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Early Changes in Biochemical Markers of Bone Formation Correlate with Improvements in Bone Structure during Teriparatide Therapy

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Context: Biochemical markers of bone turnover may reflect bone structure during anabolic treatment.

Objective: The objective was to evaluate associations between changes in biochemical markers and structural and dynamic bone parameters during teriparatide treatment.

Design: This study was a randomized, multicenter, double-blind, placebo-controlled fracture prevention trial, with 20-month median treatment duration for biopsy subset.

Setting: The trial was conducted at 11 clinical study sites.

Patients: Sixty-one postmenopausal women with osteoporosis who had paired transiliac biopsy specimens participated in the study.

Interventions: Once-daily sc injections of either placebo or teriparatide (20 or 40 μg) were administered.

Main outcome measures: The study measured: 1) serum and urinary biochemical markers of bone formation [bone alkaline phosphatase and procollagen I C-terminal propeptide (PICP)] and resorption (N-telopeptide and deoxypyridinoline); and 2) structural and dynamic analyses of bone biopsies, including two-dimensional (2D) histomorphometry and three-dimensional (3D) micro-computed tomography evaluations measured at baseline (n = 57) and 12 (n = 21) or 22 (n = 36) months.

Results: U-N-telopeptide/creatinine and serum-PICP correlated with bone structure and dynamic indices at baseline, respectively. Changes in bone alkaline phosphatase at 1 month correlated with changes at 22 months in 2D wall thickness ($r = 0.73$; $P = 0.001$), trabecular bone volume (trabecular bone volume per total volume, BV/TV) ($r = 0.58$; $P < 0.05$), marrow star volume ($r = -0.51$; $P = 0.05$); 3D trabecular thickness ($r = 0.49$; $P < 0.05$), and BV/TV ($r = 0.54$; $P < 0.05$). Changes in PICP at 1 month correlated with changes in wall thickness ($r = 0.60$; $P = 0.01$), and 2D BV/TV ($r = 0.51$; $P < 0.05$) at 22 months. Changes in markers at 6 or 12 months were not associated with changes in structural or dynamic parameters.

Conclusions: Early (1-month) changes in biochemical markers of bone formation, but not resorption, correlated with improvements in bone structure after 22 months of teriparatide therapy. **(*J Clin Endocrinol Metab* 90: 3970–3977, 2005)**

TERIPARATIDE (RECOMBINANT DNA origin) injection [recombinant human PTH (1–34)] is a bone-forming agent for the treatment of osteoporosis. In the Fracture Prevention Trial (FPT), daily self-injections of teriparatide (20 and 40 μg) reduced the risk of new vertebral and nonvertebral fractures by 65 and 53%, respectively, in postmenopausal women with advanced osteoporosis (1). In the same trial, a mean treatment time of 18 months of teriparatide therapy stimulated both trabecular and cortical bone formation, resulting in increased trabecular bone volume and connectivity, improved trabecular morphology with a shift toward a more plate-like structure, and increased cortical bone thickness (2). These changes in bone structure constitute a reversal of osteoporotic bone structural changes and may explain the reduction in fracture rates (2).

Once-daily injection of PTH induced pronounced increases in biochemical markers of bone turnover (3–8). Bone formation markers showed more rapid and larger increases than resorption markers within the first 3 months of therapy, suggesting an early imbalance of bone turnover in favor of formation that remained positive over the first 6–12 months of treatment (3–8).

There are several reports suggesting that biochemical markers of bone turnover may reflect underlying changes in bone histomorphometric parameters or bone mass (9–13). Because of a rapid and early response, the decrease of biochemical markers of bone turnover, particularly bone resorption markers, has been correlated with the effects of antiresorptive osteoporosis treatments, such as changes in bone mass or fracture rates (14–20). Previous studies have demonstrated a positive correlation between 1- to 6-month

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Abbreviations: ALP, Alkaline phosphatase; BMD, bone mineral density; BV/TV, trabecular bone volume per total volume; Cr, creatinine; 2D, two-dimensional; 3D, three-dimensional; DPD, deoxypyridinoline; DXA, dual-energy x-ray absorptiometry; FPT, Fracture Prevention Trial; μCT, micro-computed tomography; MS/BS, mineralizing surface/bone surface; NTx, N-telopeptide; PICP, procollagen I C-terminal propeptide; sLS, single tetracycline-labeled surface.

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changes in bone formation markers and bone forming effects of PTH reflected by 1- to 5-yr changes in bone mineral density (BMD) (8, 21, 22). However, mineralization transients limit the value of BMD measurements in the early phases of teriparatide treatment (1, 22, 23).

The objective of this analysis, therefore, was to investigate whether changes in biochemical markers of bone formation and resorption correlate with bone structural improvements observed after teriparatide treatment.

Subjects and Methods

Subjects

In the randomized, multicenter, double-blind, placebo-controlled FPT (1), 1637 postmenopausal women with osteoporosis were treated with once-daily injections of either placebo or teriparatide, 20 or 40 μg. The primary endpoint of this study was the number of women who experienced a new vertebral fracture. Secondary endpoints were nonvertebral fracture and BMD, assessed by dual-energy x-ray absorptiometry (DXA). The design and conduct of this trial have been previously reported (1).

A total of 102 patients from 11 sites and five countries participated in the biopsy substudy of the FPT; paired iliac crest biopsy specimens were obtained from 61 patients. Quantitative and qualitative analyses of the baseline characteristics and the efficacy response confirmed that the substudy cohort was reflective of the entire FPT cohort (1, 2). Comparisons between the teriparatide-treated groups and the placebo group from the two-dimensional (2D) histomorphometry and three-dimensional (3D) micro-computed tomography (μCT) analyses have been previously reported (2). Pairwise comparisons between 20- or 40-μg teriparatide treatment groups showed no significant difference in clinical outcome and structural variables; therefore, the teriparatide treatment groups were pooled for the analyses (1, 2).

For the current analyses, paired specimens were included if at least one parameter from 2D histomorphometry or 3D μCT could be evaluated from both specimens; 57 paired biopsies (placebo group, n = 21; teriparatide, n = 36) were adequate for evaluation. Biopsy samples were analyzed by treatment duration: 21 paired samples were obtained at 12 months and 36 paired samples at 22 months of treatment. In the placebo group, 19 patients had both 2D and 3D measurements, and 2 patients had only 2D measurements. In the combined teriparatide group, 28 patients had both 2D and 3D measurements, four patients had only 2D measurements, and four patients had only 3D measurements. One patient in the teriparatide group did not have markers of bone turnover measured at 1 month; therefore, treatment correlations with markers of bone turnover were based on the remaining 56 biopsy pairs.

Patients signed informed consent to the treatment and investigation protocol, which was approved by the Institutional Review Board for Research Involving Human Subjects, at each participating center.

Treatment

All enrolled patients were supplemented with a daily intake of 1000 mg elemental calcium and 400-1200 IU vitamin D_3. Patients self-administered a once-daily injection of placebo for 2 wk and were then randomly assigned to receive placebo or 20 or 40 μg teriparatide daily. Because of the early termination of the FPT, the treatment duration was shorter than originally planned (1). The median duration of treatment for patients in this analysis was 20 months, with a range of 17–22 months. However, for logistical reasons, there was an average 40-d period (range, 4–73 d) between the discontinuation of the study medication and all measurements for the study end closeout visit. For the purpose of these analyses, the length of the teriparatide treatment was defined by the time that the second (follow-up) iliac crest biopsy was obtained.

Histomorphometry and μCT

The transiliac crest bone biopsies were carried out using a Bordier needle trephine system, following *in vivo* double labeling with tetracycline fluorochromes given orally in a 3:12:3 day sequence as published earlier (2). The methods for biopsy specimen evaluations have been published previously (2, 24–26). Biopsies were obtained from patients at baseline, before receiving treatment. Patients were then randomized to have a follow-up biopsy from the contralateral iliac crest after either 12 months or at study end. The 12-month biopsies (mean, 12 $\pm$ 1 months; range, 11–15 months) were performed in eight placebo patients and 13 teriparatide-treated patients. The biopsy at study end (mean, 22 $\pm$ 2 months; range, 19–25 months) was performed in 13 placebo patients and 23 teriparatide-treated patients.

Static and dynamic 2D parameters were measured under light microscopy on 5-μm-thick biopsy sections stained with Goldner's trichrome. Both double tetracycline-labeled surface/BS (percent) and single tetracycline-labeled surface (sLS/BS, percent) were measured. The μCT image acquisition and analysis of the biopsy specimens were performed as described previously (2, 24–26). All measured and derived variables were expressed according to the standard nomenclature recommended by the American Society of Bone and Mineral Research nomenclature committee (25).

The following 2D dynamic parameters, characterizing bone turnover, were included: activation frequency, the probability of a remodeling event occurring along a quiescent trabecular surface; mineral apposition rate (MAR, micrometers per day, unadjusted for sLS); and mineralizing surface/bone surface (MS/BS, percent) using the correction for the so-called label escape (double tetracycline-labeled surface + 0.5 $\times$ sLS) (2).

Structural changes during teriparatide treatment were assessed using both 2D and 3D indices. The static 2D histomorphometry parameters for trabecular bone included in the current analysis were total bone volume per tissue volume [trabecular bone volume per total volume (BV/TV)], mean wall thickness (micrometers) of completed osteons, and marrow star volume (cubic millimeters) (2). Cortical thickness (micrometers), expressed as the average of evenly spaced measurements across each of the inner and outer cortices, was also measured (2). The 3D structural parameters included BV/TV, trabecular thickness (micrometers), trabecular number (per millimeter), structural model index, connectivity density, and cortical thickness (2).

Biochemical markers of bone turnover

Biochemical markers of bone formation [serum bone specific alkaline phosphatase (ALP) and serum procollagen I C-terminal propeptide (PICP)] and resorption [urinary N-telopeptide (NTx) and urinary deoxypyridinoline (DPD)] were assessed at baseline and at 1, 3, 6, and 12 months and/or at study end. Serum samples and second-void urine samples were collected in the morning, after an overnight fast.

Markers of bone turnover were measured using commercially available assays: bone ALP, Tandem-R Ostase Immunoradiometric Assay (Beckman Coulter, Inc., Brea, CA); PICP, DiaSorin PICP Equilibrium RIA (DiaSorin, Inc., Stillwater, MN); and NTx, Osteomark ELISA (Ostex International, Inc., Seattle, WA); DPD, Pyrilinks-D assay (Metra Biosystems, Mountain View, CA). Both NTx and DPD were corrected for creatinine excretion (NTx/Cr and DPD/Cr, respectively). The intra- and interassay coefficients of variation were: 4.2–6.8% and 7.4–7.9%, respectively, for bone ALP; 2.7–3.1% and 5.4–7.0% for PICP; 4.5–6.6% and 6.7–14.8% for NTx; and 5.8–7.5% and 8.6–10.1% for DPD.

Statistical analysis

Patient demographics, relevant biopsy, and biomarker variables at baseline were summarized by group mean and SD. The teriparatide treatment groups were combined for analyses, as previously described (2). Baseline analyses included both placebo- and teriparatide-treated patients. Subgroup analyses to determine the effect of treatment duration were performed based on the timing of the second biopsy at 12 or 22 months.

Percent changes from baseline in biomarkers were summarized by the descriptive statistics, median, and corresponding SE of the median (based on Bootstrap technique). Comparisons between placebo- and teriparatide-treated patients in the percentage changes of biomarkers were made using the Wilcoxon rank-sum test. To evaluate the correlation between any two variables, Pearson correlation coefficients were computed between the parameters. Tests were performed comparing each correlation coefficient with zero. The α-level for significance was 0.05.

Correlation analyses between changes in biochemical markers and

structural changes in the placebo group at any given time point were used to validate the relationships in the treatment group, *i.e.* significant correlations in the treatment group were considered valid if the comparable relationship was not significant in the placebo group.

Results

Baseline characteristics

Of 1637 randomized patients from the FPT (1), results from 21 placebo patients and 36 teriparatide-treated patients who had biochemical marker measurements and iliac crest biopsies specimens, with at least one 2D or 3D index suitable for analysis, are reported here. The baseline demographics, baseline bone structural characteristics, and biochemical markers of the subpopulation are summarized in Tables 1 and 2. There were no statistically significant differences between the treatment groups and placebo in any of the variables. Likewise, there was no difference in the baseline characteristics of the 12-month and 22-month biopsy subgroups (data not shown).

Biochemical marker analysis

The bone-forming effect of teriparatide caused rapid increases in bone ALP and PICP in the teriparatide-treated group. Median percent changes from baseline are shown over time for all four biochemical markers in Fig. 1. Bone ALP continued to increase beyond 1 month, reaching a maximum at 12 months of treatment, with a median increase of 74 ± (SE) 23%. Teriparatide treatment produced significantly larger increases in bone ALP than placebo at all time points ($P < 0.001$). PICP reached its maximum median percent change of 62 ± 10% at 1 month. The median percent increase of PICP from baseline was significantly larger than the increases for the placebo patients from 1 month through 6 months ($P < 0.001$).

This bone formation response was followed by subsequent increases in markers associated with bone resorption (Fig. 1). Significant increases in these markers, compared with placebo, commenced at 3 months. NTx/Cr reached maximum values at 12 months, with a median increase of 201 ± 95% ($P < 0.05$). DPD/Cr response was earlier, with the maximum increase occurring at 6 months (136 ± 32%; $P < 0.01$).

TABLE 1. Baseline demographics and markers of bone turnover of a subset of patients from the FPT who provided paired iliac crest biopsies and had assessment of biochemical markers of bone turnover

	n	Placebo	n	TPTD
Age (yr)	21	67.8 ± 6.5	36	67.9 ± 6.2
Years since menopause	19	20 ± 8	29	21 ± 8.0
Femoral neck DXA (g/cm^2)	19	0.64 ± 0.11	35	0.63 ± 0.09
Lumbar spine DXA (g/cm^2)	21	0.85 ± 0.19	36	0.80 ± 0.16
No. vertebral fractures	21	2.3 ± 1.7	36	2.7 ± 1.6
Bone ALP (μg/liter)	21	13.0 ± 6.4	36	14.7 ± 8.5
PICP (μg/liter)	21	117.8 ± 34.6	36	120.4 ± 57.5
NTx/Cr (nmol BCE/mmol)	19	46.2 ± 24.1	20	45.6 ± 21.4
DPD/Cr (nmol/mmol)	19	6.5 ± 2.8	20	6.8 ± 2.4

Data are presented as mean ± SD. TPTD, Combined teriparatide-treated group; bone ALP, serum bone ALP; NTx/Cr, Cr corrected urinary N-terminal type 1 collagen cross-link peptide; BCE, bone collagen equivalence; DPD/Cr, Cr corrected urinary deoxypyridinoline.

TABLE 2. Baseline 2D trabecular and cortical bone histomorphometry and microstructure

	n	Placebo	n	TPTD
Structural parameters				
2D BV/TV	18	0.18 ± 0.06	28	0.15 ± 0.06
2D W.Th (μm)	19	40.5 ± 4.1	30	41.7 ± 7.5
2D Ma.St.V (mm^3)	16	20.9 ± 27.2	27	29.6 ± 31.7
2D Ct.Th (μm)	17	919.9 ± 274.8	29	894.8 ± 403.5
3D BV/TV	19	0.14 ± 0.06	32	0.12 ± 0.06
3D Tb.Th (mm)	19	0.15 ± 0.04	32	0.15 ± 0.04
3D SMI	19	1.77 ± 0.54	32	1.95 ± 0.49
3D Tb.N (mm^{-1})	19	1.28 ± 0.21	32	1.22 ± 0.27
3D CD (mm^{-3})	19	5.64 ± 2.29	32	4.49 ± 3.48
3D Ct.Th (mm)	18	0.86 ± 0.27	25	0.81 ± 0.30
Dynamic parameters				
Ac.f (per yr)	18	0.41 ± 0.23	28	0.37 ± 0.23
MS/BS (%)	19	0.08 ± 0.05	30	0.08 ± 0.05
MAR (μm/d)	18	0.58 ± 0.10	28	0.53 ± 0.09

Data are presented as mean ± SD. TPTD, Combined teriparatide treated group; W.Th, mean wall thickness; Ma.St.V, marrow star volume; Ct.Th, cortical thickness; Tb.Th, trabecular thickness; Tb.N, trabecular number; SMI, structural model index; CD, connectivity density; MAR, mineral apposition rate; Ac.f, activation frequency.

Correlations between percent change of biochemical markers of bone formation (bone ALP, PICP) and biochemical markers of bone resorption (DPD/Cr, NTx/Cr) were calculated to assess imbalance in bone turnover during teriparatide therapy. The increase in bone ALP at 1 month did not correlate with the increase of any of the respective resorption markers. However, the increase in bone ALP at 3, 6, and 12 months significantly correlated with respective increases in both DPD/Cr ($r = 0.38$, $P < 0.05$; $r = 0.73$, $P < 0.001$; $r = 0.58$, $P < 0.001$, respectively) and NTx/Cr ($r = 0.63$, $P < 0.001$; $r = 0.61$, $P < 0.001$; $r = 0.55$, $P = 0.001$, respectively) in the teriparatide-treatment group. Increases in PICP at 1 month and 3 months did not correlate with the increases of the respective resorption markers; however, PICP at 6 and 12 months correlated with respective increases of both DPD/Cr ($r = 0.48$, $P < 0.05$; $r = 0.58$, $P < 0.001$, respectively) and NTx/Cr ($r = 0.41$, $P < 0.05$; $r = 0.44$, $P < 0.01$) in the teriparatide group. These results suggest an early transient dissociation (imbalance) between formation and resorption markers during the first 1–3 months of teriparatide treatment in favor of bone formation. No correlations were found between changes in formation and resorption markers in the placebo group.

Correlations between biochemical markers of bone turnover and 2D histomorphometry and 3D quantitative computed tomography indices

Bone turnover before teriparatide treatment was assessed by correlation of the baseline biochemical markers of bone turnover and baseline dynamic histomorphometry parameters. Baseline PICP significantly correlated with activation frequency ($r = 0.39$; $P = 0.008$) and approached significance with mineralizing surface (MS/BS) ($r = 0.28$; $P = 0.054$). Other biochemical markers did not show statistically significant correlation with the dynamic parameters at baseline.

The role of baseline bone turnover on bone structure was assessed by correlation between baseline bone markers and baseline static 2D and 3D indices. Of the four biochemical

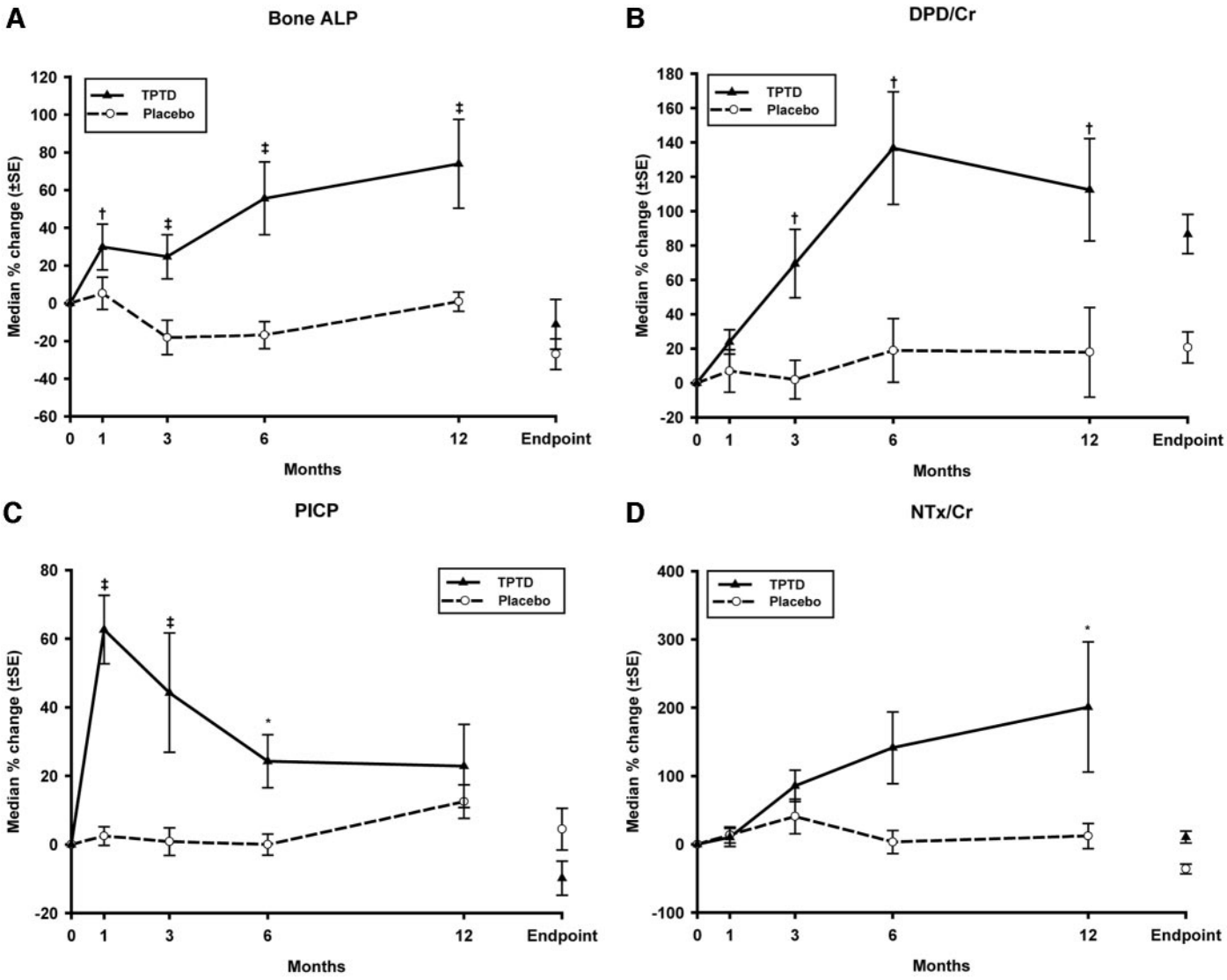

FIG. 1. The effect of teriparatide treatment on biochemical markers of bone turnover. A, Serum bone ALP; B, urinary DPD corrected for Cr (DPD/Cr); C, serum PICP; D, urinary N-terminal type I collagen cross-link peptide corrected for Cr (NTx/Cr). Data are shown as median ± SE. Placebo, *Dotted line*; teriparatide, *solid line*. *, $P < 0.05$ *vs.* placebo; †, $P < 0.01$ *vs.* placebo; ‡, $P < 0.001$.

markers measured, only NTx/Cr showed significant correlations with structural parameters. Baseline NTx/Cr was inversely correlated with baseline 2D and 3D cortical thickness (r = −0.41, $P < 0.01$; r = −0.47, $P < 0.01$, respectively) and with 3D trabecular bone volume (BV/TV) (r = −0.37; $P < 0.05$), and was positively correlated with the 2D marrow star volume (r = 0.46; $P < 0.01$). There were no significant correlations between NTx/Cr and the other structural parameters.

To further assess the role of baseline turnover in determining later structural changes during teriparatide treatment, correlations between baseline bone markers and percent change from baseline in structural and dynamic indices were also calculated. There were no significant correlations between any of the baseline markers and changes in dynamic or structural parameters at any biopsy time point.

The relationship between early biochemical marker changes and improvements in bone structure with teriparatide therapy was assessed by correlating percent change from baseline in markers to percent change from baseline in structural indices. In the 22-month subgroup, changes in bone ALP at 1 month were significantly positively correlated with changes in 2D trabecular bone volume (r = 0.58; $P < 0.05$), 2D mean wall thickness (r = 0.73; $P = 0.001$), 2D marrow star volume (r = −0.51; $P = 0.05$), 3D trabecular bone volume (r = 0.54; $P < 0.05$), and 3D trabecular thickness (r = 0.49; $P < 0.05$) (Table 3 and Fig. 2). There were no significant correlations between changes in bone ALP at 1 month and changes in the structural parameters for the 12-month group. Changes in bone ALP at 3, 6, and 12 months were not significantly correlated with changes in 2D histomorphometry or 3D μCT parameters at either biopsy time point.

Correlations between PICP at 1 and 3 months and structural indices showed less consistency. In the 22-month treatment group, change in PICP at 1 month significantly positively correlated with changes in 2D mean wall thickness (r =

TABLE 3. Correlations between changes in biochemical markers of bone formation at 1 month and changes in 2D and 3D structural parameters at study end (median of 22 months of teriparatide treatment)

	Bone ALP			PICP		
	n	r	P^a	n	r	P^a
2D BV/TV	16	0.58	<0.05	16	0.51	<0.05
2D W.Th	17	0.73	0.001	17	0.6	0.01
2D Ma.St.V	15	−0.51	0.05	15	−0.37	ns
2D Ct.Th	16	−0.14	ns	16	−0.32	ns
3D BV/TV	19	0.54	<0.05	19	0.43	ns
3D Tb.Th	19	0.49	<0.05	19	0.36	ns
3D Tb.N	19	0.31	ns	19	0.35	ns
3D Ct.Th	15	−0.2	ns	15	−0.34	ns
3D SMI	19	−0.2	ns	19	−0.28	ns
3D CD	19	0.19	ns	19	0.07	ns

Bone ALP, Serum bone ALP; W.Th, mean wall thickness; Ma.St.V, marrow star volume; Ct.Th, cortical thickness; Tb.Th, trabecular thickness; Tb.N, trabecular number; SMI, structural model index; CD, connectivity density; ns, not significant.

[a] Statistical significance for the subpopulation in whom biopsy samples were obtained at study end (22 months of teriparatide treatment).

0.60; $P = 0.01$) and 2D trabecular bone volume (r = 0.51; $P < 0.05$) (Table 3 and Fig. 3). Correlations between change in PICP at 1 month and change in 2D cortical thickness (r = 0.72; $P < 0.05$) and 3D connectivity density (r = 0.63; $P < 0.05$) were significant at 12 months but not at 22 months. Change in PICP at 3 months showed a significant inverse correlation with the change in 2D marrow star volume in the 22-month subgroup only (r = −0.61; $P < 0.05$). Changes in PICP at 6 and 12 months did not correlate with changes of any structural indices at either biopsy time point.

Changes in biochemical markers of bone resorption (NTx/Cr and DPD/Cr) did not correlate significantly with changes in any of the structural or dynamic parameters at any measured time points. Similarly, none of the above correlations were significant in the placebo group.

None of the correlations between changes in biochemical markers of bone turnover and changes in 2D dynamic parameters were significant at any measured time points. There were no statistically significant correlations between study endpoint biomarkers and dynamic parameters.

Discussion

In a subset of postmenopausal women with osteoporosis from the FPT (1), we demonstrated that early (1 month) increases in biochemical markers of bone formation correlated with improvements in bone structure after long-term (22 months) teriparatide treatment. Consistent with the findings of Delmas *et al.* (10), we have confirmed that increased bone turnover is related to the deteriorating bone structure before treatment. However, the structural improvements during teriparatide treatment seem to be independent of the increased baseline bone turnover.

In this analysis, teriparatide monotherapy resulted in time-courses of the measured biochemical markers similar to those when once-daily PTH (1–34) was administered concomitantly with estrogen (6, 8). Both histomorphometric and biochemical data support early bone formation after initiating once-daily injections of PTH (2–8, 21, 27–29). We demonstrated an early transient dissociation between biochemical markers of bone formation and resorption (5, 6, 8), with a subsequent delayed increase in bone resorption markers (4–8). We recognize that the levels of biochemical markers at study endpoint were influenced by the discontinuation of teriparatide that occurred an average of 40 d before the samples were obtained. Therefore, the true treatment effect of teriparatide on the biochemical markers obtained at this time point cannot be discerned.

Our results underscore the importance of early changes in bone turnover on the overall effects of teriparatide (30). Results from a trial of teriparatide used after antiresorptive therapy (23) and a teriparatide-alendronate comparator trial (31) show that the maximal stimulation of bone turnover takes place at 6 months and subsequently tapers off. There was a doubling of the activation frequency in teriparatide-treated patients at 6 months, with a return to the normal range by 18 months (32). Thus, with this time course for changes in dynamic parameters after teriparatide, the lack of significant correlation between biochemical markers at 1 and 3 months and biopsy parameters pertaining to bone turnover at 12 or 22 months is not unexpected. Jiang *et al.* (2) found no significant changes in trabecular bone modeling, bone resorption, and formation rates or labeling indices in a pooled analyses of the biopsies obtained. Minimal or no effect on these parameters after 12–36 months of once-daily PTH (1–34) treatment has also been demonstrated by others (2, 6, 8). The minimal effects of 15 months of teriparatide treatment on bone formation rates observed in primates support these findings (33).

We demonstrated that early 1-month changes in biochemical markers of bone formation, particularly bone ALP, were associated with improvements in indices of trabecular architecture at 22 months but not at 12 months. Others have also suggested that the early dominance of bone formation over resorption may be important for subsequent structural improvements after long-term treatment with teriparatide (8, 29, 34). These results are also consistent with findings that both BMD increase and fracture risk reduction appear to be more pronounced after longer duration of teriparatide treatment (1, 6, 35).

In contrast to trabecular bone, the early changes of bone formation markers did not correlate significantly with structural changes in cortical bone, though a significant increase in 3D cortical thickness had been observed in these patients (2). This lack of correlation may be explained by a different relationship between dynamic changes in bone turnover and bone formation within cortical and trabecular bone compartments (36, 37).

Serum levels of biochemical markers of bone turnover at 6 or12 months were less strongly related to structural improvements at 12 or 22 months. Because the major increases in PICP and bone ALP levels occur in the first 1–3 months of treatment, it is not surprising that changes in biochemical markers between 6 months and study end do not correlate with improvements in bone structure. However, it is conceivable that other biochemical markers of bone formation (*e.g.* procollagen I N-terminal propeptide) might exhibit different correlation patterns with the changes of the structural indices due to their different time courses (30).

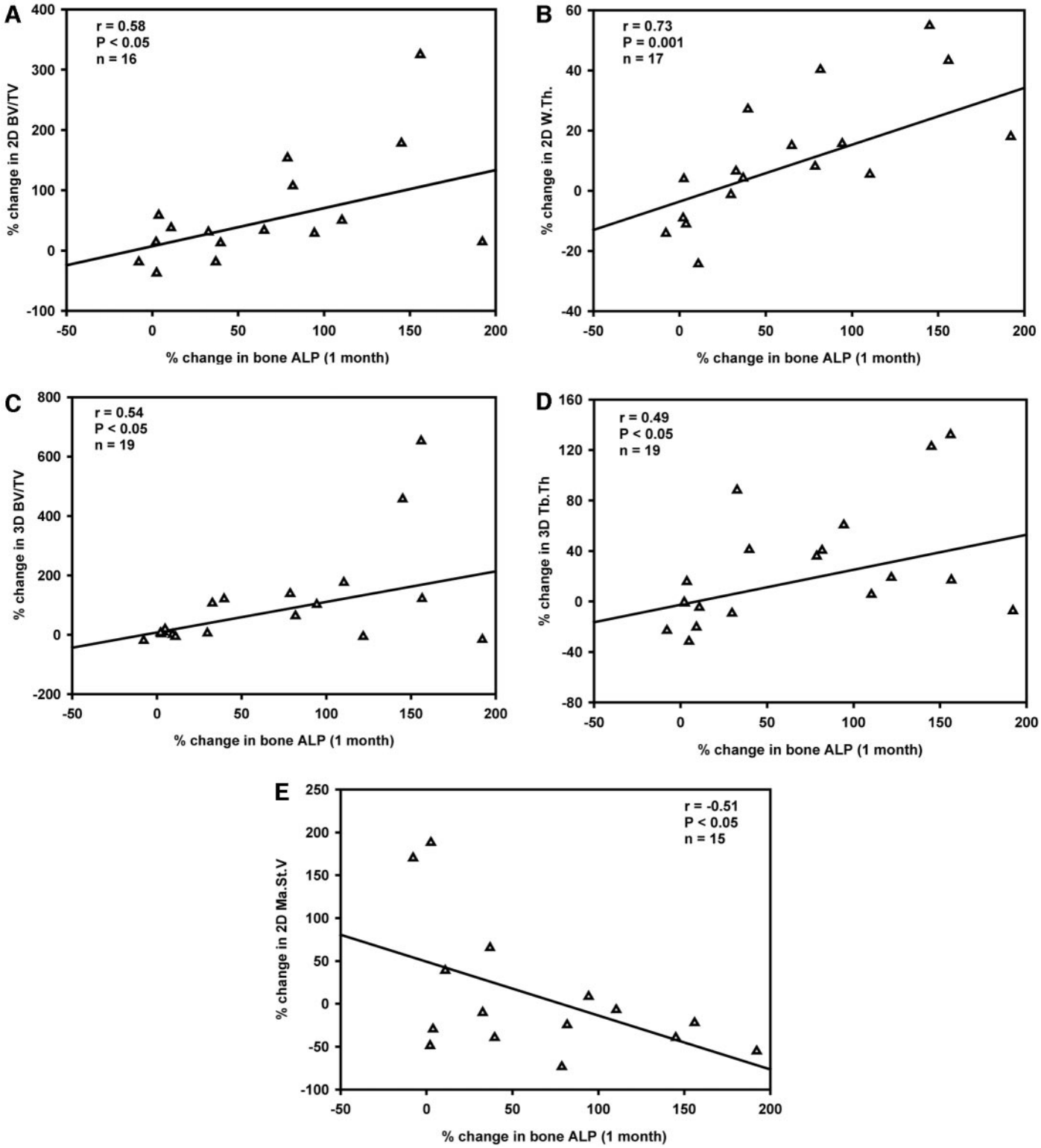

FIG. 2. Correlations between percent change in serum bone ALP from baseline to 1 month and percent change in bone structural indices from baseline to a mean of 22 months of teriparatide treatment. A, 2D trabecular bone volume (BV/TV); B, 2D wall thickness (W. Th.); C, 3D trabecular bone volume (BV/TV); D, 3D trabecular thickness (Tb.Th); E, 2D marrow star volume (Ma.St.V)

A strength of this analysis is that the subset of women who had bone biopsies and biochemical markers of bone turnover had characteristics similar to those of the overall study population of the FPT. However, several potential confounders may have limited the interpretation of these results. The bone structural changes at the iliac crest may not be representative

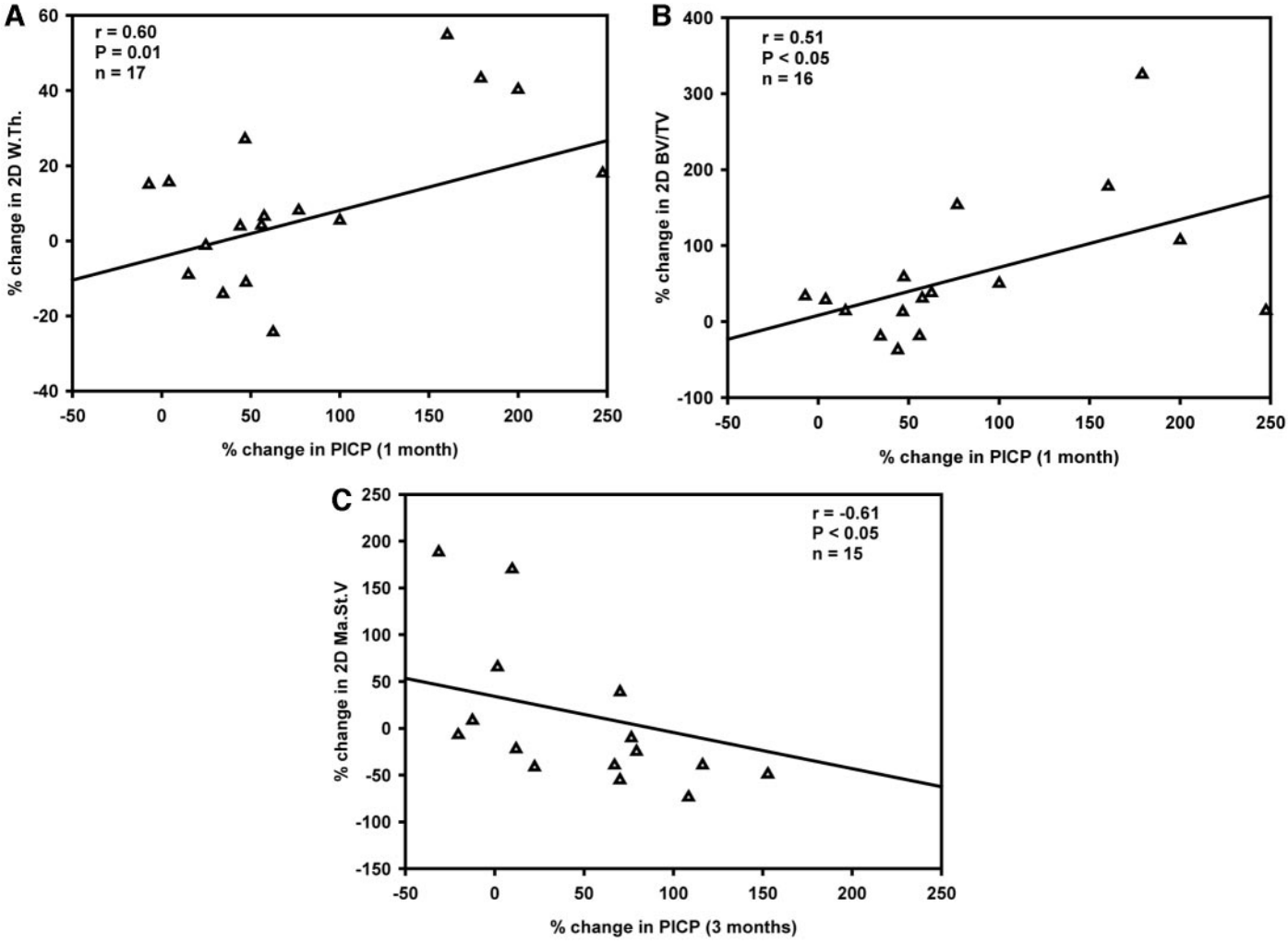

FIG. 3. Correlations between percent change in serum PICP from baseline to 1 month or 3 months and percent change in bone structural indices from baseline to a mean of 22 months of teriparatide treatment. A, 2D wall thickness (W.Th) at 1 month; B, 2D trabecular bone volume (BV/TV) at 1 month; C, 2D marrow star volume (Ma.St.V) at 3 months.

of the whole skeleton. Further, the sample size was relatively small. In addition, patients may demonstrate not only a different bone formation potential *per se* but also show differences in response kinetics; therefore, an individual peak increase in bone ALP or PICP, several weeks earlier or later than the group peak increase, could seriously weaken a correlation performed at a given time and with the number of subjects involved. The incidence of fracture in the subset of patients from whom biochemical markers were collected was not sufficiently large to assess the relationship between the observed changes in biochemical markers of bone formation and fracture risk reduction. Because the FPT was discontinued prematurely, the optimal length of teriparatide treatment has not yet been determined (1).

In conclusion, our data indicate that the early dominance of bone formation over resorption, after initiation of teriparatide therapy, may be important for the observed improvements in bone structure (29). Changes both in biochemical markers and bone structure are surrogates of clinical outcomes only; therefore, any association between those and the fracture reduction observed on teriparatide treatment remains to be evaluated. In addition, further data are needed to assess the usefulness of biochemical markers in monitoring teriparatide treatment.

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References

1. **Neer RM, Arnaud CD, Zanchetta JR, Prince R, Gaich GA, Reginster JY, Hodsman AB, Eriksen EF, Ish-Shalom S, Genant HK, Wang O, Mitlak BH** 2001 Effect of parathyroid hormone (1–34) on fractures and bone mineral density in postmenopausal women with osteoporosis. N Engl J Med 344:1434–1441
2. **Jiang Y, Zhao JJ, Mitlak BH, Wang O, Genant HK, Eriksen EF** 2003 Teriparatide [recombinant human parathyroid hormone (1–34)] improves both cortical and cancellous bone structure. J Bone Miner Res 18:1932–1941
3. **Reeve J, Bradbeer JN, Arlot M, Davies UM, Green JR, Hampton L, Edouard C, Hesp R, Hulme P, Ashby JP** 1991 hPTH 1–34 treatment of osteoporosis with added hormone replacement therapy: biochemical, kinetic and histological responses. Osteoporos Int 1:162–170
4. **Hodsman AB, Fraher LJ, Ostbye T, Adachi JD, Steer BM** 1993 An evaluation of several biochemical markers for bone formation and resorption in a protocol utilizing cyclical parathyroid hormone and calcitonin therapy for osteoporosis. J Clin Invest 91:1138–1148
5. **Hodsman AB, Fraher LJ, Watson PH, Ostbye T, Stitt LW, Adachi JD, Taves DH, Drost D** 1997 A randomized controlled trial to compare the efficacy of cyclical parathyroid hormone versus cyclical parathyroid hormone and sequential calcitonin to improve bone mass in postmenopausal women with osteoporosis. J Clin Endocrinol Metab 82:620–628
6. **Lindsay R, Nieves J, Formica C, Henneman E, Woelfert L, Shen V, Dempster D, Cosman F** 1997 Randomised controlled study of effect of parathyroid hormone on vertebral-bone mass and fracture incidence among postmenopausal women on oestrogen with osteoporosis. Lancet 350:550–555
7. **Lane NE, Sanchez S, Modin GW, Genant HK, Pierini E, Arnaud CD** 1998 Parathyroid hormone treatment can reverse corticosteroid-induced osteoporosis. Results of a randomized controlled clinical trial. J Clin Invest 102:1627–1633
8. **Lane NE, Sanchez S, Genant HK, Jenkins DK, Arnaud CD** 2000 Short-term increases in bone turnover markers predict parathyroid hormone-induced spinal bone mineral density gains in postmenopausal women with glucocorticoid-induced osteoporosis. Osteoporos Int 11:434–442
9. **Eastell R, Delmas PD, Hodgson SF, Eriksen EF, Mann KG, Riggs BL** 1988 Bone formation rate in older normal women: concurrent assessment with bone histomorphometry, calcium kinetics, and biochemical markers. J Clin Endocrinol Metab 67:741–748
10. **Delmas PD, Schlemmer A, Gineyts E, Riis B, Christiansen C** 1991 Urinary excretion of pyridinoline crosslinks correlates with bone turnover measured on iliac crest biopsy in patients with vertebral osteoporosis. J Bone Miner Res 6:639–644
11. **Riis BJ** 1991 Biochemical markers of bone turnover in diagnosis and assessment of therapy. Am J Med 91:64S–68S
12. **Roux JP, Arlot ME, Gineyts E, Meunier PJ, Delmas PD** 1995 Automatic-interactive measurement of resorption cavities in transiliac bone biopsies and correlation with deoxypyridinoline. Bone 17:153–156
13. **Garnero P, Sornay-Rendu E, Claustrat B, Delmas PD** 2000 Biochemical markers of bone turnover, endogenous hormones and the risk of fractures in postmenopausal women: the OFELY study. J Bone Miner Res 15:1526–1536
14. **Garnero P, Shih WJ, Gineyts E, Karpf DB, Delmas PD** 1994 Comparison of new biochemical markers of bone turnover in late postmenopausal osteoporotic women in response to alendronate treatment. J Clin Endocrinol Metab 79:1693–1700
15. **Akesson K, Ljunghall S, Jonsson B, Sernbo I, Johnell O, Gardsell P, Obrant KJ** 1995 Assessment of biochemical markers of bone metabolism in relation to the occurrence of fracture: a retrospective and prospective population-based study of women. J Bone Miner Res 10:1823–1829
16. **Chesnut III CH, Bell NH, Clark GS, Drinkwater BL, English SC, Johnson Jr CC, Notelovitz M, Rosen C, Cain DF, Flessland KA, Mallinak NJ** 1997 Hormone replacement therapy in postmenopausal women: urinary N-telopeptide of type I collagen monitors therapeutic effect and predicts response of bone mineral density. Am J Med 102:29–37
17. **Melton III LJ, Khosla S, Atkinson EJ, O'Fallon WM, Riggs BL** 1997 Relationship of bone turnover to bone density and fractures. J Bone Miner Res 12:1083–1091
18. **Bjarnason NH, Christiansen C, Sarkar S, Mitlak B, Knickerbocker R, Delmas PD** 1999 Six months changes in biochemical markers predict 3 year response in vertebral fracture rate in postmenopausal osteoporotic women: results from the MORE study. J Bone Miner Res 14:S157
19. **Ravn P, Clemmensen B, Christiansen C** 1999 Biochemical markers can predict the response in bone mass during alendronate treatment in early postmenopausal women. Bone 24:237–244
20. **Bjarnason NH, Christiansen C** 2000 Early response in biochemical markers predicts long-term response in bone mass during hormone replacement therapy in early postmenopausal women. Bone 26:561–569
21. **Kurland ES, Cosman F, McMahon DJ, Rosen CJ, Lindsay R, Bilezikian JP** 2000 Parathyroid hormone as a therapy for idiopathic osteoporosis in men: effects on bone mineral density and bone markers. J Clin Endocrinol Metab 85:3069–3076
22. **Reeve J, Mitchell A, Tellez M, Hulme P, Green JR, Wardley-Smith B, Mitchell R** 2001 Treatment with parathyroid peptides and estrogen replacement for severe postmenopausal vertebral osteoporosis: prediction of long-term responses in spine and femur. J Bone Miner Metab 19:102–114
23. **Ettinger B, San Martin J, Crans G, Pavo I** 2004 Differential effects of teriparatide on BMD after treatment with raloxifene or alendronate. J Bone Miner Res 19:745–751
24. **Eriksen EF** 1986 Normal and pathological remodeling of human trabecular bone: three dimensional reconstruction of the remodeling sequence in normals and in metabolic bone disease. Endocr Rev 7:379–408
25. **Parfitt AM, Drezner MK, Glorieux FH, Kanis JA, Malluche H, Meunier PJ, Ott SM, Recker RR** 1987 Bone histomorphometry: standardization of nomenclature, symbols, and units. Report of the ASBMR Histomorphometry Nomenclature Committee. J Bone Miner Res 2:595–610
26. **Jiang Y, Zhao J, Genant HK** 2002 Macro and micro imaging of bone architecture. In: Bilezikian TP, Raisz LG, Rodan GA, eds. Principles of bone biology. 2nd ed. San Diego, CA: Academic Press; 1599–1623
27. **Reeve J, Meunier PJ, Parsons JA, Bernat M, Bijvoet OL, Courpron P, Edouard C, Klenerman L, Neer RM, Renier JC, Slovik D, Vismans FJ, Potts Jr JT** 1980 Anabolic effect of human parathyroid hormone fragment on trabecular bone in involutional osteoporosis: a multicentre trial. Br Med J 280:1340–1344
28. **Hodsman AB, Steer BM** 1993 Early histomorphometric changes in response to parathyroid hormone therapy in osteoporosis: evidence for *de novo* bone formation on quiescent cancellous surfaces. Bone 14:523–527
29. **Dempster DW, Zhou H, Cosman F, Nieves J, Adachi JD, Fraher LJ, Watson PH, Lindsay R, Hodsman AB** 2001 PTH treatment directly stimulates bone formation in cancellous and cortical bone in humans. J Bone Miner Res 16:S179-
30. **McClung MR, Miller PD, Civitelli R, Warren ML, Greenspan SL, Tamayo JA, Donley DW, San Martin J** 2003 Distinctive effects of teriparatide [rhPTH(1–34)] and alendronate on bone turnover and lumbar spine bone density in women with osteoporosis. Osteoporos Int 13:S18
31. **McClung M, Miller P, Civitelli R, Warren M, Greenspan S, Haddock L, Tamayo J, Donley D, San Martin J** 2003 Differential effects of teriparatide and alendronate on markers of bone remodeling and areal and volumetric bone density in women with osteoporosis. J Bone Miner Res 18:S40
32. **Arlot M, Meunier PJ, Boivin G, Haddock L, Tamayo J, Correa-Rotter R, Jasqui S, Donley DW, Dalsky GP, San Martin J, Eriksen EF,** Differential effects of teriparatide and alendronate on bone remodeling in postmenopausal women assessed by histomorphometric parameters. J Bone Miner Res, in press
33. **Jerome CP, Burr DB, Van Bibber T, Hock JM, Brommage R** 2001 Treatment with human parathyroid hormone (1–34) for 18 months increases cancellous bone volume and improves trabecular architecture in ovariectomized cynomolgus monkeys (Macaca fascicularis). Bone 28:150–159
34. **Dempster DW, Cosman F, Kurland ES, Zhou H, Nieves J, Woelfert L, Shane E, Plavetic K, Muller R, Bilezikian J, Lindsay R** 2001 Effects of daily treatment with parathyroid hormone on bone microarchitecture and turnover in patients with osteoporosis: a paired biopsy study. J Bone Miner Res 16:1846–1853
35. **Pohl G, Wang J, Scheele W** 2003 Effect of duration of teriparatide therapy on nonvertebral fracture risk. Arthritis Rheum 48(Suppl 9):S234
36. **Balena R, Shih MS, Parfitt AM** 1992 Bone resorption and formation on the periosteal envelope of the ilium: a histomorphometric study in healthy women. J Bone Miner Res 7:1475–1482
37. **Parfitt AM** 2002 Parathyroid hormone and periosteal bone expansion. J Bone Miner Res 17:1741–1743

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