ABSTRACT

Objective: Osteoporosis is the pathological reduction of bone mineral density (BMD) and the most represented metabolic skeletal disease among population. A reduction in bone mineralization levels is associated with an increased risk of frailty fractures and of healthcare costs. Although there are many evidences bridging rheumatological diseases (such Systemic Sclerosis, Lupus Erythematosus Systemics and Rheumatoid Arthritis) with bone loss, very scarce and contradictory papers evaluate bone health in primary Sjögren’s syndrome (pSS). Aim of this retrospective study is to evaluate BMD in pSS and its relationship with inflammatory markers, Ro/SSA and La/SSB antibodies.

Methods: Fifty-three postmenopausal pSS were matched with 93 controls and studied for BMD measured by Dual Energy X-ray absorptiometry (DXA). Anti-nuclear antibodies (ANA), anti-extractable nuclear antigens (ENA), Anti-Ro/SSA, and anti-La/SSB antibodies, erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) were collected too. Mean BMDs of each explored region was compared and osteoporosis’s (OP) prevalence was assessed. Pearson’s analysis and multivariate regression models were built to highlight variables interrelations.

Results: BMD was lower in pSS compared to controls, both expressed as g/cm² and T-score considering lumbar spine and femoral neck. OP prevalence was higher among pSS compared to controls. Weight acted as the best predictor of lumbar BMD in multivariate model. No difference on BMD status was found between pSS with antibodies positivity and altered inflammatory markers with subjects displaying normal sera levels of the abovementioned variables.

Conclusions: pSS female patients in an early stage of disease have lower BMD compared to healthy controls. Anti-Ro/SSA and anti-La antibodies and inflammatory markers are not related with BMD.

Keywords: Osteoporosis; Sjögren’s syndrome; Autoantibodies; Ro-SSA; La-SSB; Bone health; Bone Mineral Density (BMD)

INTRODUCTION

Primary Sjögren Syndrome (pSS) is a chronic inflammatory autoimmune Connective Tissue Disease (CTD) of unknown etiology which is characterized by CD4 +T Helper, B cells, macrophages, and dendritic cells infiltration of target organ. Although pSS mainly involves lacrimal and salivary glands, resulting in xerostomia and xeroftalmia, many organs and system may be interested and should therefore be considered a systemic disease [1]. Osteoporosis (OP) is the most common disease affecting bone tissue and it is characterized by reduced BMD, as result of an uncoupling in bone turnover, escalating the risk of frailty and traumatic fractures [2]. Several literature evidences documented the existing association between inflammatory burden, immune dysfunction and bone loss[3-5].

Even though an altered skeletal homeostasis is widely reported in association with CTDs such as Systemic Lupus Erythematous and Systemic Sclerosis, as well as in Rheumatoid Arthritis (RA), only few papers investigated the association between osteoporosis and pSS [6-12]. Aim of this study is to evaluate BMD and prevalence of osteoporosis in pSS patients in comparison to an age-gender matched control group and to asses if Ro/SSA and La/SSB
positivity, as well as high levels of inflammatory markers, may be related to a pathological bone mineralization.

MATERIALS AND METHODS

Study population

We retrospectively evaluated BMD of 53 postmenopausal women diagnosed with pSS according to American College of Rheumatology/European League against Rheumatisms (ACR/EULAR) 2016 classification criteria in our department[13]. Exclusion criteria were the presence of any demineralizing comorbidity, such as other CTDs, diabetes, asthma, chronic obstructive pulmonary disease, malignancies, early menopause, use of corticosteroids (CS), heparin, and vitamin D. Considered cases were postmenopausal women of 55.31 (± 10.2 SD) years old with a BMI (kg/m^2) of 25.45 (± 4.1 SD). Seven of our patients were in therapy with hydroxychloroquine 400 mg per day, 12 had hypertension, 1 diabetes and 1 fibromyalgia. Mean erythrocyte sedimentation rate was 48 mm/h (± 7.9 SD) and mean C-reactive protein levels were 2 mg/dl (± 0.41 SD). We considered only women whose Dual Energy X-ray absorptiometry (DXA) was performed within 6 months from diagnosis.

Control group

Ninety-three controls were postmenopausal women of average 58.61 years old (± 3.5 SD) and had an average BMI (kg/m^2) of 24.76 (± 5.2 SD). Their BMD was evaluated by DXA in our Department for age-related issues and used as control. None of them reported significant comorbidities or were in therapy with any drug. There were no significant differences in age and anthropometric features with patient group.

Methods

Detailed medical history, weight and height were collected from patients and their sera were tested for autoimmunity with standard techniques: Anti-Nucleus Antibodies (ANA) were detected by indirect immunofluorescence on Hep2 cells (Euroimmun, Lubeck, Germany), anti-extractable nuclear antigens (ENA) using fluorescent enzyme immunoassay (FEIA-Thermofisher). Erythrocyte sedimentation rate (ESR) and CRP (C-reactive protein) were evaluated, too.

Minor salivary glands biopsy was performed according to Guevara-Gutierrez technique and analyzed using Focus Score (FS) and Chisholm and Mason (CM) grading system[14]. Samples were considered positive for pSS when FS≥1 and CM≥3.

BMD was assessed by DXA scan using a Lunar Expert® version 1.72. The following regions were evaluated: Whole body, whole left femur, neck left femur, lumbar spine (L1-L4). BMD was expressed in g/cm^2 (grams per square centimeters). Diagnosis of osteoporosis was established when T-score deviated below -2.5 standard deviations compared with reference population according to the World Health Organization (WHO) criteria.

Statistical analysis

Analysis was performed for each region based on BMD status and T-score values. The mean BMD of whole body, lumbar, femur and femur neck of each patient was matched with controls and analyzed using a two tailed t-test or a Mann-Whitney test when the population differed from a normal distribution. Mean T-score of lumbar spine and femur’s neck were evaluated using respectively a t-test for parametric data and a Mann-Whitney test for non-parametric data. A contingency analysis was carried out using Fisher’s exact test to evaluate frequency of osteoporosis at lumbar spine and femur neck between pSS patients and control group. Subsequently pSS patients BMDs were divided in subgroups based on their immunity profile: Mean BMD of lumbar spine, femur, and femur’s neck of patients with Ro/SSA and La/SSB positivity were compared with mean BMD of the same regions of Ro/SSA and La/SSB negative patients using an unpaired t-test. Mean BMD of lumbar spine and whole femur of patients with normal inflammatory markers were compared with lumbar and femoral BMD of patients with altered ones. Pearson’s analysis was performed to investigate possible existing correlation between vertebral and femoral BMD with age, weight, BMI, Ro/SSA and La/SSB titers. Subsequently a multivariate linear regression model was used to predict the value of the endogenous variable (BMD) considering age, BMI and weight as exogenous variables.

Differences were considered statistically significant when p<0.05.

RESULTS

Table 1 shows epidemiological features of our cohort of study based on sex, age, weight, eight and BMI. Considering lumbar spine and femoral neck compared to the control group we have evidenced a significantly lower BMD in pSS patients compared to controls (Table 2).

Consistently with these results, among pSS patients, mean T-score values were significantly lower considering lumbar spine and femoral neck (Table 3).

By matching BMD of lumbar spine, whole femur, femoral neck, and whole body between patients with Ro/SSA positivity and patients with Ro/SSA negativity and applying the same process for patients carrying La/SSA positivity and La/SSB negativity we did not evidence any statistically significative difference between the two groups (Table 4).

By matching BMD of lumbar spine, whole femur, femoral neck, and whole body between patients with Ro/SSA positivity and patients with Ro/SSA positivity and patients with Ro/SSA negativity and applying the same process for patients carrying La/SSA positivity and La/SSB negativity we did not evidence any statistically significative difference between the two groups as shown in Table. The patients with La/SSB positivity that were evaluated also displayed Ro/SSA positivity.

Pearson’s or Spearman’s analysis evidenced positive correlations existing between whole femur and femoral neck with BMI and weight (whole femur-BMI r=0.37, p<0.05, whole femur-weight: r=0.38, p<0.05, femoral neck-BMI: r=0.47, p<0.05, femoral neck-weight: r=0.43, p<0.05) and negative one between lumbar spine BMD and age (r=-0.28, p<0.05). No significative correlation was found with Ro/SSA and La/SSB antibodies titers with BMD. Lastly, from multivariate analysis weight acted as the best predictor of lumbar BMD in pSS (|t|=2.58, p<0.05), age (|t|=2.430, p<0.05) and BMI (|t|=2.055, p<0.05)(Table 5).

DISCUSSION AND CONCLUSION

To date, only few and contradictory papers reported an association between bone loss and pSS, therefore it is not thoroughly considered to be a CTD at high risk for OP[10,15].
We evidenced a reduced BMD at lumbar spine and femoral neck regions in a cohort of patients recently diagnosed with pSS compared to a healthy control group, according with the findings of Pasoto et al. who documented a higher rate of OP and low BMD in pSS patients[12].

Unlike other chronic autoimmune diseases, such as RA, where a high inflammatory burden plays an important pathogenetic role via Wingless pathway (WNT) imbalance, its mechanism in pSS need to be clarified yet[16]. Gravani et al. studying impaired bone health and atherosclerotic risk in pSS, discovered that a
high DKK-1 serum levels in pSS patients correlated with a lower arterial plaque prevalence and that, paradoxically, higher levels of sclerostin and DKK-1 correlated with a higher BMD, suggesting the WNT pathway involvement in the disease[11].

In our findings, no difference was assessed between patients with normal or elevated inflammatory markers, although in our cohort inflammation indices were slightly increased (Table 4).

The role of autoimmunity itself is matter of debate: Anti-Citrullinated Protein Antibodies, high titers of Double Stranded antibodies and Anti-Carboxylated Protein Antibodies positivity are proven to be related to a lower BMD and with the presence of erosive joint lesions, thus being responsible of both systemic and juxta-articular OP[5,17,18]. On the other hand, to date no paper evaluated the role of anti-Ro/SSA and anti-La/SSB in impairing bone metabolism.

Nevertheless, our data did not evidence a statistically significant difference between anti-Ro/SSA and anti-La/SSB positive and negative patients nor a correlation of the antibodies titers with BMD: The knowledge of the immunological profile concerning anti-Ro/SSA and anti-La/SSB positivity did not add any further information in bone health evaluation by DXA. This may suggest that other mechanisms, such as modifications of gut microbiota, impaired glandular function, difficult food intake, cumbersome digestion, malabsorption, reduced physical activity and an altered body composition, should be considered to explain BMD reduction in pSS.

This paper represents, to the best of our knowledge, the only study that divided in subgroups a cohort of patients affected by pSS based on their Ro/SSA and La/SSB immunity profile, comparing their BMD. Results suggest that BMD is significantly altered in patients with pSS. Frequency analysis displayed a higher prevalence of OP in pSS considering both lumbar spine (18% vs 9%) and femoral neck (11% vs 4%) compared to controls, although not statistically significantly. Lack of a significative p suggested that apparently frequency of OP is similar in pSS and healthy subjects, though the lower BMD detected in every studied region proposed a severer bone loss in Sjögren individuals.

By choosing the more similar age-matched control group we minimized the effects of confounding factors. Limits of our study lay in its retrospective nature and in the small number of considered subjects, therefore longitudinal investigations relying on a larger study sample are warranted to confirm these results, even though in our opinion a higher level of attention concerning bone health of pSS patients should be encouraged.

DECLARATIONS

Ethics and consent

This retrospective study was conducted in accordance with the Declaration of Helsinki, each patient provided consent to undergo normal clinical routine.

Conflicts of interest

The authors do not have any conflict of interest to disclose.

REFERENCES