Osteoporosis in men: a study in patients affected by chronic non-advanced liver disease

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Summary

Background. **Chronic liver disease has been shown to cause bone osteometabolic disease giving rise to osteoporosis and osteomalacia.**
Aim of this study was to evaluate bone mineral density (BMD) and bone metabolism markers in a selected series of male patients living in the same geographical area of Southern Italy, affected only by viral, non-advanced chronic liver disease, as compared to a randomly selected control group from the same geographical area.
Patients ans Methods. **Twenty-five male patients affected by biopsy-proven chronic hepatitis or liver cirrhosis only in Child's A class, virus-related, were selected in a one year period. Thirthy healthy male volunteers, living in the same geographical area, age and BMI-matched, were examined as control group.**
Bone mineral density (BMD) was measured at lumbar spine (L1-L4) and at proximal femur of non dominant leg by Dual-Energy X-ray Absorptiometry (DEXA) using a Hologic QDR 1000 densitometer. Biochemical tests of liver function and bone metabolism in patients and controls were carried out after an overnight fast.
Results. **BMD was reduced in 16 (64%) out of 25 patients: 5 patients (20%) could be classified as osteoporotic and 11 patients (44%) osteopenic on the basis of at least one measurement. Mean BMD values for lumbar spine and femoral neck were significantly decreased in patients in comparison to the controls. Using multiple linear regression analysis, no correlation was found between BMD and the biochemical and functional variables studied.**
Our patients did not show any difference, as compared to controls, in bone remodelling markers as serum PTH, 25-OHD, calcium and sexual hormones, as well as urinary hydroxyproline.
Conclusions. **Our data, obtained on the basis of severe inclusion criteria, suggest that liver disease "*per se*" independently on its severity and lasting, can reduce the bone mass. Therefore more attention has to be paid to bone metabolism in liver patients.**

KEY WORDS: osteoporosis, men, liver disease.

Introduction

Metabolic bone disease is a complication of chronic liver disease (CLD) and is well known as "hepatic osteodistrophy" (1, 2). Osteoporosis accounts for the majority of cases, whereas osteomalacia is rare in the absence of advanced liver disease and severe malabsorption (3). The prevalence is the same in men and women (4), however the published prevalence of osteoporosis considerably differs and ranges from 20% to 100%, depending on patients selection and different diagnostic criteria (3-5). Many reports are referred to a broad spectrum of liver disease of different aetiology and severity: in patients with advanced liver disease candidates for or treated with orthotopic liver transplantation, osteoporosis is prevalent and contributes to a major source of morbidity preceding and following transplantation (6, 7). Nevertheless, little is known about the prevalence among patients with non-advanced liver disease. The aetiology and pathogenesis of osteoporosis in these patients also remain undefined, even though its histology is quite similar to post-menopausal and age-related bone loss, affecting trabecular bone more rapidly than cortical bone (8, 9). Finally, there is controversy about risk factors for osteoporosis in CLD: liver cirrhosis, cholestasis, hypogonadism, corticosteroid or immunosuppressive treatment, alcohol consumption, malnutrition and malabsorption, sex, physical activity, subnormal vitamin D levels and/or vitamin D receptor genotype, insulin growth factor 1 (IGF-1) deficiency, as well as country and nationality are all reported factors affecting bone metabolism (3-5, 8-11).
However, whether non-advanced liver disease "*per se*" could be a risk factor for osteoporosis still remains uncertain; moreover there are few reports about the effects of viral liver disease on bone turnover (8).
Aim of this study was to evaluate bone mineral density (BMD) and bone metabolism markers in a selected series of male patients living in the same geographical area of Southern Italy, affected only by viral, non-advanced chronic liver disease, as compared to a randomly selected control group from the same geographical area.

Patients and methods

In a one year period we studied 98 liver patients consecutively admitted to the III Department of Internal Medicine, Federico II University, Naples, Italy. Among them we selected 25 patients affected by biopsy-proven chronic hepatitis or liver cirrhosis, virus-related, according to the following inclusion criteria: male sex, age < 60 years, body mass index (BMI) from 20 to 30 kg/m^2, normal physical activity, biopsy-proven liver cirrhosis only in Child's A class (12, 13), virus-related.

All patients lived in the same geographical area from Southern Italy. Their diet was a tipical mixed, mediterranean diet, containing milk and cheese products, without alcohol intake; only five patients consumed less than 20 g of alcohol three or four times a week. Information about dietary habits and alcohol intake were carefully obtained from a food frequency questionnaire.

Exclusion criteria were: female sex, age > 60 years, alcohol abuse, drug addiction, obesity, previous bone fractures or history of recent immobilization, autoimmune, endocrinological or rheumatological diseases, treatment with corticosteroids, diuretics, drugs affecting bone metabolism or interferon for more than three months over the past three years. Advanced liver cirrhosis (B or C Child's class), hepatocellular carcinoma, primary biliary cirrhosis, cholestatic liver diseases, genetic hemochromatosis were also considered exclusion criteria.

The selected patients had positive serological markers for viral hepatitis (2 for hepatitis B virus and 23 for hepatitis C virus) and biopsy-proven diagnosis of chronic liver disease: 12 patients were affected by chronic hepatitis and 13 patients by liver cirrhosis, A Child's class. Liver function was well preserved and renal function was normal in all patients. None exhibited indices of cholestasis nor experienced previous decompensation of cirrhosis.

Thirthy healthy male volunteers, living in the same geographical area, age, and BMI-matched, were also examined as control group.

All patients and controls received information about the study and gave their informed consent to participate.

Bone mineral density (BMD) was measured at lumbar spine (L1-L4) and at proximal femur of non dominant leg by Dual-Energy X-ray Absorptiometry (DEXA) using a Hologic QDR densitometer.

Osteoporosis was defined in analogy with the World Health Organization criteria for women (14). Individual BMD values are expressed as gr/cm^2 and T- and Z-score. Quality control was maintained by daily scanning of an anthropomorphic spine phantom. The coefficient of variation for the DEXA technique was less than 1% for the lumbar spine (LS) and 1.5% for the femoral neck (FN). The reference population adopted in this study was the international pooled sample provided by the manifacturer; their data, however, did not differ significantly for those obtained on a local sample in a study performed when the device was set up (15).

Biochemical tests of liver function and bone metabolism in patients and controls were carried out after an overnight fast. Biochemical liver function tests and serum calcium, inorganic phosphate, and magnesium were measured with an automatic technique. Serum was assayed using commercially available kits for intact Parathyroid hormone (PTH), (IRMA, Incstar Corporation), 25-hydroxyvitamin D (25-OHD), (RIA Incstar Corporation), IGF-1, (RIA, Nichols Institute, San Juan Capistrano, CA, USA), bone isoenzyme alkaline phosphatase (b-Ap) as a marker of bone formation (Tandem-T, Ostase, IRMA, Hybritech Europe, Liege, Belgium), Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH), (RIA, Biodata, Serono Diagnostics, Italy), Prolactin (HPRL), Oestrogen, Progesterone, 17-OH-Progesterone, Delta-4-Androstenedione, Testosterone (RIA, Diagnostics Products Corporation, Los Angeles, CA, USA), Adrenocorticotropic hormone (ACTH), Cortisol, Aldosterone, complete Thyroid Hormones and antibodies profile (FT3, FT4, T3, T4, TSH, Tg, TgAb, TPOAb), (RIA, Sorin Biomedica, Saluggia, Italy).

Urine was analyzed for calcium (automatic absorption spectrophotometry) and creatinine (standard automated techniques). Urinary hydroxyproline, as a marker of bone resorption, was measured with a commercial Kit (OHP Biolab, Italy) and urinary excretion was expressed in terms of urinary creatinine as the ratio of hydroxyproline to creatinine.

Statistical analysis: results were expressed as means ± SD. Student's unpaired t-test was used to compare means between the groups. The non parametric Mann-Whitney U test was used when Wilk-Shapiro test was not consistent with the Gaussian distribution of the data. Linear multiple regression analysis (using as covariates age and BMI) was used to evaluate the determinants of bone mass.

Significance was retained for $p < 0.05$.

Results

Clinical and laboratory data of patients and controls are reported in Table I.

BMD was reduced in 16 (64%) out of 25 patients: 5 patients (20%) could be classified as osteoporotic and 11 patients (44%) osteopenic on the basis of at least one measurement. Table II reports mean BMD values for lumbar spine (LS) and femoral neck (FN) in patients and controls, showing a decreased BMD in patients in both sides of measurements even if only for the LS it reaches the significance.

In order to compare the BMD of our patients with reference population data we calculated the 95% CI of the mean Z-score at the LS and at the FN. Data showed that the mean Z-score at LS but not at FN among liver patients was significantly lower as compared to the age- and sex-matched general population (lumbar spine mean Z-score 95% CI = -1.06 to -0.920; femoral neck mean Z-score 95% CI = -0.54 to + 0.53).

Using multiple linear regression analysis we investigated the relationship between BMD (gr/cm^2) and the variables considered, including the time of detection of abnormal tests of liver

Table I - Clinical features and liver function laboratory tests.

	Patients (25)	Controls (30)
Age	50.4 ± 8.4	51.2 ± 3
BMI kg/m^2	26.4 ± 4.6	26.2 ± 3.1
Albumin (g/dl)	4.0 ± 0.5	4.2 ± 0.3
GGT (U/L)	72 ± 11	30 ± 8
Prothrombin time (%)	79 ± 9	98 ± 2
AST (U/L)	90 ± 12	23 ± 6
ALT (U/L)	85 ± 10	25 ± 5
Total bilirubin (mg/dl)	0.7 ± 0.4	0.6 ± 0.4
Alkaline phosphatase (U/L)	155 ± 11	150 ± 7

Table II - BMD in liver patients and controls.

Mean values	Patients	Controls
Lumbar Spine (L1-L4) BMD (gr/cm^2) 0.08*	0.992 ± 0.18	1.11 ±
Femoral neck BMD (gr/cm^2)	0.858 ± 0.05	0.849 ± 0.15

* P = 0.0254.

function. No correlation was found between BMD and the variables studied.
Biochemical tests of bone remodelling metabolism and hormonal profiles did not differ in the two groups.

Discussion

Recently the medical community got to acknowledge that, although not as relevant as in women, male osteoporosis is an epidemiologically relevant feature of the disease and one out of three osteoporosis fractures occurs in the male population (16).
Aim of this study was to evaluate bone mineral density and bone metabolism markers in a selected series of male patients affected only by viral, non-advanced chronic liver disease. This illness is known to affect patients metabolism, possibly leading to hypermetabolism (17) affecting also the bone turnover. The eccessive bone turnover activity can lead to an alteration of bone trabeculae, and to their perforation. This fact leads to a damage of bone micro-architecture and, consequently, to the reduction of proper mechanical characteristics of the bone segment and to increased risk of fracture (18).
The present study shows that viral chronic liver disease in man is associated with reduced bone mineral density. In fact BMD at LS was significantly lower in liver patients as compared to controls ($p<0.05$) and BMD at FN showed a trend to reduction even though the difference between patients and controls was not statistically significant (Table I). Data are obtained using two different groups of controls: 1) age, BMI-matched healthy volunteers, 2) population-based reference values.
Osteoporosis has been described in patiens treated with orthotopic liver transplantation (6), as well as in subjects affected by advanced liver cirrhosis, primary biliary cirrhosis, cholestatic or alcoholic liver diseases, hemochromatosis (1-5) and in corticosteroid treated patients (1-3, 19), but little is known about bone mineral density in viral non advanced liver disease.
A recent paper on this issue (8) reported in viral cirrhosis in men a high prevalence of osteoporosis suggesting that it is a major cause of reduced bone mineral density in men. However, in this study three heterogeneous groups of patients, aged 38-74, affected by different severity of liver disease, according to Child's classification (12) and Pugh's score (13) were studied: a significant correlation between BMD at lumbar spine and severity of liver disease was found. An advanced age and a more severe liver disease could both affect the results, therefore, in the present study we examined 25 male patients selected on the basis of very restricted criteria: it was a homogeneous group, without confounding factor affecting bone metabolism, B and C Child's classes of liver cirrhosis and subsets aged > 60 years were excluded.
Previous papers reported low serum levels of PTH, 25-OHD, and calcium in advanced liver cirrhosis graded as Child's C class (5, 8, 20). In contrast we did not find any difference between patients and controls for all biochemical parameters of bone metabolism. These discrepancies may be explained by patient selection and different pathogenetic mechanisms. In fact, the pathogenesis of reduced bone density in chronic liver disease remains not clear. Reduced osteoblastic function with diminished bone formation has been suggested as the main factor responsible for osteoporosis in alcoholic liver cirrhosis: it is defined "low turnover osteoporosis" and low serum concentrations of IGF-1 are associated with this condition (8, 9). In contrast, a "high turnover osteoporosis" is characterized by normal or reduced bone formation coupled with increased resorption. In this case synthesis of matrix and its mineralization are normal, but osteoblasts are unable to fill the numerous resorption cavities. "High turnover osteoporosis" has been mainly reported in patients with chronic cholestatic liver disease as primary sclerosing cholangitis, and primary biliary cirrhosis (4, 8, 9).
In our patients serum levels of IGF-1 are normal since they are affected by non advanced liver disease and it is known that IGF-1 levels are related to the severity of liver disease (8).
Our patients did not show any difference, as compared to controls, in bone remodelling markers as serum PTH, 25-OHD, calcium and sexual hormones, as well as urinary hydroxyproline.
Neverthless, a high prevalence of osteoporosis and osteopenia was found in our selected group of male patients and bone mass reduction was more severe at LS (trabecular bone) than at FN (cortical and trabecular bone) in keeping with the above mentioned papers (8-11), probably because the rate of turnover in cortical bone is much lower than in trabecular bone.
In conclusion our data, obtained on the basis of severe inclusion criteria, excluding confounding factor affecting bone metabolism, suggest that liver disease "*per se*" indipendently on its severity and lasting, can reduce the bone mass.
The exclusion of female sex and advanced age, and the study of a very homogeneous group of male patients not alcohol consumers, never treated with steroids or drugs affecting bone mass, without clinical or laboratory signs of cholestasis suggest that osteoporosis is a frequent finding in males with non advanced viral liver disease and that more attention has to be paid to bone metabolism in liver patients.

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