The Effect of Low Dose Methotrexate on Bone Density

ANN B. CRANNEY, ROBERT J. MCKENDRY, GEORGE A. WELLS, DAYLILY S. OOI, NORDAU D. KANIGSBERG, GUNNAR R. KRAAG, and C. DOUGLAS SMITH

ABSTRACT. ***Objective.*** High dose methotrexate (MTX) has been linked with bone loss in oncology patients. However, it is unclear whether longterm low dose MTX used in the treatment of inflammatory arthritis is associated with bone loss. We compared the effect of low dose MTX on bone density in prospectively recruited patients with rheumatoid arthritis (RA) and psoriasis/psoriatic arthritis (Ps/PsA).

Methods. Thirty RA patients and 30 Ps/PsA patients taking MTX were compared to controls not taking MTX (30 with RA, 27 Ps/PsA). Bone mineral density (BMD) of the radius, lumbar spine, trochanter, and femoral neck was measured using Lunar dual energy x-ray absorptiometry. Student t tests were used to detect differences in bone density (using Z scores) of the MTX group versus controls for both the RA and Ps/PsA groups. Analysis of covariance was used to examine for confounders including disease duration, disease activity, age, and sex.

Results. BMD of the radius/femoral neck/trochanter did not differ significantly between the MTX treated groups and controls when analyzed by Z scores. The mean difference between the MTX group and controls of the femoral neck was 0.040 (95% CI –0.40, 0.12) and 0.060 (95% CI –0.30, 0.15) for the RA and Ps/PsA groups, respectively. The absolute BMD of the lumbar spine (L2–L4) was higher in the RA MTX group than in controls. Analysis of covariance did not reveal an effect of study group on bone density.

Conclusion. This study suggests that low dose MTX does not have a negative effect on bone density, at either cortical or trabecular sites. (J Rheumatol 2001;28:2395–9)

Key Indexing Terms:
METHOTREXATE OSTEOPOROSIS ARTHRITIS BONE DENSITY

Methotrexate (MTX) is a folate antagonist commonly used for the treatment of malignancy and, more recently, in low doses to control inflammatory arthritis and psoriasis. Animal studies have revealed that MTX decreases osteoblast activity, but not the production of osteoblasts[1]. Scheven, *et al* have shown that MTX is a potent inhibitor of osteoblast proliferation but does not alter biochemical markers[2]. In another animal study, Suzuki, *et al* found that in rats with adjuvant induced arthritis, MTX at a dose of 3 mg/kg restored osteogenic activity to normal, resulting in an increase in periarticular bone mineral density (BMD) and suppression of arthritis[3].

From the Division of Rheumatology, the Division of Dermatology, and the Department of Pathology and Laboratory Medicine, Ottawa Hospital; and the Department of Epidemiology and Community Medicine, University of Ottawa, Ottawa, Ontario, Canada.

Supported by a research grant from Physicians' Services Incorporated Foundation. Dr. Cranney is the recipient of Scholarship 99112 from The Arthritis Society.

A. Cranney, MD, MSc; R.J. McKendry, MD; G.R. Kraag, MD; C.D. Smith, MD, Division of Rheumatology, Ottawa Hospital; G. Wells, PhD, Department of Epidemiology and Community Medicine, University of Ottawa; D.S. Ooi, MB, Department of Pathology and Laboratory Medicine; N.D. Kanigsberg, MD, Division of Dermatology, Ottawa Hospital.

Address reprint requests to Dr. A. Cranney, Ottawa Hospital — Civic Campus, Room 461, 737 Parkdale Avenue, Ottawa, Ontario K1Y 1J8, Canada.

Submitted November 15, 2000; revision accepted May 31, 2001.

At high doses, MTX used for treatment of childhood leukemia and breast cancer has been shown to cause osteopathy characterized by osteopenia and associated stress fractures of the lower tibia/fibula[4-6].

Our review of the literature on the effect of low dose MTX on the human skeleton revealed few case reports and cross sectional studies[7-10]. In postmenopausal women with either rheumatoid arthritis (RA) or psoriatic arthritis (PsA), increased stress fractures were seen in association with a loss of cortical bone density. One cross sectional study suggested that MTX may diminish cortical bone mass[11], while other similar studies failed to show a reduction in cortical or trabecular bone mass in comparison to controls[12-15]. Three prospective studies failed to show any loss of bone density with low dose MTX[16-18]. A limitation of all these studies is that there are many confounding variables including age, sex, disease duration, and disease activity that can directly affect bone health. RA is known to be associated with periarticular and generalized bone loss[19,20], and frequent use of corticosteroids in the management of arthritis is associated with bone loss in the trabecular region. With the increasing use of MTX in rheumatology, gastroenterology, and dermatology, there is a need to ascertain its effect on bone, especially in patients with RA who are already at increased risk for osteoporosis.

Our primary objective was to determine the effect of low

dose MTX by comparing BMD in patients taking MTX to disease controls not treated with MTX.

Our *a priori* hypothesis was that MTX induces osteopenia in patients with RA or psoriasis/psoriatic arthritis (Ps/PsA).

MATERIALS AND METHODS

A prospective survey consisting of 4 groups was conducted 1997–99 at Ottawa Hospital. The weekly dose of MTX ranged from 7.5 to 20 mg per week. Our goal was to recruit 30 patients to each of the 4 groups: (1) RA patients treated with low dose MTX, (2) RA patients never treated with low dose MTX, (3) Ps/PsA patients taking low dose MTX, and (4) Ps/PsA patients never treated with MTX. Patients who had been taking osteoporosis medication, other than calcium or vitamin D, or who had a history of myeloma or hyperparathyroidism were excluded. Patients taking steroids at a dose > 10 mg prednisone daily were excluded.

BMD of the lumbar spine (L2–L4), femoral neck, trochanter, and distal and proximal radius was measured using Lunar DXA. Femoral neck and proximal radius BMD were used to evaluate the effect on cortical bone. Biochemical variables evaluated included serum bone-specific alkaline phosphatase (BSAP), calcium, 25-hydroxyvitamin D_3, and parathyroid hormone (PTH).

Disease activity was assessed by the 28 joint count[21], and for the psoriasis patients the Psoriasis Area Severity Index (PASI) score was used[22]. We evaluated function using the Health Assessment Questionnaire (HAQ). Current dose of corticosteroid was expressed as an equivalent dose of prednisone.

The sample size was based on detecting an absolute difference in BMD of 0.138 g/cm^2. This difference was based on a study comparing MTX and no MTX in patients with primary biliary cirrhosis[11]; using an alpha of 0.05 and power of 80%, a sample size of 30 per group was determined.

Bone-specific alkaline phosphatase was measured using Hybritech's Tandem-R Ostase, 25-OH vitamin D_3 using Diasorin's 25-Hydroxyvitamin D ^{125}I radioimmunoassay, PTH using Diasorin's Intact PTH immunoradiometric assay, and serum calcium with Roche Diagnostics Roche/Hitachi 917 using the manufacturer's reagents.

Student t tests and 95% confidence intervals (CI) were used to assess differences between the MTX group and controls for both RA and Ps/PsA groups. Since matching patients for age and sex was difficult, we controlled for age and sex in the analysis. Analysis of covariance was used to examine various confounders including disease duration, age and sex. Further, multiple linear regression analysis was performed to estimate the effect of cumulative MTX on BMD (using Z scores) after adjustment for age, sex, and disease duration.

RESULTS

We recruited a total of 120 patients. One patient was excluded based on results that were consistent with hyperparathyroidism and 2 patients were excluded because they did not complete the bone density testing.

Tables 1 and 2 outline the demographic baseline characteristics of each of the 4 study groups. The groups were comparable in terms of disease related characteristics. There were differences in sex distribution and percentage postmenopausal, and we controlled for this in the analysis. All patients but one were Caucasian.

The mean disease duration was longer in the RA control patients, although this difference was not clinically significant. Disease duration was similar in the psoriasis MTX group and psoriasis controls. The mean weekly MTX dose was 10.1 mg in the RA group and 12.4 mg in the Ps/PsA group. The cumulative dose of MTX was higher in the Ps/PsA group than in the RA group (3571 vs 2810 mg). As shown in Tables 1 and 2, the duration of MTX use was similar for MTX RA and Ps/PsA patients (6.11 and 6.65 yrs, respectively).

There was no significant difference for absolute femoral neck BMD between MTX treated RA and RA controls (mean difference 0.040, 95% CI –0.40, 0.12) or between the 2 psoriasis groups (mean difference 0.060, 95% CI –0.30, 0.15). Similarly, there was no significant difference between MTX treated patients and controls at the femoral neck, trochanter, or radius for either PsA or RA groups (Tables 1 and 2). At the lumbar spine, the MTX treated RA patients did show a significantly higher BMD than controls, with a mean difference of 0.101 ($p = 0.029$). However, this was not seen with the Ps/PsA group. We also analyzed differences in Z scores between the groups for each bone density site, and found no significant difference between respective groups.

Due to the difference in menopausal status between RA MTX treated and RA controls, we stratified by menopausal status and carried out the Wilcoxon sign-rank test because of small numbers. Using this nonparametric test, a significant difference was found in Z scores of the femoral neck between the 2 RA groups when evaluating subjects who were postmenopausal ($p = 0.024$), with the women experiencing significantly higher BMD in the MTX group. There was no difference for women who were premenopausal. For the 2 psoriasis groups (MTX versus controls) there was a difference in the percentage of subjects undergoing hormone replacement therapy (HRT) and the percentage of women. When analyzed by Wilcoxon sign-rank test and stratifying according to HRT use or sex, there was no significant difference in bone density between MTX treated patients and controls.

No serum biochemical marker of calcium or bone metabolism showed a significant difference between the MTX treated and control groups. Serum BSAP, serum calcium, 25-hydroxyvitamin D_3, and intact PTH were not different between treatment and control groups.

Analysis of covariance was conducted using the covariates age, sex, disease activity, and disease duration; only age and sex revealed a significant relationship with BMD. Together, age and sex of these factors explained between 31% of variability of proximal forearm BMD in the RA group and 56% of variability in the psoriasis group. We adjusted for disease activity, duration, age, sex, and function using the HAQ. Age, sex, and the HAQ score were significant predictors of BMD.

The amount of variability in BMD of the femoral neck and trochanter (using Z scores) explained by cumulative MTX dose was 6% and 11%, respectively, using multiple regression analysis.

Table 1. Characteristics of RA patients.

Variable	RA Control (n = 30), Mean	SD	RA Taking MTX (n = 30), Mean	SD
Age, yrs	58.40	9.22	55.93	19.93
Duration of disease, yrs	17.30	12.07	13.93	8.42
Body mass index, kg/m^2	27.85	5.05	27.11	5.76
Current steroid (prednisone) dose, mg	5.15	4.07 (n = 10)	8.46	9.61 (n = 14)
Cumulative prednisone use, mean, mg	1264		3451	
% Currently on calcium (% vitamin D)	37 (20)		30 (16)	
Mean dietary calcium intake, mg	459.83		553.1	
% Female	67.0		77.0	
% Postmenopausal females	90.0		60.0	
% Females on HRT	35.0		31.8	
Years postmenopausal	11.07	9.83	15.1	11.42
Cumulative MTX, mg (range)	NA		2810 (540–8060)	
MTX duration, yrs (weekly dose, mg)	0		6.11 (10.08) 71	
% on folic acid				
Activity level, %				
Sedentary	20		10	
Mild	47		46	
Moderate	30		36	
Athletic	3		7	
28 Tender joint count	6.67	5.97	6.53	4.34
ESR, mm/h	26.0	25.42	20.17	19.67
HAQ score	1.01	0.79	1.07	0.70
BMD proximal radius, g/cm^2	0.63	0.14	0.65	0.11
Mean Z score	–0.90	1.41	–0.36	1.33
BMD trochanter, g/cm^2	0.76	0.20	0.80	0.14
Mean Z score	–0.30	1.53	0.07	1.36
BMD spine L2-L4, g/cm^2	1.08	0.18	1.18	0.17 $p < 0.02$
Mean Z score	–0.12	1.39	0.47	1.64
BMD femoral neck, g/cm^2	0.86	0.15	0.90	0.14
Mean Z score	–0.29	1.01	0.04	1.13
Bone-specific ALP, μg/l	11.67	4.75	12.07	5.35
Serum PTH, pmol/l	3.30	1.19	3.83	2.16
Serum testosterone, nmol/l	13.08	1.86	15.43	3.62
25 Hydroxy-vitamin D_3, nmol/l	62.83	23.03	65.23	24.73

DISCUSSION

This study suggests that MTX given in a continuous low dose has no negative impact on cortical or trabecular bone density. Our study agrees with recent findings that MTX does not have a deleterious effect on axial bone mass[12,13,16–18]. It is possible that MTX has a positive effect on bone through its effect on disease activity. El Miedany, *et al* evaluated the effect of low dose MTX on bone in patients with RA[23]. After 3 months of MTX, there was a significant decrease in bone resorption markers and an increase in bone formation markers, suggesting that low dose MTX may exert a protective effect on bone due to suppression of disease activity. MTX decreases production of the cytokine interleukin 1, which stimulates bone resorption[24].

Folinic acid supplementation has been shown to prevent MTX induced toxicity in osteoblast-like cells, and it may also act to prevent osteopenia in patients taking MTX[25].

Our study design has limitations, since it was not conducted in a longitudinal fashion. We assessed bone density and biochemical markers at only one time point. In addition, many of the subjects were postmenopausal women, which would limit the generalizability of our results. BMD is a surrogate outcome measure and we were not able to follow patients taking MTX over time to see if they developed more stress fractures than controls.

The bone density of the radius in patients with arthritis may be affected due to local inflammation at the wrist, resulting in periarticular osteopenia. However, we did not find significantly lower bone density of the trochanter or femoral neck regions in patients taking MTX.

Another potential limitation is that the majority of our subjects used lower doses of MTX than is common current practice. Physicians are now using larger weekly doses of MTX to treat inflammatory arthritis, and a prospective evaluation of BMD of patients taking larger cumulative doses of MTX would be an interesting area of further research.

We did not find a negative effect on bone density in arthritis patients taking low dose MTX in comparison to

Table 2. Characteristics of PsA patients.

Variable	Control (n = 27), 11 Psoriasis/16 PsA Patients		Taking MTX (n = 30), 10 Psoriasis/20 PsA Patients	
	Mean	SD	Mean	SD
Age, yrs	53.22	14.43	49.30	10.56
Duration of disease, yrs	17.74	11.88	18.5	14.66
Body mass index, kg/m^2	29.15	5.13	30.21	5.68
Current prednisone dose, mg	9.33	15.19 (n = 6)	7.56	4.52 (n = 8)
Cumulative prednisone use, mg	290		347	
% Taking calcium (% vitamin D)	8 (0)		23 (20)	
Mean dietary calcium intake, mg	459		438.72	
% Female	70.4		56.7	
% Postmenopausal females	63		64.7	
% Females on HRT	11		47	
Years postmenopausal	16.78	11.55	10.20	8.57
Cumulative MTX, mg (range)	NA		3571 (620–11,960)	
MTX duration, yrs (weekly dose, mg)	0		6.65 (12.42)	
% on folic acid			47	
Activity level, %				
Sedentary	4		10	
Mild	26		57	
Moderate	48		30	
Athletic	22		3	
28 Tender joint count	3.78	4.58	4.88	6.01
ESR, mm/h	21.40	23.65	23.31	24.50
HAQ score	0.47	0.55	0.55	0.69
BMD proximal radius, g/cm^2	0.70	0.09	0.72	0.12
Mean Z score	0.001	0.98	–0.17	1.05
BMD trochanter, g/cm^2	0.85	0.12	0.87	0.16
Mean Z score	0.14	0.93	0.05	1.15
BMD spine L2-L4, g/cm^2	1.22	0.13	1.26	0.15
Mean Z score	0.41	1.17	0.40	1.29
BMD femoral neck, g/cm^2	0.95	0.15	1.01	0.17
Mean Z score	–0.01	1.01	0.11	1.05
Bone-specific ALP, μg/l	12.38	3.87	12.61	5.63
Serum PTH, pmol/l	3.17	0.84	3.83	2.16
Serum testosterone, nmol/l	13.51	1.49	14.89	4.85
25 Hydroxy-vitamin D_3, nmol/l	71.04	34.52	80.83	33.93

those not taking MTX. However, we were not able to determine whether higher doses of cumulative MTX may have a negative effect on bone mineralization.

ACKNOWLEDGMENT

We thank Kathy Drouin, RN, for her assistance with this study.

REFERENCES

1. May KP, Mercill D, McDermott MT, West SG. The effect of methotrexate on mouse bone cells in culture. Arthritis Rheum 1996;39:489-94.
2. Scheven BA, van der Veen MJ, Damen CA, et al. Effects of methotrexate on human osteoblasts in vitro: modulation by 1,25-dihydroxyvitamin D_3. J Bone Miner Res 1995;10:874-80.
3. Suzuki Y, Nakagawa M, Masuda C, et al. Short term low dose methotrexate ameliorates abnormal bone metabolism and bone loss in adjuvant induced arthritis. J Rheumatol 1997;24:1890-5.
4. Schwartz AM, Leonidas JC. Methotrexate osteopathy. Skeletal Radiol 1984;11:13-6.
5. Stanisavljevic S, Babcock AL. Fractures in children treated with methotrexate for leukemia. Clin Orthop Rel Res 1977;125:139-44.
6. Bruning PF, Pit MJ, de Jong-Bakker M, van den Ende A, Hart A, van Enk A. Bone mineral density after adjuvant chemotherapy for premenopausal breast cancer. Br J Cancer 1990;61:308-10.
7. Singwe M, Legars L, Karneff A, Prier A, Kaplan G. Multiple stress fractures in a scleroderma patient on methotrexate therapy. Rev Rhum Engl Ed 1998;65:508-10.
8. Preston SJ, Diamond T, Scott A, Laurent MR. Methotrexate osteopathy in rheumatic disease. Ann Rheum Dis 1993;52:582-5.
9. Zonneveld IM, Bakker WK, Dijkstra PF, et al. Methotrexate osteopathy in long-term low dose methotrexate treatment for psoriasis and rheumatoid arthritis. Arch Dermatol 1996;132:184-7.
10. Maenaut K, Westhovens R, Dequeker J. Methotrexate osteopathy, does it exist? J Rheumatol 1996;23:2156-9.
11. Blum M, Wallenstein CJ, Luckey M. Effect of methotrexate on bone in postmenopausal women with primary biliary cirrhosis. J Bone Miner Res 1996;11:T545.
12. Ide M, Suzuki Y, Ichikawa Y, Mizushima Y. Influence of long-term low-dose methotrexate therapy on periarticular and generalized osteoporosis in rheumatoid arthritis. Jap J Rheumatol 1999;9:75-85.
13. Carbone LD, Kaeley G, McKown KM, Cremer M, Palmieri G,

Kaplan S. Effects of long-term administration of methotrexate on bone mineral density in rheumatoid arthritis. Calcif Tissue Int 1999;64:100-1.
14. Katz JN, Leboff MS, Wade JP, Brown EM, Liang MH. Effect of methotrexate on bone density and calcium homeostasis in rheumatoid arthritis [abstract]. Clin Res 1989;37:509A.
15. Bianchi ML, Bardare M, Cimaz R, Galbiati E, Corona F, Cherubini R. Does methotrexate have an effect on bone mass? [abstract]. J Bone Miner Res 1996;11:S225.
16. Buckley LM, Lieb ES, Cartularo KS, Vacek PM, Cooper SM. Effects of low dose methotrexate on the bone mineral density of patients with rheumatoid arthritis. J Rheumatol 1997;24:1489-94.
17. Pascaretti Ch, Perroux-Goumy L, Masson C, Legrand E, Bregreon C, Audran M. Methotrexate osteopathy in rheumatoid arthritis: a two year longitudinal and prospective study [abstract]. J Bone Miner Res 1996;11:S513.
18. Mazzantini M, DiMunno O, Incerti-Vecchi L, Pasero G. Vertebral bone mineral density changes in female rheumatoid arthritis patients treated with low-dose methotrexate. Clin Exp Rheumatol 2000;18:327-31.
19. Ferraccioli G, Casatta L, Bartoli E. Increase of bone mineral density and anabolic variables in patients with rheumatoid arthritis resistant to methotrexate after cyclosporin A therapy. J Rheumatol 1996;23:1539-42.
20. Dequeker J, Maenaut K, Verwilghen J, Westhovens R. Osteoporosis in rheumatoid arthritis. Clin Exp Rheumatol 1995;13 Suppl 12: S21-6.
21. Smolen JS, Breedveld FC, Eberl G, et al. Validity and reliability of the 28-joint count for the assessment of rheumatoid arthritis activity. Arthritis Rheum 1995;38:38-43.
22. Marks R, Barton SP, Shuttleworth D, et al. Assessment of disease progress in psoriasis. Arch Dermatol 1989;125:235-40.
23. El Miedany YM, Abubakr IH, El Baddini M. Effect of low dose methotrexate on markers of bone metabolism in patients with rheumatoid arthritis. J Rheumatol 1998;25:2083-7.
24. Suzuki Y, Tanihara M, Ichikawa Y, et al. Periarticular osteopenia in adjuvant arthritis: Role of interleukin-1 in decreased osteogenic and increased resorptive potential of bone marrow cells. Ann Rheum Dis 1995;54:484-90.
25. Preston SJ, Clifton-Bligh P, Laurent MR, Jackson C, Mason RS. Effect of methotrexate and sulphasalazine on UMR 106 rat osteosarcoma cells. Br J Rheumatol 1997;36:178-84.