull, face, hands, and feet, pathological fractures due to malignancy, atypical femoral fractures, periprosthetic fractures, and healed fracture (17). Summary-level data for height as well as fracture in women and men were obtained from the second wave of analyses of the UK Biobank from Neale Lab (http://www.nealelab.is/uk-biobank) with 360 388 individuals for height, 193 291 individuals (20 242 cases and 173 049 noncases) for fracture in women and 165 950 individuals (14 538 cases and 151 412 noncases) for fracture in men.

Conclusion: The present study supports a role for IGF-1 in preventing fracture, possibly and partly mediated by greater bone mineral density.

Key Words: insulin-like growth factor-1, bone mineral density, fracture, causal inference, mendelian randomization
after excluding those of non-European ancestry, closely related individuals (or at least one of a related pair of individuals), individuals with sex chromosome aneuploidies, and individuals who had withdrawn consent. All genetic association estimates were adjusted for age, sex (where applicable), and genetic principal components.

Statistical analysis

Univariable and multivariable inverse-variance-weighted methods with random effects were employed as the main analyses. In the multivariable MR analysis, we adjusted for height because height might be a pleiotropic factor in the associations of genetically proxied IGF-1 levels with eBMD and fracture. In addition, we adjusted for eBMD in the analysis of fracture to assess whether any association of IGF-1 with fracture risk may be mediated through eBMD. Four complementary MR methods were used, including the inverse-variance-weighted method with fixed effects as well as the weighted median (19), MR-PRESSO (Mendelian Randomization Pleiotropy RESidual Sum and Outlier) (20), and contamination mixture (21) methods. The weighted median model can provide consistent estimates if more than 50% of the weight in the analysis comes from a valid instrumental variable (19). The MR-PRESSO test can detect possible outliers and provides estimates after removal of outliers, thereby correcting for horizontal pleiotropy (20). The distortion test in MR-PRESSO can distinguish the differences between estimates derived from models with and without outlier’s correction for fracture (20). The contamination mixture model provides a robust and efficient estimate when using hundreds of genetic instruments for an exposure (21). We also performed sensitivity analyses excluding SNVs associated with eBMD and fracture below the loci significance level ($P < 1 \times 10^{-5}$). The $I^2$ statistic was used to measure the heterogeneity across used SNVs (22). Possible pleiotropic effect was estimated by the intercept in MR-Egger regression model ($P$ for intercept < .05) (23). All statistical analyses were 2-sided and performed using the mrrobust package (24) in Stata/SE 15.0 and MR (25) package in R 3.6.0 software.

Results

Higher genetically predicted levels of IGF-1 were associated with greater eBMD (Fig. 1) and a decreased risk of fracture (Fig. 2). For 1-SD increase in IGF-1, the change of eBMD levels was 0.04 g/cm$^2$ (95% CI, 0.01-0.07; $P = .011$) and the odds ratio of fracture was 0.94 (95% CI, 0.91-0.98; $P = .003$). Results remained consistent in sensitivity analyses (see Figs. 1 and 2). There was significant heterogeneity among used SNVs in both analyses of eBMD ($I^2 = 92\%$) and fracture ($I^2 = 40\%$). We detected possible pleiotropy in the analysis of fracture (intercept = −.003; $P = .012$) but not in the analysis of eBMD (intercept = .002; $P = .056$). Four and eight possible SNV outliers were detected and removed in the MR-PRESSO analyses of fracture and eBMD, respectively. There was no difference between estimates derived from MR-PRESSO with and without outlier’s correction for fracture ($P = .515$), but a significant difference was detected for eBMD ($P = .002$). After outlier removal, the consequence was improved precision for the association between IGF-1 and eBMD. The associations of IGF-1 with eBMD (change 0.02; 95% CI, 0.00-0.03) and fracture risk (OR$_{SD}$ 0.97; 95% CI, 0.91-1.00) were observed in the contamination mixture model. In the analysis excluding SNVs associated with fracture (2 SNVs) and eBMD (57 SNVs) below the loci significance level, the association remained for fracture (OR$_{SD}$ 0.94, 95% CI, 0.91-0.98), but not for eBMD (change 0.006; 95% CI, −0.012-0.024) in the inverse-variance weighted model. The adjustment for height in the multivariable MR analysis did not alter the associations of IGF-1 levels with eBMD and fracture (see

![Figure 1](https://academic.oup.com/jcem/article-lookup/106/4/e1552) Association between genetically proxied increased insulin-like growth factor-1 (IGF-1) levels and estimated bone mineral density (eBMD). IVW, inverse-variance weighted; OR, odds ratio. The estimate of multivariable mendelian randomization analysis was adjusted for height.
After adjustment for eBMD, the association of IGF-1 with fracture was somewhat attenuated (odds ratio 0.96; 95% CI, 0.92-0.99; P = .012).

Genetically predicted high levels of IGF-1 were associated with fracture risk in both women and men (Table 1). The associations were directionally consistent in the weighted median model and persisted after adjustment for height and eBMD (women). The association with the adjustment for eBMD in men became borderline, albeit with almost identical magnitude in univariable MR analysis. We noticed moderate heterogeneity in the inverse-variance-weighted models, and the P for intercept in MR-Egger regression analysis was less than .05 for women, but no outlier was detected.

Discussion

This MR study found that genetically proxied higher serum IGF-1 levels were associated with a decreased fracture risk and possibly greater eBMD both in women and men. The associations remained after adjustment for height and eBMD expectedly partly mediated the association between IGF-1 and fracture.

Our findings were in line with some previous observational studies that found an inverse association between high levels of IGF-1 and lower risk of bone mineral density loss (3) and fracture (4, 5, 26). Our study additionally showed that the inverse associations were independent of height. The protective effect of IGF-1 on fracture risk was revealed to be partly independent of bone mineral density in a previous observational study (5), and our MR study corroborated this finding. With regard to bone mineral density, the null finding from sensitivity analysis excluding SNV-associated eBMD below the loci significance level might weaken the certainty of a causal association between higher IGF-1 levels and higher eBMD. However, this null finding could not rule out a potential effect of IGF-1 on bone mineral density for 2 reasons. First, whether it is biological plausible to exclude these SNVs deserves investigation given that certain SNVs might control IGF-1 and bone mineral density in a shared biological pathway. Second, the null findings might be caused by inadequate power. Thus, the association between IGF-1 and bone mineral density needs verification in more studies.

Sex-specific associations of IGF-1 with bone mineral density and fracture were revealed in observational studies.
The present study did not observe any sex difference in the association of genetically predicted high levels of IGF-1 with fracture risk. Even though the association between IGF-1 and fracture in men became borderline after adjusting for eBMD, the identical magnitude of the association compared to that in univariable MR analyses suggested that a larger $P$ value might be attributed to a reduced power due to a smaller number of cases when the analyses were confined to men.

Several underlying mechanisms explaining the protective role of IGF-1 against bone mineral density loss and fracture have been proposed (1). IGF-1 helps the proliferation of cells of osteoblastic lineage, enhances the function of the mature osteoblast, and plays a fundamental role in the stimulation of osteoblastic function and bone formation. In addition, IGF-1 can increase collagen synthesis and decrease its degradation, thereby maintaining appropriate levels of bone matrix and bone mass. IGF-1 may also influence osteoclasts in a manner protecting bone, but the evidence regarding this pathway is not clear. High IGF-1 might also increase and help maintain muscle mass, thereby reducing falls and fracture risk.

There are several strengths and limitations of the present study. The major strength was the MR design that reduced the bias derived from residual confounding and reverse causality, and therefore strengthened the causal inference in the association of IGF-1 with bone mineral density and fracture. In addition, the genetic instruments for IGF-1 had good validity and were used in previous MR studies, which guaranteed the robustness of our results. Our study was confined to individuals of European origin and adjustment was made for genetic principal components. Thus, population structure bias unlikely affected our results. However, the population specification might limit the generalizability of our findings to other populations. Another limitation is the used sources for the exposure and outcomes were from one sample. However, the set of genetic instruments for IGF-1 had a great strength with an $F$ statistic of greater than 10, thereby minimizing the possibility that the causal estimate would be biased in the direction of the observational association (27). The present study used more than 400 SNVs as instruments for IGF-1, which increased the possibility that our results were biased by invalid instruments. Nonetheless, the supportive results from the contamination mixture model along with consistent estimates from other sensitivity analyses diminished this possibility and validated our findings. The epigenetic phenomenon, such as methylation, and interaction between gene and environmental exposure might also influence the associations of IGF-1 with bone mineral density and fracture. However, we were unable to assess these effects in the present MR study. We estimated the association between IGF-1 and bone mineral density based on eBMD data, which was measured by the heel quantitative ultrasound method. This method was sensitive to the operator, homogeneous application of the ultrasound test gel for each patient, site of measurement, and environmental factors, in particular with regard to temperature even though a high consistency between SNVs identified for bone mineral density and eBMD was observed (21). Furthermore, we used genetic instruments for total IGF-1 levels rather than the free and bioavailable IGF-1 fraction, which might be more strongly associated with bone mineral density and fracture.

In conclusion, the present MR study supports a role for IGF-1 in the prevention of fracture possibly and partly through increased bone mineral density. Given that IGF-1 levels may be modified by protein intake (28-30), vitamin D supplementation (31), diet regimens (32), endurance exercises (33), and certain drugs, such as metformin (34) and dehydroepiandrosterone (35), our findings may strengthen several measures against bone mineral density loss and fracture. However, interventions to increase circulating IGF-1 levels need to be weighed against potential adverse health effects such as increased risk of type 2 diabetes and certain cancers (15, 16).

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Author Contributions: S.Y. and S.C.L. designed the study. S.Y. performed the statistical analyses, drafted the manuscript, and created the figures. S.Y., Z.H.W., S.L.C., K.M., and S.C.L. interpreted the data and edited the manuscript. All authors have given final approval of the version to be published.

Additional Information

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Disclosures: The authors have nothing to disclose.

Data Availability: All data used in the present study are based on publicly available summary statistics data provided by genetic consortia and are available on request to the corresponding author.

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