Preventing fragility fractures in patients with hypercholesterolemia: the importance of REMS technology

Chiara Stomaci [1], Fabiola Rosa Contaldo [1*], Marco Di Paola [2*], Roberto Franchini [2*], Fiorella Anna Lombardi [2*], Giusy Peluso [3*], Paola Pisani [2*], Francesco Conversano [2], Sergio Casciaro [2]

[1] Department of Biological and Environmental Sciences and Technologies, University of Salento, Lecce, Italy; [2] Institute of Clinical Physiology, National Research Council, Lecce, Italy; [3] Department of Innovation Engineering, University of Salento, Lecce, Italy; * Equal contributor in alphabetic order

ABSTRACT

Purpose: Hypercholesterolemia (HC) is associated with altered bone metabolism, potentially increasing osteoporosis and fracture risk. This study evaluates bone quality in HC patients using the REMS-derived Fragility Score (FS), and compares it with that of an age- and BMI-matched healthy population.

Methods: A retrospective study was conducted in 184 Caucasian patients (105 HC patients, 79 healthy controls) from "A. Galateo" Hospital (Lecce, Italy). REMS ultrasound scans were performed at the lumbar spine and proximal femur using the EchoStation device to assess T-score and FS. The Shapiro-Wilk test was used to assess normality of distribution, and the Mann-Whitney test to compare the means of the two groups. To evaluate the diagnostic performance of FS in identifying patients with deteriorated bone quality, a receiver operating characteristic curve analysis was performed. $p < 0.05$ was always considered statistically significant.

Results: HC patients showed significantly higher FS and significantly lower T-scores than controls. Median lumbar spine FS and T-score were 73.12 [65.7 – 77.4] and -2.30 [-2.75 – (-1.70)] in HC patients vs. 25.01 [19.7 – 27.1] and -1.60 [-2.2 – (-1.1)] in controls ($p < 0.0001$). Similar differences were observed at the proximal femur ($p < 0.0001$). FS showed good discriminative ability for bone quality, with area under the curve values of 0.74 (lumbar, $p < 0.001$) and 0.81 (femur, $p < 0.001$).

Conclusions: HC negatively impacts bone quality, increasing the probability of fragility fracture risk. REMS-derived FS effectively distinguishes compromised HC patients, supporting its use in bone health monitoring and early intervention.

KEYWORDS

Hypercholesterolemia, bone quality, REMS, fragility score.

Introduction

Cholesterol is a key component in various physiological processes. This lipid plays a fundamental role in cellular membranes, where it regulates membrane structure, permeability, and fluidity [1]. It also serves as a precursor for oxysterols, which are further metabolized into bile acids [2]. Moreover, it forms the backbone of all steroid hormones, including estrogens, progesterone, testosterone, cortisol, and aldosterone, as well as vitamin D [3,4]. Cholesterol can be obtained from the diet or synthesized endogenously by nearly all cell types. The liver is the primary organ responsible for cholesterol synthesis, accounting for approximately 50% of its total production [5]. Blood cholesterol levels can vary depending on factors such as age, sex, diet, lifestyle, and underlying diseases [6].

Hypercholesterolemia (HC), or high cholesterol, is a condition characterized by elevated cholesterol levels in the bloodstream [6]. While no universal cutoff values for cholesterol levels have been established [6], a total cholesterol level above 200 mg/dL (5.2 mmol/L) is commonly used as a diagnostic threshold for HC, with values ≥ 250 mg/dL indicating a concerning pathological condition [6]. Abnormal cholesterol levels are associated with various diseases, including atherosclerotic cardiovascular disease, obesity, diabetes, cancer, neurodegenerative disorders, osteoporosis, and viral infections [7]. HC can be primary, resulting from genetic mutations as in familial HC, or secondary, due to factors like diet, obesity, or underlying conditions such as diabetes and hypothyroidism [6]. Beyond its well-established effects on cardiovascular health, HC has also been linked to alterations in bone metabolism, potentially increasing the risk of osteoporosis [7], a disease characterized by reduced bone mineral density (BMD) [8]. It is commonly encountered in postmenopausal women, in whom either impaired bone formation or excessive bone resorption leads to increased fracture risk [8]. Very often osteoporosis arises as a consequence of other diseases, and in this case, it is referred to as secondary

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Contact
Sergio Casciaro; casciaro@ifc.cnr.it
Institute of Clinical Physiology, National Research Council, Ecotekne Campus, via Provinciale Lecce-Monteroni, Lecce, Italy.

osteoporosis [8]. Over the past decades, multiple epidemiological studies have identified high plasma cholesterol levels as a risk factor for osteoporosis [9-12]. Notably, an analysis of data from the National Health and Nutrition Examination Survey (NHANES), which included 10,038 U.S. patients (aged 20-59 years) from 2011 to 2018, revealed a negative correlation between BMD and total cholesterol levels [12].

The relationship between lipid metabolism and bone physiology remains complex and not yet fully understood, but several mechanisms have been proposed to explain it. Bone homeostasis is maintained by the coordinated activity of osteoblasts, which are responsible for bone formation, and osteoclasts, which mediate bone resorption [13]. Osteoblasts produce the receptor activator of nuclear factor kappa-B ligand (RANKL), a key factor that binds to the receptor activator of nuclear factor kappa-B on osteoclast precursors, promoting their differentiation and activation [13]. This process is counterbalanced by osteoprotegerin (OPG), a decoy receptor produced by osteoblasts that inhibits RANKL activity, thereby preventing excessive bone resorption [13].

Several *in vitro* experiments have demonstrated that cholesterol strongly influences osteoclast differentiation and survival [14]. High cholesterol levels have been shown to enhance osteoclast activity by increasing the expression of pro-osteoclastogenic factors such as interleukin-1α (IL-1α) and RANKL, while reducing OPG levels, ultimately leading to an imbalance in bone remodeling [14,15]. In rat models, high dietary cholesterol levels induced alveolar bone resorption, an effect counteracted by vitamin C administration, suggesting a possible oxidative stress-related mechanism [16]. Additionally, cholesterol-lowering drugs such as statins have been found to reduce osteoclast activity and enhance bone formation by modulating the RANKL/OPG pathway, further supporting the role of cholesterol in bone health [17,18].

According to a study carried out in 2022, osteoporosis is currently a significant public health concern, with a global prevalence of 19.7%, while the global prevalence of its precursor, osteopenia, is 40.4% [19]. Given these rates, early diagnosis and effective patient management are crucial.

In recent years, radiofrequency echographic multi-spectrometry (REMS) has emerged as an effective, non-ionizing method for assessing bone density at axial sites. REMS calculates BMD and T-score values by processing raw ultrasound signals obtained through ultrasound scans of the lumbar vertebrae and/or femoral neck [20,21]. Besides the quantitative BMD, T- and Z-score parameters, an additional REMS-based parameter known as the Fragility Score (FS) has been implemented as an indicator of skeletal fragility. Ranging from 0 to 100, the FS is a dimensionless value that reflects the similarity between a patient's bone ultrasound profile and the spectral model of a fractured bone, independently of BMD. It assumes that fragile bone has microstructural features that influence the spectral characteristics of the acquired ultrasound signal, being different from those reflecting a robust bone structure. This allows the quality of bone micro-architecture to be assessed at the lumbar spine or femoral neck, with higher scores indicating poorer bone quality [22].

The present study aims to assess bone quality using the REMS-derived FS in hypercholesterolemic patients, and to compare it with that of an age- and BMI-matched healthy population.

Methods

Study design and participants

This retrospective study focuses on a population of 184 patients from "A. Galateo" Hospital in San Cesario di Lecce (Lecce, Italy). Included subjects were selected on the basis of the following criteria: Caucasian ethnicity, both genders, and absence of previous fragility fractures. Based on their conditions, the enrolled patients were separated into two sub-groups as follows: patients diagnosed with HC (n=105), i.e., patients with total cholesterol levels ≥ 250 mg/dL, regardless of whether their HC was primary or secondary (multifactorial hypercholesterolemia), and control patients (n=79), i.e., healthy patients with total cholesterol levels < 200 mg/dL and no pathological conditions who underwent preventive bone health screening. The two sub-groups were matched for age and BMI. Prior to commencement, the study protocol was approved by the hospital's ethics review board and all participants provided written informed consent before enrollment.

T-score and Fragility Score assessment

REMS scans were performed using an EchoStation device (Echolight S.p.a., Lecce, Italy) equipped with a convex transducer operating at a nominal frequency of 3.5 MHz. Scans were performed at two sites—the proximal femur and the lumbar spine—by positioning the probe parallel to the target site, with its indicator facing the patient, in order to capture standardized anatomical views. Once the target bone interface was visualized on the monitor, the operator adjusted the scan depth and focus to optimize the image quality. Each scan consists of multiple (up to 100) images, and for each one, a specific portion of the radiofrequency signal is automatically identified (region of interest, ROI) using the dedicated Echolight software described in Casciaro *et al*. [23]. Then on the basis of the correlation between its frequency spectrum and each of the two age-, BMI- and sex-matched models stored in a reference database, each image is classified as "frail" or "non-frail". The FS value is defined as the percentage of the ROI classified as "frail" in the segmented images [24].

Statistical analysis

The collected data were assessed for normality of distribution using the Shapiro-Wilk test (α=0.05), while an unpaired non-parametric Mann-Whitney test (α=0.05) was used to compare the two populations (with $p < 0.05$ taken as statistically significant). All the data are presented as mean ± standard deviation (SD) and as median [first quartile-third quartile]. RStudio was used to generate all the graphs and perform the statistical analyses [25]. The diagnostic performance of FS in identifying patients with poor bone quality was evaluated by receiver operating characteristic (ROC) curve analysis using the De Long test [26], with p-values considered significant if below the threshold of 0.05. The ROC curve analysis and graph were

generated using MedCalc® Statistical Software version 22.021 (MedCalc Software Ltd, Ostend, Belgium; https://www.medcalc.org; 2024).

Results

The study included 105 patients with HC and 79 healthy controls. Mean age and BMI were similar between the two groups: 67.6 ± 7.2 years and 25.7 ± 4.12 kg/m² in the HC patients, and 63.1 ± 8.0 years and 25.06 ± 4.88 kg/m² in the controls. The characteristics of the two subgroups are detailed in Table I.

REMS analysis of the proximal femur and lumbar spine showed significant differences between the two populations.

At the lumbar spine, the HC population showed a median FS of 73.12 [65.7 – 77.4], versus 25.01 [19.7 – 27.1] in the healthy controls. At the femur, the HC population showed a median FS of 77.6 [70.8 – 81.7], and the healthy control group a median value of 23.8 [21.5 – 26.1]. Both FS analyses (lumbar spine and femur) showed significant differences between the two populations ($p < 0.0001$), as shown in Figure 1.

The median T-score at the lumbar spine was -2.3 [-2.8 – (-1.7)] in the HC population, versus -1.6 [-2.2 – (-1.1)] in the healthy control group. At the femur, the HC population showed a median T-score of -2.2 [-2.7 – (-1.7)], versus -1.9 [-2.4 – (-1.4)] in the healthy controls. Both T-score analyses (lumbar spine and femur) showed significant differences between the two groups ($p < 0.0001$), as represented in Figure 2.

The area under the curve (AUC) for the FS, measured at the lumbar spine and proximal femur and used to discriminate between patients with and without deteriorated bone quality, was 0.74 ($p < 0.001$) and 0.81 ($p < 0.001$), respectively (Figure 3).

Discussion

High blood cholesterol (or hypercholesterolemia) has been linked to alterations of bone metabolism, thus potentially increasing the risk of osteoporosis. Knowledge about the com-

Table I Characteristics of the 2 sub-groups (Hypercholesterolemic and Healthy patients) that underwent T-score and Fragility Score assessment at both the proximal femur and lumbar spine.

FEATURES	HYPERCHOLESTEROLEMIC (*n* = 105)	HEALTHY (*n* = 79)
Age (years)	67.6 ± 7.2	63.1 ± 8.0
Ethnicity	100% Caucasian	100% Caucasian
BMI (kg/m²)	25.7 ± 4.12	25.06 ± 4.88
Patients with traumatic fractures	42 (40%)	2 (2.5%)
Sex distribution	86.7% (women) - 13.3% (men)	75.4% (women) - 24.6% (men)
Total cholesterol (mg/dL)	≥ 250	< 200

Figure 1 Fragility Score (FS) measured at the lumbar spine and femoral neck in healthy and hypercholesteraemic (HC) patients. The healthy and HC population median for the spine is 25.01 [19.7 – 27.1] and 73,12 [65.7 – 77.4], respectively. For the femur the median is 23.8 [21.5 – 26.1] and 77.6 [70.8 – 81.7] for healthy and HC population, respectively. Data are presented as median [first quartile-third quartile] (n = 105 in HC group and n = 79 in control group; statistical significancy **** = $p < 0.0001$).

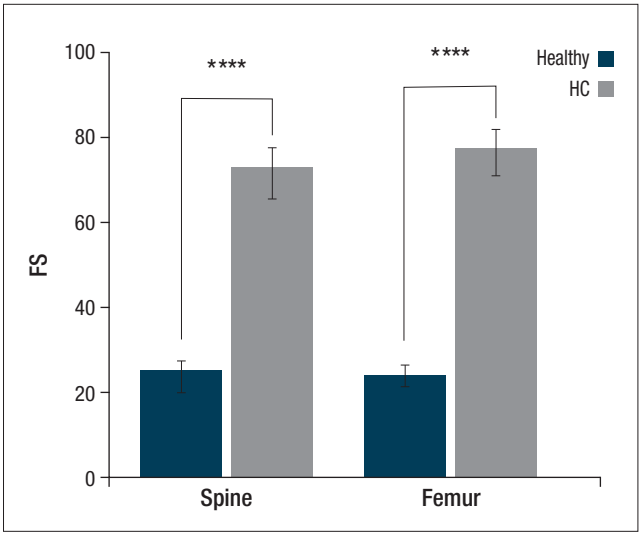


Figure 2 T-score measured at the lumbar spine and femoral neck in healthy and hypercholesteraemic (HC) patients. The healthy and HC population median for the spine is -1.60 [-2.2 – (-1.1)] and -2.30 [-2,75 – (-1.70)], respectively. For the femur the median is -1.90 [-2.35 – (-1.37)] and -2.2[-2.7 – (-1.7)] for healthy and HC population, respectively. Data are presented as median [first quartile-third quartile] (n = 105 in Hc group and n = 78 in control group; statistical significancy **** = $p < 0.0001$).

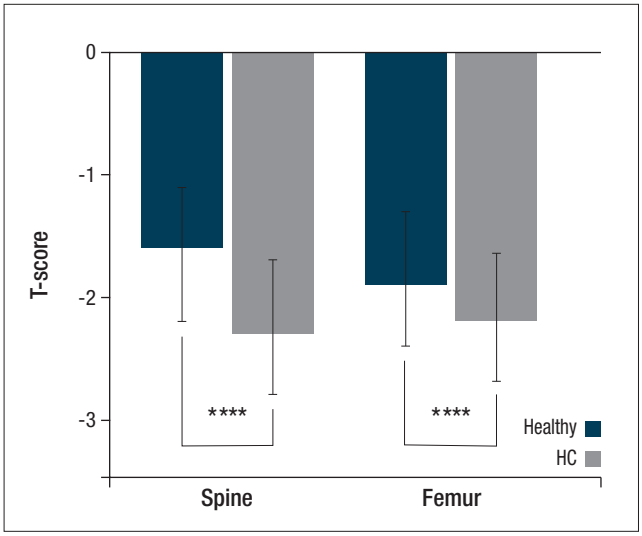
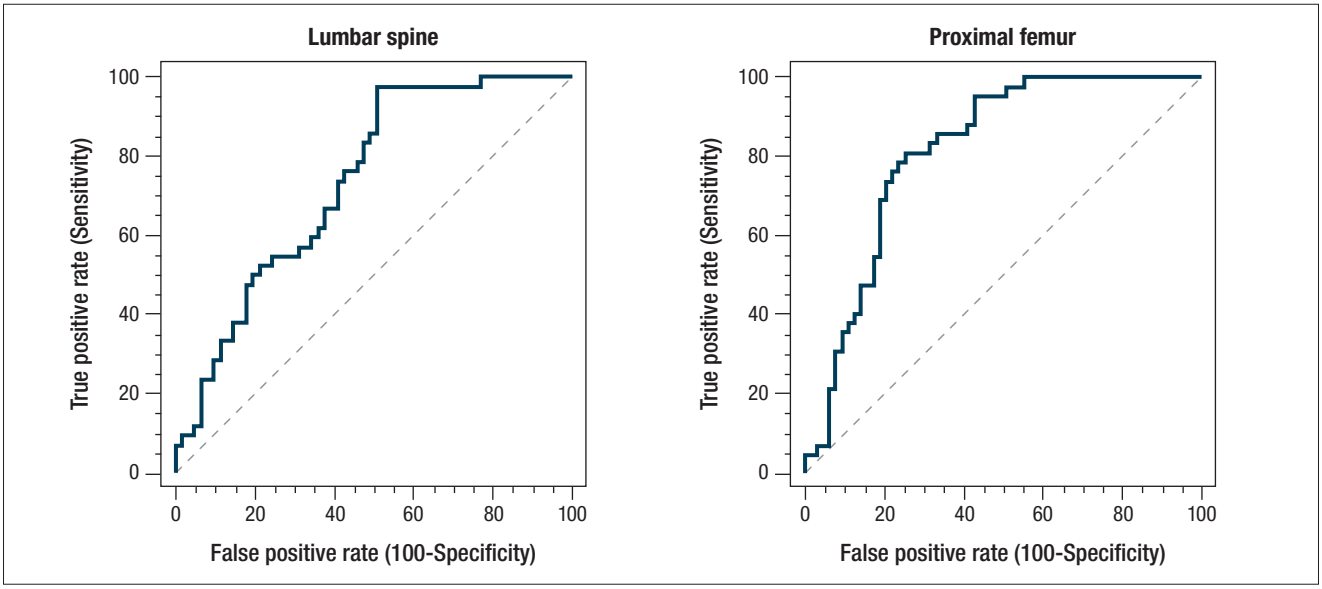


Figure 3 ROC curves showing sensitivity and specificity of REMS-FS for HC patients deteriorated bone quality. A black diagonal line representing the null hypothesis (area under the curve, AUC = 0.5).



plex relationship between this lipid and bone physiology is currently poor. Therefore, the present study, by means of REMS technology, aimed to assess bone quantity and quality in patients affected by HC compared with healthy controls.

As expected, the findings confirmed a significant association between HC and impaired bone health, as demonstrated by REMS-derived FS and T-score values. Patients with HC, compared with the control group, exhibited significantly higher FS values and lower T-scores at both the lumbar spine and the femur. These results indicate that HC is associated with worse bone quality and reduced BMD, aligning with previous epidemiological studies that suggested a negative correlation between cholesterol levels and bone integrity [9-12].

Several biological mechanisms could explain these findings. Cholesterol plays a crucial role in osteoclast differentiation and activation, enhancing bone resorption through increased expression of pro-osteoclastogenic factors, such as IL-1α and RANKL, while reducing OPG levels [13-15]. This imbalance in bone remodeling is likely to contribute to the higher FS and lower T-scores observed in HC patients. Additionally, oxidative stress, which is known to be exacerbated by elevated cholesterol levels, may further contribute to bone degradation [16]. The observation that statins (cholesterol-lowering drugs) exert a protective effect on bone health by modulating the RANKL/OPG pathway further supports the association between lipid metabolism and bone homeostasis [17,18].

The clinical implications of these findings are substantial. Given the high prevalence of HC and osteoporosis, early identification of at-risk individuals is crucial for effective intervention. Traditional bone density assessment methods, such as dual-energy X-ray absorptiometry, provide valuable insights into BMD but do not capture bone quality aspects [19]. In this context, REMS technology emerges as a promising tool for evaluating bone fragility beyond mere BMD measurements [20,21]. The significantly higher FS values observed in HC patients highlight the utility of REMS in assessing bone microarchitecture deterioration, which may not be detectable using conventional methods [22]. Furthermore, the AUC clearly showed the reliability of the FS to discriminate between HC patients with impaired bone health and healthy control subjects, thus proposing the FS parameter as an indicator of possible future fragility fractures also in HC cohorts. In this context, it is worth noting that traumatic fractures, unlike fragility fractures, do not affect the internal bone microarchitecture, thus preserving the use of FS as an indicator of bone quality.

In conclusion, this study demonstrated the detrimental impact of HC on bone health, as shown by increased FS values and decreased T-scores in HC patients compared with controls. These results reinforce the importance of considering lipid metabolism as a key factor in bone physiology and suggest that cholesterol management could play a role in osteoporosis prevention strategies [7]. However, to better explore the association, future research should focus on the impact that specific types of cholesterol (resulting from genetic mutations, diet, obesity, pathological conditions, etc.) may exert on bone deterioration. Furthermore, the application of REMS technology offers a non-invasive and highly sensitive approach for evaluating bone quality in patients with metabolic disorders [20,21]. Future research should explore potential therapeutic strategies targeting cholesterol-related pathways to mitigate osteoporosis risk in HC populations.

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