Age-Dependent Changes in Bone Mineral Density and Body Composition in Middle-aged and Elderly Women: Role of Growth Hormone, Insulin-like Growth Factor-I and Insulin-like Growth Factor-binding Protein-3

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Abstract The present study was performed to investigate age-dependent changes in bone mineral density (BMD) and body composition, and the role of growth hormone (GH), insulin-like growth factor-I (IGF-I) and IGF-binding protein-3 (IGFBP-3) in these changes in healthy middle-aged and elderly Japanese women. BMD and body composition [bone mineral content (BMC), lean body mass (LBM) and fat] were measured by dual energy X-ray absorptiometry. BMD, BMC and LBM declined with aging, while fat mass did not. Plasma IGF-I and IGFBP-3 levels also declined with aging. Urinary GH, plasma IGF-I and IGFBP-3 levels were positively correlated with BMD and BMC, even when BMD was adjusted for age and body size. The plasma IGF-I level was positively correlated with LBM. GH and IGFBP-3 levels also showed the tendency to be correlated with LBM. The urinary GH level was negatively correlated with fat mass, whereas plasma IGF-I and IGFBP-3 levels were positively correlated with fat mass. The GH-IGF-I-IGFBP-3 axis positively regulates bone and muscle mass, while GH and IGF-I-IGFBP-3 negatively and positively regulate fat mass, respectively.

Key words: GH, IGF-I, IGF-binding protein-3, bone mineral density, body composition

Introduction

Aging is associated with body compositional changes such as a decrease in bone mass. Growth hormone (GH) secretion as well as plasma levels of insulin-like growth factor-I (IGF-I), an anabolic factor for bone, and IGF-binding protein-3 (IGFBP-3), a modulator

of IGF action, also decrease with aging (1). Although there is evidence that GH plays an important role in the maintenance of bone mass in adulthood and participates in the process of bone remodeling partly via IGF-I (2-7), it is unclear how the GH-IGF-I-IGFBP-3 axis is involved in the age-dependent decrease in bone mass. On the other hand, the recent development of dual energy X-ray absorptiometry (DEXA) has made it possible to analyze body composition such as lean body mass (LBM) and fat mass easily and precisely

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(8). Our previous study indicated that DEXA is useful for the analysis of body composition at different ages and in both sexes (9). Although GH acts on bone, muscle and fat, it is unknown how the GH-IGF-I-IGFBP-3 axis is involved in age-dependent changes in body composition. The present study was performed to investigate age-dependent changes in bone mineral density (BMD) and body composition, and the role of GH, IGH-I and IGFBP-3 in these changes in healthy middle-aged and elderly Japanese women.

Subjects and Methods

Subjects

The subjects were 165 Japanese healthy women, aged 43 to 88 years, who visited our out-patient clinic for bone mass examination. They agreed to participate in this study and gave informed consent. None were known to have liver or renal dysfunction, nutritional disorders or metabolic diseases which might cause changes in body composition. All subjects were free of drugs known to influence bone and calcium metabolism.

Measurements of BMD and Body Composition

Total and lumbar BMD as well as bone mineral content (BMC), LBM and fat were measured by DEXA (QDR-2,000, Hologic). The percentage of BMC, LBM and fat was calculated by dividing each absolute value of body composition by total body mass.

Biochemical Measurements

GH concentration divided by the early morning urinary creatinine level was used as

an index of endogenous GH secretion. Urinary GH levels were measured by sandwich enzyme immunoassay (10, 11). Plasma IGF-I levels were measured by radioimmunoassay (RIA) after acid-ethanol extraction (12, 13). Plasma IGFBP-3 and 2 levels were also measured by RIA (14).

Statistical Analysis

Data were expressed as the mean $\pm$ SEM. Statistical analysis was performed using Student's t-test. p-Values less than 0.05 were considered significant. Regression analysis was performed using the computer program of Abacus Concepts, Statview (Abacus Concepts, Inc., Berkely, CA). Simple regression analysis was performed to assess the linear relationships between the study parameters, and Pearson's correlation coefficients were calculated.

Results and Discussion

Role of GH-IGF-I-IGFBP-3 Axis in Age-Dependent Bone Loss

Fig. 1 shows changes in total BMD with aging in middle-aged and elderly women. The total BMD clearly decreased with aging. Plasma levels of IGF-I and IGFBP-3 also decreased with age (Fig. 2), which is consistent with a previous report (1). In contrast, the plasma IGFBP-2 level increased with age. Since there is evidence that the plasma IGFBP-2 level increases under conditions where there is insufficient IGFBP-3 to bind the available IGFs (15), it may function as an additional IGF carrier, when IGFBP-3 level is inadequate.

We next examined whether the GH-IGF-I-

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IGFBP-3 axis was involved in an age-dependent decrease in BMD. As shown in Fig. 3, the GH, IGF-I and IGFBP-3 levels were all positively correlated with total BMD, while the plasma IGFBP-2 level was negatively correlated with total BMD. These findings indicate an important role for the GH-IGF-I-IGFBP-3 axis in the regulation of bone mass, which is consistent with a previous report (5). Since BMD and the GH-IGF-I-IGFBP-3 axis are influenced by aging and/or body size, we next examined the relationship between BMD and the GH-IGF-I-IGFBP-3 axis when the effects of aging and body size were excluded. As shown in Fig. 4, the Z score of BMD (standard deviation of average BMD at each age) was also positively correlated with plasma IGF-I and IGFBP-3 levels. BMD/height, the most convenient index of body size adjustment (16), was also still positively correlated with GH, IGF-I and IGFBP-3 levels (Fig. 5). BMD does not always reflect bone strength, although it is considered to account for about 75 to 85 % of the variance in ultimate strength of bone

Fig. 1 Changes in total BMD with aging.

Fig. 2 Relationship between age and plasma IGF-I or IGFBPs levels.

Fig. 3 Relationship between total BMD and urinary GH, plasma IGF-I or IGFBPs levels.

tissue (17). We also examined the relationship between the susceptibility to osteoporotic fractures and plasma IGF-I or IGFBP-3 levels. The subjects were classified by decade to exclude the influence of aging. As shown in Fig. 6, plasma levels of IGF-I and IGFBP-3 were significantly lower in subjects with spinal fractures, compared to those without fractures in any decade, while the plasma IGFBP-2 level was not significantly different between these two groups. These findings indicate that the GH-IGF-I-IGFBP-3 axis is important to maintain bone mass quantitatively as well as qualitatively, and that aging

negatively regulates the GH-IGF-I-IGFBP-3 axis, resulting in bone loss.

Role of GH-IGF-I-IGFBP-3 Axis in Age-Dependent Body Compositional Changes

We next analyzed body compositional changes with aging in middle-aged and elderly women. As shown in Fig. 7, BMC, % BMC and LBM declined with aging, while neither % LBM fat nor % fat significantly changed with aging. In order to clarify whether the GH-IGF-I-IGFBP-3 axis is involved in age-dependent body compositional changes, the relationship between body composition and GH, IGF-I and

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Fig. 4 Relationship between Z scores of lumbar spine BMD and plasma IGF-I or IGFBPs levels.

IGFBP-3 levels was examined. As shown in Fig. 8, the GH level was positively correlated with BMC, % BMC and % LBM, while the GH level was negatively correlated with fat and % fat. As shown in Fig. 9, the plasma IGF-I level was positively correlated with BMC, % BMC, LBM, fat and % fat, and negatively correlated with % LBM. As shown in Fig. 10, the plasma IGFBP-3 level was also positively correlated with BMC, % BMC, fat, and % fat, and negatively correlated with % LBM in the same manner as the plasma IGF-I level. The relationship between the plasma IGFBP-3 level and fat was higher, compared to that between the plasma IGF-I level and fat. These findings are summarized as follows: the GH-IGF-I-IGFBP-3 axis positively regulates bone and muscle mass; aging negatively regulates the GH-IGF-I-IGFBP-3 axis, resulting in bone and muscle loss; GH positively regulates bone and muscle mass in part via the IGF-I-IGFBP-3 pathway; GH positively regulates % LBM, while IGF-I-IGFBP-3 negatively regulate % LBM. These counterregulatory actions of GH and IGF-I-IGFBP-3 on % LBM might partly explain why aging affects LBM but not % LBM. From our data, it is not clearly why the IGF-I-IGFBP-3 system negatively affects % LBM, but one reason might be that IGF-I and IGFBP-3 (especially IGFBP-3) affect fat mass more than muscle mass. The GH level is negatively correlated with fat and % fat, which is consistent with previous reports suggesting a reciprocal relationship between adiposity and GH secretion (18, 19). In contrast, the plasma IGF-I and IGFBP-3 levels are positively correlated with fat and % fat. These data suggest that GH directly acts on fat not via the IGF-I-IGFBP-3 pathway, but that IGF-I-IGFBP-3

Fig. 5 Relationship between total BMD/height and urinary GH, plasma IGF-I or IGFFBPs levels.

regulates fat mass in a manner independently of GH. These counterregulatory actions of GH and IGF-I-IGFBP-3 on fat might partly explain why aging did not affect fat mass. Copeland, *et al.* reported that the negative correlation between plasma IGF-I level and adiposity existed in men but not in women (20). This controversy might be partly explained by sex and / or racial differences.

In conclusion, the present study demonstrated age-dependent changes in BMD and body composition in middle-aged and elderly Japanese women. GH and IGF-I-IGFBP-3 play an important role in age-dependent changes in BMD and body composition. The GH-IGF-I-

Fig. 6 Differences in plasma IGF-I and IGFFBPs levels in subjects with and without spinal fracture according to decade of age.

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Fig. 7 Changes in body composition with aging.

Fig. 8 Relationship between urinary GH level and body composition.

Fig. 9 Relationship between plasma IGF-I level and body composition.

Fig. 10 Relationship between plasma IGFBP-3 level and body composition.

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IGFBP-3 axis positively regulates bone and muscle mass. In contrast, GH negatively regulates fat mass, while IGF-I-IGFBP-3 positively regulates fat mass.

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