Different Mechanism of Bone Loss in Ageing Women and Men in Khon Kaen Province

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Abstract

The changes of vitamin D status and biochemical markers of bone turnover have been reported with aging. In this study we determined age-related levels of vitamin D and biochemical markers of bone turnover in the general adult population between the ages of 20 and 84 years who were living in Khon Kaen province in northeastern Thailand. Serum 25 hydroxyvitamin D was determined as an indicator of vitamin D status. Serum total alkaline phosphatase and N-terminal mid fragment osteocalcin were measured as biochemical markers of bone formation and serum C-terminal fragment of type I collagen was measured as a marker of bone resorption.

The levels of serum 25 hydroxyvitamin D were high in the Khon Kaen population. Men had higher levels of 25 hydroxyvitamin D than did women. However, there were no changes with age in either sex. In women, all biochemical markers of bone turnover increased with age after the fourth decade. The sharp increase was observed in the sixth decade which was around the menopausal age. In contrast, in men all biochemical markers of bone turnover except serum total alkaline phosphatase had a tendency to decrease with age.

Conclusion : There was no evidence of vitamin D deficiency in a Khon Kaen population. In addition, serum vitamin D levels did not decline with ageing. Women and men showed different changes of biochemical markers of bone turnover with ageing indicating gender difference in the pathogenesis of osteoporosis.

Key word : Vitamin D, Bone Markers, Bone Loss

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Bone mass at any point in life represents a balance between the amount of bone resorption and the amount of bone formation. When bone resorption exceeds formation, bone loss will occur. The reduction in bone mass could be a consequence of the changes which occur in both sexes with ageing although a variety of factors including genetic, nutrition, hormonal, lifestyle, vitamin D status and other variables are known to influence bone mass and bone loss[1,2].

Vitamin D is of paramount importance for bone health. It has been suggested that the decrease in vitamin D stores with aging is a significant contributory cause of age-related osteoporosis. The circulating level of 25 hydroxyvitamin D, the major storage form of vitamin D, has been considered to be a good biochemical indicator of vitamin D status[3] since it reflects vitamin D storage in the body[4,5]. Inadequate dietary intake of calcium and vitamin D may contribute to the high prevalence of bone loss [6]. Bone loss occurs in both men and women after peak bone mass is attained[7-9]. The rate of bone loss can be determined by assessing the biochemical markers of bone turnover which include markers of bone formation and resorption. A higher bone marker level is associated with a higher bone turnover rate[10,11].

Khon Kaen is a province which is situated in the northeastern part of Thailand. Factors known to contribute to bone mass or bone loss such as seasonal variation, lifestyle and nutritional habits of the population are different from the central part of Thailand. The pattern of changes in vitamin D status and bone turnover is necessary as basic knowledge for prevention of bone loss and osteoporosis in the elderly. However, this information is lacking. Thus, in the present study, we evaluated the circulating levels of 25 hydroxyvitamin D, biochemical markers of bone formation (total alkaline phosphatase, N-terminal mid fragment osteocalcin) and bone resorption (carboxy terminal telopeptide fragment of type I collagen) in a healthy Thai population who were living in Khon Kaen province. These values may be used as being representative of the rural population in northeastern Thailand and may provide clinically relevant information about skeletal remodeling for predicting future bone loss.

MATERIAL AND METHOD

Subjects

Subjects consisted of 251 rural adults living in two subdistricts (Nong Toom and Kok Si) of Muang district, Khon Kaen province. The participants were randomly selected from a list of members of each household and were invited to participate in the study. There were 125 women and 126 men, aged 20 - 84 years. Eighty-three per cent were farmers. The menopausal age in the females was 47.4 ± 4.7 (mean ± SD). All were in good health. The participants were excluded from the study if they had any chronic disease, or were taking a corticosteroid or calcium supplement, or were receiving hormone replacement therapy. None had signs or symptoms of metabolic or hormonal disease known to affect bone or calcium metabolism or had a history of bone fracture. The study was approved by the ethics committee of the Faculty of Medicine, Ramathibodi Hospital, Mahidol University, and informed consent was obtained from all study participants. The participants were stratified into 6 groups according to their age.

Laboratory assays

Blood samples were collected between 8.00 a.m. and 9.00 a.m. after an overnight fast. Sera were stored at -80°C before analysis. Serum 25 hydroxyvitamin D (25 OHD) was measured by radioimmunoassay (DiaSorin Inc., Stillwater, USA). Serum total alkaline phosphatase (AP) was measured by automated enzymatic method (Dimension R x L Clinical Chemistry System, Dade Behring Inc., USA). Serum N-terminal mid fragment osteocalcin (N-mid OC) and serum carboxy terminal telopeptide fragment of type I collagen (CTx) were measured by electrochemiluminescence immunoassay (Roche Diagnostics, Germany). The within-assay coefficient of variations (CVs) of serum 25 OHD, total AP, N-mid OC and CTx were, respectively, 15.8 per cent, 1.4 per cent 1.0 per cent, 3.4 per cent and of between-assay CVs were 14.4 per cent, 3.1 per cent 5.1 per cent, 4.1 per cent, respectively, at mean concentrations of 83.4 ng/ml, 79 U/L, 20.5 ng/ml, and 0.41 ng/ml.

Statistical analyses

All data are presented as mean ± SE. Comparisons between groups were analyzed by Student's unpaired *t*-test or Mann-Whitney U test. The statistical significance among age groups in the same sex was determined with one-way analysis of variance followed by Scheffe F test or with Kruskal-Wallis test. All analyses were performed using SPSS/PC release 10.0.

Table 1. Serum vitamin D and biochemical markers of bone turnover levels in women.

Age	n	25 OHD (ng/ml)	Total AP (U/L)	N-mid OC (ng/ml)	CTx (ng/ml)
20 - 29	21	42.41 ± 2.05	76.90 ± 3.79	17.88 ± 1.68	0.36 ± 0.04
30 - 39	21	37.51 ± 2.03	65.90 ± 3.47	12.77 ± 1.11	0.22 ± 0.02
40 - 49	21	40.48 ± 3.36	76.14 ± 4.47	16.17 ± 0.87	0.24 ± 0.01
50 - 59	21	38.12 ± 1.71	99.95 ± 4.89	22.45 ± 2.23	0.42 ± 0.05
60 - 69	20	40.83 ± 2.01	99.4 ± 5.14	26.47 ± 1.98	0.44 ± 0.03
70 - 84	21	47.26 ± 2.53	105.38 ± 4.27	31.66 ± 5.66	0.47 ± 0.05

Value are mean ± SE

Table 2. Serum vitamin D and biochemical markers of bone turnover levels in men.

Age	n	25 OHD (ng/ml)	Total AP (U/L)	N-mid OC (ng/ml)	CTx (ng/ml)
20 - 29	21	52.02 ± 2.78	92.52 ± 2.78	26.40 ± 2.38	0.52 ± 0.04
30 - 39	21	51.43 ± 3.06	80.86 ± 2.85	18.44 ± 1.13	0.40 ± 0.03
40 - 49	21	57.12 ± 3.17	91.19 ± 4.04	19.68 ± 2.69	0.41 ± 0.04
50 - 59	21	49.53 ± 3.34	93.43 ± 4.82	17.88 ± 1.08	0.33 ± 0.03
60 - 69	21	56.57 ± 3.65	96.33 ± 4.14	15.85 ± 1.32	0.30 ± 0.02
70 - 84	21	53.67 ± 3.31	101.29 ± 6.58	22.26 ± 3.24	0.39 ± 0.04

Value are mean ± SE

RESULTS

The levels of serum 25 OHD, total AP, N-mid OC and CTx of each age group in both sexes are presented in Table 1 and Table 2.

In women, serum 25 OHD levels decreased in the fourth decade. Thereafter, it showed a small change until the eighth and ninth decades (Fig. 1). None of these values differed significantly. Serum total AP levels were lowest in the fourth decade followed by an increase after the fifth decade. A marked increase of total AP was found in the sixth decade. A significant increase from the fourth decade of total AP was detected after the fifth decade. The levels of serum N-mid OC and CTx significantly decreased in the fourth decade and significantly increased thereafter. However, the increase of serum CTx in the fifth decade was not significant.

In men, serum 25 OHD level showed little change with age (Fig. 1). The lowest level was observed in the sixth decade, but it was not significantly different from the third decade. Serum total AP decreased in the fourth decade and gradually increased until the eight decade. However, these changes were not statistically different from the third decade. Serum N-mid OC and CTx levels decreased gradually from the third through seventh decade followed by an increase in the eighth decade. All decreases of serum N-mid OC were significantly different from the third decade, whereas, a significant decrease from the third decade in serum CTx was found only in the seventh decade.

Compared with women in the same age groups, men had higher levels of serum 25 OHD than women at all age groups but the higher levels of 25 OHD in men at the eighth decade did not significantly differ from women. The level of serum total AP, N-mid OC and CTx were significantly higher in men than in women until the fifth decade. After the fifth decade, they were lower than in women. However, serum N-mid OC and CTx levels were significantly lower than in women only in the seventh decade.

DISCUSSION

A number of previous studies have demonstrated changes of serum vitamin D with increasing age[12-15]. The majority of these studies have been conducted in countries where there is less sunlight and less pervasive food fortification. In addition, most elderly populations are more likely to be

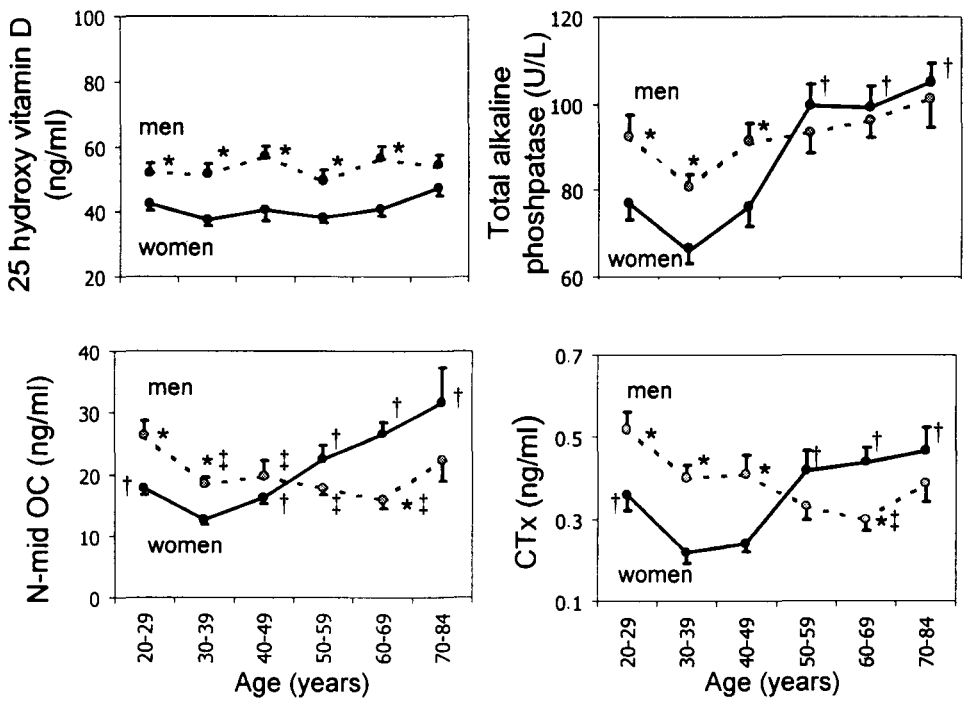

Fig. 1. Changes in serum 25 OHD, total AP, N-mid OC and CTx with age in women and men. Values are mean ± SE. * p < 0.05 *vs* women at the same age group, † p < 0.05 *vs* the fourth decade in women, ‡ p < 0.05 *vs* the third decade in men.

housebound and have low daily sunshine exposure (16). In contrast, the levels of serum 25 OHD in both sexes in the Khon Kaen population were higher than in many other reports(17-20). Moreover, serum 25 OHD levels were almost stable throughout life in both sexes. The discrepancy between our data and other results may be explained by the location of Khon Kaen which is nearer to the equator. In addition, the majority of the population in this study, including the elderly, had numerous outdoor activities working in the fields leading to adequate sunlight exposure. Our result was similar to other studies which also had greater sunlight(21,22). Furthermore, men had a higher level of serum 25 OHD than women. This may be because women in our country were more likely to be housebound than men because of being housewives. These data suggested that vitamin D deficiency in Khon Kaen is not a major risk factor of bone loss.

Regarding biochemical markers of bone turnover, both markers of bone formation and bone resorption have been reported to be high during childhood and decrease after puberty(11,23). Our results also demonstrated a decrease in biochemical markers of bone formation and bone resorption in the fourth decade. In women, all biochemical markers of bone turnover increased progressively with age after the fourth decade. A rapid increase in serum total AP and CTx was found in the sixth decade which is around the menopausal age as has been observed in other studies(24-28). The rapid increase in the levels of biochemical markers of bone turnover after the onset of menopause indicated a rapid rate of bone remodeling activity in these subjects. The increased levels of biochemical markers of bone turnover may increase fracture risk independent of current BMD(29-32). In addition, the increase of all biochemical markers of bone turnover continued with ageing although progressively attenuated after menopause. This finding was similar to the report by Garnero et al(26) who found that both bone formation and bone resorption rate con-

tinued to increase in elderly women even after 75 years of age.

Wishart et al[33] reported a decrease in bone formation and bone resorption in men with age. Our results showed that serum N mid OC and CTx in men had a tendency to decrease with age after the fourth decade, whereas, AP still increased moderately. A major factor of bone loss in men has been reported to be reduction in osteoblastic function with age[34-37]. However, the observation that serum total AP level in men did not decrease with age may probably be explained by a rise in the liver fraction of AP with age[38]. Serum total AP level is generally considered non-bone specific because many other organs contribute to its level. Among the several tissues containing alkaline phosphatase, the liver and bone isoenzymes are the major contributors to serum levels, each accounting for about half of the total[39,40]. Thus, total AP can be a good bone marker if there is no increase of hepatic isoenzyme. However, all markers in this study slightly increased in the eighth decade. This may be caused by the lower mobility in these elderly men [41]. In addition, men had higher levels of biochemical markers of bone turnover than women. This may be due to greater bone mass in men[42]. Because of the absence of menopause-equivalent with associated acceleration of bone loss in men, after the fifth decade all of these markers in men were lower than in women. These results confirmed that elderly women had a higher bone turnover rate than men and bone turnover in men tended to decline with ageing.

This study confirmed that, as in Bangkok [22], vitamin D deficiency is not a problem in Khon Kaen. It also underscored different mechanisms of bone loss in ageing men and women.

SUMMARY

Serum 25 OHD levels were high in the Khon Kaen population. There were no changes of 25 OHD with age in either sex. However, women and men showed different age-related changes in bone metabolism. The levels of biochemical markers of bone turnover in women tended to increase with age and had a high bone turnover rate at the onset of menopause. In contrast, bone turnover in men tended to decline with age.

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ความแตกต่างของกลไกการสูญเสียมวลกระดูกที่เกี่ยวข้องกับอายุในเพศหญิงและเพศชายในจังหวัดขอนแก่น

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จากการที่มีรายงานในต่างประเทศและในภาคกลางของประเทศไทยว่าระดับวิตามินดีและดัชนีทางชีวเคมีที่บ่งถึงการสร้างและสลายมวลกระดูกมีการเปลี่ยนแปลงตามอายุ คณะผู้วิจัยจึงทำการศึกษาถึงการเปลี่ยนแปลงระดับวิตามินดีและดัชนีที่บ่งถึงการสร้างและสลายมวลกระดูกกับอายุระหว่าง 20 ถึง 84 ปี ในประชากรที่อาศัยอยู่ในจังหวัดขอนแก่นซึ่งตั้งอยู่ทางภาคตะวันออกเฉียงเหนือของประเทศไทย ดัชนีที่ใช้ในการศึกษาครั้งนี้ได้แก่ เซรั่ม 25 hydroxyvitamin D ซึ่งเป็นตัวบ่งถึงระดับวิตามินดีที่มีอยู่ในร่างกาย เซรั่ม total alkaline phosphatase และ N terminal mid-fregment osteocalcin ซึ่งเป็นดัชนีของการสร้างมวลกระดูก และ C-terminal fragment of type I collagen ซึ่งเป็นดัชนีของการสลายมวลกระดูก

การศึกษาพบว่าระดับ 25 hydroxyvitamin D ของประชากรในจังหวัดขอนแก่นไม่มีการเปลี่ยนแปลงตามอายุทั้งในเพศชายและเพศหญิง เพศชายมีระดับ 25 hydroxyvitamin D สูงกว่าเพศหญิง ในเพศหญิงเมื่อมีอายุมากกว่า 30 ปี ดัชนีทางชีวเคมีที่บ่งถึงการสร้างและสลายมวลกระดูกมีระดับเพิ่มขึ้นตามอายุ ดัชนีเหล่านี้มีการเพิ่มขึ้นอย่างรวดเร็วเมื่อเข้าสู่วัยหมดประจำเดือน ในทางตรงกันข้าม ในเพศชายดัชนีทางชีวเคมีที่บ่งถึงการสร้างและสลายมวลกระดูกมีระดับลดลงตามอายุ ยกเว้นเซรั่ม total alkaline phosphatase ที่มีระดับสูงขึ้นตามอายุ

สรุปได้ว่าประชากรชาวขอนแก่นไม่ขาดวิตามินดี และระดับวิตามินดีไม่ลดลงตามอายุ เพศหญิงและเพศชายมีการเปลี่ยนแปลงของดัชนีทางชีวเคมีที่บ่งถึงการสร้างและการสลายมวลกระดูกตามอายุต่างกัน แสดงว่ากลไกในการสูญเสียมวลกระดูกในเพศชายและในเพศหญิงมีความแตกต่างกัน

คำสำคัญ : วิตามินดี, ดัชนีของกระดูก, การสูญเสียมวลกระดูก

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