Serum Levels of Insulin-Like Growth Factor I and the Density, Volume, and Cross-Sectional Area of Cortical Bone in Children

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ABSTRACT

Insulin-like growth factor I (IGF-I) is a major regulator of bone growth during childhood. However, beyond knowledge that IGF-I influences longitudinal growth, its associations to changes in the cross-sectional dimensions, the volume, or the material density of bone during growth are unknown. We assessed the relationships between serum IGF-I and measurements of cross-sectional area, cortical bone area, and cortical bone density at the midshaft of the femur in 197 normal healthy white children and adolescents (103 boys and 94 girls; aged 7.8–18.2 yr). Bone determinations were obtained using computed tomography, and levels of IGF-I were measured by RIA after an extraction procedure. IGF-I correlated significantly with both cross-sectional area ($r = 0.49; P < 0.0001$) and cortical bone area ($r = 0.50; P < 0.0001$), but did not correlate with the material density of cortical bone ($r = −0.08$). Multiple regression analyses showed that circulating levels of IGF-I were associated with cross-sectional area ($P = 0.03$) and cortical bone area ($P = 0.04$) values, even after correcting for the confounding effects of age, gender, weight, and femoral length. We conclude that IGF-I is a major determinant of the cross-sectional properties of bone, but does not influence the material density of bone, in the appendicular skeleton. (J Clin Endocrinol Metab 84: 2780–2783, 1999)

Insulin-like growth factors (IGF) have been the focus of extensive investigation since they were discovered to stimulate cartilage sulfation approximately 2 decades ago (1). Considerable data are available regarding the role of insulin-like growth factor I (IGF-I) as a promoter of cell growth and differentiation (2). Studies have found IGF-I to be an important regulator of osteoblastic differentiation, proliferation, and collagenase activity, and consequently a major determinant of bone matrix apposition (3–8). In addition, it has been found that IGF-I stimulates the activation and formation of osteoclasts (9, 10), and that osteoblasts mediate IGF-I-stimulated formation of osteoclasts (11). IGF-I may thus modulate osteoblast-osteoclast interactions and serve as a coupling agent in the bone-remodeling cycle (12).

Compared to the comprehensive investigations on the role of IGF-I in regulating bone growth, information regarding its affects on bone mass and/or bone density is limited and conflicting. In elderly subjects with osteoporosis, levels of IGF-I are consistently lower than those in controls (13–15), but a clear relationship between IGF-I measurements and bone mineral density values has not been established; some investigators report an association (13, 16–20), whereas others do not (21, 22). Increases in skeletal and serum IGF-I levels during childhood parallel increases in bone mass during growth, suggesting a relationship between elevated IGF-I levels and bone gains (23–25). More recent studies in experimental animals and humans have suggested that IGF-I asserts its affect on bone mass acquisition mainly by increasing bone size. Rosen et al. demonstrated that rats treated with IGF-I had greater bone size but lower bone density than untreated animals (26). Similarly, studies by Bachrach and colleagues indicate that childhood onset of GH resistance, which is associated with a marked deficiency of IGF-I, results in smaller bones, but normal bone density (27). However, the structural basis for the relationship between increases in bone mass and serum IGF-I levels in healthy children has not yet been definitively established.

The recent adaptation of quantitative computed tomography (CT) to accurately measure the cross-sectional dimensions, volume, and density of bone in the appendicular skeleton has significantly improved our ability to noninvasively quantify the various structural components of bone mass. This study was designed to determine the possible association between variations in IGF-I levels and these skeletal parameters during childhood.

Subjects and Methods

Study subjects

The study subjects were 197 healthy white children and adolescents (103 boys and 94 girls) between the ages of 7.8–18.2 yr, who were recruited from schools of Los Angeles County. The investigational protocol was approved by the institutional review board for clinical investigations at Childrens Hospital Los Angeles, and informed consent was obtained from all subjects and/or their parents. Subjects were excluded if they had any chronic illness, if they had been ill for longer than 2 weeks during the previous 6 months, or if they...
had taken any medications, vitamin preparations, or calcium supplements within the previous 6 months. No subject had any physical impairment to limit their physical activity.

Candidates underwent a physical examination by a pediatrician, and measurements of total height and trunk height were obtained to the nearest 0.25 cm using the Harpenden stadiometer and the Harpenden Sitting Table (Holtain Ltd., Crymmych, Wales), respectively, and measurements of weight were obtained to the nearest 0.1 kg using the Scale-Tronix (Scale-Tronix, Inc., Wheaton, IL). Children in whom either height or weight was not within the 5th and 95th percentiles for the mean age-adjusted normal values were excluded from further evaluation (28). Thereafter, body surface area and body mass index were calculated, as previously described (29). The length of the femur was calculated as the distance between the greater trochanter and the distal lateral condyle.

**IGF-I measurements**

After an overnight fast, blood was drawn for determination of IGF-I. Levels of serum IGF-I were measured at Quest Diagnostics, Inc./Nichols Institute Diagnostics (San Juan Capistrano, CA) after acid-ethanol extraction to eliminate binding to serum proteins, using a RIA with rabbit anti-IGF-I and [125I]IGF-I. The assay was calibrated to the WHO First International Reference Preparation, 87/518. The bound-free separation was achieved using goat antirabbit γ-globulin and polyethylene glycol. Samples were stored at −70 C until assayed in batches of approximately 25–50. The sensitivity of the assay was 1 ng/mL; interassay variability was lower than 9.2%, and intraassay variability was lower than 7.9%.

**CT bone measurements**

All CT bone measurements were performed with the same scanner (model CT-T 9800, General Electric Co., Milwaukee, WI) and mineral reference phantom for simultaneous calibration (CT-T bone densitometry package, General Electric). The scanning site was located by physical examination, and measurements of cortical bone density, cross-sectional area, and cortical bone area were obtained from a single 1.5-mm thick imaging scan at the midportion of the distance between the greater trochanter and the lateral condyle using 120 kVp, 70 mA, and 2 s. The outer and inner boundaries of the cortex, representing the periosteal and endosteal surfaces of the bone, respectively, were identified at the place of the maximum slope of the femoral profile through the bone. The area within the outer cortical shell represented the femoral cross-sectional area, and the area between the outer and inner cortical shells represented the cortical bone area (30). For the purposes of this study, measurements of cortical bone area reflect the volume of bone, and the density of cortical bone is defined as the amount of bone per pixel (milligrams per cm³) at the midshaft of the femur. Because of the thickness and the relative lack of porosity of cortical bone in the femur, CT values reflect the material or true density of the bone (the amount of collagen and mineral in a given volume of bone) (30). These measurements are analogous to in vitro determinations of the intrinsic mineral density of bone, which are commonly expressed as the ash weight per unit volume of bone (31).

The coefficients of variation for repeated CT measurements of cross-sectional area, cortical bone area, and cortical bone density were calculated to be between 0.6–2.5%. The time required for the procedure was approximately 10 min, and the radiation exposure was approximately 100 mrem (1.0 mJ/kg) localized to 1.5 mm at the midportions of the femurs; the effective radiation dose was approximately 1 mrem (32).

**Statistical analysis**

The results are expressed as the mean (sd), unless otherwise stated. Normal distribution of the data was ascertained using the Shapiro-Wilk W test. Body mass index and IGF-I were not normally distributed; therefore, in all the analyses their log-transformed values were used. The relationships among age, anthropometric variables, IGF-I serum levels, and CT bone measurements were assessed using simple correlation coefficient, as for a null hypothesis $\rho = 0$. To test whether IGF-I exerted an independent effect on bone size measurements, multivariate analyses were performed. Gender, age, weight, and femur length were considered to be confounding variables, whereas IGF-I was the dependent variable, and femoral cortical bone area and cross-sectional area were the dependent variables. All tests were conducted at the $\alpha = 0.05$ level and were two tailed. The statistical software JMP IN (SAS Institute, Inc., Cary, NC) was used for the analyses.

**Results**

Table 1 summarizes the ages, anthropometric measurements, quantitative CT bone measurement values, and IGF-I serum levels in 197 children and adolescents.

The correlation coefficients for age, anthropometric measurements, and femoral CT measurements are shown in Table 2. The cross-sectional and the cortical bone areas at the midshaft of the femur correlated strongly with age, weight, height, femur length, trunk height, and body surface area. In contrast, the material density of cortical bone at this location showed no correlation with age or any anthropometric variable. IGF-I serum levels showed close correlations with age ($r = 0.39$), weight ($r = 0.48$), and other anthropometric measurements, including CT values for femoral cortical bone area ($r = 0.50$; $P < 0.0001$) and femoral cross-sectional area ($r = 0.49$; $P < 0.0001$). IGF-I serum levels did not correlate with the material density of bone (r = −0.08; Table 2).

The cross-sectional dimensions of the femur were used as dependent variables in multivariate analyses to ascertain whether the relationship observed between the cross-sectional properties and circulating IGF-I levels was independent of changes in age and anthropometry. The model was built to correct for differences in gender and age; we also accounted for the confounding effect of body size and included weight and femoral length in the model (Table 3). The model chosen explained 80% of the variability in femoral cortical bone area ($r = 0.89$; $P < 0.0001$) and 78% of the variability in femoral cross-sectional area ($R = 0.88$; $P < 0.0001$). IGF-I, therefore, exerts an effect on the cross-sectional properties of the femur that is independent of gender, age, and femoral length.

**Discussion**

The objective of this study was to examine the possible relationships between changes in serum IGF-I values and increases in the cross-sectional dimensions and the density of bone in the appendicular skeleton during growth. We found that in healthy white children there is a direct relationship between levels of IGF-I and the cross-sectional area and the cortical bone area of the femur, but no association with mea-

### Table 1. Age, anthropometric measurements, femoral bone measurements, and IGF-I serum levels in 197 healthy children

<table>
<thead>
<tr>
<th>Measurements</th>
<th>Mean (sd)</th>
<th>Range</th>
</tr>
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<tbody>
<tr>
<td>Age (yr)</td>
<td>13.00 (1.94)</td>
<td>7.85–18.18</td>
</tr>
<tr>
<td>Wt (kg)</td>
<td>49.3 (11.4)</td>
<td>21.5–81.3</td>
</tr>
<tr>
<td>Ht (cm)</td>
<td>155.0 (11.7)</td>
<td>119.1–180.8</td>
</tr>
<tr>
<td>Femur length (cm)</td>
<td>41.7 (3.9)</td>
<td>26.2–52.9</td>
</tr>
<tr>
<td>Trunk ht (cm)</td>
<td>89.6 (5.6)</td>
<td>63.5–94.0</td>
</tr>
<tr>
<td>SA (m²)</td>
<td>1.44 (0.21)</td>
<td>0.83–1.94</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>20.2 (2.9)</td>
<td>14.7–29.0</td>
</tr>
<tr>
<td>CBD (g/cm³)</td>
<td>1.98 (0.07)</td>
<td>1.71–2.16</td>
</tr>
<tr>
<td>CBA (cm²)</td>
<td>3.49 (0.71)</td>
<td>1.88–5.28</td>
</tr>
<tr>
<td>CSA (cm²)</td>
<td>4.54 (0.90)</td>
<td>2.40–7.57</td>
</tr>
<tr>
<td>IGF-I (ng/mL)</td>
<td>505.7 (163.5)</td>
<td>90.0–1098.0</td>
</tr>
</tbody>
</table>

SA, Surface area; BMI, body mass index; CBD, cortical bone density; CBA, cortical bone area; CSA, cross-sectional area.
measurements of femoral size

TABLE 3. Multiple regression models of IGF-I levels on healthy children

<table>
<thead>
<tr>
<th></th>
<th>Age</th>
<th>Wt</th>
<th>Ht</th>
<th>trunk ht</th>
<th>Femur length</th>
<th>SA</th>
<th>BMI</th>
<th>CBD</th>
<th>CBA</th>
<th>CSA</th>
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<tbody>
<tr>
<td>Femur length</td>
<td>0.64</td>
<td>0.75</td>
<td>0.76</td>
<td>0.64</td>
<td>0.64</td>
<td>0.66</td>
<td>0.66</td>
<td>0.34</td>
<td>0.33</td>
<td>0.39</td>
</tr>
<tr>
<td>SA</td>
<td>0.66</td>
<td>0.99</td>
<td>0.82</td>
<td>0.77</td>
<td>0.77</td>
<td>0.77</td>
<td>0.77</td>
<td>0.32</td>
<td>0.32</td>
<td>0.39</td>
</tr>
<tr>
<td>BMI</td>
<td>0.34</td>
<td>0.82</td>
<td>0.36</td>
<td>0.34</td>
<td>0.34</td>
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<td>0.34</td>
<td>0.34</td>
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</tr>
<tr>
<td>CBD</td>
<td>0.15</td>
<td>0.10</td>
<td>0.04</td>
<td>0.07</td>
<td>0.07</td>
<td>0.07</td>
<td>0.07</td>
<td>0.07</td>
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</tr>
<tr>
<td>CBA</td>
<td>0.71</td>
<td>0.84</td>
<td>0.85</td>
<td>0.83</td>
<td>0.78</td>
<td>0.78</td>
<td>0.78</td>
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<tr>
<td>CSA</td>
<td>0.66</td>
<td>0.83</td>
<td>0.85</td>
<td>0.79</td>
<td>0.79</td>
<td>0.79</td>
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<tr>
<td>IGF-I</td>
<td>0.39</td>
<td>0.48</td>
<td>0.59</td>
<td>0.56</td>
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CBD, Cortical bone density; CBA, cortical bone area; SA, cross-sectional area; SA, surface area; BMI, body mass index.

a P < 0.0001.
b P < 0.001.

In conclusion, the action of IGF-I at the cellular level and its association with bone mass have justified the interest in IGF-I as a bone formation-stimulating drug in the treatment of osteoporosis. The findings in the present study corroborate previous evidence of a positive effect of IGF-I on bone mass. Our results in children further indicate that, at least in the appendicular skeleton, IGF-I exerts its effect on bone mass by increasing the cross-sectional and cortical areas of the long bones.

References


