CONCISE REPORT

Increase in bone mineral density of patients with spondyloarthropathy treated with anti-tumour necrosis factor α

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Objective: To determine the changes in bone mineral density (BMD) in patients with spondyloarthropathy (SpA) treated with infliximab.

Patients and methods: 29 patients (six women; 23 men) aged 22–68 years, with persistently active SpA despite a high dose of non-steroidal anti-inflammatory drug and/or treatment with methotrexate or sulfasalazine, were studied. Median duration of disease was 13 years (range 3–30). Twenty five patients were treated with 5 mg/kg and four with 3 mg/kg of infliximab at weeks 0, 2, 6 and then received either no infusion (n=3), or additional infusion of infliximab every other month (n=6), and the remainder received one infusion only in the case of a relapse. Lumbar and femoral BMD was measured by dual energy *x* ray absorptiometry at baseline and six months later. Serum osteocalcin and urinary deoxypyridinoline were measured in 19 patients at weeks 0, 2, 24, and in 13 patients at all visits.

Results: In six months there was a significant increase in BMD at the spine (3.6%, p=0.001), total hip (2.2%, p=0.0012), and trochanter (2.3%, p=0.0012). A trend for increase (1.1%) was observed at the femoral neck. There was an increase in osteocalcin between baseline and week 6 (third infusion)—median 1.45 µg/l (p=0.013). No change in marker of bone resorption was observed at the same time. There was no change in biochemical markers between baseline and final visits. There was a trend for a correlation between the decrease at six months in erythrocyte sedimentation rate, and lumbar spine BMD change (r_s=−0.35, p=0.06).

Conclusion: These data suggest that a benefit of anti-tumour necrosis factor α therapy on BMD in patients with SpA may be through an uncoupling effect on bone cells.

I t has been recognised for a long time that osteoporosis is commonly associated with ankylosing spondylitis (AS). Among possible mechanisms, inflammatory mediators released during the course of AS, and decreased mobility of patients, might have a role in the occurrence of osteoporosis, by directly affecting bone remodelling. Longitudinal studies in early AS have shown that spine and hip bone mineral density (BMD) decrease predominantly in patients with active disease.[1][2] This bone loss might be related to an increase in bone resorption, and correlations have been established between some biochemical markers of bone resorption on the one hand, and erythrocyte sedimentation rate (ESR), and serum C reactive protein (CRP) level on the other.[1][3] Among factors which can influence bone resorption and osteoclast activity, tumour necrosis factor α (TNFα) has a prominent role. This cytokine has been shown to mediate the increase of bone resorption both in systemic osteoporosis related to oestrogen deficiency,[4] and in periarticular or periprosthetic bone erosions. In a model of transgenic mice expressing soluble TNF receptor to neutralise TNFα, animals were protected from oestrogen deficiency related bone loss.[5] TNFα is also a powerful inhibitor of bone formation.[6] However, data on bone formation in AS are controversial, with some studies, but not others, showing a decrease in bone formation.[7][8]

Infliximab is a human/mouse neutralising chimeric monoclonal antibody of IgG1κ isotype with specificity and high affinity for TNFα. It has been successfully used in the treatment of rheumatoid arthritis, Crohn's disease, and spondyloarthropathy (SpA).[9–11] Thus this prospective study aimed at evaluating changes in BMD of patients affected with SpA, and treated with infliximab. Moreover, we investigated bone turnover by using biochemical markers of bone formation and resorption, and we studied the relationship between BMD, markers of bone turnover, and response to the treatment.

PATIENTS AND METHODS

Twenty nine patients (six women; 23 men) affected with SpA according to European Spondylarthropathy Study Group criteria, and requiring anti-TNFα therapy because of persistently active disease despite a high dose of non-steroidal anti-inflammatory drug and/or treatment with methotrexate or sulfasalazine, were prospectively included in this study. Patients receiving or having ever received bisphosphonates or hormone replacement therapy were excluded. Twenty three patients had SpA without associated condition (including 17 with inflammatory back pain as the leading symptom), two had psoriatic arthritis, and four inflammatory bowel disease. Informed consent for anti-TNFα treatment was obtained from all treated patients. Their age ranged between 22 and 68 years (median 35), and their disease duration between 3 and 30 years (median 13). Their body mass index was in the range of 17.3–36.7 kg/m² (median 24). One patient was menopausal at baseline. Five were taking calcium (1 g/day) and vitamin D (800 IU/day). Fourteen had received corticosteroids at a mean daily dose of 12.6 mg; four of them were still receiving steroids during the study (average 7.5 mg/day). Sixteen patients were receiving methotrexate during the course of the study at mean weekly dose of 11.4 mg (range (7.5–20). The activity and severity of the disease were assessed by a visual analogue scale for global pain, Bath Ankylosing Spondylitis Disease Activity Index (BASDAI), and Bath Ankylosing Spondylitis Functional

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Activity Index (BASFI). At baseline, the median (range) scores were respectively 70 (45–100), 57.6 (20–95), and 42 (2–96). At baseline median (range) ESR and CRP were respectively 41 mm/1st h (2–84) and 42 mg/l (5–116). ESR and CRP were assessed monthly during the study.

BMD (g/cm^2) was determined, at baseline and six months later, at the lumbar spine (second to fourth vertebrae, antero-posterior view) and the upper extremity of the left femur, by dual energy x ray absorptiometry (DXA) (QDR 2000, Hologic, Waltham, USA). For the femur the following sites were assessed: trochanter, femoral neck, and the whole femoral extremity (total hip). In one patient measurement of the second hip was not performed owing to a hip replacement. Quality control of the device was performed by daily measurement of a spine phantom.

T scores (number of standard deviations (SDs) from the normal mean obtained from young healthy adults) were calculated. Osteopenia was defined as a T score between −1 and −2.5 SD and osteoporosis as a T score ⩽−2.5 SD. References values have been previously published for women; for men, manufacturer's reference values were used. The mean T score was −1.99 (range −4.41 to 2.33) and −1.59 (range −5.18 to 1.11) at the lumbar spine and total femur, respectively. Seven patients were osteoporotic and 12 were osteopenic at either spine or femur.

At baseline, serum parathormone and 25-hydroxycholecalciferol D levels were 23.6 µg/l (4.1–32.9) and 23.9 µg/l (9–42.4), respectively. Serum osteocalcin (using a competitive enzyme linked immunosorbent assay (ELISA) method, normal values 5–25 µg/l) and urinary total deoxy-pyridinoline ((D-Pyr), by an ELISA method based on a mono-clonal antibody that recognises the free forms of D-Pyr cross links; normal values corrected for urinary creatinine levels 2–7 nmol/mmol) were measured in 19 patients at weeks 0, 2, 24, and in 13 patients at all visits. Median (range) of osteocalcin and D-Pyr/creatinine levels was 18.7 µg/l (6–32.7) and 7 nmol/mmol (0.1–10.2), respectively, at baseline. Serum osteo-calcin and D-Pyr/creatinine were increased in 6/19 (32%) and 10/19 (53%) patients, respectively. In 4/19 (21%) patients both D-Pyr/creatinine and osteocalcin were above the upper normal limit.

Twenty five patients received 5 mg/kg and four received 3 mg/kg of infliximab at weeks 0, 2, and 6. Subsequently, some of those patients received no infusion (n=3), some received one infusion every other month (n=6), and the remainder received one infusion only in the case of a relapse. After the first three infusions, all clinical parameters improved dramatically in all patients except one (data not shown). During follow up, a relapse was seen in 13 patients, with a mean duration of 3.7 months (range 2–4.5) after the third perfusion. In the whole group, the mean number of infusions was 4.5 (range 3–5).

Statistical analysis

We described changes during this six month follow up in BMD, osteocalcin, D-Pyr/creatinine, ESR, and CRP. Data were compared using the Wilcoxon signed rank sum test. Same comparisons were also performed between baseline and week 6. Although absolute changes were measured, data are presented as relative (%) changes. Correlations between BMD changes and changes in biochemical parameters were also tested using the Spearman correlation coefficient. Analyses were performed using SAS version 8.0 (SAS Institute, Cary, NC) and S-Plus 2000 (MathSoft Inc, Seattle, WA) software.

RESULTS

In six months a significant increase in BMD at the spine (mean (SD) 3.6 (5.9)%, p=0.001), total hip (2.2 (3.3)%, p=0.0012), and trochanter (2.3 (3.4)%, p=0.0012) was seen. A non-significant increase (1.1%, p=0.19) was seen at the

femoral neck. Mean (SD) change in body weight was 0.7 (2.6) kg (range −5 to 6) during the study. Comparable results were obtained in men and women, and in patients with or without methotrexate (data not shown). In the four patients receiving corticosteroids during the study, there was no change in BMD.

There was no statistically significant difference between baseline and six month values for osteocalcin and D-Pyr/creatinine. However, there was an increase in osteocalcin between baseline and week 6 (third infusion) (median 23.8 µg/l (6.3–66.1)); the median change was 1.45 µg/l (−2.10 to 42.10), p=0.013. This increase was not related to the baseline value (data not shown). D-Pyr/creatinine did not change during the same period.

Neither osteocalcin nor D-Pyr/creatinine changes correlated with the change in BMD. There was a decrease in ESR and CRP between baseline and the final visit (p=0.008 and p=0.005, respectively), and between baseline and the six week assessment (p<10^{-4} for both). There was a trend for a correla-tion between six month changes in ESR and lumbar spine BMD change (r_s=−0.35, p=0.06); the correlation coefficient between changes in CRP and lumbar spine BMD was of the same order (r_s=−0.28), although not significant.

DISCUSSION

In this study an increase in lumbar spine and femoral BMD was seen in patients with SpA, six months after initiation of infliximab. In our opinion, this increase was not due to confounding factors, such as syndesmophytes or interpedicu-lar ankylosis, which cannot explain the six months changes, additional treatments (only five patients received calcium and physiological doses of vitamin D supplements, none received treatments for osteoporosis), or drift of the DXA device (qual-ity control was performed daily). We checked the positioning of patients carefully before each scan; only slight changes might have occurred between the two scans, due to the decrease in spine and pelvic pain.

These results may be of clinical importance, and may increase the knowledge of mechanisms of SpA associated bone loss. The cause of SpA associated osteoporosis remains controversial. It has been suggested that local or systemic inflammatory cytokine release may be implicated in bone loss. A negative relation between serum osteocalcin and serum inflammatory parameters,[12] and a positive relation between bone resorption markers and serum inflammatory parameters,[3 8 12 13] have been described. In two longitudinal studies a decrease in BMD was seen only in patients with SpA with persistent active disease.[1 2] Finally, TNFα has been shown to increase bone resorption and decrease bone formation.[4 6] This study, in which inhibition of TNFα resulted in a decrease in systemic inflammation, and in an increase in bone density, adds some evidence in favour of the hypothesis of bone loss mainly due to systemic inflammation through direct effects of TNF on bone. However, it must be pointed out that only a trend for a correlation was seen between the six month changes in ESR and BMD, and no correlation was found between CRP and BMD changes. This result might have been due to low statistical power, or might have been different if inflammatory parameters had been evaluated throughout the follow up; but it suggests that other factors may explain the observed increase in BMD. Particularly, an increased mobility of patients might have been an additional cause for increased bone density. Immobility is a well known risk factor for bone loss, but some authors found low bone density in patients taking regular exercise, or did not find any relation between bone loss and baseline functional index or spine mobility.[2] However, owing to confounding factors, evaluation of the relationship between the level of physical activity and BMD remains difficult in SpA.

The changes in biochemical parameters of bone turnover and BMD were not correlated. Moreover, we did not observed

a change in measures of bone turnover at six months, as compared with baseline. Patients were given treatment infusions at baseline, two and six weeks, then according to various designs (including three with no further infusion). Thus, it is difficult to comment on the biological results obtained at the six months' determination. However, the results at week 6 were obtained on a more homogeneous group of patients, all having received infusion six and four weeks before. These results suggest an uncoupling effect on bone turnover, bone formation being increased, while bone resorption remained unchanged. In ovariectomised rats and mice, it has been shown that the administration of TNF binding proteins stimulated bone formation.[14] The mechanism of anti-TNFα induced bone formation might be increased osteoblast activity or recruitment of osteoblasts or activation of resting lining cells. Bone resorption, as assessed by D-Pyr/creatinine, did not decrease. However bisphosphonates, which are potent anti-osteoclastic drugs, and can cause an increase in BMD, have no effect on the urinary excretion of free D-Pyr. Whether peptide bound cross link markers may have different behaviour during anti-TNFα therapy needs further study. On the other hand, anti-TNF may have indirect effects on bone through the cytokine cascade. Some inflammatory cytokines, including TNFα, stimulate the production of osteoprotegerin, a potent inhibitor of osteoclast activity, by human bone marrow stromal cells.[15] Thus, the positive effects of inhibition of TNFα on bone resorption might be decreased by down regulation of osteoprotegerin. Additional results are needed to assess the bone cell effect of anti-TNFα therapy.

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