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The effect of raloxifene on bone marrow adipose tissue and bone turnover in postmenopausal women with osteoporosis

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ABSTRACT

In patients with postmenopausal osteoporosis low bone volume is associated with high bone marrow adipose tissue (MAT). Moreover, high MAT is associated with increased fracture risk. This suggests an interaction between MAT and bone turnover, however literature remains equivocal. Estrogen treatment decreases MAT, but the effect of raloxifene, a selective estrogen receptor modulator (SERM) registered for treatment of postmenopausal osteoporosis, on MAT is not known. The aim of this study is 1] to determine the effect of raloxifene on MAT and 2] to determine the relationship between MAT and bone turnover in patients with osteoporosis.

Bone biopsies from the MORE trial were analyzed. The MORE trial investigated the effects of raloxifene 60 or 120 mg per day versus placebo on bone metabolism and fracture incidence in patients with postmenopausal osteoporosis. We quantified MAT in iliac crest biopsies obtained at baseline and after 2 years of treatment ($n = 53$; age 68.2 ± 6.2 years).

Raloxifene did not affect the change in MAT volume after 2 years compared to baseline (placebo: 1.89 ± 10.84%, raloxifene 60 mg: 6.31 ± 7.22%, raloxifene 120 mg: −0.77 ± 10.72%), nor affected change in mean adipocyte size (placebo: 1.45 (4.45) μm, raloxifene 60 mg: 1.45 (4.35) μm, raloxifene 120 mg: 0.81 (5.21) μm). Adipocyte number tended to decrease after placebo treatment (−9.92 (42.88) cells/mm^2) and tended to increase during raloxifene 60 mg treatment (13.27 (66.14) cells/mm^2) while adipocyte number remained unchanged in the raloxifene 120 mg group, compared to placebo (3.06 (39.80) cells/mm^2, Kruskal-Wallis $p = 0.055$, post hoc: placebo vs raloxifene 60 mg $p = 0.017$). MAT volume and adipocyte size were negatively associated with osteoclast number at baseline ($R^2 = 0.123$, $p = 0.006$ and $R^2 = 0.098$, $p = 0.016$ respectively). Furthermore adipocyte size was negatively associated with osteoid surface ($R^2 = 0.067$, $p = 0.049$). Finally, patients with vertebral fractures had higher MAT volume (50.82 (8.80)%) and larger adipocytes (55.75 (3.14) μm) compared to patients without fractures (45.58 (12.72)% $p = 0.032$, 52.77 (3.73) μm $p = 0.004$ respectively).

In conclusion, raloxifene did not affect marrow adipose tissue, but tended to increase adipocyte number compared to placebo. At baseline MAT volume and adipocyte size were associated with bone resorption, and adipocyte size was associated with osteoid surface, suggesting an interaction between bone marrow adipocytes and bone turnover. In addition, we found that high MAT volume and larger adipocyte size are associated with prevalent vertebral fractures in postmenopausal women with osteoporosis, indicating that adipocyte size affects bone quality independent of bone volume.

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Abbreviations: MAT, marrow adipose tissue; BMD, bone mineral density; MSC, mesenchymal stem cell; SERM, selective estrogen receptor modulator; MORE, Multiple Outcomes of Raloxifene Evaluation; RANKL, receptor activator of nuclear factor κ-B ligand.

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1. Introduction

High bone marrow adipose tissue (MAT) is associated with low bone mineral density (BMD) and MAT is increased in patients with osteoporosis [1–3]. MAT is also associated with vertebral fractures [4,5], independently of BMD in some studies [6].Two potential mechanisms for the association between MAT and bone have been proposed. First, mesenchymal stem cells (MSCs) may differentiate into either an adipocyte or an osteoblast [7]. Therefore, a shift in the lineage allocation of the MSCs towards the adipocyte would decrease the number of osteoblasts and thus bone formation. Secondly, in vitro studies have shown that adipocytes may directly influence osteoblast and osteoclast differentiation and function, through secretion of adipokines and free fatty acids [8–12], suggesting a direct effect of MAT on bone turnover. Indirect evidence for this hypothesis comes from animal studies showing a positive correlation of marrow adiposity with bone resorption [13], and a negative correlation with bone formation [14] in ovariectomized rats. Clinical studies have indicated a negative association between MAT and bone formation in healthy premenopausal women, but MAT was not associated with bone formation or resorption in premenopausal women with osteoporosis [15]. In contrast, in elderly men and women with hip fractures MAT was negatively associated with bone resorption but not with bone formation [16]. Altogether, clinical studies on the association between MAT and bone turnover are heterogeneous and have shown different results.

Hormone replacement therapy in postmenopausal women effectively improves bone mass by decreasing bone turnover and decreasing MAT [17]. However, estrogen combined with medroxyprogesterone, is no longer recommended as a treatment for postmenopausal osteoporosis due to the increased risk of breast cancer and venous thromboembolism [18,19]. Raloxifene, a selective estrogen receptor modulator (SERM), has been developed as an osteoporosis therapy to specifically relay the beneficial effects of estrogen on bone, without the adverse side effects [20]. In vitro raloxifene increased lipid deposition in differentiating 3T3-L1 cells [21] and in vivo, in rats, raloxifene decreased MAT [22]. Clinical data on the effect of raloxifene on MAT are lacking.

The first aim of this study is to determine the effect of raloxifene on MAT in patients with postmenopausal osteoporosis. We hypothesized that raloxifene decreases MAT, similar to estradiol [17]. The second aim is to explore the association between MAT, bone volume, bone turnover and vertebral fracture in postmenopausal women with osteoporosis and the interaction with raloxifene treatment. We hypothesized that MAT is inversely associated with histomorphometric measures of bone volume and bone turnover and that MAT is increased in vertebral fracture patients.

2. Materials and methods

2.1. Study design and subjects

The present study analyzed marrow adiposity in bone biopsies previously obtained for an ancillary histomorphometry study [23] of the MORE trial, conducted between 1994 and 1999 to examine the effect of raloxifene on bone mineral density and fracture risk [20]. The MORE study was a multi-center, double-blind, randomized, placebo-controlled clinical trial. In the main study, 7705 postmenopausal women with osteoporosis were randomly assigned to treatment with placebo, 60 mg or 120 mg of raloxifene hydrochloride daily in addition to daily supplements of 500 mg of calcium and 400–600 IE of vitamin D during 36 months. Of the total, 88 participants were included in the ancillary bone histomorphometry study which was conducted at two centers in the US and two centers in Europe and included bone biopsies before start of treatment and after 24 months of treatment.

Eligible women were at least two years postmenopausal and had osteoporosis, defined by either femoral neck or lumbar spine bone mineral density (BMD) T-score < -2.5 (study group 1) or a] one or more moderate to severe (25–40% reduction from expected vertebral height) or two or more mild (20–25% reduction from expected vertebral height) vertebral fractures and low BMD or b] two or more moderate to severe vertebral fractures regardless of BMD (study group 2). Exclusion criteria were severe or long-term disabling conditions; metabolic bone diseases; endocrine conditions requiring hormonal therapy (except stable hypothyroidism and type 2 diabetes mellitus); use of systemic estrogen, progestogen, or androgen during the previous 6 months; a known, suspected, or history of breast cancer, endometrial cancer, or abnormal uterine bleeding; thromboembolic events or stroke during the past 10 years; any type of cancer besides superficial skin cancer in the previous 5 years; active renal lithiasis; abnormal hepatic function or consumption of more than four alcoholic drinks per day.

All subjects provided written informed consent. Approval was obtained from all local institutional review boards. The trial was registered (clinicaltrials.gov NCT 00670319).

2.2. Measurements

2.2.1. Histomorphometry

Bone biopsies were obtained at baseline and after 24 months of treatment by a transverse biopsy from the anterior iliac crest following double tetracycline fluorescent labeling. Biopsies were cut with a Jung microtome into 5–8 μm sections and stained with Goldner's trichrome stain or tartrate-resistant acid phosphatase stain. The biopsy procedure and handling of the samples and bone histomorphometric measurements have been described in more detail previously [23]. All assessments of the sections were performed blinded to the treatment assignment and to the timing of the biopsies (baseline or post-treatment).

2.2.2. Marrow adiposity parameters

For the adipocyte parameters, we used sections with Goldner's trichrome staining. The following MAT outcome parameters were measured and calculated: 1] MAT volume as a percentage of the tissue volume (total adipose tissue volume: Ad.V/TV; %), 2] MAT volume as a percentage of the marrow volume (marrow adipose tissue volume: Ad.V/Ma.V; %), 3] mean adipocyte diameter (Ad.Dm; μm), calculated using the formula: $2^{*}\sqrt{\frac{Ad.Ar/N}{\pi}}$, assuming that all adipocytes are essentially circular, representing adipocyte size, 4] adipocyte density (Ad.Dn; cells/mm^2 marrow area) representing adipocyte number. These measurements were performed by semi-automatically tracing out individual adipocytes 'ghosts' in all the fields analyzed. Adipocyte ghosts appear as distinct, translucent, yellow ellipsoids in the marrow space. A standardized area in the secondary spongiosa was measured in 1–4 sections per biopsy including a mean marrow area of 15.24 mm^2 (range 2.88–62.83 mm^2; SD $\pm$ 6.82 mm^2) per biopsy. A watershed algorithm was used to separate the individual adipocytes. Fields were captured using a Nikon Microscope (Eclipse E 800), a DS-U1 camera (Nikon) and NIS Elements software (version 2.34, Nikon) at 40× magnification. Adipocyte analysis was performed using a semi-automated measurement program on ImageJ [24] based image analysis software adapted from the OsteoidHisto package [25]. All assessments of the sections were performed by examiners who were blinded to the treatment assignment and to the timing of the biopsies (baseline or post-treatment).

2.2.3. Bone turnover parameters

Cancellous bone volume as percentage of tissue volume (BV/TV; %), osteoid surface as a percentage of the total bone surface (OS/BS; %), number of osteoclasts per tissue area (N.Oc/T.Ar, in number of cells/mm^2) and bone formation rate (BFR/BV; referenced to bone volume, in %/year) were measured as secondary outcomes according to the guidelines of the ASBMR nomenclature committee [26] and were published previously [23].

2.2.4. Bone mineral density and vertebral fracture status

Lumbar spine and total hip BMD were measured by dual-energy x-ray absorptiometry (DXA) (Hologic, Bedford, MA, USA or Norland, White Plains, NY USA). A conventional radiography of the spine was obtained at baseline, and assessed for vertebral fractures by two radiologists [20].

2.3. Statistical analysis

The statistical analysis was performed with IBM SPSS Statistics for Windows (version 24; SPSS Inc., Chicago, IL, USA). The mean and standard deviation (±SD) or the median and interquartile range (IQR) are reported depending on the distribution of the data. To compare the treatment groups at baseline and to assess the effect of treatment on changes in MAT parameters, ANOVA or Kruskall-Wallis tests were used depending on the distribution of the data. To compare treatment groups (post hoc) and correct for multiple testing, we used Tukey tests or Mann-Whitney *U* tests. To assess the association between MAT parameters and bone turnover at baseline and follow up, linear regression models were used. At baseline, only MAT parameters were included as predictors. At follow up, MAT parameters, treatment and their interaction were included as predictors. In case assumptions were violated (normally distributed residuals, equal variances), the outcome variable was rank transformed. To compare MAT and bone parameters in patients with and without vertebral fractures at baseline, independent *t*-tests or Mann-Whitney U tests were used depending on the distribution of the data. All statistical tests were two-sided and a *p*-value of 0.05 was considered significant.

3. Results

3.1. Subjects

In the MORE trial ancillary histomorphometry study, 88 subjects were included at baseline and 65 paired biopsies were available for bone parameter analysis at 24 months. In the present study, 59 biopsies at baseline and 53 paired biopsies after 24 months were of sufficient quality to analyze the bone marrow adipocyte parameters.

As expected from randomization, no differences were observed between the treatment groups at baseline. Table 1 shows the baseline characteristics of the subjects. BMD and bone histomorphometric changes have been previously reported [23], Table 2 shows the changes in bone variables in the subset of patients specifically analyzed in this study.

3.2. Effect of raloxifene on marrow adiposity

2 years of raloxifene treatment did not affect the change in MAT volume compared to baseline, neither when expressed as a percentage of the marrow volume (Ad.V/Ma.V; mean ± SD change per group: placebo 1.89 ± 10.84%, raloxifene 60 mg 6.31 ± 7.22%, raloxifene 120 mg −0.77 ± 10.72%); nor when expressed as percentage of the tissue volume (Ad.V/TV; mean ± SD change per group: placebo 1.35 ± 10.61%, raloxifene 60 mg 6.25 ± 7.45%, raloxifene 120 mg −0.13 ± 8.97%) compared to placebo ($p = 0.138$ and $p = 0.137$ respectively; Fig. 1A). Also adipocyte size (Ad.Dm) remained unchanged after treatment (median (IQR) change per group: placebo 1.45 (4.45) μm, raloxifene 60 mg 1.45 (4.35) μm and raloxifene 120 mg 0.81 (5.21), $p = 0.301$; Fig. 1B). Adipocyte number (Ad.Dn) tended to increase after raloxifene 60 mg (median Ad.Dn (IQR): 13.27 (66.14) cells/mm^2) compared to placebo (median Ad.Dn (IQR): −9.92 (42.88) cells/mm^2) whereas adipocyte number remained unchanged after raloxifene 120 mg (median Ad.Dn (IQR): 3.06 (39.80) cells/mm^2, $p = 0.055$, post hoc: placebo vs raloxifene 60 mg $p = 0.017$; Fig. 1C).

Table 1
Baseline characteristics.

	Placebo	Raloxifene 60 mg	Raloxifene 120 mg
N	26	17	16
Biopsy at 24 months (N)	22	16	15
Age (years)	68 ± 6	70 ± 6	68 ± 7
Years postmenopausal (years)	18 ± 6	23 ± 8	20 ± 9
BMD lumbar spine (g/cm^2)	0.806 ± 0.127	0.812 ± 0.131	0.841 ± 0.105
BMD total hip (g/cm^2)	0.716 ± 0.070	0.698 ± 0.088	0.713 ± 0.103
Vertebral fracture (N)	9	6	4
Bone volume (BV/TV, %)	18 ± 6	19 ± 4	16 ± 5
Osteoid surface (OS/BS, %)	8 (11)	8 (6)	14 (13)
Osteoclast number (N.Oc/T.Ar, cells/mm^2)	0.65 ± 0.32	0.88 ± 0.58	0.89 ± 0.58
Bone formation rate (BFR/BV, %/y)	23 (23)	24 (28)	31 (27)
Total adipose tissue volume (Ad.V/TV, %)	41 ± 7	36 ± 7	39 ± 8
Marrow adipose tissue volume (Ad.V/Ma.V, %)	50 ± 8	44 ± 7	47 ± 8
Adipocyte size (Ad.Dm, μm)	54 ± 4	53 ± 4	54 ± 3
Adipocyte number (Ad.Dn, cells/mm^3 Ma.V)	218 ± 34	202 ± 25	205 ± 20

Baseline characteristics of study patients per treatment group. Mean ± standard deviation or median (IQR).
N = number of subjects; BMD = bone mineral density.

3.3. Marrow adiposity and bone turnover

At baseline, higher MAT volume and smaller adipocytes were both associated with lower osteoclast number ($R^2 = 0.123$, $B = -0.022$, $p = 0.006$, Fig. 2A and: $R^2 = 0.098$, $B = -0.040$, $p = 0.016$ respectively). Furthermore, larger adipocytes were associated with lower osteoid surface ($R^2 = 0.067$, $p = 0.049$; Fig. 2C), but MAT was not associated with bone formation rate ($R^2 = 0.036$, $p = 0.166$).

After 2 years raloxifene treatment, MAT volume and adipocyte size were no longer associated with osteoclast number ($R^2 = 0.013$, $p = 0.502$, Fig. 2B and $R^2 = 0.009$, $p = 0.634$, respectively). Adipocyte size was no longer associated with osteoid surface ($R^2 = 0.038$, $p = 0.325$, Fig. 2D). No significant effects of treatment nor significant interaction effects were found for the associations between MAT parameters and bone turnover parameters at follow-up.

3.4. Vertebral fracture and marrow adiposity

At baseline, MAT volume was higher (Ad.V/Ma.V: 50.82 (8.80)%; Ad.V/TV: 43.11 (8.35)%) and adipocytes were larger (55.75 ± 3.14 μm) in patients with vertebral fractures independent of BMD, compared to patients without vertebral fractures and a T-score < −2.5 (Ad.V/Ma.V 45.58 (12.72)%, $p = 0.032$; Ad.V/TV: 37.17 (11.77)%, $p = 0.042$; Ad.Dm: 52.77 ± 3.73 μm, $p = 0.004$; Fig. 3). There were no differences in bone volume or lumbar spine BMD between de patients with and without vertebral fractures (data not shown).

Table 2
Changes in bone parameters.

N	Placebo	Raloxifene 60 mg	Raloxifene 120 mg
	22	16	15
ΔBMD lumbar spine (g/cm^2)	0.002 ± 0.043	0.021 ± 0.038	0.014 ± 0.033
ΔBV/TV (%)	0 ± 7	−2 ± 6	−1 ± 4
ΔBFR/BV (%/y)	−7 ± 17	−8 ± 16	−13 ± 21
ΔN.Oc/T.Ar (cells/mm^2)	0.18 (0.49)	−0.11 (0.53)	−0.01 (1.21)

Mean changes ± standard deviation or median changes(IQR) in bone parameters in the subset of patients analyzed in this study. N = number of subjects, BMD = bone mineral density.

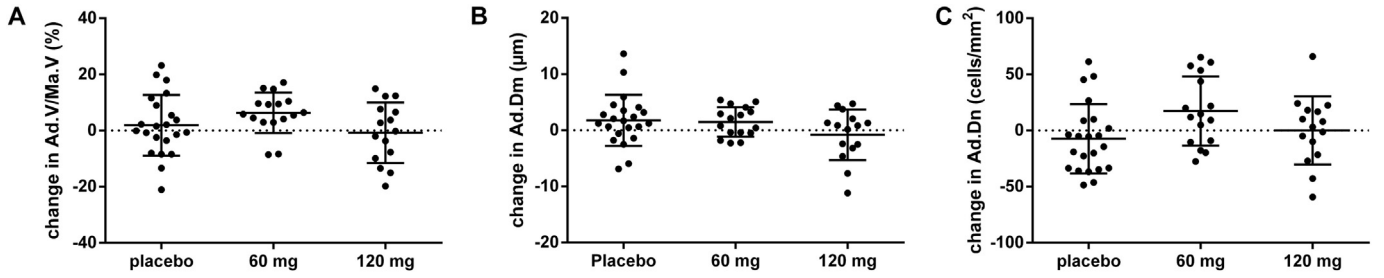

Fig. 1. Changes in A) MAT volume, B) adipocyte size and C) adipocyte number during the 24 months study period. Trend for adipocyte density ($p = 0.055$); post hoc: placebo vs raloxifene 60 mg $p = 0.017$.

4. Discussion

The present study showed that raloxifene treatment had no effect on marrow adipose tissue volume or adipocyte size, however it tended to increase the number of marrow adipocytes. At baseline, both marrow fat content and adipocyte size were inversely associated with osteoclast number. Adipocyte size was inversely associated with the bone formation parameter osteoid surface, but none of the MAT parameters were associated with bone formation rate. Patients with prevalent vertebral fractures had higher MAT and larger marrow adipocytes compared to postmenopausal women with osteoporosis without vertebral fractures.

4.1. Effects of Raloxifene on MAT in postmenopausal osteoporosis

In the current study, raloxifene did not decrease MAT volume and had no effect on adipocyte size, however, raloxifene tended to increase adipocyte number when comparing raloxifene 60 mg to the placebo treated group. Previous research by Syed et al. showed that estradiol decreased MAT volume by both a decrease in adipocyte size and number in postmenopausal women with osteoporosis compared to placebo [17]. While both studies included postmenopausal women, there were some methodological differences. First, our subjects were on average 5 years older (69 versus 64 years). Second, the effect of estradiol was evaluated after 1 year, while raloxifene treatment lasted for 2 years. Third, all subjects in the Syed paper had one or more vertebral fractures and low BMD, while the subjects in the MORE trial had either low BMD without vertebral fractures or vertebral fractures regardless of BMD. As only 17 patients, divided over the 3 treatment groups, had vertebral fractures in our study, groups were too small for further analysis. In these 17 subjects, MAT increased after 2 years of treatment in the placebo and the raloxifene 60 mg group, and MAT decreased after 2 years of raloxifene 120 mg ($n = 4$). A fourth methodological difference was found in vitamin D supplementation. While it was discontinued 6 months prior to the start of the trial in the subjects of the Syed paper, all subjects in the MORE trial started vitamin D and calcium supplementation at the beginning of the trial. However, it is unlikely that the vitamin D supplementation is the cause of the difference between the two studies since, in mice, vitamin D was shown to decrease MAT [27]. Thus vitamin D supplementation would rather mask, than cause a difference between the two studies. A last notable difference between the two studies is the increase in MAT in the placebo groups. As in the Syed paper MAT showed a relative increase of approximately 20% in

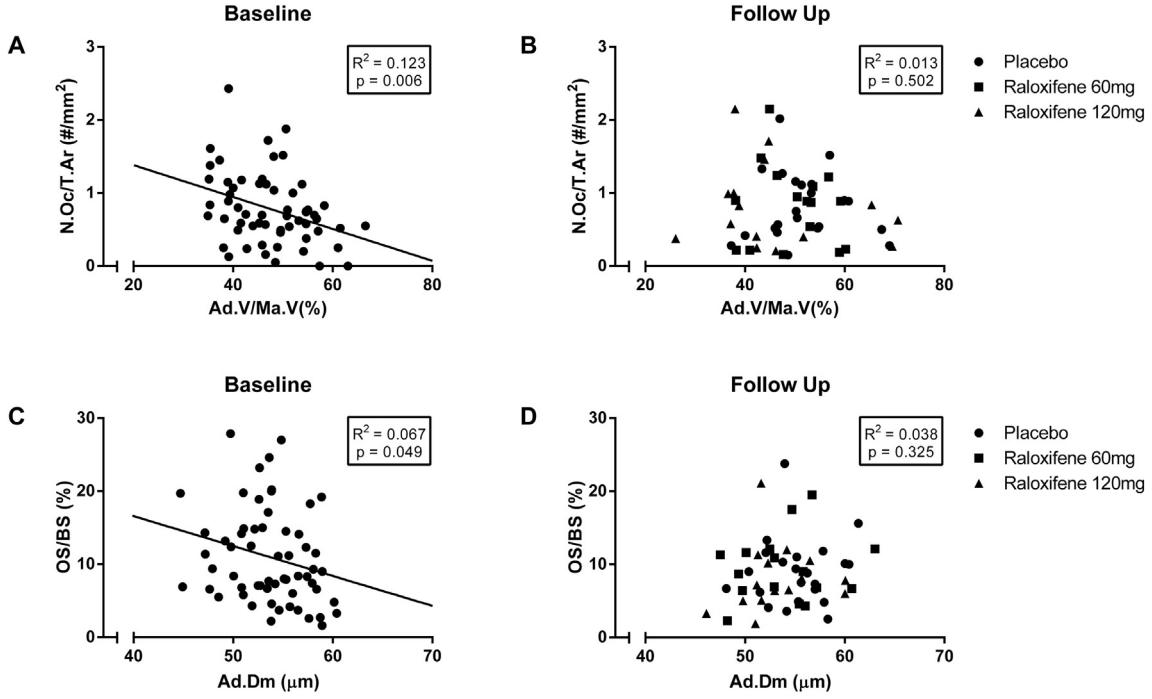

Fig. 2. The association between MAT parameters and bone turnover. A) Significant negative association between MAT volume and osteoclast number at baseline, but not at follow up (B). C) Significant negative association between adipocyte size and osteoid surface at baseline, which was no longer significant after treatment (D).

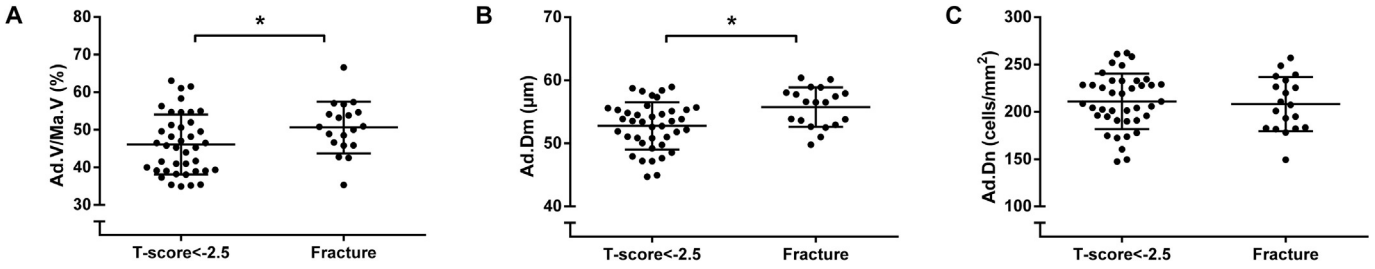

Fig. 3. A) MAT volume was higher and B) adipocyte size was larger in patients with vertebral fractures at baseline compared to patients with low BMD (T-score < −2.5) without vertebral fractures. C) Adipocyte number was similar in both groups.

1 year versus 4% in 2 years in our placebo group. Longitudinal studies on yearly increases in MAT in postmenopausal osteoporosis are lacking, but in heathy subjects, MAT increases approximately 1% per year [28,29].

Apart from methodological differences, raloxifene could have a different effect than estradiol in specific tissues. A possible explanation for this different effect could come from the complex interactions between raloxifene, estrogen receptors, coactivators and corepressor proteins [30–32] within the heterogeneous cell types in the bone microenvironment. The results of previous preclinical research on the effects of raloxifene on MAT are inconsistent. Murase et al. showed that, in vitro, raloxifene dose dependently increased lipid deposition in differentiating 3T3-L1 cells [21]. But since MAT appears to be a distinct adipose tissue [33,34], it is unknown if the results also apply on MAT. In vivo research by Somjen et al. showed that MAT decreased after raloxifene treatment in ovariectomized rats [22]. Since these rats were ovariectomized during growth and MAT was quantified directly under the growth plate, thus including the primary spongiosa, the results could be related to modeling activity rather than remodeling activity, which makes extrapolation to the clinical situation of postmenopausal osteoporosis difficult.

Raloxifene did not show a dose dependent effect on MAT parameters; on MAT volume and adipocyte size raloxifene 60 and 120 mg seem to have contradictory effects. These different effects of 60 and 120 mg of raloxifene were also observed on bone parameters [23]. A possible explanation for this difference could be receptor kinetics, for example desensitization or saturation, but this has never been investigated so far.

4.2. MAT and bone turnover before and after raloxifene treatment

Our results show a negative association between MAT volume and bone resorption in postmenopausal women with osteoporosis. Several reports have indicated that bone marrow pre-adipocytes express receptor activator of nuclear factor κ-B ligand (RANKL) which stimulates osteoclast differentiation and activation [35,36]. RANKL could therefore be a mediator between marrow adipocytes and bone resorption.

Furthermore, marrow adipocyte size was negatively associated with bone resorption and with osteoid surface, suggesting a role for adipocyte size in bone turnover. Previous research showed that large subcutaneous adipocytes secrete more adipokines with a shift towards pro-inflammatory adipokines than smaller adipocytes [37]. It is unknown if large bone marrow adipocytes also secrete more pro-inflammatory adipokines than smaller ones. But this could mediate the association between marrow adipocyte size and bone turnover. A second possible explanation could be lipid transfer from marrow adipocytes to osteoclasts and osteoblasts. A recent study on the interaction of marrow adipocytes with leukemia cells in the bone marrow, showed that adipocytes may transfer their lipids to leukemic cells [38] supporting their maintenance. Whether marrow adipocytes are also capable of transferring lipids to bone cells and what their role is, remains to be determined. On the other hand, in vitro research showed that fatty acids secreted by subcutaneous adipocytes can decrease osteoblast differentiation and function [8–10] and increase osteoclast formation [12]. Since the size of an adipocyte is mainly determined by its lipid content, these lipids might exert the negative effect on bone cells.

We measured osteoclast number (N.Oc/T.Ar) as a measure of bone resorption, although osteoclast number does not provide information on osteoclast function, which is a limitation of histomorphometric studies in general. Another histomorphometric measure of bone resorption is eroded surface, but we did not choose to use this parameter in this study due to greater inter-observer variability compared to osteoclast number [39]. Moreover, resorption pits were scarce in these biopsies and therefore were not representative of the overall resorption rate [23].

4.3. MAT and vertebral fracture

We showed, for the first time, that besides MAT volume, adipocyte size is associated with vertebral fractures; vertebral fracture patients have larger marrow adipocytes compared to patients without vertebral fracture. Interestingly, there was no difference in bone volume or BMD between the patients with and without fracture, possibly indicating that the negative effect of the larger marrow adipocytes could be independent of bone volume and BMD and suggesting a role for MAT in bone quality. Previous studies showed that high MAT is associated with vertebral fractures [4–6]. These studies used magnetic resonance imaging (MRI) techniques to quantify MAT, consequently marrow adipocyte size and number could not be determined [4–6].

A possible explanation for increased marrow adipocyte size in patients with vertebral fractures could be contributed to differences in secretion of pro-inflammatory adipokines secreted by different sizes of marrow adipocytes, as occurs in larger subcutaneous adipocytes [37]. Again, it is unknown if this also applies for marrow adipocytes, but it could indicate that smaller adipocytes have a more favorable profile of adipokines secreted locally and possibly exerting a positive influence on bone metabolism [8–11]. Accordingly, potent anti-osteoporosis therapies such as estrogen, teriparatide and bisphosphonates also decrease marrow adipocyte size, accompanied by an increase in BMD and a decrease in fracture risk [17,40,41]. Thus it seems that decreasing marrow adipocyte size has a bone-favorable effect.

5. Conclusions

This study shows that raloxifene does not decrease marrow adipose tissue and that it tended to increase adipocyte number in postmenopausal women with osteoporosis. MAT volume was only associated with bone resorption, while adipocyte size was associated with both bone resorption and bone formation, which may point to an effect of the marrow adipocytes on bone turnover. Finally, we showed for the first time that the higher MAT volume in patients with vertebral fractures compared to patients without vertebral fractures was associated with larger bone marrow adipocytes, which supported the tenet that

larger adipocytes indeed have a negative effect on bone cells, possibly by secreting adipokines or fatty acids. This will be an important question for further research, since targeting adipocyte size might represent a future therapy for bone disease and fracture prevention.

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Author contributions

KB, AV, PB and NB designed the study; KB, AO, SO, RH and PL acquired the data; KB, AV and MT analyzed the data; KB, AV, PB and NB interpreted the data; KB, AV, PB and NB drafted the manuscript; all authors critically revised the manuscript and approved the final version of the manuscript to be submitted.

Conflicts of interest

The authors declare no conflicts of interest.

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