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Utility of REMS-Derived Fragility Score and Trabecular Bone Score in evaluating Bone Health in Type 2 Diabetes Mellitus

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A mia Mamma, a mio Papà, a mio Fratello, a Leonardo:

Il senso di ogni cosa.

Utility of REMS-Derived Fragility Score and Trabecular Bone Score in Evaluating Bone Health in Type 2 Diabetes Mellitus

1. Introduction

Diabetes mellitus

Diabetes mellitus (DM) is a group of metabolic disorders characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. Today, DM is considered a true global epidemic. Approximately 422 million people worldwide are affected by DM, and 1.5 million deaths are directly attributable to diabetes each year **(1)**. According to the World Health Organization (WHO), the prevalence of DM has been steadily increasing over the past decades, particularly type 2 diabetes mellitus (T2DM), which is strongly associated with excess body weight and unhealthy lifestyle habits (poor diet, physical inactivity) **(2)**. Based on ISTAT 2020 data, the prevalence of DM in Italy is approximately 5.9%, corresponding to more than 3.5 million individuals, with a slowly increasing trend in recent years. Prevalence rises with age, reaching 21% among individuals aged 75 years or older **(3)**.

DM can be classified into several clinically distinct entities:

-Type 1 diabetes mellitus (T1DM): caused by autoimmune destruction of pancreatic β-cells, usually leading to absolute insulin deficiency.

-Type 2 diabetes mellitus (T2DM): characterized by peripheral insulin resistance and impaired insulin secretion.

- Diabetes due to specific genetic defects of β-cell function or insulin action, or secondary to pancreatic diseases (such as cystic fibrosis or pancreatitis) or to the use of certain drugs or chemicals (e.g., glucocorticoids, HIV/AIDS therapy, or post-organ transplantation).

- Gestational diabetes mellitus (GDM): glucose intolerance occurring in some women during pregnancy, particularly in the second or third trimester.

There is also a subgroup of individuals whose glucose levels do not meet the diagnostic thresholds for DM but exceed normal ranges. Two conditions have therefore been identified:

Impaired fasting glucose (IFG), with fasting plasma glucose between 100 and 126 mg/dL;

Impaired glucose tolerance (IGT), with 2-hour plasma glucose during an oral glucose tolerance test between 140 and 199 mg/dL.

These states are defined as prediabetes and identify individuals at higher risk of developing DM. IFG and IGT are not clinical entities per se but rather risk factors for diabetes and cardiovascular disease. They are often associated with other metabolic abnormalities such as abdominal obesity, dyslipidemia (hypertriglyceridemia and/or low HDL cholesterol), and hypertension **(1)**.

Type 1 Diabetes Mellitus

Type 1 diabetes mellitus results from autoimmune destruction of pancreatic β-cells, leading to absolute insulin deficiency. It accounts for 5–10% of all diabetes cases. T1DM occurs most commonly during childhood but may develop at any age. The onset is highly variable—typically rapid in infants and children and slower in adults.

Some children and adolescents may present with diabetic ketoacidosis as the first manifestation, whereas others may initially show mild fasting hyperglycemia that can progress rapidly to severe hyperglycemia and ketoacidosis in the presence of infection or stress. In other individuals, particularly adults, residual β-cell function may persist for years, sufficient to prevent ketoacidosis.

Although insulin secretion is markedly reduced, brief periods of near-normal glucose homeostasis and insulin release may occur shortly after diagnosis. This transient phase is defined as the "honeymoon period", which may last 6 to 12 months **(4)**.

Type 2 Diabetes Mellitus

Type 2 diabetes mellitus, also referred to as non–insulin-dependent diabetes or adult-onset diabetes, is characterized in its prediabetic phase by early insulin resistance and compensatory hyperinsulinemia, followed by an eventual decline in insulin secretion, resulting in hyperglycemia and overt diabetes.

Its pathogenesis involves a complex interaction among genetic, metabolic, and environmental factors, leading to insulin resistance and β-cell dysfunction. Family history of diabetes, advancing age, unhealthy diet, obesity, and physical inactivity represent major risk factors.

In a healthy individual who develops insulin resistance, insulin secretion rises to maintain normal glucose metabolism. However, in genetically predisposed individuals, both insulin resistance and impaired insulin secretion contribute to the development of hyperglycemia. A small percentage of individuals with T2DM exhibit minimal insulin resistance but significant β-cell deficiency. Conversely, others show profound insulin resistance with only a mild defect in insulin secretion **(5)**.

Diagnostic Criteria

The diagnostic criteria for diabetes are as follows:

1. A1C ≥ 6.5%, or

2. Fasting plasma glucose ≥ 126 mg/dL, or

3. Plasma glucose ≥ 200 mg/dL during an oral glucose tolerance test, or

4. Random plasma glucose ≥ 200 mg/dL in a patient with classic symptoms of hyperglycemia (polyuria, polydipsia, fatigue, weight loss) **(1)**.

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“Diabetic Osteopathy”: a New Complication of Diabetes Mellitus

Individuals with diabetes mellitus (DM) are exposed to numerous severe complications, both acute and chronic.

Acute Complications

Diabetic ketoacidosis and hyperosmolar hyperglycemic state represent the two most severe acute metabolic complications of diabetes mellitus. These conditions may occur in both type 1 and type 2 diabetes. The mortality rate for diabetic ketoacidosis is below 5% in specialized centers, whereas the mortality associated with hyperosmolar hyperglycemic state remains high, around 15%.

Chronic Complications

Diabetes is associated with severe, debilitating, and potentially life-threatening chronic complications, which are categorized as *microvascular* and *macrovascular*.

Microvascular complications involve peripheral nerves (neuropathy), the kidneys (nephropathy), and the retina (retinopathy).

Macrovascular complications result from the deposition of atherosclerotic plaques in the arteries and therefore include stroke, myocardial infarction, and peripheral arterial disease.

Hyperglycemia is central to the development of complications in both type 1 and type 2 diabetes. The prevention or delay of onset and progression of chronic complications depends on sustained glycemic control and correction of metabolic abnormalities.

Bone tissue is also adversely affected by DM. For this reason, the term “**diabetic osteopathy**”has been introduced.

This term describes alterations in bone mineral density, bone formation and remodeling processes, and the increased risk of fractures associated with both type 1 and type 2 diabetes. Growing evidence supports the concept that bone should be considered a target organ in diabetes and that bone fragility should be recognized as a potential complication of the disease.

Fragility fractures are extremely frequent in individuals with DM and represent major causes of morbidity and mortality in both T1DM and T2DM.

A recent meta-analysis **(1)** showed that the relative risk (RR) of hip fracture is doubled in diabetic individuals compared with non-diabetics, with a particularly pronounced increase in T1DM (RR 5.3, CI 3.4–8.3) compared with T2DM (RR 1.6, CI 1.4–1.8). Case–control studies have also reported an increased risk of postoperative cardiovascular events in patients with DM and hip fracture, which contributes to prolonged hospitalization **(2)** and a 1.4-fold higher mortality in diabetics compared with non-diabetics **(3)**.

Overall, the risk of death following hip fracture is 28% higher in men with T2DM and 57% higher in women with T2DM **(4)**. For all these reasons, fractures represent a negative event not only for the individual patient but also for society, constituting a significant healthcare burden **(5)**.

Mechanisms Underlying Bone Fragility in Diabetes

The mechanisms contributing to skeletal fragility in diabetic individuals are multifactorial and vary according to the underlying pathophysiology of each type of diabetes. In T1DM, fragility is secondary to β-cell failure and consequently to low IGF-1 levels, typically manifesting at a young age **(6)**. In T2DM, by contrast, bone quality is predominantly affected. Fractures occur in later stages of the disease. Altered bone quality is associated with chronic hyperglycemia, accumulation of advanced glycation end-products (AGEs), proinflammatory cytokines and adipokines, and microvascular bone damage leading to osteocyte dysfunction, impaired bone turnover, and altered collagen properties **(7)**. Low bone mineral density (BMD) is observed in 22–37% of patients with T1DM **(8)**, often correlating with microvascular complications **(9)**. In T2DM, BMD levels are generally higher than in non-diabetic individuals **(10)**. This increase is more evident in younger patients and in men, and paradoxically in those with higher HbA1c levels **(11)**. Conversely, trabecular bone score (TBS) at the lumbar spine is reduced in T2DM **(12)**.

Pathogenesis of Impaired Bone Quality

Chronic Hyperglycemia and AGEs

Whereas reduced lumbar and femoral BMD may partially explain bone fragility in T1DM, BMD in T2DM is typically normal or even increased **(13)(14)**, despite a significantly higher fracture risk **(15–18)**. This discrepancy is attributed to several factors, including reduced bone strength, poorer bone quality, alterations in bone remodeling, microarchitectural defects, and changes in matrix composition and mineral components. Chronic hyperglycemia impairs bone quality both directly—by adversely affecting osteoblasts **(19)**—and indirectly through mechanisms such as plasma hyperosmolarity, acute acidosis **(20)**, and AGE formation. AGEs, formed by non-enzymatic reactions involving proteins (notably collagen), accumulate progressively in the bone matrix, disrupting cellular homeostasis **(21)**. Through activation of their receptor (RAGE), AGEs increase oxidative stress by promoting reactive oxygen species (ROS) production and inducing chronic inflammation **(22)**. They interfere with osteoblast differentiation, function, and collagen binding **(23–25)**. Glycation results in the formation of cross-links that weaken the mechanical properties of bone **(26)**, despite increased BMD in T2DM. Higher AGE levels have been correlated with vertebral fractures **(27)**. Pentosidine, a major AGE, shows associations between elevated urinary or serum levels and vertebral fracture risk **(28)(29)**. Correlation also exists between fracture risk and HbA1c levels, disease duration, and presence of complications **(30)**, indicating that poor glycemic control contributes to impaired bone quality.

Insulin, IGF-1, and Amylin

Patients with diabetes often exhibit reduced levels of insulin, IGF-1, and amylin, all of which are involved in bone metabolism. Serum IGF-1 levels are inversely associated with vertebral fractures in postmenopausal women with diabetes, independent of age, renal function, insulin secretion, and lumbar BMD **(31)**. Amylin acts on both osteoblasts and osteoclasts—stimulating the former and inhibiting the latter **(32)**. Its deficiency in T1DM may contribute to impaired bone metabolism, although further studies are needed to clarify this relationship and evaluate the role of amylin analogues as potential anti-osteoporotic therapies **(33,34)**. In T2DM, insulin levels decline in later disease stages, and its role appears more closely related to insulin resistance and reduced muscle strength secondary to decreased glucose uptake **(35)**.

Microvascular Bone Damage

Microvascular disease leads to impaired skeletal vascularization, particularly at the cortical level, reducing bone strength through decreased cortical density and increased cortical porosity, especially detectable at the radial site **(36)**.

Loss of the Incretin Effect

Incretins, including GIP and GLP-1, are gastrointestinal hormones responsible for the incretin effect. GLP-1 receptors are expressed on bone marrow stromal cells and immature osteoblasts **(37)**. GLP-1 primarily enhances insulin secretion and β-cell function **(38,39)**. Although incretins appear to exert favorable effects on bone, this remains an area requiring further investigation.

Chronic Inflammation and Obesity

Diabetes is characterized by a chronic low-grade inflammatory state, evidenced by persistently elevated proinflammatory cytokines, which exert anti-osteoblastic and pro-resorptive effects **(40)**. High cytokine levels may explain the increased fracture risk in obese diabetic individuals, who consequently lose the mechanical protective effect typically associated with higher adiposity. Excess adipose tissue may impair bone homeostasis through adipokines and other proinflammatory mediators **(41,42)**. Adiponectin, produced by adipose tissue, shows a complex and not fully clarified relationship with BMD. Levels are generally low in T2DM **(43)**. In vitro studies suggest anabolic effects on osteoblasts and inhibitory effects on osteoclasts **(44)**. Clinical correlations remain inconsistent, with studies reporting both inverse **(45)** and positive associations **(46)**. Leptin, produced by white adipose tissue and bone marrow adipocytes, is also reduced in T2DM. It regulates appetite, promotes osteoblastogenesis, and suppresses adipogenesis. One Japanese study found a negative correlation between serum leptin and urinary NTX levels and a positive correlation with radial Z-score, suggesting differential effects on cortical versus trabecular bone **(46,47)**.

Microstructural Bone Alterations

Assessment of bone microarchitecture and fracture resistance is essential for accurate fracture risk evaluation in diabetic bone disease. High-resolution peripheral quantitative CT (HR-pQCT) has yielded variable findings—some studies indicate greater cortical porosity, whereas others report no significant differences between diabetic and non-diabetic subjects **(48)**. Given its limited availability, more practical tools are needed.

The trabecular bone score (TBS), derived from lumbar spine DXA images, serves as a useful predictor of fracture risk. TBS is lower in T2DM patients than in controls and correlates with impaired microarchitecture and increased fracture risk **(49)**.

Altered Bone Turnover

Numerous studies demonstrate significantly reduced bone turnover in both T1DM and T2DM. Turnover abnormalities can be detected by evaluating serum and urinary bone turnover markers. In T2DM, reduced levels of CTX, TRAP5b, and osteocalcin have been reported **(50,51)**. This may reflect a state of relative hypoparathyroidism, potentially due to impaired calcium sensing or chronic hypomagnesemia in T2DM. Although data are heterogeneous—given variability in disease duration, age, ethnicity, and glycemic control—no definitive predictive value has been established for turnover markers on BMD or fracture risk. Osteocalcin, in both its carboxylated and undercarboxylated forms, is lower in T2DM patients than in non-diabetic controls **(52)**. The osteocalcin/alkaline phosphatase ratio correlates inversely with vertebral fractures in men with T2DM, and this association remains significant even after adjustment for confounders. A meta-analysis confirmed reduced osteocalcin and increased alkaline phosphatase levels, along with decreased CTX **(53)**. Sclerostin, an inhibitor of osteoblast activity, is elevated in T2DM and correlates positively with vertebral fractures **(54)**. This relationship is most evident in postmenopausal women with prior fractures, who also exhibit thinner cortical bone on quantitative CT. Sclerostin levels increase with weight loss, which is a primary therapeutic recommendation for T2DM. A 10% reduction in body weight corresponds to increased bone resorption and decreased bone mass **(55)**. Histomorphometric studies in T2DM reveal reduced osteoblast numbers and decreased osteoid tissue **(56)**. Biopsies from T2DM—but not T1DM—show reduced mineralization surfaces and decreased bone formation rates in trabecular, intracortical, and cortical bone **(57)**. Post-2015 biopsy analyses demonstrate reduced activation frequency of bone remodeling units and increased bone mineralization and pentosidine accumulation, which correlate with HbA1c levels. Taken together, these findings confirm the relatively low bone turnover characteristic of T2DM **(58)**.

Increased Risk of Falls

Older adults with T2DM experience more frequent falls than their non-diabetic counterparts. Risk factors include retinopathy, cerebrovascular cognitive decline, cardiovascular disease, neuropathy, and hypoglycemia related to insulin or insulin-secretagogue therapy. A linear relationship has also been observed between deterioration in renal function and fall frequency.

Reduced vitamin D levels, common in T2DM, contribute to decreased muscle strength and neuropathy **(59)**.

This clinical context has stimulated increasing interest in the development of easy-to-use and reliable diagnostic methods for assessing qualitative and structural skeletal alterations, as well as fracture risk, in patients with T2DM **(60)**. Among these, the most widely adopted tool has been the trabecular bone score (TBS).

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Trabecular Bone Score (TBS)

TBS is a texture-based index obtained from lumbar spine DXA images that captures variations in pixel gray levels, thereby offering an indirect assessment of trabecular bone microarchitecture. It is computed by constructing a variogram from the projected DXA image within the region of interest, which quantifies the sum of squared differences in gray levels between pixels separated by a specified distance.The slope of the log–log transformed variogram is then used to derive the TBS value. **(1)** By analyzing experimental variograms derived from projected DXA images, TBS is able to detect differences in three-dimensional (3D) bone microarchitecture even among two-dimensional (2D) DXA measurements that appear similar. Higher TBS values are associated with superior skeletal texture, reflecting a more preserved microarchitecture, whereas lower TBS values indicate poorer skeletal texture, consistent with microarchitectural deterioration. The association between TBS-derived texture parameters and 3D microarchitectural features has been demonstrated in several ex vivo studies, which have shown significant correlations between TBS and bone microstructural parameters assessed using micro–computed tomography**(2) (3).** From a clinical perspective, TBS has been demonstrated to predict both prevalent and incident fragility fractures in primary osteoporosis. It serves as a valuable complement to BMD and established clinical risk factors for fracture identification, risk stratification, and treatment monitoring. Moreover, multiple studies have highlighted the utility of TBS in secondary forms of osteoporosis, a context in which DXA-derived BMD often shows limited sensitivity for fracture risk prediction. Notably, in conditions such as diabetes mellitus or prolonged glucocorticoid exposure, the increased fracture risk is not adequately explained by the conventional relationship between DXA-measured BMD and fracture risk **(4).**

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TBS and Diabetes

A growing number of studies have assessed TBS in various conditions known to increase the risk of fragility fractures **(1)** and many of them have corroborated the ability of TBS to predict such fractures in patients with secondary osteoporosis **(2).** In general, compared to control subjects, TBS was reported to be lower in patients with diabetes **(3-4)**, primary hyperparathyroidism **(5)**, acromegaly **(6)**, anorexia nervosa **(7)**, hypercortisolism **(8)**. Previous studies have consistently demonstrated a strong association between diabetes and bone fragility, with clear evidence that both type 1 and type 2 diabetes mellitus adversely affect skeletal health, resulting in an increased risk of fractures **(9)**. Interestingly, a paradoxical relationship exists between type 2 diabetes (T2DM), bone mineral density, and fracture risk. “Despite having BMD values that are often comparable to or higher than those of the general population, individuals with T2D exhibit an increased risk of fragility fractures across all skeletal sites (10) despite having comparable or even higher BMD values measured by DXA. Alterations in skeletal properties or bone quality are possible explanations for this T2DM-related skeletal fragility **(11)**, and TBS could be useful for fracture risk assessment in these patients **(12)**. In 2013, Leslie and cols. **(13)** were the first to examine the association between TBS and the incidence of fractures in 29,407 women above the age of 50 years from the province of Manitoba, Canada, including 2,356 with diabetes (mostly T2DM). Interestingly, compared to controls, women with diabetes had higher baseline BMD at all sites, but lower TBS, even after adjusting for multiple confounding variables. Furthermore, over a mean of 4.7 years of follow-up, the incidence of MOF was greater in women with diabetes (7.4%, n = 175) than in non-diabetics (5.5%, n = 1,493; $p < 0.001$). They reported that TBS predicted MOF independently of BMD in women with diabetes (HR: 1.27, 95% CI: 1.10-1.46), similar to those without diabetes (HR: 1.31, 95% CI: 1.24-1.38). Other studies have also confirmed that despite greater BMD values, those with T2DM have lower TBS than controls **(14).** Another study found a greater prevalence of morphometric vertebral fractures in postmenopausal women with T2DM (34.3%) than in controls (18.7%, $p = 0.01$) (111). Vertebral fractures were associated with lower values of TBS (area under the curve, AUC: 0.69; $p < 0.0001$) and FN-BMD (AUC: 0.63; $p < 0.004$). A recent meta-analysis of 40,508 individuals (35,546 women and 4962 men; 4,269 patients with diabetes) showed that, overall, T2DM was associated with decreased TBS (more pronounced in women). However, there was evidence of substantial heterogeneity among studies – most of them used unadjusted TBS values, and only a few adjusted for parameters that may directly affect TBS, such as age, BMI, LS-BMD, and the TBS software. In summary, the relationship between T2DM and TBS is mixed.**(15)** Several studies **(16,17,18,19)** have shown that TBS is lower in patients with diabetes, especially in those with poor glycemic control, disease complications, and/or longer duration of disease. Recently, studies reported that the effect of abdominal soft tissue thickness (STT) should be considered when interpreting TBS in patients with T2DM in whom increased abdominal adiposity may artifactually reduce TBS values **(20)**. Another study examined TBS in 119 patients with type 1 diabetes (T1DM) (59 males, 60 premenopausal females; mean age = 43.4 years) and 68 matched healthy controls and found that TBS was comparable in T1DM patients and non-diabetic controls, but was lower in T1DM patients with existing clinical fractures (n = 24) than in controls **(21)**. Using a multivariate model, TBS ($p = 0.049$) and HbA1c ($p = 0.036$) were found to be independently associated with prevalent fractures in T1DM patients. A few other studies have examined the differences in TBS between T1DM patients and healthy controls and have reported heterogeneous results **(22)**.

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REMS (Radiofrequency Echographic Multi-Spectrometry)

In recent years, a novel technique has attracted considerable interest among osteoporosis experts worldwide: Radiofrequency Echographic Multi-spectrometry

(REMS). REMS is a non-ionizing method that assesses bone status through the analysis of raw, unfiltered ultrasound signals—referred to as radiofrequency (RF) signals—acquired during ultrasound examinations of the lumbar spine and proximal femur. The analysis of these native RF signals enables the extraction of information related to bone tissue characteristics. Bone density is subsequently estimated by comparing the spectral features of the acquired signals with previously established reference spectral models **(1).**

REMS enables the evaluation of bone health and the estimation of fracture risk through a rapid ultrasound examination of key axial skeletal sites, namely the lumbar spine and proximal femur. The technique employs a transducer that emits ultrasound waves toward the target area; the reflected signals are subsequently received and processed to reconstruct B-mode images of the region of interest. An automated analysis of the radiofrequency signals is then performed to identify bone interfaces and regions of interest (ROIs), as well as to assess the characteristics of the internal bone microarchitecture. Using this approach, essential anatomical structures—such as individual vertebral bodies, the femoral neck, femoral head, and greater trochanter—can be accurately recognized, allowing for the measurement and quantification of both bone mineral density (BMD) and bone quality **(2).**

Automated recognition of the target bone structures is accomplished through a series of image-processing procedures applied to each image frame. These steps include the organization of image data features into rectangular matrices, brightness masking, contrast enhancement, image smoothing, histogram equalization, thresholding, and morphological analysis (1). One of the advantages of REMS compared with DXA is its ability to automatically account for artefacts arising from calcifications, osteophytes, vertebral fractures, metallic implants, and similar factors, potentially leading to more accurate BMD measurements, as demonstrated in several recent studies. Measurements can be performed rapidly at both the femoral neck (40 s) and lumbar spine (80 s).

The radiofrequency signal is subjected to spectral analysis, and the resulting waveform is compared with data from reference populations, including both normal and osteoporotic subjects. This comparison allows the calculation of quantitative parameters such as BMD values, as well as T-scores and Z-scores, which are comparable to those obtained with DXA. BMD measurements are generated for each lumbar vertebra, the femoral neck, the total hip, and the greater trochanter. REMS-derived BMD values are based on spectral models originally developed using a reference population that also underwent DXA for osteoporosis classification. These data were subsequently reviewed by experienced operators to minimize potential errors—such as incorrect patient positioning, imprecise data analysis, or the presence of artefacts—that could otherwise lead to unreliable BMD measurements **(3)**

REMS showed good performance in identifying osteoporosis based on BMD. Sensitivity was 91.5% at the femoral neck and 91.7% at the lumbar spine, while specificity reached 91.8% and 92.0%, respectively. Agreement with reference measurements was 88.2% at the femoral neck and 88.8% at the lumbar spine when no T-score tolerance was applied, and increased to 98.0% and 97.4%, respectively, when a tolerance of 0.3 T-score was allowed. REMS also demonstrated high precision, with low intra-operator variability. Inter-operator variability was only slightly higher, at 0.48% for the femoral neck and 0.54% for the lumbar spine. Owing to its high precision and repeatability, REMS BMD measurements are suitable for short-term therapeutic monitoring, overcoming the limitations of other densitometric techniques that typically require at least one year between consecutive scans. **(2) (3) (4)**

The Fragility Score is a REMS-derived indicator of skeletal fragility, reflecting bone microarchitecture independently of BMD, and is assessed at the spine and femoral neck. It ranges from 0, indicating normal bone structure, to 100, representing maximal structural fragility. This parameter is calculated based on the proportion of ultrasound scan lines whose spectral profiles show a stronger correlation with a "fragile" (i.e., fractured) bone spectral model compared with a normal bone spectral model. The Fragility Score is subsequently used to estimate fracture risk over a five-year period through predictive models developed from a proprietary database containing data from both fractured and non-fractured individuals. The diagnostic accuracy of the Fragility Score for predicting incident fragility fractures over a five-year period has been validated against BMD T-scores obtained using both DXA and REMS, using the actual occurrence of major osteoporotic or hip fragility fractures as the reference gold standard. **(5) (6)**

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2. Materials and Methods

The primary objective of this single-center study was therefore to evaluate REMS-derived parameters (BMD and FS) in a large cohort of male and female patients with T2DM, and to explore their associations with clinical characteristics and the presence of osteoporotic fractures.

2.1. Study Population

A total of 300 consecutive Caucasian outpatients with type 2 diabetes mellitus (T2DM), including 129 men and 171 women, were recruited between January 2024 and June 2025 from the Diabetes Unit of the Department of Internal Medicine at the University Hospital of Siena. All participants provided written informed consent, and all data were anonymized prior to statistical analysis.

Eligibility criteria were as follows: age between 50 and 80 years; postmenopausal status for women; body mass index (BMI) ranging from 18.5 to 39.9 kg/m^2; age at T2DM diagnosis >30 years; and glycated hemoglobin (HbA1c) <7.5%. Exclusion criteria included prior or current anti-osteoporotic treatment (with the exception of calcium and/or vitamin D supplementation), the presence of malignant or metabolic bone diseases (such as cancer, multiple myeloma, or hyperparathyroidism), and the use of medications known to affect bone metabolism.

The control group consisted of 120 consecutive non-diabetic individuals (53 men and 67 women) referred to the outpatient clinic of the same department during the same study period, along with a small number of healthy volunteers recruited from hospital staff. Inclusion criteria for controls mirrored those of the T2DM group (age 50–80 years, BMI 18.5–39.9 kg/m², and postmenopausal status for women). Non-diabetic subjects with comorbidities or treatments potentially interfering with bone metabolism were excluded.

For all participants, a detailed personal and family medical history was collected, including smoking status, alcohol consumption, duration of menopause, duration of T2DM, and the presence of concomitant comorbidities. Anthropometric measurements were obtained under standardized conditions: height and weight were recorded, and BMI was calculated as weight (kg) divided by height squared (m²).

Overall, 52 individuals (40 with T2DM and 12 controls) were excluded for failure to meet inclusion criteria or withdrawal of consent, and an additional 43 participants (37 with T2DM and 6 controls) were excluded due to inadequate quality of BMD or REMS scans. The final analysis therefore included 223 participants with T2DM and 102 control subjects.

2.2. Dual-Energy X-Ray Absorptiometry (DXA)

Bone mineral density (BMD) of the lumbar spine (LS), femoral neck (FN), and total hip (TH) was measured in all participants using dual-energy X-ray absorptiometry (DXA; Discovery W, Hologic, Waltham, MA, USA), in accordance with standardized acquisition protocols. Diagnostic classification followed World Health Organization (WHO) criteria, with osteoporosis defined by a T-score ≤ −2.5 and osteopenia by a T-score between −1.0 and −2.5. T-scores were calculated using sex-specific Italian reference data.

To obtain additional information on bone microarchitecture, trabecular bone score (TBS) was calculated from standard anteroposterior lumbar spine DXA images using TBS iNsight software (version 2.2.0.0, Medimaps SA, Bordeaux, France). TBS values were generated through a fully automated, operator-independent process

2.3. Radiofrequency Echographic MultiSpectrometry (REMS)

Bone mineral density was additionally evaluated using radiofrequency echographic multispectrometry (REMS; EchoStation, Echolight SpA, Lecce, Italy) with a 3.5-

MHz convex ultrasound transducer. The REMS methodology, including its accuracy, precision, and reproducibility, has been described in detail elsewhere. In brief, the probe was positioned over the abdomen or hip to visualize the anatomical site of interest, with imaging depth and focal settings adjusted as required. Raw ultrasound signals were processed to generate a patient-specific spectral profile, which was automatically compared with reference spectral models matched for sex, age, anatomical site, and body mass index within a dedicated database.

Skeletal fragility was further assessed using the REMS-derived Fragility Score (FS), an index reflecting bone microarchitecture independently of BMD at both the lumbar spine and femoral neck. The FS ranges from 0, indicating normal microarchitecture, to 100, indicating maximal skeletal fragility, and is calculated as the proportion of scan lines more closely resembling a fragile (fractured) bone model rather than a normal bone model. The FS has been validated as a predictor of 5-year fragility fracture risk in both women and men. Lower FS values indicate preserved bone microarchitecture and reduced fracture risk, whereas higher values reflect compromised microarchitecture and increased skeletal fragility.

2.4. Laboratory Tests and Fractures Assessment

After an overnight fast of at least 12 hours, venous blood samples were collected to assess glycated hemoglobin, creatinine, calcium, phosphate, parathyroid hormone (PTH), C-terminal telopeptide of type I collagen (CTX),Sclerostin,Adiponectin, Myostatin and serum 25-hydroxyvitamin D (25OHD) concentrations. Routine biochemical parameters were measured using colorimetric methods (Autoanalyzer, Falcor 350, Menarini, Florence, Italy). Serum PTH levels were determined by an immunoradiometric assay (Total Intact PTH, Antibodies Lab Inc., Santee, CA, USA), with intra- and inter-assay coefficients of variation (CVs) of 3.6% and 4.9%, respectively. Sclerostin was quantified by enzyme-linked immunosorbent assay (ELISA). Adiponectin levels were measured using an ELISA method. Serum 25-hydroxyvitamin D (25OHD) concentrations were quantified using a chemiluminescence immunoassay (LIAISON 25OHD Total Assay, DiaSorin Inc., Stillwater, MN, USA), which showed intra- and inter-assay CVs of 6.8% and 9.2%, respectively.

2.5. Fracture Assessment

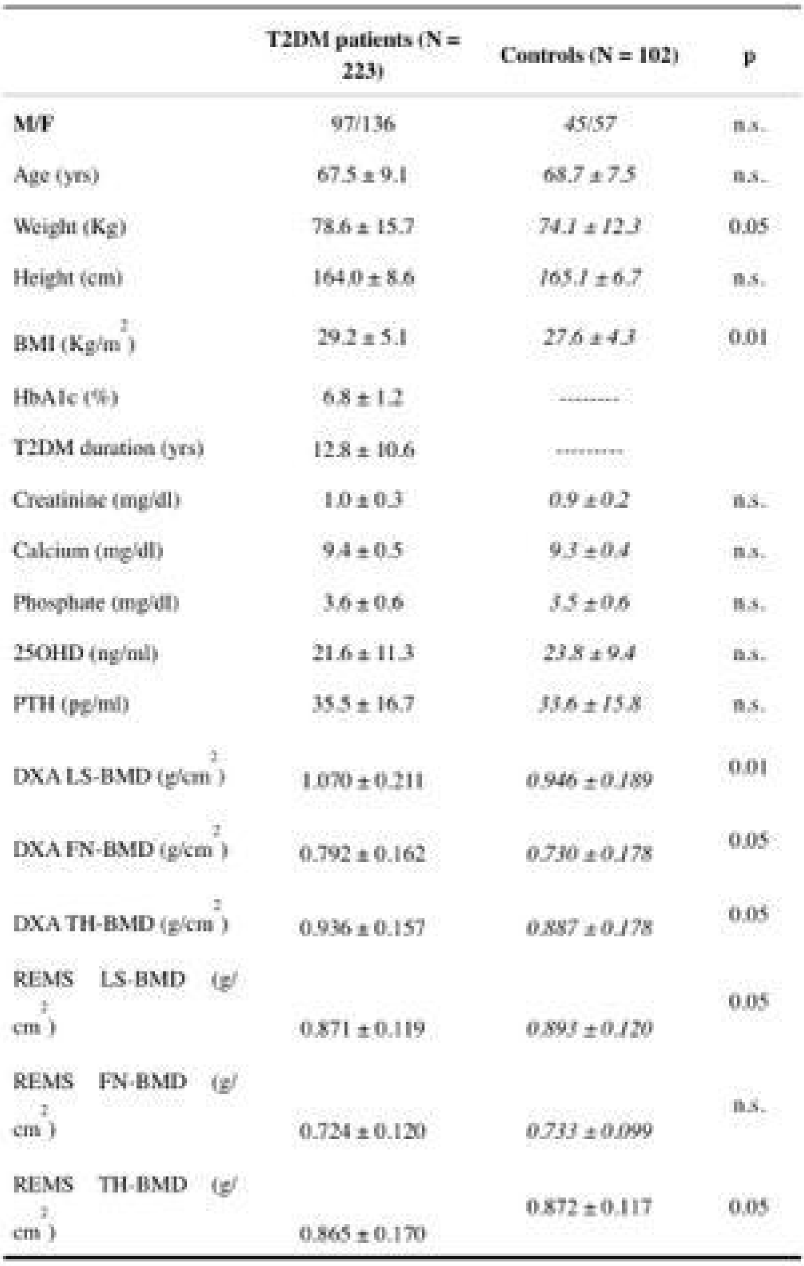
Prior Major Osteoporotic Fractures (MOF)—including those of the hip, spine, wrist, and humerus—were assessed in the T2DM group via both self-report and subsequent verification through clinical and radiological records.

2.6. Statistical Analysis

The values in the study are presented as “mean ± standard deviation (SD). The normality of the distribution of outcome variables was assessed using the Kolmogorov–Smirnov test. Clinical data and initial values of the measured variables in the study groups were compared using the Student’s *t*-test and Mann–Whitney U-test, depending on the appropriateness of the data distribution. Categorical variables were subjected to comparison using the Chi-square test or Fisher’s exact test, as deemed appropriate. Associations between different parameters were examined through Pearson’s correlation or Spearman’s correlation, as appropriate, or via partial correlation analysis. All statistical analyses were performed using the SPSS statistical package for Windows version 16.0 (SPSS Inc., Chicago, IL, USA).

3. Results

The T2DM and control groups were matched for age, height, PTH, and 25OHD levels **(Table 1)**. A significant difference was observed for BMI, which was higher in the T2DM cohort ($p < 0.01$). The mean diabetes duration was 12.8 ± 10.6 years. BMD findings diverged significantly by measurement technique: DXA showed higher BMD across all sites in T2DM patients, with LS-BMD and TH-BMD differences reaching significance ($p < 0.01$). In sharp contrast, REMS recorded lower BMD values at all sites in T2DM patients, with a significant reduction (*p* < 0.05) noted solely for LS-BMD.



	T2DM patients (N = 223)	Controls (N = 102)	p
M/F	97/136	*45/57*	n.s.
Age (yrs)	67.5 ± 9.1	*68.7 ± 7.5*	n.s.
Weight (Kg)	78.6 ± 15.7	*74.1 ± 12.3*	0.05
Height (cm)	164.0 ± 8.6	*165.1 ± 6.7*	n.s.
BMI (Kg/m^2)	29.2 ± 5.1	*27.6 ± 4.3*	0.01
HbA1c (%)	6.8 ± 1.2	---------	
T2DM duration (yrs)	12.8 ± 10.6	----------	
Creatinine (mg/dl)	1.0 ± 0.3	*0.9 ± 0.2*	n.s.
Calcium (mg/dl)	9.4 ± 0.5	*9.3 ± 0.4*	n.s.
Phosphate (mg/dl)	3.6 ± 0.6	*3.5 ± 0.6*	n.s.
25OHD (ng/ml)	21.6 ± 11.3	*23.8 ± 9.4*	n.s.
PTH (pg/ml)	35.5 ± 16.7	*33.6 ± 15.8*	n.s.
DXA LS-BMD (g/cm^2)	1.070 ± 0.211	*0.946 ± 0.189*	0.01
DXA FN-BMD (g/cm^2)	0.792 ± 0.162	*0.730 ± 0.178*	0.05
DXA TH-BMD (g/cm^2)	0.936 ± 0.157	*0.887 ± 0.178*	0.05
REMS LS-BMD (g/cm^2)	0.871 ± 0.119	*0.893 ± 0.120*	0.05
REMS FN-BMD (g/cm^2)	0.724 ± 0.120	*0.733 ± 0.099*	n.s.
REMS TH-BMD (g/cm^2)	0.865 ± 0.170	0.872 ± 0.117	0.05

Table 1.

Anthropometric, clinical and densitometric characteristics of the T2DM patients and the controls.

As illustrated in **Figure 1**, a clear and statistically significant disparity was observed between the two measurement modalities. The mean T-scores for BMD determined by REMS were substantially lower than the corresponding DXA T-scores at the lumbar spine (*p* < 0.001) and the total hip (*p* < 0.01).

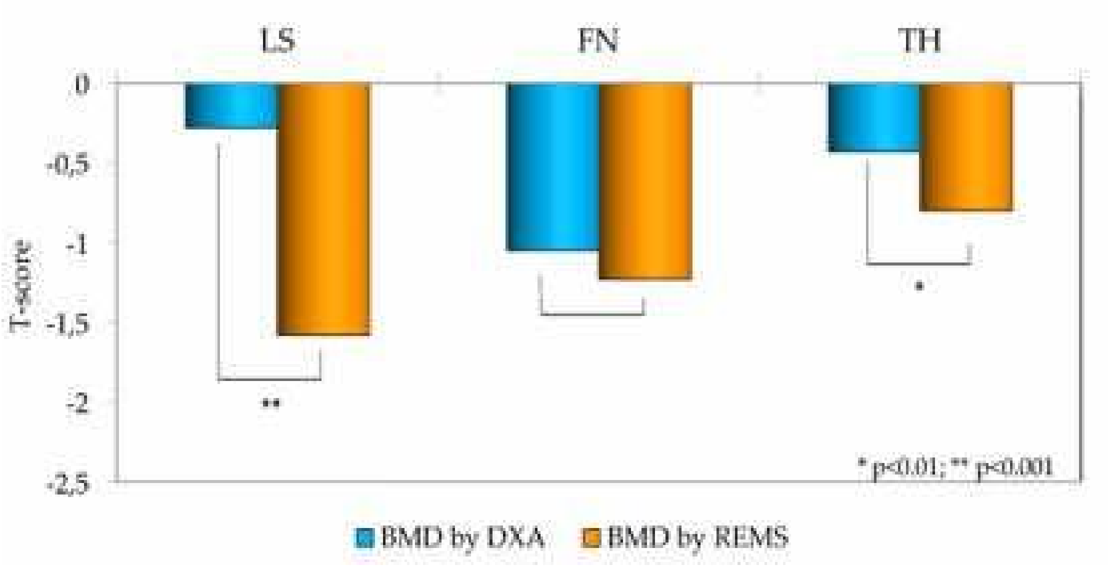


Figure 1.

Bone Mineral Density (BMD) T-scores at the Lumbar Spine (LS), Femoral Neck (FN), and Total Hip (TH) in T2DM patients, measured by DXA and REMS technique.

Figure 2 shows the percentage of T2DM men (A) and T2DM women (B) classified as “osteoporotic”, “osteopenic” or “normal” on the basis of BMD T-score values obtained by DXA and REMS technique, respectively. Regarding the male population, it is evident that the REMS technique allows a greater number of T2DM patients to be classified as osteoporotic and osteopenic than DXA (22.8% and 47.4% vs. 7.0% and 38.6%, respectively). Moreover, classification of bone status showed a marked difference between the two techniques, particularly in the T2DM group. Among T2DM men, the percentage classified as normal was substantially higher by DXA (54.4%) than by REMS (29.8%). The female population exhibited a similar pattern of reclassification. Specifically, REMS classified a significantly greater proportion of T2DM women as osteoporotic (43.9%) compared to DXA (17.8%). Conversely, the percentage of T2DM women categorized as osteopenic or normal was higher when assessed by DXA (46.7% and 35.5%, respectively) compared to REMS (40.7% and 15.4%, respectively).

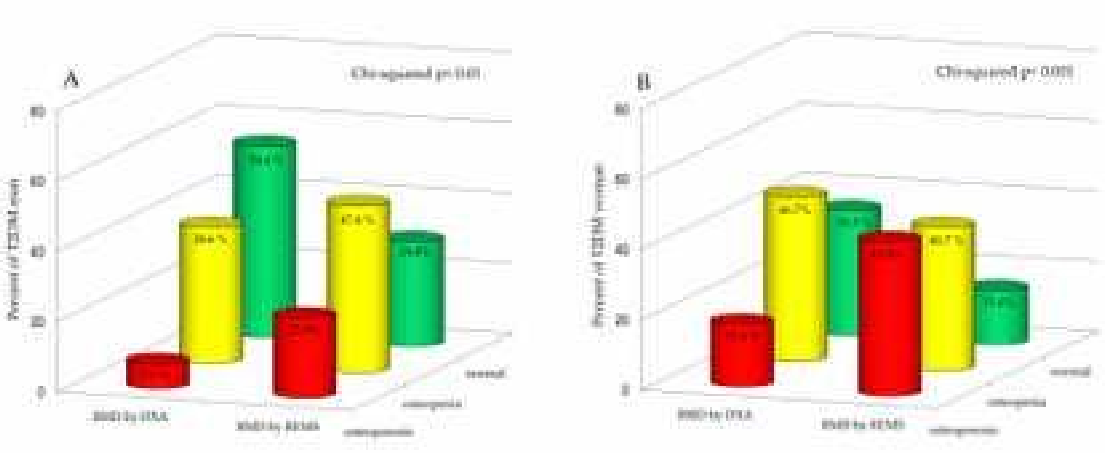


Figure 2.

Percentage of Type 2 Diabetes Mellitus (T2DM) men (A) and women (B) categorized as osteoporotic, osteopenic, or normal based on BMD T-score values obtained by DXA and REMS techniques.

A history of major osteoporotic fractures was reported in 42 (=18.8%) T2DM patients. More specifically, this was observed in 11 (=11.3%) males and 31 (=22.8%) females. Values of BMD expressed as T-score at lumbar spine (LS) and at total hip (TH) by DXA and REMS technique in T2DM patients with or without MOF are shown in **Figure 3.** As expected, the T2DM patients with previous MOF presented significantly lower values of T-score both BMD-LS and BMD-TH by DXA and T-score BMD-LS and BMD-TH by REMS with respect to those without fractures; however, the value at the T-score BMD LS by REMS technique, demonstrated higher statistical significance ($p < 0.01$).

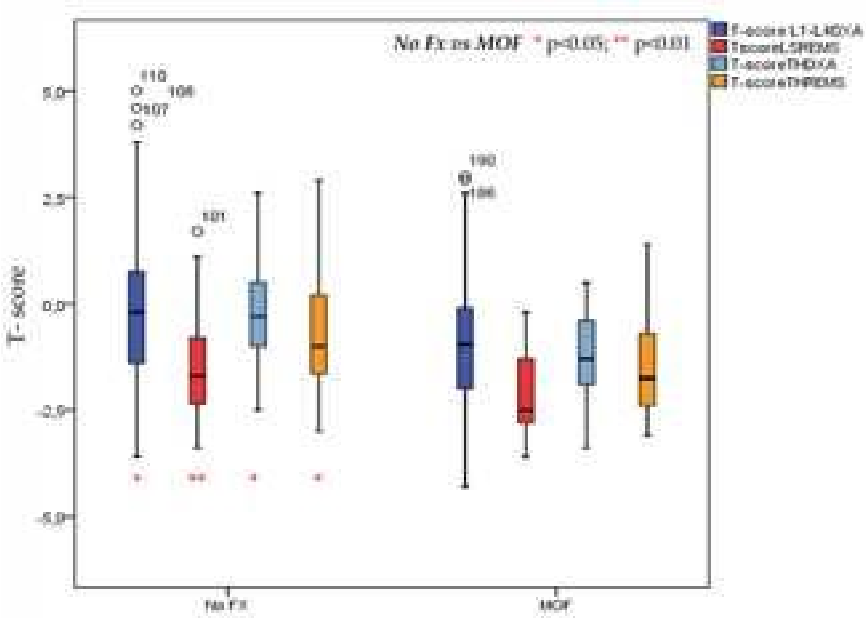


Figure 3.

Values of BMD expressed as T-score at lumbar spine (LS) and at total hip (TH) by DXA and REMS technique in T2DM patients with or without MOF.

Figure 4 illustrates the Trabecular Bone Score, measured by DXA, and the Fragility Score, measured by REMS, in patients with T2DM with or without MOF. As expected, patients with a history of MOF had significantly lower TBS values and significantly higher FS values at both lumbar and femoral sites compared with those without fractures ($p < 0.05$).

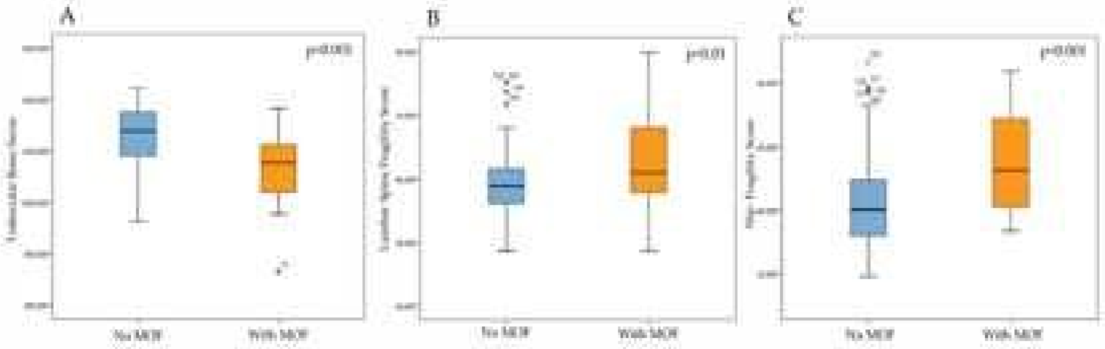


Figure 4.

Trabecular Bone Score by DXA (A), Fragility Score at lumbar spine (B) and Fragility score at hip (C) by REMS technique in DM2 patients with or without MOF.

4. Discussion

In the present study, we evaluated for the first time the clinical relevance of REMS-derived parameters, namely bone mineral density (BMD) and Fragility Score (FS), in a large cohort of male and female patients with type 2 diabetes mellitus (T2DM), comparing these measures with BMD and trabecular bone score (TBS) obtained by DXA. We found that REMS classified twice as many men as women with T2DM as osteoporotic. In addition, FS showed a discriminative ability comparable to that of TBS in distinguishing T2DM patients with and without major osteoporotic fractures. A growing body of evidence indicates that skeletal fragility and fragility fractures—particularly at the hip and distal radius—are frequent and clinically significant complications in individuals with T2DM. **(1,2).** This phenomenon is commonly referred to as the ‘diabetic bone paradox,’ as individuals with T2DM often present with normal or even increased BMD values. The apparent dissociation between BMD and fracture risk in T2DM is largely attributed to qualitative alterations in bone tissue, including microarchitectural deterioration, accumulation of advanced glycation end products (AGEs) within the collagen matrix, impaired bone remodeling, and changes in bone material properties, which collectively result in reduced bone strength despite preserved or increased bone mass **(3)**.

Unfortunately, bone quality is not readily quantifiable, as the most accurate techniques for assessing bone quality and strength—such as bone biopsy and microindentation—are invasive procedures and therefore not routinely applicable in clinical practice. **(3,4,5).**

High-resolution peripheral quantitative computed tomography (HR-pQCT) enables the acquisition of high-quality three-dimensional images of bone and represents a valuable non-invasive technique for the assessment of volumetric bone mineral density and other structural parameters. Nevertheless, owing to its technical complexity and limited availability, HR-pQCT is not suitable for routine clinical practice. Furthermore, the ability of HR-pQCT–derived parameters to reliably predict fracture risk in patients with T2DM remains uncertain. **(3).** In parallel, growing interest has been directed toward quantitative ultrasound (QUS) techniques. QUS is particularly appealing as it assesses bone properties through the attenuation and reflection of ultrasound waves and offers several practical advantages over DXA, including lower cost, portability, and the absence of ionizing radiation. **(6).** Nonetheless, studies evaluating QUS in patients with T2DM have produced inconsistent results, and QUS has not demonstrated sufficient ability to discriminate between diabetic individuals with and without fractures. **(7).** In addition, QUS is subject to several important limitations:

measurements are confined to peripheral skeletal sites, and the wide heterogeneity of currently available devices—each relying on different measurement techniques and parameters—limits the comparability and reproducibility of results. **(3,8).**

Radiofrequency Echographic Multi-Spectrometry represents an innovative, ultra-sound- based technology that enables the quantitative assessment of bone quality and strength without exposure to ionizing radiation **(9)**. The present study, conducted in a large and well-characterized cohort of men and women with T2DM, demonstrated that BMD values assessed by REMS were significantly lower in patients with T2DM compared with age- and sex-matched healthy controls. Moreover, the proportion of diabetic participants classified as osteoporotic according to REMS-derived BMD was substantially higher than that identified using DXA-derived BMD criteria. These findings are particularly noteworthy, as they contrast with the majority of previous studies based on DXA in T2DM populations, which have generally reported normal or even mildly increased BMD values compared with non-diabetic controls **(10,11)**. The present study corroborates the results of a preliminary investigation by Caffarelli et al. conducted in a cohort of postmenopausal women with T2DM. In that study, BMD values measured by DXA were higher in diabetic patients than in non-diabetic controls, whereas BMD assessed by REMS was significantly lower in the diabetic group. This divergence led to a substantial discrepancy in osteoporosis prevalence, with REMS classifying 47% of patients as osteoporotic compared with only 28% when osteoporosis was defined using DXA-derived BMD. **(12)**. The discrepancy between REMS and DXA measurements may be explained, at least in part, by the ability of REMS to minimize or exclude artifacts frequently observed in patients with T2DM—such as osteophytes, degenerative joint changes, vascular calcifications, and diffuse idiopathic skeletal hyperostosis—which are known to result in an overestimation of BMD when assessed by DXA**(8,9,13,14)**. Thanks to its capacity to recognize and exclude these confounding factors, REMS may provide a more accurate assessment of true skeletal status in this patient population. An additional and clinically meaningful observation from this study is that BMD values measured by REMS were significantly lower in T2DM patients with a documented history of major osteoporotic fractures compared with those without fractures. This association underscores the potential of REMS to capture alterations in bone microarchitecture and strength that are not adequately reflected by DXA-derived BMD measurements **(8,9).** The capability of REMS to detect individuals at increased risk of fracture has been extensively documented **(9,15,16).** In particular, a longitudinal study involving a large cohort of Caucasian women demonstrated that the REMS T-score represented a reliable and independent predictor of incident fragility fractures over a follow-up period of up to five years **(15)**.

Taken together, these findings suggest that REMS may share similarities with the trabecular bone score in its ability to capture alterations in bone quality in patients with T2DM. In this regard, Shevroja et al. reported that multiple studies involving more than 40,508 individuals, including 4,269 patients with diabetes, consistently showed lower TBS values in diabetic subjects compared with controls, despite higher BMD values in the diabetic group. Collectively, these observations support the role of TBS as a valuable tool for fracture risk assessment in patients with diabetes. **(17).** However, in one study, the difference in TBS between diabetic and non-diabetic individuals was statistically significant in women but not in men **(18)**. Several studies have further evaluated the ability of TBS to discriminate between T2DM patients with fractures and control subjects, showing that TBS values are reduced in diabetic patients and that TBS is associated with fracture risk **(19)**. In this regard, several studies have confirmed the role of TBS as an instrument that enhances diagnostic accuracy in distinguishing fragility fractures within the context of T2DM-related secondary osteoporosis **(20,21,22)**. Nevertheless, several important limitations constrain the clinical applicability of TBS. As it is derived from lumbar spine DXA images, TBS is inherently affected by the same sources of error and artifacts that influence BMD measurements, including degenerative spinal changes, osteophytes, and vascular calcifications. In addition, body mass index and abdominal adiposity are known to impact TBS values, frequently resulting in an underestimation of trabecular bone quality in overweight or obese individuals. This limitation is particularly relevant in patients with T2DM, among whom obesity is highly prevalent **(23).** However, the most recent versions of the TBS software seem to be less affected by the regional fat and BMI **(23,24).** A further limitation of TBS is that, as it cannot be applied to the femur, it fails to provide adequate information regarding the structural characteristics of cortical bone.

The Fragility Score is a dimensionless REMS-based measure of skeletal fragility that evaluates bone microarchitecture independently of BMD, specifically at the spine and femoral neck **(25)**. Several investigations have demonstrated that FS exhibits a significant association with both prevalent and incident fragility fractures **(25,26)**. Another interesting finding of this study is that FS values at both lumbar and femoral sites, were significantly higher in T2DM patients with a history of MOF. This observation appears to confirm previous investigations demonstrating that the FS shows a significant association with both prevalent and incident fragility fractures, independently of BMD values **(26,27)**. In their study, Pisani et al. evaluated a cohort of 1,989 Caucasian men and women aged 30 to 90 years to determine fracture incidence over a follow-up period of up to five years. Their results demonstrated a strong predictive performance of the Fragility Score (FS) in identifying individuals at increased risk of incident fragility fractures, with an area under the curve (AUC) of 0.811 in women and

0.780 in men. Notably, the discriminative ability of FS for fracture risk was superior to that of both REMS-derived BMD T-scores and DXA-derived BMD T-scores. **(26)**. Similarly, Lalli et al. reported, in a cohort of 175 patients with either primary or disuse osteoporosis, that FS values were significantly higher among individuals with a history of fractures than among those without previous fracture events **(27).** These findings further support the clinical utility of the FS as a reliable tool for fracture risk stratification, potentially enhancing the accuracy of current assessment methods based solely on densitometric parameters.

An important advantage of FS, especially at the femoral site, lies in its ability to provide clinically relevant information on skeletal fragility in a substantial proportion of diabetic patients with a BMI > 40 Kg/m^2, in whom TBS cannot be reliably assessed **(23)**.

Taken together, the results of the present study suggest that REMS may complement and integrate TBS in the evaluation of bone status in patients with T2DM.

This study has several limitations. First, the cross-sectional design precludes the establishment of causal relationships among the variables examined. Second, the relatively small number of participants with major fragility fractures may limit the generalizability of the findings.

Nevertheless, the study also has notable strengths. To our knowledge, this is the first investigation to specifically assess REMS-derived parameters, including FS and BMD, in male patients with diabetes. In addition, the single-center design allowed fracture status to be verified directly through radiological reports, thereby ensuring methodological consistency and diagnostic accuracy.

Conclusions

In conclusion, the findings of this study support the potential of REMS technology and FS as a valuable tool to improve the diagnosis of osteoporosis and the assessment of fracture risk in patients with type 2 diabetes mellitus.

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