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ASSOCIATION BETWEEN LOW BONE MINERAL DENSITY, METABOLIC SYNDROME AND SEX STEROIDS DEFICIENCY IN MEN

F.L. Popa[1,3], M. Stanciu[2,4,*], A. Banciu[5], M. Berteanu[6]

"Lucian Blaga" University of Sibiu, Faculty of Medicine, [1]Dept. of Rehabilitation Medicine, [2]Dept. of Endocrinology, Emergency Academic Hospital, [3]Dept. of Rehabilitation Medicine, [4]Dept. of Endocrinology, Sibiu, Romania, [5]Klinik Michelsberg, Social Foundation Bamberg, Dept. of Neurology, Bamberg, Germany, [6]"Carol Davila" University of Medicine and Pharmacy, Dept. of Physical Medicine and Rehabilitation, Bucharest, Romania

Abstract

Objective. To analyze the association between low bone mineral density (BMD), metabolic syndrome (MS) and sex hormones deficiency in men.

Methods. We included in this retrospective study 199 men with osteoporosis or osteopenia and 167 men with normal BMD as controls, aged between 55-85 years old. Patients' evaluation included: medical history and physical examination, X-ray of thoracic and lumbar spine, measuring BMD at hip and lumbar spine, serum glucose and lipid profile, serum levels of total testosterone (tT), free testosterone (fT) and estradiol (E2).

Results. The results revealed a significant association between low BMD and MS (p=0.011). Vertebral fractures were more frequently associated with MS (p=0.041). Patients with MS had lower vertebral BMD (p=0.037) and lower E2 levels (p=0.024) compared with those without MS. In men with MS, E2 deficiency can predict the value of vertebral and hip BMD. fT deficiency can predict only the value of hip BMD.

Conclusions. A significant association between MS, low BMD, vertebral fractures and sex steroids deficiency, in particular E2 and fT was found. The presence of MS and sex hormones deficit can predict the reduction of BMD.

Key words: sex steroids, bone mineral density, men, metabolic syndrome.

INTRODUCTION

Current research suggests that metabolic syndrome (MS) is frequently associated with osteoporosis (1, 2) and a common risk factor: sex steroids deficiency (3, 4).

The effects of MS on bone mass appear to be inconsistent. In a US population-based study, femoral neck bone mineral density (BMD) was associated with abdominal obesity, and femoral neck BMD increased along with additional components of MS (5). Two meta-analyses suggest that MS has no clear influence on BMD (6) and it is not clearly associated with prevalence or incidence of fractures (7). Contrarily, Szulc *et al*. (8) revealed that men with MS have lower BMD, but lower fracture risk in the MINOS study. In another study, incidence of osteoporotic non-vertebral fractures was higher in subjects with MS (9).

Relationships between sex steroids and MS or BMD are unclear. Low testosterone is considered a risk factor for the development of MS and type 2 diabetes in men (10). However, MS has also been associated with higher estradiol (E2) levels, which is bone protective (11). Another study revealed that obesity and low E2 negatively affect bone mass in men (12). Some studies support the hypothesis that low sex hormones levels are positively associated with reduced BMD (13), but other researches have failed to confirm these observations (14).

It follows that the association between MS, BMD reduction and sex steroids deficiency has been researched, but the results are not conclusive. In addition, there are few data on Romanian subjects. Therefore, the aim of our research was to analyse the relationship between low BMD, MS and sex hormones deficiency in Romanian men included in the study.

METHODS

This clinical retrospective study was conducted between 2008 and 2012 and included 199 men who were diagnosed by dual-energy X-ray absorptiometry method (DXA) with osteoporosis or osteopenia (study group)

*Correspondence to: Mihaela Stanciu MD, "Lucian Blaga" University Sibiu, Faculty of Medicine, Dept. of Endocrinology, 2A Lucian Blaga street, Sibiu, 550169, Romania, E-mail: mihaela.stanciu@yahoo.com

and a control group that included 167 men with normal BMD. Patients' age ranged between 55 and 85 years old. All patients from study group were hospitalized and major diseases linked to osteoporosis were excluded. In the control group were included hospitalized patients or outpatients. All subjects were evaluated after having given their informed consent.

Inclusion criteria for the study group were the diagnosis of osteoporosis or osteopenia established by DXA method and for the control group, normal values of BMD.

Exclusion criteria for both groups were neoplasia regardless of stage or location and psychiatric disorders that would affect cognitive ability and patients' compliance.

Patients' evaluation included: medical history and physical examination. The following anthropometric measurements were carried out: body mass and height to calculate the body mass index (BMI) expressed as a body mass (kg)/height(m^2). Height, weight, and waist circumference were measured in subjects while they were wearing a light robe and no shoes. Waists were measured using the World Health Organization (WHO) waist circumference measurement guidelines. Blood pressure was measured with a manual sphygmomanometer in a sitting position on at least two different occasions after 10 min of rest.

X-ray of thoracic and lumbar spine, measuring BMD at hip and lumbar spine by DXA was assessed in all patients included in the study. BMD was measured at the lumbar spine and at the femoral neck using a LUNAR DPX-NT densitometer (Medtel, Australia). The DXA scans were obtained by standard procedures by the manufacturer for scanning and analysis. Patients' BMD was measured at the lumbar spine (anteroposterior projection at L1-L4) and at the femurs (i.e., femoral neck, trochanter, and total hip). The WHO classification system was applied defining osteoporosis as T score ≤ -2.5 and osteopenia as -2.5 < T score < -1.

Blood for laboratory investigations was collected from the median cubital vein between 7.00 and 9.00 a.m. after an overnight bed-rest, with last meal on the preceding day at 6 p.m. We evaluated the serum glucose, triglycerides and HDL-cholesterol for all subjects. Blood chemistry was measured by an enzymatic technique using an ARCHITECT c8000 analyzer (Abbott Diagnostics, USA).

Serum concentrations of total testosterone (tT), free testosterone (fT) were measured by electrochemiluminescence immunoassay (ECLIA). E2 serum concentration was measured by Enzyme-linked Immunosorbent Assay (ELISA). The hormonal investigations were carried out using a COBAS 6000 analyzer (Roche Diagnostics, USA). Sex steroid deficiency was considered at below normal levels for tT (below 2.25 ng/mL), fT (below 5.472 ng/L) and E2 (below 28 pmol/L).

MS was defined by the International Diabetes Federation and included abdominal obesity (abdominal circumference over 94 cm for European men and over 80 cm for European women) plus any two of following factors: elevated fasting plasma glucose (over 100 mg/dL), elevated triglycerides (over 150 mg/dL), elevated blood pressure (over 130/85 mmHg), low HDL-cholesterol (below 40 mg/dL in men and below 50 mg/dL in women) (15).

Statistical analysis was processed using International Business Machines-Statistical Package for the Social Sciences version 18.0, 2010 (IBM-SPSS 18.0, Armonk, New York, USA). The value of statistical significance was set at $p<0.05$. The mathematical values of tests were declared for significant comparisons. If tests were insignificant ($p\geq0.05$), only the value of significance (p) was reported.

For tabulation of scale data, we reported the mean and standard deviation expressed to 2 decimals and ordinal data were tabulated as percentage per group and presented with one decimal.

For parametric comparison, we used t-test. For ordinal or non-parametric data, we used Mann-Whitney test for comparing 2 groups. Chi-square test was used in tables 2 * 2 with ordinal / nominal data.

Multivariate analyses were performed using multiple logistic regression method, with the calculation of OR (95%Cl).

RESULTS

We found no significant differences between the two groups regarding mean age (p=0.218), BMI (0.295), median triglycerides (p=0.442) and mean tT (p=0.244) values. Control group has significantly lower median values of HDL cholesterol (r=0.22, p<0.001) compared to study group, but the size difference is considered small (Table 1).

Mean waist circumference (p=0.0054), fasting glucose (p<0.0001), systolic and diastolic blood pressure (p<0.0001) were significantly higher in study group. Mean fT (r=0.23, p<0.001) and E2 (r=0.16, p=0.002) were significantly lower in study group (Table 1).

Sex steroids deficiency was 3.61 times more frequent in study group compared to control group

Table 1. Clinical, biochemical, hormonal and DXA characteristics of study and control group

Variable		Study group		Control group	p
Age (years)		69.72 ± 7.81		68.75 ± 7.074	0.218
BMI (kg/m^2)		26.21 ± 3.71		25.83 ± 2.96	0.295
Waist (cm)		86.57 ± 7.70		84.58 ± 5.50	0.0054*
Systemic blood pressure (mmHg)		129.86 ± 7.34		124.43 ± 8.22	<0.0001*
Diastolic blood pressure (mmHg)		81.62 ± 6.16		77.68 ± 6.16	<0.0001*
Fasting glucose (mg/dL)		91.33 ± 13.53		85.99 ± 7.31	<0.0001*
Triglycerides (mg/dL)	Mean	118.29 ± 71.88	Mean	102.39 ± 42.64	
	Median	89.00	Median	87.00	0.442**
HDL-cholesterol (mg/dL)	Mean	52.49 ± 6.53	Mean	50.65 ± 4.78	
	Median	54.00	Median	50.00	<0.001**
tT (ng/mL)		4.73 ± 1.94		5.09 ± 1.70	0.244
fT (ng/L)		8.06 ± 4.01		9.74 ± 4.20	< 0.001*
E2 (pmol/L)		41.98 ± 17.46		48.83 ± 10.74	0.002*
Hip BMD (g/cm^2)		0.82 ± 0.07		0.98 ± 0.01	< 0.001*
Vertebral BMD (g/cm^2)		0.92 ± 0.07		1.16 ± 0.04	< 0.001*

tT total testosterone, fT free testosterone, E2 estradiol. Values are expressed as mean ± SD;
* Difference is significant at the 0.01 level.
** Non-parametric tests were applied and medians were compared.

Table 2. Comparison of vertebral BMD, hip BMD and mean sex steroids values in men with or without metabolic syndrome

	With Metabolic syndrome				Without Metabolic syndrome				p
	Study		Control		Study		Control		
	Mean	Standard Deviation	Mean	Standard Deviation	Mean	Standard Deviation	Mean	Standard Deviation	
Vertebral BMD (g/cm²)	0.90	0.08	1.16	0.03	0.93	0.07	1.17	0.04	0.281
Hip BMD (g/cm²)	0.83	0.09	0.98	0.02	0.82	0.07	0.99	0.02	0.154
tT value (ng/mL)	4.62	2.38	4.39	2.03	4.76	1.82	5.18	1.65	0.244
fT value (ng/L)	8.72	4.19	8.60	3.73	7.89	3.96	9.88	4.25	0.092
E2 value (pmol/L)	37.75	19.68	44.50	12.56	43.07	16.74	49.36	10.43	0.918

tT total testosterone, fT free testosterone, E2 estradiol.

(OR=3.61; 95%CI=2.329-5.599; $\chi^2(1)=34,20$, p<0.001). The risk of having sex steroids deficiency is 1.75 times more likely for individuals diagnosed with low BMD (RR=1.75; 95%CI=1.446-2.131).

Our study demonstrated a significant association between low BMD and MS. MS is 2.14 times more frequent in study group (20.6%) compared to control group (10.8%) (OR=2.14; 95%CI=1.182-3.905; $\chi^2(1)=6.48$, p=0.011). The risk of having MS is 1.35 times more likely for the individuals diagnosed with low BMD (RR=1.35; 95%CI=1.104-1.651).

Hypertension (40.2% in study group, 25.7% in control group, p=0.004), high triglycerides (20.1% in study group, 11.4% in control group, p=0.024) and hyperglycaemia (11.1% in study group, 3% in control group, p<0.05) are more frequently in study group. No differences were found between groups regarding the frequency of low HDL cholesterol values (14.1% in study group, 9.0% in control group, p=0.132).

Osteoporotic fractures were recorded in 68.3% (28 cases) of males with MS and in 51.9% (82 cases) of patients without MS from the study group, but the difference was not significant (p=0.060). A significant association was found between MS and the presence of vertebral fractures (VF) ($\chi^2(1)=4.19$; p=0.041; RR=1.755; 95% Cl=1.024-3.008). No association was detected between MS and the presence of hip (p=0.570) or peripheral (p=0.939) fractures.

A two-way between groups analysis of variance was conducted to explore the impact of study/control group and the presence or absence of MS on several variables (Table 2).

For vertebral BMD, the interaction between study group and presence of MS was not significant (p=0.281). However, patients with MS had a significantly lower vertebral BMD compared with those without MS, the vertebral BMD difference between groups being of - 0.19 (p=0.037, 95%CI (-0.038; -0.001). The interaction between study group and presence of MS was not significant for hip BMD (p=0.154) (Table 2).

For tT (p=0.244), fT (p=0.092) and E2 (p=0.918) the interaction between study group and presence of MS was not significant. Instead patients

Table 3. Prediction model for vertebral BMD value

	Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B	
	B	Std. Error	Beta			Lower Bound	Upper Bound
(Constant)	0.910	0.025		37.066	0.000	0.861	0.958
Metabolic syndrome	0.017	0.013	0.099	1.319	0.189	-0.009	0.044
Low tT	-0.031	0.018	0.142	-1.736	0.084	-0.066	0.004
Low fT	-0.012	0.012	0.081	-1.014	0.312	-0.035	0.011
Low E2	-0.036	0.011	0.226	-3.158	0.002	-0.059	-0.014

Dependent Variable: lumbar BMD (g/cm^2).
tT total testosterone, fT free testosterone, E2 estradiol.

Table 4. Prediction model for hip BMD value

	Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B	
	B	Std. Error	Beta			Lower Bound	Upper Bound
(Constant)	0.893	0.024		19.279	0.000	0.846	0.940
Metabolic syndrome	-0.017	0.013	-0.089	-1.349	0.179	-0.042	0.008
Low tT	-0.002	0.017	-0.009	0.123	0.902	-0.036	0.032
Low fT	-0.046	0.011	-0.293	4.237	0.000	-0.067	-0.025
Low E2	-0.069	0.011	-0.406	6.527	0.000	-0.090	-0.048

Dependent Variable: lumbar BMD (g/cm^2).
tT total testosterone, fT free testosterone, E2 estradiol.

with MS had significantly lower E2 levels compared with those without MS, the E2 mean difference between groups being of - 5.08 (p=0.024, 95%CI (-9.496;-0.678) (Table 2).

Multiple regression was used to assess the ability of diagnosis of MS and the specific deficit of sex steroids to predict the value of vertebral BMD. This model was statistically significant (F(4.186)=6.61, p<0.001) and the variance explained by the model was 12.7%. Only one measure contributed significantly to the model: low E2 classification (beta=0.226, p=0.002) (Table 3).

The same analysis was made for hip BMD. This model was statistically significant (F(4.198)=19.03, p<0.001) and the variance explained by the model was 28.2%. Two measures contributed significantly to the model: low E2 classification (beta=-0.406, p<0.001) and low fT classification (beta=-0.293, p<0.001) (Table 4).

DISCUSSION

We investigated the relationship between serum sex steroids, BMD and MS in men.

We found a significant association between low BMD and MS (20.6%). Other studies showed similar percentages between 20-24% (16) and the results of a meta-analysis suggest that MS is a risk factor for developing osteoporosis in men (1).

Our results revealed that patients with MS had lower vertebral BMD (but not femoral neck BMD) compared with those without MS. A study conducted in Korean men and postmenopausal women MS was associated with a lower femoral neck BMD (only measured) especially in men (16).

Our analysis showed that there was no significant association between the presence and location of osteoporotic fractures and MS, except for VF. The results of other studies are discordant (7). The Third National Health and Nutrition Examination Survey reported no difference in the prevalence of non-vertebral fractures between people with and without MS (5). Ahmed *et al*. found that MS have a significant protective effect on fracture risk in the Tromsø Study (17). Szulc *et al*. found that men with MS have lower BMD but lower fracture risk in the MINOS study (8). A meta-analysis suggests that MS has no clear influence on BMD, or its influence may be beneficial (7). In the Rancho Bernardo study, von Muhlen *et al*. found that the incidence of nonvertebral fractures was higher in individuals with MS (9).

There is convincing evidence that testosterone deficiency is a risk factor for the development of MS (10). The relation between E2 and MS in men is unclear. Some studies revealed that obese men have higher total and free E2 levels than the non-obese (11). On the contrary, conditions of oestrogen deficiency (such as congenital aromatase deficiency) are accompanied by hyperglycaemia and dyslipidemia (18). Our results showed that patients with low BMD and MS had significant lower E2 levels compared with patients without MS.

We revealed that in men with MS, low levels of E2 can predict low vertebral and hip BMD. Low levels

of fT can predict only low hip BMD. This finding is in accordance with the results from the Swedish Osteoporotic Fractures in Men Study (MrOS) (19). In this research, hip BMD was correlated with both fT and free E2 in elderly men. In the United States, part of MrOS vertebral BMD was correlated with fT and free E2 (20); hip BMD was correlated only with bioavailable E2 in the men over 65 years old (21).

Our study has some limitations. The cross-sectional design does not allow assessing the causality between low BMD, MS and sex hormones deficiency. We did not investigate the free estradiol and the sex hormone binding globulin levels, which restricted the analysis of the role of these factors on BMD and on the development of MS in men.

In conclusion, based on significant association between MS, low BMD and sex steroids deficiency in men, in particular E2 and fT, our data sustained the importance of evaluation of osteoporosis in patients with MS. MS is significantly associated with VF. In patients with MS, low levels of E2 can predict the reduction of vertebral and hip BMD. Low levels of fT can predict only the reduction of hip BMD.

Conflict of interest

The authors declare that there is no conflict of interest.

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