Sequential Changes of Bone Metabolism in Normal and Delayed Union of the Spine

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Abstract Time-dependent changes in bone markers in delayed or nonunion of vertebral fracture were compared with those of normal union. Thirty-three patients with a fresh vertebral fracture were enrolled. Urinary Type I collagen C-terminal telopeptide, pyridinoline, deoxypyridinoline, serum C-terminal telopeptide, and N-midportion of osteocalcin (OC_{N-mid}) were determined at the time of hospital admission (within 24 hours after the fracture event in all cases) and at 2, 4, 12, 24, and 48 weeks thereafter. Subjects were divided into two groups according to the results of MR images taken 48 weeks after fracture. Twenty-four were normally united (Group N) and nine had delayed or nonunion (Group D) of the spine. No differences between values of bone resorption markers in Group N and Group D were observed at any time. Serum OC_{N-mid} in Group N started to increase at 2 weeks and reached the peak value at 24 weeks (180%); however, serum OC_{N-mid} in Group D increased at most 120% from baseline to 4 weeks. Values of serum OC_{N-mid} in Group N were higher

at 24 and 48 weeks than those in Group D. Impairment of fracture healing was strongly associated with a deficit in the increase of osteocalcin in the later stage of fracture repair.

Level of Evidence: Level II, prognostic study. See the Guidelines for Authors for a complete description of levels of evidence.

Introduction

The number of patients with osteoporosis is increasing with the demographic trend toward an aging population. Osteoporosis is characterized by reduced bone quantity and quality, which can lead to hip fracture or vertebral fracture after a simple fall. A fractured vertebra sometimes can result in delayed or nonunion despite appropriate conservative treatment. Delayed union or nonunion plays a role in the development of prolonged back pain and even neurologic deficit depending on the degree of vertebral collapse with spinal cord or cauda equina compression.

Almost all vertebral compression fractures can be identified on plain radiographs; however, subtle vertebral deformity caused by fracture or fine fracture lines sometimes can be missed on plain radiographs. Plain radiography is able to show the degree of wedge or concave deformity, but it is difficult to identify fine fracture lines in a fractured vertebral body resulting from osteopenia or sometimes fracture lines may be hidden by colonic gas. Moreover, plain radiography cannot differentiate a fresh vertebral fracture from an old one. Magnetic resonance imaging can accurately evaluate bone marrow in the vertebrae and can discriminate a fresh fracture from an old one by the changes in signal intensity in the fractured

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vertebrae. Also, MRI can assess details of the healing process of fractured vertebra more easily than plain radiography [28]. Magnetic resonance imaging is the gold standard for diagnosis of delayed or nonunion of fractured vertebrae because it can detect not only the shape of the vertebra but also quality inside the vertebra.

To date, changes in bone metabolism after fracture have been well examined. Bone resorption and formation markers change dramatically after fracture of long bones because of the fracture healing and repair process [2, 12–14, 21]. Bone resorption markers increased soon after a fracture event and bone formation markers increased gradually thereafter. Also, changes in bone resorption and bone formation markers for patients with nonunion of long bones have been reported [8, 15, 16, 19, 20, 22]. However, little is known regarding the changes in bone metabolism in the case of delayed or nonunion of the spine. Numerous antiosteoporosis drugs are available. However, it is controversial whether antiosteoporosis agents are useful for delayed or nonunion of vertebral compression fracture because details of bone metabolism in patients with delayed or nonunion of the spine are not clear. If alterations in bone metabolism are associated with the development of delayed or nonunion of the spine, a better understanding of the pathophysiology may help guide pharmaceutical interventions in patients with delayed or nonunion of the spine to facilitate fracture healing.

In this study, we investigated time-dependent changes in bone resorption and bone formation markers and bone mineral densities (BMDs) for patients with fresh single spinal compression fractures during a 48-week period. The primary purposes of this study were to elucidate (1) how often vertebral compression fractures are missed using only radiographs, (2) how biochemical markers and BMDs change after a single vertebral fracture, and (3) whether levels of biochemical parameters are affected by bed rest after a single vertebral fracture. The secondary research question of this study is whether time-dependent changes in metabolic bone markers and BMDs in cases of delayed or nonunion of vertebral fractures, as determined by MRI, are different from fractures that progress to normal union. These differences may help clarify the pathologic bone metabolism that leads to delayed union or nonunion of osteoporotic vertebral fractures.

Materials and Methods

From October 2000 to June 2002, 58 patients with fresh single vertebral compression fractures were admitted to our hospital within 24 hours after fracture. Among these, 25 patients did not return for followup after the first or second

visit to the hospital. Therefore, 33 patients with a fresh vertebral compression fracture were considered for this study. All subjects sustained a vertebral compression fracture resulting from a simple fall from a standing height or less. All patients were treated conservatively because no neurologic deficits of the lower extremities and no bladder or bowel dysfunction were involved at the time of the injury or during the study. Patients had anteroposterior and lateral plain radiographs of the lumbar or thoracic spine, or both, and MRI of the affected vertebra. After bed rest for 0 to 3 weeks to alleviate pain, a flexible corset was fitted and patients were allowed to walk. The corset was worn thereafter for approximately 3 months depending on each patient's symptoms.

Nonsteroidal antiinflammatory drugs were administered orally to all patients until spontaneous back pain disappeared. No pharmacotherapeutic intervention for osteoporosis such as vitamin D or bisphosphonate was administered during the study. Patients were excluded if they met any of the following criteria at least 2 years before the fracture: (1) they were admitted with a pathologic fracture; (2) they sustained a previous fracture; (3) they had a thyroid disorder; (4) they abused alcohol; (5) they had taken supplements of calcium, vitamin D, bisphosphonate, vitamin K, thiazide, or glucocorticoids; (6) they had osteomalacia after gastrectomy; (7) they had undergone orchidectomy; (8) they were undergoing hormone replacement therapy for prostate disease; (9) they were unable to walk; or (10) they had pseudarthrosis of the fracture site. Informed consent was obtained from each patient at the outpatient clinic. Blood and urine samples taken between 9:00 AM and 11:00 AM were obtained at the time of admission. Thereafter, samples were obtained 2, 4, 12, 24, and 48 weeks after admission. Samples were stored at -30°C until analysis. Bone mineral density of two regions of the left proximal femur (femoral neck and trochanter) and two regions of the left forearm (distal one-third and ultradistal radius) were measured at admission and 24 and 48 weeks after admission. Magnetic resonance imaging was performed with an MR scanner (GEYMS MRvector Fast 0.5-Tesla system; GE Yokogawa Medical, Tokyo, Japan). Sagittal T1-weighted spin-echo (TE 20 ms/TR 500 ms) and sagittal and axial T2-weighted spin-echo (TE 100 ms/TR 3000 ms) sequences were performed. Section parameters were 5 mm thick with a 2-mm interslice gap; a 160×256 acquisition matrix was used for sagittal T1 images, 256×224 for sagittal T2 images, and 224×256 for axial images; and a 300-mm field of view was used for sagittal images and 200 mm for axial images. Morphometry was performed on lateral radiographs of the spine defining the anterior (ha), posterior (hp), and middle (hm) vertebral heights [9]. A vertebral compression fracture was defined using ratios of vertebral heights: anteroposterior ratio (APR) = ha/hp ; middle-posterior ratio (MPR) = hm/hp ;

and posterior-posterior (adjacent vertebra) ratio (HPR) = hp/hp' . A vertebral fracture was defined by a reduction of greater than 20% in APR, MPR, or HPR. If a fracture was not clear on plain radiography, although a vertebral fracture was strongly suspected from the patient's symptoms, it then was defined by MRI. All MR images were viewed by two individuals (TO, DS) who were blinded to the results. A fracture was considered healed when normointensity was seen in the previously fractured vertebra on T1- and T2-weighted MR images taken 48 weeks after admission. Delayed or nonunion of the fractured vertebra was defined by signal intensity inside the affected vertebral body from an MR image taken 48 weeks after admission according to Hasegawa et al. [10]. Delayed or nonunion was judged if a fluid-containing space on T2-weighted images or a lineal low-intensity area on T2-weighted images was visible on MRI at 48 weeks [3, 18].

For monitoring biochemical changes, we measured urinary and serum Type I collagen C-terminal telopeptide (CTX) and urinary pyridinoline (PYR) and deoxypyridinoline (DPD) as the bone resorption markers and serum N-midportion of osteocalcin (OC_{N-mid}) as the bone formation marker. Urinary CTX was measured by a CrossLapsTM enzyme-linked immunosorbent assay (ELISA) kit (Nordic Bioscience Diagnostics, Herlev, Denmark) according to Bonde et al. [5]. The CTX is based on an immobilized synthetic peptide with an amino acid sequence specific to part of the C-telopeptide of the αI chain of Type I collagen (Glu-Lys-Ala-His-Asp-Gly-Arg = CrossLapsTM peptide). The intraassay and interassay coefficients of variation were less than 6% and 8%, respectively.

Serum CTX was measured by ELISA according to Bonde et al. [4]. The assay developed for measurement of Type I collagen degradation products in sera is based on an immobilized synthetic peptide with an isomerized amino acid sequence specific for part of the C-terminal peptide of the αI chain of Type I collagen (EKAHD- β -GGR). The intraassay and interassay coefficients of variation were 6.7% and 9.2%, respectively.

Urinary PYR and DPD were measured by high-performance liquid chromatography after acid hydrolysis using a Gilson ASPEC (automated sample preparation with extraction column; Gilson Inc, Middleton, WI) according to Pratt et al. [23]. In our protocol, the intraassay and interassay coefficients of variation were 6.4% and 5.9% for PYR and 6.0% and 6.0% for DPD, respectively. Values of urinary CTX, PYR, and DPD were corrected for urinary creatinine concentration, which was measured on an autoanalyzer.

The serum concentration of osteocalcin (OC_{N-mid}) was measured by ELISA (two-site N-MIDTM Osteocalcin; Osteometer BioTech A/S, Ballerup, Denmark) [24]. The intraassay and interassay coefficients of variation were less

than 4.2% and 4.0%, respectively, and the detection limit was 2.0 $\mu g/L$.

Bone mineral density in grams per square centimeter was determined for two regions of the left proximal femur (femoral neck and trochanter) and two regions of the left forearm (distal one-third and ultradistal radius) using dual-energy x-ray absorptiometry from an Expert 5000 instrument (Lunar, Madison, WI). The precision of the integral calcium hydroxyapatite standard was less than 0.4% with time.

Data were analyzed using a Statview 5.0 (SAS Institute Inc, Cary, NC) program on a Macintosh computer (Apple, Inc, Cupertino, CA). Longitudinal data were presented as percentage changes from the baseline to allow for relative comparison. Two-way analysis of variance followed by the Scheffe-F test for two related samples (longitudinal changes of biochemical markers and BMDs) were used to evaluate differences. Spearman's correlation coefficient by rank was used for correlation between the period of bed rest and values of biochemical markers. Differences in the baseline data, including characteristics of the patients, BMDs of the proximal femur and forearm, and biochemical parameters between the two groups, were analyzed using a nonparametric test followed by a Mann-Whitney U test. Analysis of variance followed by the Scheffe-F test was used for comparison of the biochemical and BMD data between groups. If differences were observed between groups by analysis of variance, a nonparametric test followed by a Mann-Whitney U test was used for comparison of the biochemical data at each time between groups. All values except biochemical parameters were expressed as mean $\pm$ standard deviation. Values of biochemical parameters were expressed as mean $\pm$ standard error. Statistical significance was defined as a p value less than 0.05.

Results

According to the morphometric analysis of plain radiography, only one vertebral fracture among all fractures was not identified by radiography. This particular vertebra (that of an 86-year-old woman) was not deformed, but abnormal signal intensity was seen on MRI.

All patients in this series with acute compression fractures had an increase in some of the measured bone resorption markers (urinary and serum CTX, urinary PYR, and urinary DPD) 2 weeks after admission with peak values at 2 to 4 weeks. These started to decrease from 4 to 12 weeks (Figs. 1, 2). Twenty-four weeks after admission, the measured levels of these bone resorption markers essentially had returned to the initial values measured on admission. Serum OC_{N-mid} started to increase 2 weeks after admission and continued to increase up to 24 weeks

Fig. 1A–B Percentage changes from the baseline values of (A) urinary and (B) serum Type I collagen C-terminal telopeptide for all subjects are shown. The data are shown as mean $\pm$ standard error. Urinary and serum C-terminal telopeptide increased at 2 and 4 weeks after vertebral fracture compared with the baseline values. ** = $p < 0.01$ compared with the baseline values.

(Fig. 3). Values of serum OC_{N-mid} at 48 weeks were still higher than the initial values ($p = 0.002$ at 48 weeks). Percentage changes in femoral neck and trochanter BMDs at 24 and 48 weeks after admission were lower than the initial BMDs (Table 1). However, the distal one-third and ultradistal radius BMDs did not change from the baseline values.

The period of bed rest correlated with percentage changes in urinary CTx ($r = 0.499$, $p = 0.035$), serum CTx ($r = 0.520$, $p = 0.014$), urinary PYR ($r = 0.725$, $p = 0.001$), and urinary DPD ($r = 0.676$, $p = 0.003$) at 4 weeks and with percentage changes in urinary PYR ($r = 0.558$, $p = 0.043$) at 12 weeks. However, no correlation was observed between the period of bed rest and percentage changes in serum OC_{N-mid} at any time.

Of the 33 vertebral fractures studied, 24 were united normally (Group N) (Fig. 4A) and nine failed to heal (Group D) (Fig. 4B–C) within the 48-week study period judged by MRI taken 48 weeks after admission. Of nine vertebrae in Group D, three collapsed vertebrae had localized low signal intensity on a T1-weighted image and high signal intensity on a T2-weighted image of fluid collection in the vertebra, which meant nonunion (Fig. 4B). The remaining six vertebrae had lineal low-intensity signal on

Fig. 2A–B Percentage changes from the baseline values of (A) urinary pyridinoline and (B) deoxypyridinoline for all subjects are shown. The data are shown as mean $\pm$ standard error. Urinary pyridinoline increased at 2, 4, and 12 weeks and deoxypyridinoline increased at 2 and 4 weeks after vertebral fracture compared with baseline values. * = $p < 0.05$ and ** = $p < 0.01$ compared with baseline values.

Fig. 3 Percentage changes from the baseline values of serum levels of OC_{N-mid} for all subjects are shown. The data are shown as mean $\pm$ standard error. Serum OC_{N-mid} increased at 4, 12, 24, and 48 weeks after vertebral fracture compared with the baseline values. * = $p < 0.05$ and ** = $p < 0.01$ compared with the baseline values.

T2-weighted images that meant delayed union (Fig. 4C). There were no differences in the anthropometric indices, including age, height, weight, body mass index, period of

Table 1. Percentage changes in bone mineral density from baseline for all subjects

Period after fracture	Femoral neck	Trochanter	Distal one-third radius	Ultradistal radius
24 weeks	$-2.98 \pm 1.12^\dagger$	$-4.43 \pm 1.62^\dagger$	-0.20 ± 0.89	$+2.61 \pm 2.42$
48 weeks	$-5.03 \pm 1.42^\dagger$	$-4.66 \pm 1.96^*$	-1.21 ± 1.14	$+1.59 \pm 2.39$

Data are shown as mean $\pm$ standard error (%); * $p < 0.05$ versus baseline values; $^\dagger p < 0.01$ versus baseline values.

Fig. 4A–C T2-weighted MR images of the fractured vertebra 48 weeks after fracture in (A) the normally united group (Group N) and (B, C) the nonunion or delayed union group (Group D) are

shown. (B) The arrowhead indicates fluid collection in the vertebra, which means nonunion. (C) The arrow indicates lineal low-intensity signal, which means delayed union.

bed rest, BMDs of the proximal femur and forearm, and biochemical data between groups (Tables 2, 3). Changes in bone resorption markers (urinary and serum CTx, urinary PYR, and urinary DPD) in each group were the same in all subjects (Figs. 5, 6). No differences between values in Group N and Group D were observed at any time. Serum OC_{N-mid} in Group N started to increase at 2 weeks and continued to increase with the peak value at 24 weeks (180%) (Fig. 7). However, serum OC_{N-mid} in Group D increased at most 120% from the baseline values at 4 weeks after admission. Values of serum OC_{N-mid} in Group N were higher at 24 and 48 weeks than those in Group D ($p = 0.049$ at 24 weeks and $p = 0.009$ at 48 weeks). Femoral neck and

trochanter BMDs at 24 and 48 weeks after admission decreased compared with the baseline values in Group N ($p = 0.026$ at 24 and 48 weeks for neck BMD; $p = 0.005$ at 24 weeks and $p = 0.019$ at 48 weeks for trochanter BMD) (Fig. 8). However, forearm BMD did not change in Group N (Fig. 9). Bone mineral densities of all regions, including the proximal femur and forearm in Group D, did not change compared with the baseline values (Figs. 8, 9). No differences in BMDs of the two regions of the proximal femur and two regions of the forearm were observed between groups at 24 and 48 weeks after admission. Back pain in eight of 24 patients (33%) in Group N and seven of nine patients (78%) in Group D remained at the end of the study.

Table 2. Anthropometric and bone mineral density data for all subjects and each group

Data	All subjects	Group N	Group D
Number (male/female)	33 (8/25)	24 (5/19)	9 (3/6)
Age (years)	71.8 ± 11.6	72.3 ± 9.8	70.2 ± 16.1
Height (cm)	149.9 ± 7.5	150.7 ± 7.3	147.5 ± 8.0
Weight (kg)	48.7 ± 9.5	48.3 ± 8.8	49.8 ± 12.0
Body mass index (kg/m^2)	21.8 ± 4.1	21.4 ± 4.2	22.8 ± 3.8
Bed rest (days)	9.5 ± 9.5	9.5 ± 8.8	9.6 ± 11.6
Femoral neck BMD (g/cm^2)	0.623 ± 0.122	0.606 ± 0.127	0.673 ± 0.097
Trochanteric BMD (g/cm^2)	0.517 ± 0.130	0.506 ± 0.127	0.549 ± 0.140
Distal one-third radius BMD (g/cm^2)	0.463 ± 0.110	0.449 ± 0.106	0.502 ± 0.121
Ultradistal radius BMD (g/cm^2)	0.229 ± 0.075	0.226 ± 0.076	0.239 ± 0.075

Data are shown as mean $\pm$ standard deviation; BMD = bone mineral density.

Table 3. Biochemical data at baseline for all subjects and each group

Biochemical marker	All subjects	Group N	Group D
Urinary CTx ($\mu\text{g}/\text{mmol creatinine}$)	332.8 ± 86.2	382.4 ± 86.2	206.0 ± 30.0
Serum CTx (pmol/L)	4350 ± 612	4663 ± 811	3551 ± 651
Urinary PYR ($\text{nmol}/\text{mmol creatinine}$)	37.7 ± 5.0	39.7 ± 6.8	32.6 ± 4.3
Urinary DPD ($\text{nmol}/\text{mmol creatinine}$)	7.6 ± 1.1	8.2 ± 1.4	5.8 ± 0.9
Serum OC _{N-mid} (ng/mL)	13.1 ± 1.0	13.3 ± 1.2	12.7 ± 2.1

Data are shown as mean $\pm$ standard error; CTx = Type I collagen C-terminal telopeptide; PYR = pyridinoline; DPD = deoxypyridinoline; OC_{N-mid} = N-midportion of osteocalcin.

Discussion

Only one fracture was missed on plain radiography according to the morphometric criteria used in this study. Results suggest these morphometric criteria for plain radiographs were mostly successful in the diagnosis of vertebral compression fractures. Signal intensities on MRI in fractured vertebrae vary depending on the period after fracture events. All fractures less than 3 months old showed hypointensity on T1-weighted imaging and hyperintensity on T2-weighted imaging [28]. Abnormal intensities did not return to normal intensities until at least 7 months after vertebral fracture [27]. Therefore, a lineal low-intensity line on a T2-weighted image at 48 weeks

after fracture or fluid-containing space on MRI was considered a delayed union or nonunion of the vertebrae in this study. However, one of the limitations of this study is intraobserver or interobserver variations of radiographic measurements, including bone morphometry on plain radiography and MRI, were not performed.

To date, long-term changes in bone resorption and formation markers in subjects with vertebral compression fractures have not been reported. In our study, bone resorption markers started to increase 2 weeks after vertebral fracture, with peak values at 2 to 4 weeks, and started to decrease from 4 to 12 weeks. Serum OC_{N-mid}

Fig. 5A–B Percentage changes from the baseline values of (A) urinary C-terminal telopeptide and (B) serum C-terminal telopeptide in Groups N and D are shown. The data are shown as mean $\pm$ standard error. No differences were observed between the groups.

Fig. 6A–B Percentage changes from the baseline values of (A) urinary pyridinoline and (B) deoxypyridinoline in Groups N and D are shown. The data are shown as mean $\pm$ standard error. No differences were observed between the groups.

Fig. 7 Percentage changes from the baseline values of serum levels of OC_{N-mid} in Groups N and D are shown. The data are shown as mean $\pm$ standard error. Serum OC_{N-mid} at 24 and 48 weeks after vertebral fracture was higher in Group N than in Group D. * = $p < 0.05$ and ** = $p < 0.01$ compared with Group D.

Fig. 8A–B Percentage changes from the baseline values in the BMD of (A) the femoral neck and (B) the trochanter in Groups N and D are shown. The data are shown as the mean $\pm$ standard error. Bone mineral density of the femoral neck and trochanter at 24 and 48 weeks after vertebral fracture in Group N decreased compared with the baseline values. * = $p < 0.05$ and ** = $p < 0.01$ compared with the baseline values.

started to increase 2 weeks after fracture, and values at 48 weeks were still higher than the initial values. This pattern of changes in bone resorption and formation

Fig. 9A–B Percentage changes from the baseline values in the BMD of (A) the distal one-third radius and (B) the ultradistal radius in Groups N and D are shown. The data are shown as the mean $\pm$ standard error. No differences were observed compared with the baseline values in each group.

markers was mostly consistent in the case of hip fracture [21].

Comparing normal union with delayed or nonunion of the spine, bone resorption markers did not differ between Group N and Group D. However, the bone formation marker in Group D did not increase up to the level in Group N throughout the study. Changes in bone resorption and formation markers in the case of nonunion of long bones have been reported [8, 15, 16, 19, 20, 22]. Levels of bone resorption markers in the delayed union group did not differ from those in the normally united group for patients with tibial shaft fractures. However, levels of bone formation markers, including osteocalcin, Type I collagen C-terminal propeptide, and bone-specific alkaline phosphatase, were lower in the delayed union group than in the normally united group [16, 20]. These results in previous studies were in good agreement with our results. However, among bone formation markers, a pattern of sequential changes of de novo Type I collagen products (Type I collagen C-terminal propeptide) or osteocalcin for patients with tibial shaft fractures differs from that of bone-specific alkaline phosphatase. Low levels of serum OC_{N-mid} in Group D compared with those levels in Group N were only detectable at 24 and 48 weeks after the fracture event, at the later stage of the healing process in our study. However, patients with delayed union of an intramedullary fixed tibial fracture had lower levels of bone-specific alkaline

phosphatase between 4 and 7 weeks, in the early phase after a fracture event, than did patients with normal healing [8]. In the process of normal fracture healing, bone-specific alkaline phosphatase may be associated with osteoblastic activity during the early stage of fracture healing, whereas osteocalcin may be associated with mineralization of the woven bone during the late stage of the fracture healing process. Therefore, osteocalcin may reflect an aspect of poorly healing vertebra different from that of bone-specific alkaline phosphatase. Unfortunately, bone-specific alkaline phosphatase was not measured in our patients. Regarding the pathologic aspects, the presence of fibrous or avascular granulation tissue, necrotic bone, or serous fluid in non-united vertebral bodies was observed under the microscope [10, 25]. From the pathologic evidence, we concluded impairment of fracture healing resulting from a decrease of osteoblast number and mineralization activity in the delayed or nonunited vertebra was strongly associated with a deficit of OC_{N-mid} at the later stage of the healing process in Group D (Fig. 7).

Bone mineral densities of the neck and trochanter of the proximal femur decreased after vertebral compression fracture and were not restored up to the baseline levels in all subjects by the end of the study. However, the BMDs of the distal one-third and ultradistal radius did not change and the mean values of ultradistal radius BMD even increased at 24 and 48 weeks. Disuse bone atrophy or immobilization resulting from long-term bed rest might be associated with the decrease of BMDs in the lower extremities. A vertebral compression fracture did not affect the disuse bone atrophy of the forearm, which was a non-weightbearing bone. According to published reports, BMD in the contralateral proximal femur did not recover at 48 or 52 weeks after a hip or ankle fracture [11, 13]. From those previous results and our data, recovery from disuse bone atrophy could not be expected 48 weeks after fracture. Because bone formation surpassed bone resorption 48 weeks after fracture according to our biochemical data, BMD might be expected to increase thereafter.

The factors that affect bone metabolism after fracture are the invasiveness of surgery, the period of bed rest, the degree of fracture, immobilization, and the process of fracture healing [7, 12, 17, 26, 31]. In our study groups, there were no differences in the period of bed rest and BMD of the femoral neck. This means there was no bias from the effect of immobilization and disuse bone atrophy between groups. We especially focused on the effect of bed rest on the values of bone markers. Bone resorption markers correlated with the period of bed rest after a fracture event; however, bone formation markers did not correlate with the period of bed rest for the study group. This result suggests changes in bone resorption markers are affected not only by fracture per se but also by disuse bone

atrophy resulting from bed rest. Therefore, one must be careful when interpreting values of bone resorption markers of patients during long-term bed rest.

Delayed union or nonunion plays a role in the development of prolonged back pain and leads to numerous inconveniences in daily activities. In this study, 78% in Group D reported back pain 48 weeks after a vertebral compression fracture, whereas 33% reported back pain in Group N. This result indicates prolonged back pain clearly is associated with instability in poorly united vertebra. An operative procedure to restore the stability of the spine might be necessary for patients with delayed or nonunion of the spine.

No antiosteoporosis drugs were administered to the subjects throughout this study to avoid the modifications of antiosteoporosis drugs on levels of biochemical markers and BMDs. Because one study showed bisphosphonate did not influence fracture healing of the appendicular bone unfavorably, antiosteoporosis drugs should be administered to the patients with fragility fracture as soon as possible after the fracture event to prevent further bone loss [30].

Finally, the only therapeutic option for nonunion of the spine is operative intervention. No pharmacotherapeutic intervention has been developed to date. Vitamin K_2 is one of the drugs that promote bone formation. Vitamin K is a cofactor of γ -carboxylase that converts undercarboxylated osteocalcin to carboxylated osteocalcin; it also modulates proliferation of osteoblasts and the uptake of carboxylated osteocalcin into bone [1]. Also, we believe parathyroid hormone can be used as an antiosteoporosis drug to stimulate bone formation. Intermittent parathyroid hormone administration has been used to investigate if BMDs improve in the patients with osteoporosis [29]. Once-daily administration of parathyroid hormone enhances the morphometric and mechanical properties of fracture callus [6]. This treatment is very attractive to patients with delayed union or nonunion of the spine because of its effectiveness. From our results, if serum levels of bone formation marker measured 24 weeks after vertebral compression fracture do not increase compared with baseline values, antiosteoporosis agents that promote bone formation such as vitamin K_2 or parathyroid hormone might be useful to prevent delayed union or nonunion of the vertebrae or to treat patients with delayed union or nonunion of the spine.

Bone metabolism in patients with delayed union or nonunion of the spine differs from that of patients with normal union. Although the degree of bone resorption and loss of BMD after fracture is the same for both groups, the sequential change in bone formation markers is quite different between the groups. Measurement of serum osteocalcin after fractures may be useful to investigate the ability of a fracture to heal normally in patients with vertebral compression fractures.

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