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Denosumab Treatment Effects on Structural Damage, Bone Mineral Density, and Bone Turnover in Rheumatoid Arthritis

A Twelve-Month, Multicenter, Randomized, Double-Blind, Placebo-Controlled, Phase II Clinical Trial

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Objective. **RANKL is essential for osteoclast development, activation, and survival. Denosumab is a fully human monoclonal IgG2 antibody that binds RANKL, inhibiting its activity. The aim of this multicenter, randomized, double-blind, placebo-controlled, phase II study was to evaluate the effects of denosumab on structural damage in patients with rheumatoid arthritis (RA) receiving methotrexate treatment.**

Methods. **RA patients received subcutaneous placebo (n = 75), denosumab 60 mg (n = 71), or denosumab 180 mg (n = 72) injections every 6 months for 12 months. The primary end point was the change from baseline in the magnetic resonance imaging (MRI) erosion score at 6 months.**

Results. **At 6 months, the increase in the MRI erosion score from baseline was lower in the 60-mg denosumab group (mean change 0.13; $P = 0.118$) and significantly lower in the 180-mg denosumab group (mean change 0.06; $P = 0.007$) than in the placebo group (mean change 1.75). A significant difference in the modified Sharp erosion score was observed as early as 6 months in the 180-mg denosumab group ($P = 0.019$) as compared with placebo, and at 12 months, both the 60-mg ($P = 0.012$) and the 180-mg ($P = 0.007$) denosumab groups were significantly different from the placebo group. Denosumab caused sustained suppression of markers of bone turnover. There was no evidence of an effect of denosumab on joint space narrowing or on measures of RA disease activity. Rates of adverse events were comparable between the denosumab and placebo groups.**

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Conclusion. **Addition of twice-yearly injections of denosumab to ongoing methotrexate treatment inhibited structural damage in patients with RA for up to 12 months, with no increase in the rates of adverse events as compared with placebo.**

Bone destruction is a central feature of rheumatoid arthritis (RA). Increased osteoclast activity contributes to local and systemic abnormalities of bone remodeling, including bone erosions and focal and systemic osteoporosis (1). RANKL is essential for osteoclast formation, function, and survival (2–5), and it is a key mediator of increased osteoclast activity in RA (6–10).

Denosumab (formerly, AMG 162) is a fully human monoclonal antibody that binds and inhibits RANKL, resulting in suppression of bone resorption (11,12). Denosumab specifically binds to human RANKL and therefore cannot be evaluated in animal models. Osteoprotegerin is an endogenous inhibitor of RANKL–RANK interactions; in animal models, a fusion protein of osteoprotegerin showed antierosive effects in rats with adjuvant- or collagen-induced arthritis (8,9) and inhibited inflammatory bone loss and erosions in tumor necrosis factor (TNF)–transgenic mice (13,14).

Clinical studies have demonstrated that when administered subcutaneously once every 6 months, denosumab decreases bone turnover and increases bone mineral density (BMD) in postmenopausal women with low BMD, and it is currently being investigated as a treatment for osteoporosis (11,12). Denosumab has also been shown to decrease bone turnover in patients with multiple myeloma and bone metastases from breast cancer (15).

Herein, we report on a phase II study that evaluated the ability of denosumab to decrease the progression of structural damage in patients with RA who were receiving methotrexate treatment. In RA, structural damage can be measured by various techniques, including radiography, ultrasonography, computed tomography, and magnetic resonance imaging (MRI). Although radiography remains the imaging standard for assessing structural damage, MRI may be more sensitive to structural changes in the joints, particularly in early RA (16). Accumulating data suggest that MRI may reflect structural damage in RA better than radiography because of its greater sensitivity to changes in erosion (17,18). This study is the first large multicenter trial in RA to include MRI scores for erosions as the primary end point.

PATIENTS AND METHODS

Study participants. Participants were recruited at 39 centers in the US and Canada. Key inclusion criteria were the presence of RA for ≥24 weeks, as diagnosed according to the American College of Rheumatology (ACR; formerly, the American Rheumatism Association) 1987 criteria (19), a stable dosage of methotrexate at 7.5–25 mg/week for ≥8 weeks, and ≥6 swollen joints (66-joint count, excluding the distal interphalangeal joints). The presence of either erosive disease (≥3 definite erosions on radiographs of the hands and feet) or both a C-reactive protein level ≥2.0 mg/dl and anti–cyclic citrullinated peptide antibodies was required.

Key exclusion criteria consisted of the following: glucocorticoid dosage >15 mg/day (prednisone or equivalent); scheduled surgery or joint replacement in the hands, wrists, or feet; pregnancy; or use of a biologic agent for RA (e.g., etanercept, infliximab) or leflunomide within 8 weeks before study randomization. Use of biologic agents ≥8 weeks before randomization was permitted. Patients with contraindications to whole-body MRI, such as claustrophobia, pacemakers, aneurysm clips, or intraocular metallic fragments, were also excluded.

The institutional review boards and independent ethics committees of the participating medical centers approved the protocol and amendments. Each patient gave written informed consent before any study-related procedures were initiated. The study was conducted in accordance with the principles of the Declaration of Helsinki.

Study design. This 12-month, multicenter, randomized, double-blind, placebo-controlled, phase II study included 3 treatment groups: denosumab 60 mg, denosumab 180 mg, and placebo. Study drug was administered by subcutaneous injections every 6 months in 2 doses (at baseline and at 6 months). All patients were to take daily supplements containing 0.5–1.0 gm of elemental calcium and 400–800 IU of vitamin D. Changes in the dosage of methotrexate or the addition of leflunomide, hydroxychloroquine, or sulfasalazine (individually or in combination), as well as changes in the dosage or the addition of steroids or nonsteroidal antiinflammatory drugs were allowed at any time throughout the study, except within 2 weeks of the study visit. Patients were allowed to take bisphosphonates, and rescue therapy with an anti-TNF agent was allowed after 6 months.

The randomization schedule was generated before the study and was stratified according to the current use of glucocorticoids and the prior use of a biologic agent.

Study assessments. MRIs of both hands (metacarpophalangeal [MCP] joints) and wrists were obtained as 2 images, using a standardized procedure, at baseline and 6 months. All images were acquired with 1.5T whole-body MRI scanners. Scans were performed with the arm of interest lying at the patient's side or over the patient's head, and with either a circumferential wrist coil or a surface coil, depending on the capabilities of the imaging center; the same positioning and technique were used for all serial examinations of each patient. Two types of MRI sequences were used. One type consisted of the following parameters: a coronal 3-dimensional (3-D) fast gradient-recalled echo (FGRE) sequence, with a repetition time (TR)/echo time (TE) of 24.9/7.7 msec (minimum full), a flip angle of 20°, number of signals averaged 1, a matrix of 512 × 192 over a field of view (FOV) of 120 × 120 mm, and 24 contiguous slices of 1.5 mm in thickness. The other type consisted of the following parameters: a coronal 2-dimensional (2-D) short-tau inversion recovery (STIR) sequence, with

TR/TE/inversion time of 4,000/30/150 msec, an echo train length of 8 msec, number of signals averaged 2, a matrix of 256 × 192 over a FOV of 120 × 120 mm, and 14 contiguous slices of 3 mm in thickness. The 3-D FGRE and 2-D STIR sequences were used for each anatomic location (hand and wrist) on each side of the body, amounting to 8 scans per patient per visit, not including the localizer scans. Including time for the localizer scan and subject positioning, the total examination time was typically about 90 minutes per visit.

Staff members at the imaging sites were trained in MRI protocol. Image quality was monitored, and images were analyzed centrally (Synarc, San Francisco, CA). All images were scored for bone erosion using a variation (20) of the RA MRI Scoring (RAMRIS) method originally developed by OMERACT (21–23). In the original RAMRIS method, 23 sites in each hand (both sides of MCP joints 2–5) and wrist (trapezium, trapezoid, capitate, hamate, triquetrum, pisiform, lunate, scaphoid, distal radius, distal ulna, and both sides of carpometacarpal joints 1–5) are scored for erosion on a scale of 0–10, with each increment representing 10% loss of articular bone. The variation used in this study included both sides of the first MCP joint and expanded the original 11-point scale of the RAMRIS erosion score to 21 points, with increments of 0.5 (5%) instead of 1 (10%). Incremental changes in the MRI score are based largely on a volumetric increase in erosion as a percentage of the articular bone involved. Erosion scores from all locations in both hands and wrists were summed to give a total erosion score of 0–500 for each subject.

Two radiologists who were experienced in MRI assessment and dedicated to clinical trials imaging independently read the images. Each reader was blinded to the other reader's assessment results, examination visit order, treatment allocation, and the patient's identity. Serial images from each patient were displayed side-by-side and evaluated simultaneously. Reader agreement with the expanded RAMRIS method was evaluated using images from 10 patients from the study data set. The intraclass correlation coefficients (ICCs) for erosion scores from these 10 patients were 0.87 at baseline, 0.84 at 6 months, and 0.79 for the change from baseline to 6 months. Initially, each reader read half of the scans from the remaining patients using the same method. Prior to unblinding of the readers, the decision was made to have both readers score all MRI scans and calculate the average score of the 2 readings for each patient to provide more robust data and to be consistent with the methods for the secondary radiographic analyses. The ICC values for the post hoc analysis of MRI erosion scores were 0.89 at baseline, 0.90 at 6 months, and 0.68 for the change from baseline to 6 months.

Radiographs of the hands/wrists and feet were obtained at baseline and at 6 and 12 months according to a standardized procedure, and the films were sent to a central facility for analysis using the modified Sharp/van der Heijde method (24,25). Two qualified physicians experienced in scoring radiographic changes of RA (PAO and JTS) each read the images from all patients; the final score was the average of the scores from the 2 readers. ICC values for the static modified total Sharp scores and modified Sharp erosion scores between the 2 readers were all ≥0.98 at baseline, 6 months, and 12 months. ICC values for change from baseline in the modified total Sharp scores were 0.73 at 6 months and 0.88 at 12 months; ICC values for change from baseline in the modified Sharp erosion score were 0.72 at 6 months and 0.86 at 12 months.

Screening radiographs to determine the presence of the number of erosions needed for study eligibility were read locally by a radiologist or an experienced rheumatologist.

Dual x-ray absorptiometry assessments of BMD of the lumbar spine (L1 through L4), total hip, femoral neck, and trochanter were performed at baseline and at 1, 6, and 12 months using bone densitometers from GE Healthcare (Madison, WI) and Hologic (Bedford, MA).

Blood and urine samples were obtained at baseline and at 1, 3, 6, and 12 months after a fast of ≥8 hours. The baseline and 6-month samples were obtained before administration of the study drug. A central laboratory performed serum chemistry and hematology tests. Blood and urine samples were assayed in specialty laboratories for bone and cartilage markers, including serum C-telopeptide of type I collagen (CTX-I), serum N-propeptide of type I collagen (PINP), and urine C-telopeptide of type II collagen (CTX-II). CTX-I and CTX-II assays were conducted using kits from Nordic Biosciences (Chesapeake, VA) and PINP assays were conducted using kits from Orion Diagnostica (Espoo, Finland).

Clinical assessments recorded at each study visit included the following: 66-joint count for swollen joints, 68-joint count for tender joints, physician's global assessment of disease activity by visual analog scale (VAS; 0 = no disease; 100 = most severe disease), patient's global assessment of disease activity by VAS (0 = no disease; 100 = most severe disease), pain score by VAS (0 = no pain; 100 = worst pain), and the Health Assessment Questionnaire (HAQ; 0 = no difficulty; 3 = unable to do) (26). Using these outcomes, ACR responses (27) were determined and 28-joint count Disease Activity Scores (DAS28) (28) were calculated.

Safety was assessed based on reports of adverse events, including RA flares, changes in vital signs, and changes in laboratory values. Antibodies detected by validated electrochemiluminescence immunoassay were screened for denosumab-neutralizing activity, as previously described (12).

Study end points. The primary efficacy end point was the change in MRI erosion score from baseline to 6 months. The key secondary efficacy end point was the change in the total modified Sharp score from baseline to 12 months. Other secondary efficacy end points included changes from baseline in the modified Sharp erosion score and the modified Sharp joint space narrowing score at 6 and 12 months; percentage change in bone and cartilage markers at 3, 6, and 12 months; percentage change in BMD at 12 months; and mean change in nonlaboratory outcomes at 6 and 12 months.

Statistical analysis. All statistical tests were 2-sided. A sample size of 198 patients was estimated to have 81% power to detect a treatment-group difference at a 2-sided significance level of 0.025, assuming a 0.6585 probability that the change in MRI score was lower in the denosumab group than in the placebo group.

Efficacy analyses included all randomized patients who received at least 1 dose of study treatment and had a baseline evaluation and at least 1 postbaseline evaluation. Data were imputed by linear interpolation for MRI scores outside the study window for the 6-month visit (±7 days) and for radiographic scores outside the study windows for the 6-month (±30 days) and 12-month (±30 days) visits. Categorical end points

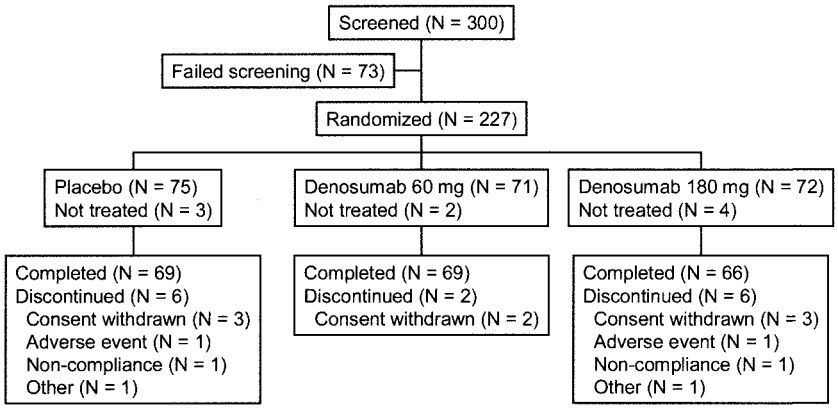

Figure 1. Disposition of the study patients from screening to study end. Most patients completed the 12-month treatment period. The reasons for early withdrawal from the study were comparable among the 3 treatment groups.

were summarized by the number and percentage of patients. Continuous variables were summarized using mean and standard deviation values. The van Elteren stratified rank test, accounting for the use of glucocorticoids and the previous use of biologic agents, was used to compare treatment groups for the primary efficacy analysis and key secondary efficacy analyses. Additional sensitivity analyses were performed after adjusting for both the baseline MRI erosion score (or radiographic score) and strata.

Hierarchical testing procedures and the Hochberg method (29) were used to protect the overall Type I error at 0.05 for multiple comparisons for the primary end point and key secondary end point. The Hochberg method was used to account for multiple comparisons (i.e., comparisons of each of the 2 denosumab dosage groups versus placebo) with the primary efficacy end point; statistical inference of the treatment effects on the key secondary end point was made only if the statistical inference of the treatment effect on the primary efficacy end point was statistically significant. Other secondary efficacy end points (bone and cartilage markers, BMD, and

Table 1. Demographic and clinical characteristics of all enrolled patients at baseline, by treatment group

Characteristic*	Placebo (n = 78)	Denosumab 60 mg (n = 73)	Denosumab 180 mg (n = 76)
Women, no. (%)	62 (79)	51 (70)	53 (70)
Age, mean ± SD years	57.0 ± 11.1	57.3 ± 11.4	58.0 ± 11.0
Disease duration, mean ± SD years	9.7 ± 8.1	10.5 ± 7.2	12.9 ± 11.3
Rheumatoid factor positive, no. (%)	61 (78)	55 (75)	60 (79)
Weekly methotrexate dose, mean ± SD mg	16.3 ± 4.1	15.7 ± 4.3	16.3 ± 4.8
Baseline corticosteroid use, no. (%)	28 (36)	27 (37)	29 (38)
Previous biologic therapy, no. (%)	17 (22)	14 (19)	16 (21)
Time between the last dose of anti-TNF agent and the start of the study, mean ± SD days	584 ± 521	884 ± 621	633 ± 468
Bisphosphonate use before and during study, no. (%)	21 (27)	13 (18)	15 (20)
MRI erosion score, range 0–500			
Mean ± SD	32.1 ± 26.5	41.2 ± 37.4	46.7 ± 42.5
Median (range)	27.1 (0–101)	32.8 (1–165)	35.9 (2–199)
Total modified Sharp score, range 0–448			
Mean ± SD	29.9 ± 34.7	40.0 ± 40.1	51.3 ± 59.8
Median (range)	17.5 (0–149)	29.0 (0–164)	26.3 (0–249)
Modified Sharp erosion score, range 0–280			
Mean ± SD	16.6 ± 17.2	22.2 ± 22.0	29.9 ± 35.2
Median (range)	10.5 (0–67)	15.0 (0–94)	16.3 (0–150)
Modified Sharp JSN score, range 0–168			
Mean ± SD	13.3 ± 18.9	17.8 ± 20.3	21.4 ± 26.2
Median (range)	5.0 (0–94)	11.0 (0–73)	10.3 (0–105)

* Anti-TNF = anti–tumor necrosis factor; MRI = magnetic resonance imaging; JSN = joint space narrowing.

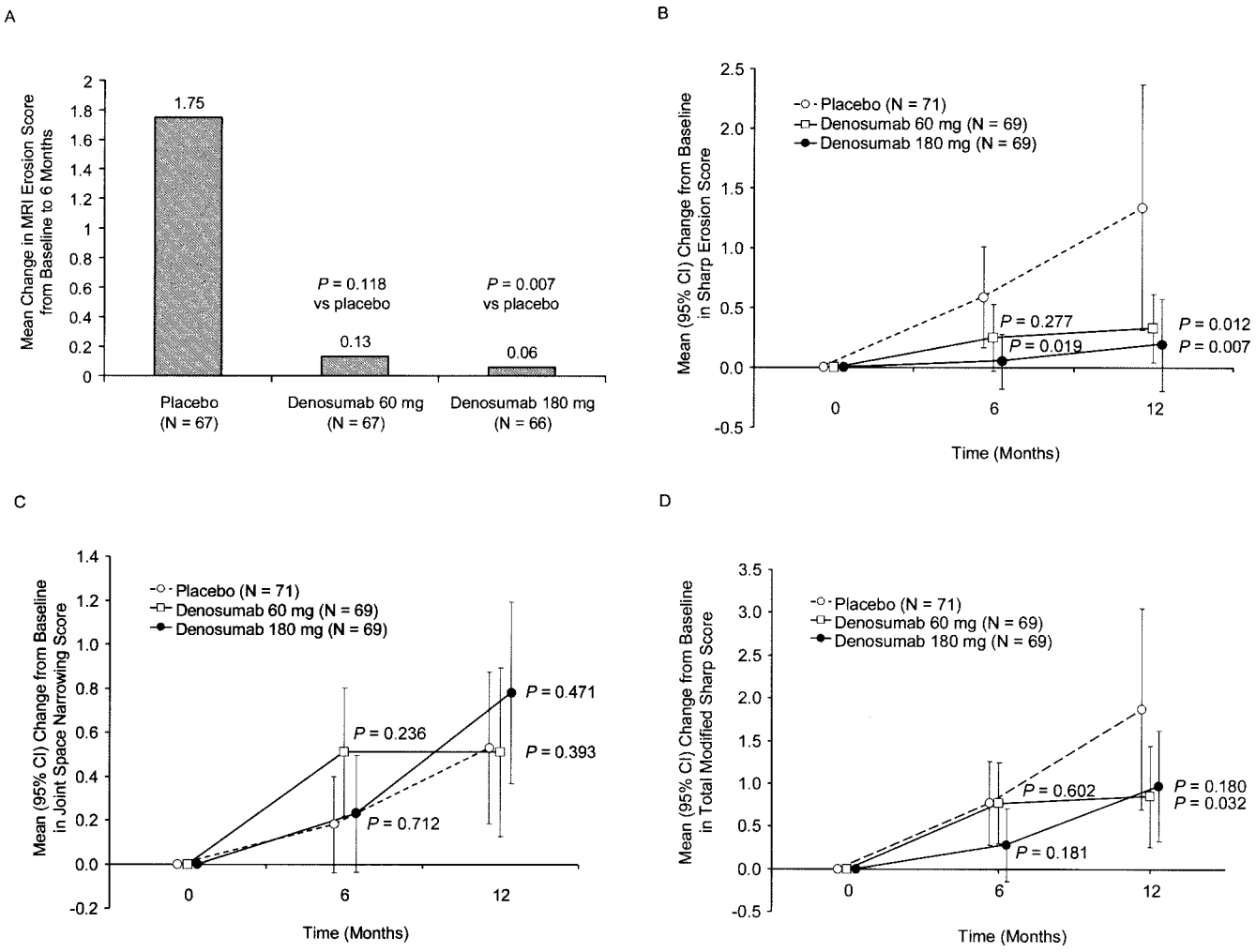

Figure 2. Mean change from baseline in the magnetic resonance imaging (MRI) erosion score and the modified Sharp scores in the 3 treatment groups. **A,** The mean change from baseline in the MRI erosion score at 6 months (the primary efficacy end point) was significantly different between the denosumab 180-mg group and the placebo group. **B,** The mean increase in the modified Sharp erosion score at 12 months was lower in both denosumab treatment groups as compared with the placebo group. **C,** The mean change in the modified Sharp joint space narrowing score at 12 months was not significantly different between either of the active treatment groups and the placebo group. **D,** The mean change in the total modified Sharp score at 12 months was lower in the denosumab 60-mg group than in the placebo group. 95% CI = 95% confidence interval. The time points in **B–D** are slightly displaced for readability.

clinical parameters) were summarized without statistical comparisons between treatment groups.

Safety analyses included all randomized patients who received at least 1 dose of study treatment. Adverse events and laboratory values were summarized by treatment group.

RESULTS

Subjects. Of the 300 patients screened, 227 patients were enrolled, and 218 patients received study treatment (Figure 1). Most of the patients (94%) completed study treatment. The most common reason for discontinuation was withdrawal of consent. Demographic and baseline characteristics (Table 1) were generally comparable between treatment groups. Disease duration, MRI erosion score, and total modified Sharp score tended to be greater in the denosumab groups than in the placebo group at baseline, but the differences were not statistically significant.

Efficacy. *Changes in MRI erosion scores.* The change in the MRI erosion score from baseline to 6 months was lower in the denosumab groups than in the

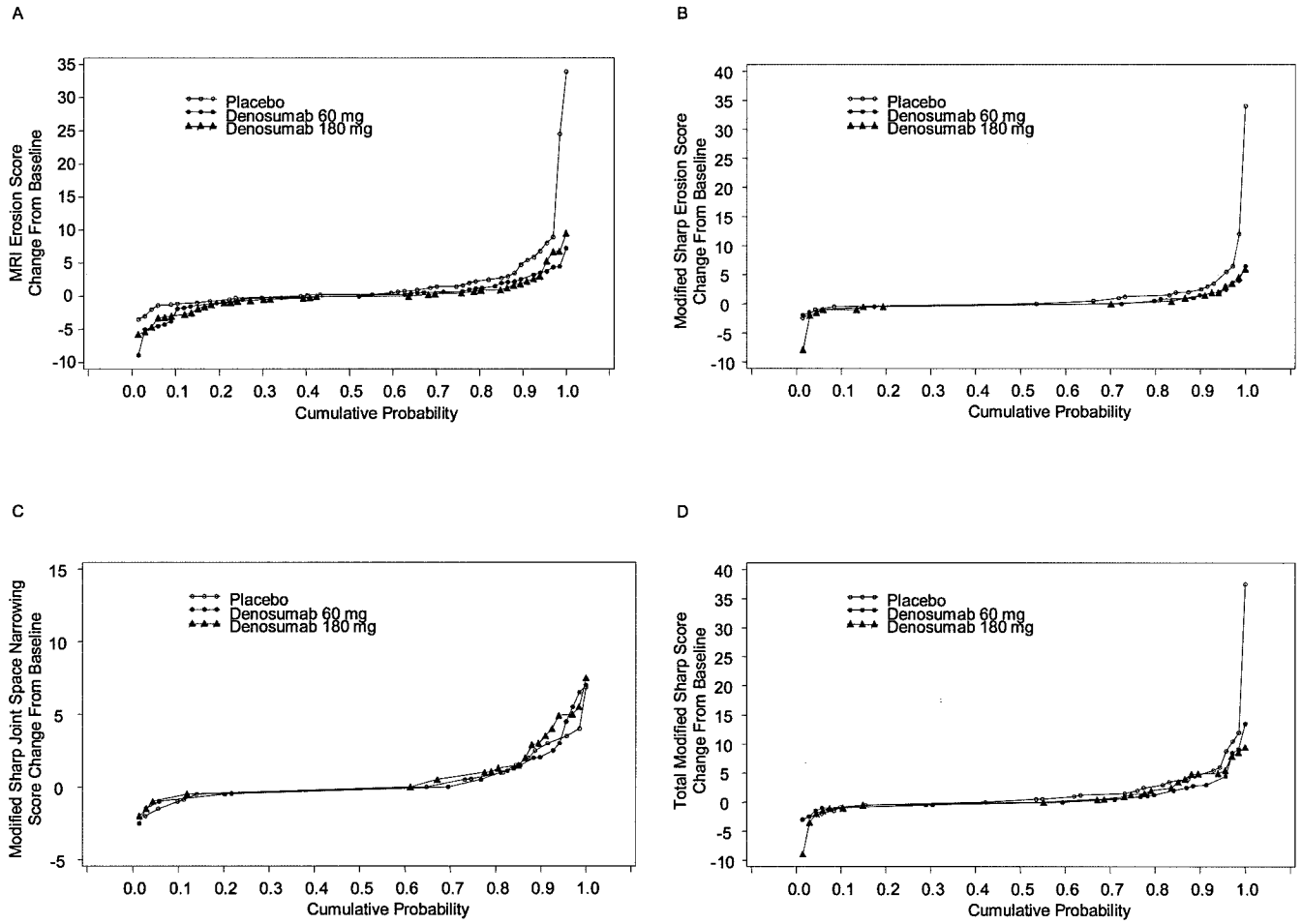

Figure 3. Probability plots of changes from baseline in **A,** the magnetic resonance imaging (MRI) erosion score, **B,** the modified Sharp erosion score, **C,** the modified Sharp joint space narrowing score, and **D,** the total modified Sharp score in individual patients in each of the 3 treatment groups.

placebo group (Figure 2A). The mean change at 6 months was 1.75 (median 0.25) in the placebo group, 0.13 (median 0.00) in the 60-mg denosumab group, and 0.06 (median 0.00) in the 180-mg denosumab group. The observed difference between the 180-mg denosumab group and the placebo group was statistically significant ($P = 0.007$); the difference between the 60-mg denosumab group and the placebo group was not statistically significant ($P = 0.118$). The percentages of patients with stable or decreased MRI erosion scores at 6 months in the placebo, 60-mg denosumab, and 180-mg denosumab groups were 39%, 51%, and 64%, respectively.

The cumulative probability plot for the change in MRI erosion scores from baseline in each patient in each of the 3 treatment groups is shown in Figure 3A.

Radiographic progression. Statistical analyses of secondary efficacy end points were made only if the primary analysis of the MRI data showed statistically significant differences for both of the denosumab doses as compared with placebo. Because the primary analysis did not achieve statistical significance for the 60-mg denosumab group, statistical testing of the radiographic results was exploratory.

Modified Sharp erosion scores increased more with placebo treatment than with denosumab treatment (Figure 2B). The mean increases from baseline in the modified Sharp erosion scores in the placebo, 60-mg denosumab, and 180-mg denosumab groups were as follows: 0.59, 0.25 ($P = 0.277$), and 0.05 ($P = 0.019$), respectively, at 6 months, and 1.34, 0.33 ($P = 0.012$), and

Table 2. Percentage change from baseline in markers of bone turnover and cartilage in all treated patients, by treatment group*

Marker	Placebo (n = 71)	Denosumab 60 mg (n = 70)	Denosumab 180 mg (n = 71)
Serum CTX-I			
3 months	0.30 ± 51.55	−74.23 ± 18.43	−72.71 ± 17.92
6 months	1.64 ± 53.86	−36.28 ± 46.43	−55.14 ± 37.85
12 months	−11.90 ± 46.54	−36.96 ± 47.04	−49.99 ± 56.83
Serum PINP			
3 months	−6.64 ± 31.21	−55.45 ± 19.86	−50.77 ± 26.81
6 months	−16.42 ± 39.16	−45.69 ± 27.23	−35.60 ± 111.44
12 months	−13.97 ± 36.84	−27.48 ± 47.84	−44.42 ± 19.40
Urine CTX-II/creatinine			
3 months	26.14 ± 122.23	−10.76 ± 73.75	−23.39 ± 69.15
6 months	28.50 ± 86.03	32.88 ± 93.06	52.12 ± 123.75
12 months	110.82 ± 333.66	83.32 ± 152.34	97.26 ± 193.18

* Values are the mean ± SD percentage change from baseline. CTX-I = C-telopeptide of type I collagen; PINP = N-propeptide of type I collagen; CTX-II = C-telopeptide of type II collagen.

0.19 ($P = 0.007$), respectively, at 12 months. The median increase in modified Sharp erosion scores was 0.00 for all treatment groups at 6 months and at 12 months.

There was no evidence of an effect of denosumab on the modified Sharp joint space narrowing score (Figure 2C). Mean increases from baseline in joint space narrowing scores in the placebo, 60-mg denosumab, and 180-mg denosumab groups were as follows: 0.18, 0.51 ($P = 0.236$), and 0.23 ($P = 0.712$), respectively, at 6 months, and 0.53, 0.51 ($P = 0.393$), and 0.78 ($P = 0.471$), respectively, at 12 months.

Total modified Sharp scores increased more in the placebo group than in the denosumab groups (Figure 2D). Mean increases in total modified Sharp scores in the placebo, 60-mg denosumab, and 180-mg denosumab groups were as follows: 0.77, 0.77 ($P = 0.602$), and 0.28 ($P = 0.181$), respectively, at 6 months, and 1.87, 0.85 ($P = 0.032$), and 0.97 ($P = 0.180$), respectively, at 12 months.

Cumulative probability plots for changes in the modified Sharp erosion score, the modified Sharp joint space narrowing score, and the total modified Sharp

Table 3. Adverse events reported by ≥5% of all treated patients, by treatment group*

Adverse event	Placebo (n = 75)	Denosumab 60 mg (n = 71)	Denosumab 180 mg (n = 72)
Any adverse event	67 (89)	60 (85)	56 (78)
Rheumatoid arthritis flare	25 (33)	21 (30)	21 (29)
Upper respiratory tract infection	6 (8)	11 (15)	9 (13)
Sinusitis	8 (11)	4 (6)	8 (11)
Arthralgia	2 (3)	6 (8)	4 (6)
Nasopharyngitis	9 (12)	5 (7)	5 (7)
Influenza†	0 (0)	2 (3)	7 (10)
Bronchitis	3 (4)	3 (4)	4 (6)
Cough	5 (7)	6 (8)	1 (1)
Urinary tract infection	1 (1)	4 (6)	3 (4)
Serious adverse event	7 (9)	3 (4)	6 (8)
Infection requiring hospitalization	1 (1)	1 (1)	2 (3)
Neoplasm	2 (3)	1 (1)	1 (1)
Discontinuation due to adverse event	1 (1)	0 (0)	1 (1)
Treatment-related adverse event	7 (9)	9 (13)	9 (13)
Treatment-related serious adverse event	0 (0)	0 (0)	0 (0)
Death	0 (0)	0 (0)	0 (0)

* Values are the number (%) of patients.
† Patient-reported event; no case definition was applied.

score from baseline in each patient in each of the 3 treatment groups are shown in Figures 3B–D, respectively.

The potential influence of differences in the baseline MRI score and the total modified Sharp score between the placebo and denosumab groups was assessed by a sensitivity analysis. After adjusting for these differences, the results were similar to the primary and key secondary efficacy analyses (data not shown).

Changes in markers of bone and cartilage turnover and BMD. Denosumab treatment resulted in a sustained decrease in markers of bone turnover and an increase in BMD over baseline values. Substantial suppression of markers of bone turnover (serum CTX-I and PINP) was apparent in the denosumab treatment groups at 3, 6, and 12 months (Table 2). Decreases from baseline levels in the cartilage turnover marker (urine CTX-II/creatinine) were observed in the denosumab groups at 3 months, but not at 6 or 12 months (Table 2).

Mean percentages of change from baseline in the BMD values at 12 months in the placebo, 60-mg denosumab, and 180-mg denosumab groups were as follows: for the lumbar spine, 0.9%, 3.0%, and 4.0%, respectively; for the total hip, −0.3%, 1.6%, and 1.7%, respectively; for the trochanter, −0.3%, 2.0%, and 2.1%, respectively; and for the femoral neck, −0.5%, 1.3%, and 1.6%, respectively. Positive effects on BMD were observed in both denosumab dosage groups compared with placebo ($P < 0.05$ for each comparison) at 12 months.

ACR responses and changes in HAQ and DAS28 scores. Denosumab treatment had no effect on the ACR response or any components of the ACR response (data not shown). ACR20/50/70 responses at 12 months were as follows: 25%/15%/6% in the placebo group, 23%/9%/1% in the 60-mg denosumab group, and 22%/11%/3% in the 180-mg denosumab group. The mean ± SD change from baseline in the HAQ score at 12 months was also comparable across treatment arms: −0.06 ± 0.51 in the placebo group, −0.13 ± 0.45 in the 60-mg denosumab group, and −0.09 ± 0.43 in the 180-mg denosumab group. Similarly, denosumab treatment did not produce improvements in the DAS28. The mean ± SD change from baseline in the DAS28 at 12 months was −0.60 ± 1.37 in the placebo group, −0.44 ± 1.01 in the 60-mg denosumab group, and −0.69 ± 1.06 in the 180-mg denosumab group.

Concomitant medications. Eleven patients in the placebo group, 8 in the 60-mg denosumab group, and 7 in the 180-mg denosumab group increased their dosage of methotrexate during the study. Of those whose RA required the addition of steroids or increases in the steroid dosages, 20 were in the placebo group, 12 were in the 60-mg denosumab group, and 22 were in the 180-mg denosumab group. The number of patients in whom a disease-modifying antirheumatic drug (DMARD) (i.e., leflunomide, hydroxychloroquine, or sulfasalazine) was added to the treatment regimen was low across the treatment groups (2 in the placebo group and 4 in the 180-mg denosumab group). Only 7 patients (2 receiving placebo, 1 receiving 60 mg of denosumab, and 4 receiving 180 mg of denosumab) required rescue therapy with anti-TNF agents after 6 months.

Safety. Rates of adverse events were comparable among the 3 treatment groups (Table 3). The most commonly reported adverse events during 12 months of treatment and evaluation were RA flare, upper respiratory tract infection, and sinusitis. No patient had a treatment-related serious adverse event (SAE). The incidences of neoplasms and SAEs involving infections were low and were balanced across the 3 groups (Table 3). The 2 SAEs involving infections (pyelonephritis and urosepsis) in the 180-mg denosumab group occurred in 1 patient 6 weeks apart, and only 1 event (disseminated histoplasmosis) occurred in the placebo group and 1 event (cellulitis) in the 60-mg denosumab group. One patient in the placebo group (1%), no patients in the 60-mg denosumab group (0%), and 1 patient in the 180-mg denosumab group (1%) discontinued the study because of an adverse event.

No clinically relevant changes in the laboratory results, vital signs, or parathyroid hormone levels were observed among patients in the 3 treatment groups. No neutralizing antibodies against denosumab were observed during the study.

DISCUSSION

In this 12-month, multicenter, randomized, double-blind, placebo-controlled, phase II clinical trial, subcutaneous injection of denosumab at 60 mg or 180 mg once every 6 months inhibited the progression of bone erosion scores in patients with active, erosive RA who were currently receiving treatment with methotrexate. The antierosive effects of 180 mg of denosumab were greater than those of placebo at 6 months, as measured by erosion scores on MRI scans and radiographs (modified Sharp erosion scores). The reduction of structural damage in the 60-mg denosumab group was also greater than that in the placebo group, as measured by the total modified Sharp score and the modified Sharp erosion score at 12 months. Consistent with

reported preclinical findings (8,13), denosumab did not have an effect on RA disease activity, as measured by the ACR response criteria, the DAS28 scores, and the occurrence of RA flares. As reported in previous clinical trials (12,15), rates of SAEs, particularly events of medical interest (i.e., neoplasms and SAEs involving infections) (30), were low and were balanced between the denosumab and placebo groups.

A recent report from a single-center, proof-of-concept study of 39 patients receiving methotrexate showed that the bisphosphonate zoledronic acid might also be effective in inhibiting bone erosion (31). Treatment with zoledronic acid resulted in a change of 61% in the MRI erosion scores at 6 months compared with methotrexate alone. In the present study, the change in erosion scores at 6 months was 93% lower in the 60-mg denosumab group and 97% lower in the 180-mg denosumab group relative to the placebo group (receiving methotrexate alone). While bisphosphonates act as antiresorptive agents, mainly by their action on osteoclasts, denosumab directly targets osteoclastogenesis by its specific action on the RANKL pathway.

In RA, inflammatory changes result in erosion of both bone and soft tissue in the joints. Bone resorption is regulated by RANKL, the primary mediator of osteoclast formation, function, and survival (32). Binding of RANKL to its receptor RANK promotes osteoclastogenesis, whereas the endogenous glycoprotein osteoprotegerin is produced predominantly by osteoblasts, and it competitively binds RANK and inhibits osteoclastogenesis. Patients with active RA have higher levels of RANKL than do healthy adults or patients with inactive RA (6), and the balance between levels of RANKL and osteoprotegerin is correlated with the extent of bone erosion (7). Administration of osteoprotegerin to animals with collagen-induced arthritis has been shown to reduce bone loss and cartilage destruction (8,9). Denosumab also inhibits RANKL, which results in a decrease in osteoclastogenesis, but it is a fully human monoclonal antibody that selectively binds RANKL.

Denosumab therapy did not affect HAQ scores at 12 months. Correlations between joint destruction and long-term clinical outcomes (primarily HAQ scores) have generally been significant (33,34), but in short-term studies, the correlation is usually not significant. In patients with RA, early disability is usually due to active inflammation, whereas long-term disability is due to structural damage. Until now, all DMARDs (including biologic agents) that have had an effect on joint destruction have also had important effects on the signs and symptoms of RA. Denosumab represents a new treatment strategy that protects against the destructive aspects of this disease, but lacks any known effect on inflammation. While it appears self-evident that any joint destruction is deleterious, there are no clear data that provide the precise levels or rates of destruction that lead to overt loss of function or other clinical manifestations. Furthermore, there are few data to date that suggest the relative importance of cartilage preservation compared with erosive damage.

The concordance between MRI and radiography results provides independent measures that highlight the antierosive effects of denosumab in this population. The MRI erosion score at 6 months showed the same trend as the modified Sharp erosion score at both 6 months and 12 months, particularly in the 180-mg denosumab group.

In this study, the rate of radiographic progression of erosions in the placebo (plus methotrexate) group was similar to the reported rates in methotrexate-treated patients in previous clinical trials (35,36). Notably, the effect of denosumab treatment on the rate of progression of erosions, based on radiographic assessments, was similar to the effects of treatment with anti-TNF agents (35,36).

Although patients in the denosumab treatment groups had greater levels of MRI erosions and radiographic damage at baseline compared with the placebo group, this imbalance had no effect on the findings of the primary and key secondary efficacy analyses (data not shown). In addition, this imbalance would be expected to be biased against the effects of denosumab, since high levels of erosions at baseline are usually a predictor of more aggressive future erosive disease. Alternatively, it is also possible that patients with greater levels of MRI erosions and radiographic damage at baseline had higher levels of osteoclast activity and, therefore, would be more sensitive to RANKL inhibition. One would expect that these patients would also have markedly elevated levels of RANKL compared with patients in the placebo group. However, we did not observe differences in RANKL levels between treatment groups (data not shown).

Denosumab treatment had no impact on the rate of joint space narrowing. This observation may reflect the mechanism of action of denosumab or the insufficiency of the dosages used in this study.

In addition to suppression of markers of bone turnover, both doses of denosumab also suppressed CTX-II/creatinine at 3 months, indicating the potential of denosumab to reduce cartilage erosion as well. However, the suppression of CTX-II/creatinine was not

maintained at 6 months or 12 months, suggesting that the dosages may not have been sufficient to reduce cartilage erosion over the 6-month interval.

Although increases in BMD were observed in the denosumab groups as compared with the placebo group, the effects were not as pronounced as the results from another study of postmenopausal women with low BMD (12). There are several possible reasons for this observation. First, the study populations are different. Although RA is a risk factor for osteopenia and osteoporosis (37), eligibility criteria for this study did not require the presence of low BMD scores at baseline, and most of the enrolled patients had normal BMD values; therefore, modest improvement should not be surprising. Second, a subset of patients in this study (n = 49) were concomitantly treated with bisphosphonates for osteoporosis (either postmenopausal or steroid-induced). Gains in BMD with denosumab treatment compared with placebo within this subset of patients were similar to the observed BMD results in the total study population. Interestingly, gains in BMD with bisphosphonate treatment in the placebo group were consistently <2% at 12 months in all measurement sites, whereas denosumab-treated patients showed gains with bisphosphonate treatment that were typically 2–4-fold greater than with bisphosphonate treatment alone.

In summary, the results of this study demonstrated that the addition of twice-yearly injections of denosumab to ongoing treatment with methotrexate inhibited structural damage, improved BMD, and suppressed bone turnover in RA patients, without increasing the rate of adverse events as compared with placebo treatment. Additional clinical studies of denosumab therapy in patients with RA are warranted.

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AUTHOR CONTRIBUTIONS

Dr. Cohen had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study design. Cohen, Peterfy, van der Heijde, Tsuji, Newmark.

Acquisition of data. Cohen, Dore, Lane, Ory, Peterfy, Sharp, Zhou, Tsuji, Newmark.

Analysis and interpretation of data. Cohen, Lane, Ory, Peterfy, van der Heijde, Zhou, Tsuji, Newmark.

Manuscript preparation. Cohen, Dore, Lane, Peterfy, van der Heijde, Zhou, Tsuji, Newmark.

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ROLE OF THE STUDY SPONSOR

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