Associations Between Bone Impact Microindentation and Clinical Risk Factors for Fracture

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Impact microindentation (IMI) measures bone material strength index (BMSi) *in vivo*. However, clinical risk factors that affect BMSi are largely unknown. This study investigated associations between BMSi and clinical risk factors for fracture in men. BMSi was measured using the OsteoProbe in 357 men (ages 33 to 96 years) from the Geelong Osteoporosis Study. Risk factors included age, weight, height, body mass index (BMI), femoral neck bone mineral density (BMD), parental hip fracture, prior fracture, type 2 diabetes mellitus (T2DM), secondary osteoporosis, smoking, alcohol consumption, sedentary lifestyle, medications, diseases, and low serum vitamin D levels. BMSi was negatively associated with age ($r = -0.131$, $P = 0.014$), weight ($r = -0.109$, $P = 0.040$), and BMI ($r = -0.083$, $P = 0.001$); no correlations were detected with BMD ($r = 0.000$, $P = 0.998$) or height ($r = 0.087$, $P = 0.10$). Mean BMSi values for men with and without prior fracture were 80.2 ± 6.9 vs 82.8 ± 6.1 ($P = 0.024$); parental hip fracture, 80.1 ± 6.1 vs 82.8 ± 6.9 ($P = 0.029$); and T2DM, 80.3 ± 8.5 vs 82.9 ± 6.6 ($P = 0.059$). BMSi did not differ in the presence vs absence of other risk factors. In multivariable models, mean (± SD) BMSi remained associated with prior fracture and parental hip fracture after adjusting for age and BMI: prior fracture (80.5 ± 1.1 vs 82.8 ± 0.4, $P = 0.044$); parental fracture (79.9 ± 1.2 vs 82.9 ± 0.4, $P = 0.015$). No other confounders were identified. We conclude that in men, BMSi discriminates prior fracture and parental hip fracture, which are both known to increase the risk for incident fracture. These findings suggest that IMI may be useful for identifying men who have an increased risk for fracture. **(*Endocrinology* 160: 2143–2150, 2019)**

Fractures in men contribute substantially to morbidity, decreased quality of life, and increased mortality (1–3). It is evident that areal bone mineral density (BMD) measured using dual-energy x-ray absorptiometry (DXA) does not fully elucidate fracture risk, given that the highest burden of fractures originates from people with a moderate reduction in bone mass (4–6). This indicates that determinants of bone strength other than BMD may play a role in bone fragility. Some of these determinants include alterations in bone microarchitecture, matrix composition, and intrinsic material properties of the bone.

Clinical risk factors such as age, weight, height, prior fracture, parental fracture, smoking (7), a sedentary lifestyle (8, 9), androgen deprivation therapy (ADT) (10, 11), selective serotonin reuptake inhibitors (SSRIs) (12), anticonvulsants (13), and vitamin D deficiency (14–17) contribute independently to the risk of fracture. Hence, the integration of multiple risk factors into fracture assessment tools such as the Garvan nomogram (18), fracture risk assessment tool [FRAX (7)], and our own fracture risk (FRISK) score (19) can improve fracture risk assessment (7) and inform management decisions.

ISSN Online 1945-7170

Received 31 May 2019. Accepted 9 July 2019.
First Published Online 16 July 2019

Abbreviations: 25OHD, 25-hydroxyvitamin D; ADT, androgen deprivation therapy; BMD, bone mineral density; BMI, body mass index; BMSi, bone material strength index; DXA, dual-energy x-ray absorptiometry; IMI, impact microindentation; SSRI, selective serotonin reuptake inhibitor; T2DM, type 2 diabetes mellitus.

Impact microindentation (IMI) allows *in vivo* evaluation of bone material strength index (BMSi), a property of cortical bone (20). Although some previous studies have reported an association between low BMSi and cortical porosity (21), high prevalence of fracture (22, 23), and decreased areal BMD (24), the ability of BMSi to discriminate fracture risk remains uncertain (24, 25). Further, little is known about how clinical risk factors for fracture affect BMSi measurements. Moreover, although some studies have shown no correlation between BMD and BMSi (26–28), differences in BMSi have been demonstrated in populations where discrepancies exist between BMD and fracture propensity, such as type 2 diabetes mellitus (T2DM) (29, 30), patients infected with HIV (31), glucocorticoid treatments (32), and chronic kidney disease (33).

In this study, we aimed to investigate associations between BMSi and age, anthropometrics [weight, height, body mass index (BMI)], BMD, history of fracture (parental hip fracture, prior fracture), T2DM, secondary osteoporosis (oral glucocorticoids, anticonvulsants, SSRIs, ADT, hyperparathyroidism, rheumatoid arthritis, and gastrointestinal diseases), lifestyle factors (smoking, alcohol consumption, sedentary lifestyle), and low vitamin D status.

Methods

Participants

Participants for this study were men from the Geelong Osteoporosis Study, a population-based cohort study situated in a geographically well-defined region in south-eastern Australia, known as the Barwon Statistical Division (34). The male arm of the Geelong Osteoporosis Study commenced in 2001 with recruitment of 1540 men aged 20 to 92 years. Participants are reassessed every few years, and data for this cross-sectional analysis were generated from the first 501 men assessed in the current follow-up phase (ages 33 to 96 years). The study was approved by the Human Research Ethics Committee at Barwon Health. All participants provided written informed consent.

Impact microindentation

IMI using the OsteoProbe RUO (Active Life Technologies, Santa Barbara, CA) was conducted to measure BMSi on the anterior surface of the midtibia (35). The indentation site was determined by measuring the midpoint from the medial border of the tibial plateau to the distal edge of the medial malleolus. Following disinfection of the area and administration of local anesthesia, the OsteoProbe was inserted through the skin and periosteum until reaching the surface of the bone at the anterior face of the midtibia. The probe has a radius of 10 μm or less, and is ~15 mm long. Participants are therefore excluded when the probe does not reach the periosteum without contact of the probe guide with the skin. At least 11 indentations were performed for each participant, of which the first measurement was systematically disregarded followed by 10 valid test indentations. The first measurement is disregarded to ensure that there is sufficient penetration of the probe through the periosteum. Two trained operators conducted the IMI measurements. The procedure was conducted according to internationally recognized recommendations for using the OsteoProbe (35). We have previously reported the feasibility and tolerability of IMI measurements in our cohort (36).

Other measures

Height and weight were measured to the nearest 0.1 cm and 0.1 kg, respectively. BMI (kg/m^2) was calculated. Areal BMD (g/cm^2) was measured at the femoral neck using DXA (Lunar; Prodigy, Madison, WI). Quality control was maintained through daily measurements of a Lunar DXA phantom.

Medical history and lifestyle factors

All participants completed comprehensive questionnaires detailing medical history, medication use, and lifestyle behaviors. A parental hip fracture referred to at least one maternal or paternal hip fracture. A participant's prior fracture was defined as any low-trauma fracture equivalent to a fall from a standing height or less, excluding fractures of the toe, skull, finger, and face, occurring during adulthood (age ≥ 20 years). Fractures were radiologically verified where possible. Median (interquartile range) time elapsed since prior fracture was 12 (2 to 25) years. Current smoking referred to use of tobacco at least daily and excessive alcohol consumption as three or more units daily on average. Mobility was categorized as: active (moves, walks and works energetically, participates in vigorous or light exercises) or inactive/sedentary (sedentary, limited walking, no appreciable exercise, not able to walk without considerable assistance, or not able to walk). Secondary osteoporosis included current use of oral glucocorticoids (n = 3), anticonvulsants (n = 11), SSRIs (n = 12), ADT (n = 3), and history of hyperparathyroidism (n = 4), rheumatoid arthritis (n = 5), or gastrointestinal disease (n = 6).

T2DM status

Diabetes was classified as fasting plasma glucose ≥ 7.0 mmol/L (126 mg/dL) and/or a self-report of diabetes and/or use of antihyperglycemic agents. T2DM was determined by examination of medical records.

Vitamin D status

A blood sample was collected after an overnight fast, and serum 25-hydroxyvitamin D (25OHD) was measured using Siemens assay (37).

Statistical analysis

The distribution of continuous data was visually assessed for normality using histograms. Categorical data were considered as binary variables. Associations between clinical risk factors and BMSi values were identified using Pearson's correlation for continuous variables and two-sample *t* tests for categorical variables. Multiple linear regression models were used to determine whether identified differences in BMSi were independent of other

factors. The models were tested for interaction terms. Serum 25OHD was assessed as both a continuous and categorical variable (<28, 28–50, 51–75, and >75 nmol/L).

The necessary assumption for linear regression model was investigated using Q-Q plots. A sensitivity analysis was performed after excluding suspected outliers. Statistical analyses were performed using Minitab V.18 (State College, PA).

Results

Of 501 potential participants in this current follow-up, 357 underwent IMI testing. Reasons for nonmeasurement in 144 men were needle phobia (n = 18), existing skin infections (n = 39), excessive soft tissues around midtibia region (n = 78), discomfort (pressure, not pain) after the first indentation (n = 5), inability to provide informed consent (n = 2), and two participants did not provide any reasons for declining. Compared with participants, nonparticipants were older (mean age ± SD: 65.6 ± 1.3 vs 58.3 ± 0.9 years, $P = 0.163$) and had greater BMI (30.8 ± 7.8 vs 26.7 ± 3.1 kg/m^2, $P < 0.001$). Blood test results were available for 300 participants. There were three bisphosphonate users in this population, which was too small for any subgroup analyses.

Table 1. Participant Characteristics (n = 357)

	Mean (± SD)
Age (y)	63.2 ± 13.8
Weight (kg)	81.4 ± 11.4
Height (cm)	174.1 ± 6.9
BMI (kg/m^2)	26.8 ± 3.2
Femoral neck BMD (g/cm^2)	0.958 ± 0.128
BMSi	82.6 ± 7.2
Parental hip fracture, n (%)	34 (9.5%)
Prior fracture, n (%)[a]	38 (11.9%)
T2DM, n (%)	44 (12.3%)
Secondary osteoporosis, n (%)[b]	44 (12.32%)
Smoking, n (%)	21 (5.9%)
Alcohol consumption, n (%)[c]	60 (16.8%)
Sedentary, n (%)	76 (21.3%)
Serum 25OHD (nmol/L)[d]	63.94 ± 20.10
25OHD (<28 nmol/L)	7 (2.33%)
25OHD (28–50 nmol/L)	72 (24.0%)
25OHD (51–75 nmol/L)	132 (44.0%)
25OHD (>75 nmol/L)	89 (29.7%)

[a]Fractures were 2 vertebra, 2 hip, 2 foot, 3 elbow, 4 ankle, 5 humerus, 8 tibia, and 12 rib.

[b]Current use of oral glucocorticoids, anticonvulsants, SSRIs, ADT, and presence of hyperparathyroidism, rheumatoid arthritis, or gastrointestinal diseases.

[c]Consumes three or more units of alcohol daily.

[d]Missing data for 57 men.

Unadjusted data

Participant characteristics and prevalence of risk factors are presented in Table 1. BMSi was negatively correlated with age ($r = -0.131$, $P = 0.014$), weight ($r = -0.109$, $P = 0.040$), and BMI ($r = -0.083$, $P = 0.001$). No associations were detected between BMSi and BMD ($r = 0.000$, $P = 0.998$), height ($r = 0.087$, $P = 0.10$), or serum 25OHD ($r = 0.009$, $P = 0.883$) (Fig. 1A–1D). No differences were detected between subgroups when 25OHD was assessed as a categorical variable.

BMSi in the presence or absence of risk factors were as follows (mean ±SD; Fig. 2): parental hip fracture, 80.1 ± 6.1 vs 82.8 ± 6.9 ($P = 0.029$); prior fracture, 80.2 ± 6.9 vs 82.8 ± 6.1 ($P = 0.024$); T2DM, 80.3 ± 8.5 vs 82.9 ± 6.6 ($P = 0.059$); secondary osteoporosis, 82.6 ± 5.4 vs 82.6 ± 7.1 ($P = 0.944$); smoking, 83.7 ± 5.5 vs 82.8 ± 5.7 ($P = 0.286$); high alcohol consumption, 82.0 ± 7.4 vs 82.7 ± 6.8 ($P = 0.490$); and sedentary lifestyle, 82.0 ± 7.9 vs 82.7 ± 6.6 ($P = 0.433$).

BMD did not differ in the presence or absence of parental hip fracture (0.936 ± 0.12 vs 0.959 ± 0.1, $P = 0.302$) or prior fracture (0.944 ± 0.1 vs 0.958 ± 0.1, $P = 0.507$).

Multivariable models

In multivariable models, mean BMSi remained associated with prior fracture and parental hip fracture after adjusting for age and BMI (prior fracture: $\beta = -2.21$, $P = 0.057$; parental fracture: $\beta = -2.95$, $P = 0.015$). BMD did not contribute to either model. After age adjustment, the association between BMSi and T2DM was attenuated ($\beta = -1.07$, $P = 0.403$).

Sensitivity analysis

After excluding four potential outliers, BMSi was again correlated with age ($r = -0.119$, $P = 0.026$), BMI ($r = -0.192$, $P < 0.001$), parental hip fracture (mean ± SD, 83.1 ± 6.4 vs 80.1 ± 6.1, $P = 0.009$), and prior fracture (83.2 ± 6.3 vs 80.2 ± 6.1, $P = 0.007$). The patterns observed with multivariable models remained unchanged (data not shown).

Discussion

We report that BMSi was negatively correlated with age, weight, and BMI, and that mean BMSi values were lower for men with prior fracture or parental hip fracture. No differences in mean BMSi were detected on the basis of smoking, sedentary lifestyle, secondary osteoporosis, alcohol consumption, or low vitamin D status.

In terms of fracture history, our findings are similar to those of Duarte Sosa and Fink Eriksen (28), who

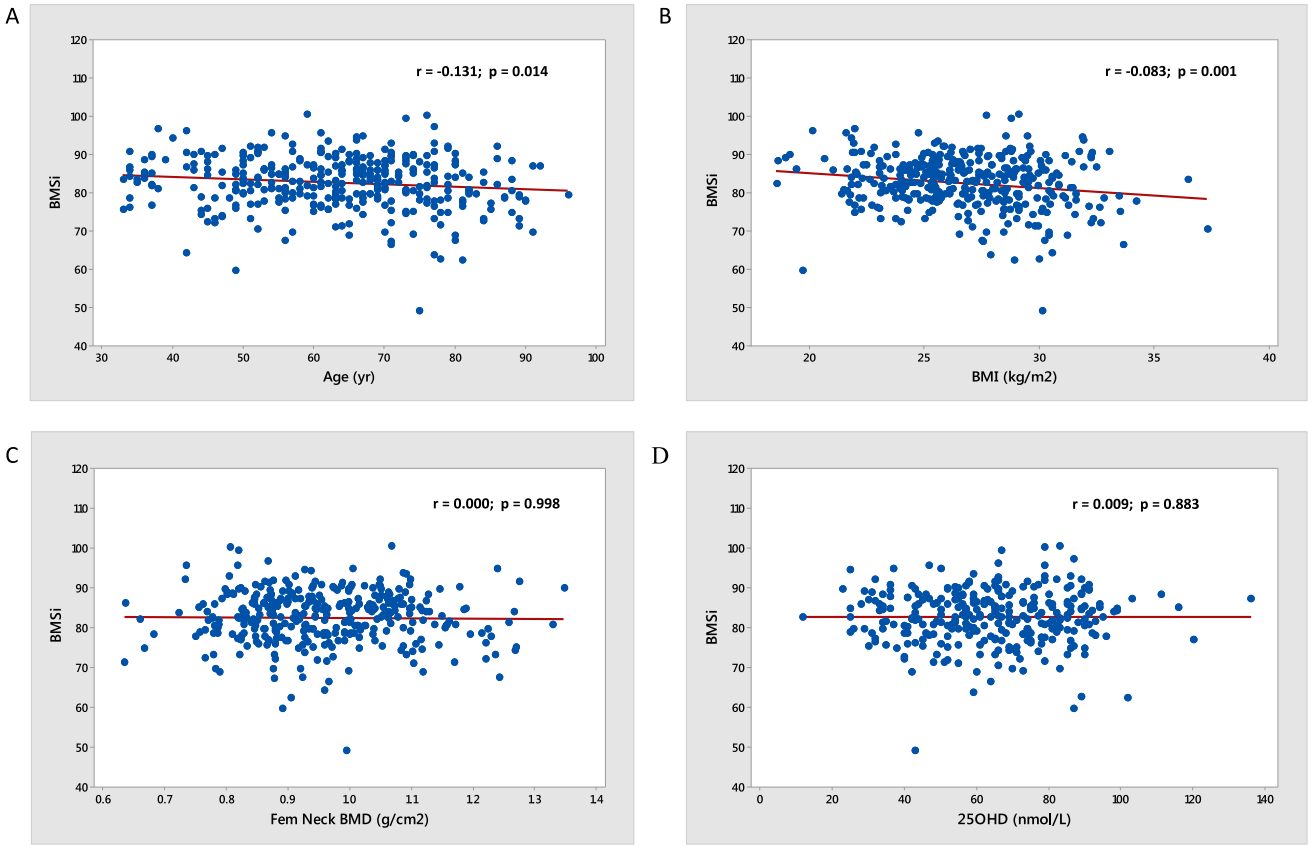

Figure 1. Scatterplots showing BMSi and (A) age; (B) BMI; (C) femoral (Fem) neck BMD; and (D) serum 25OHD.

reported a lower mean BMSi in 30 Norwegian women (aged 19 to 85 years) with prior stress fractures compared with 30 controls. They excluded women with a recorded history of low-energy fracture sustained at ≥ 45 years or a BMD *T*-score of ≤ −2.5 for at least one skeletal site. Similarly, Malgo *et al.* (26) observed a lower BMSi in 63 patients (24 men, aged 40 to 85 years) with a fragility fracture compared with nonfracture peers, despite similar BMD. All participants had low BMD (osteopenia or osteoporosis as diagnosed by DXA), whereas exclusion

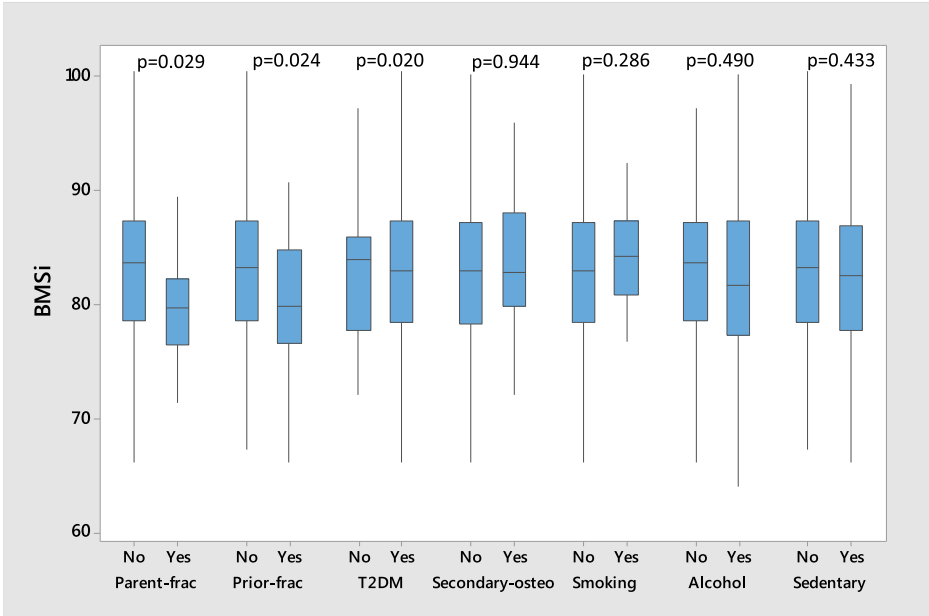

Figure 2. Boxplot of unadjusted BMSi values for participants, showing differences in BMSi values in participants with and without risk factors for fracture, assessed using two-sample *t* tests. frac, fracture; osteo, osteoporosis.

criteria included metabolic bone disorders other than osteoporosis, low serum 25OHD, kidney impairment, current use of glucocorticoids, aromatase inhibitors, chemotherapy, ADT, and other bone-acting agents.

Conversely, Johansson *et al.* (38) and Rudäng *et al.* (24) did not find differences in BMSi between fracture and nonfracture participants in a Swedish population-based study of 472 women (aged 75 to 80 years) and 211 women (mean age 78 years), respectively, although some participants had been exposed to antiosteoporosis drugs. It is well established that there is an increased risk of subsequent fracture in individuals who have previously experienced a fracture (39). Previous fractures may suggest defects in the microarchitecture of bone, and the presence of nonskeletal influences (such as increased risk of falls or decreased protective responses including changes in posture and reflexes) that increase the risk of fracture. Moreover, in the Swedish studies, the participants were of advanced age, when bone density and structure are substantially deteriorated and, in these cases, BMSi may have a limited contribution to the final sum of material properties, density, and microarchitecture in determining bone strength. Similarly, a parental history of fracture has been linked to an increased fracture risk in individuals, suggesting the importance of genetic factors in the determination of fracture risk (40). The mechanism for the increased risk in individuals has been sparsely documented but, irrespective of the mechanism, our results suggest that bone material properties are altered in men with a prior or parental fracture and that BMSi captures elements of bone fragility.

Our findings on the association between age and BMSi are similar to that of Malgo *et al.* (41) where they reported an inverse association between age and BMSi for 90 patients (male and female) with low bone mass ($r = -0.539$; $P < 0.001$). In contrast, Duarte Sosa *et al.* (27) reported no association between age and BMSi in 42 Norwegian and 46 Spanish women and among 30 women with previous stress fractures and 30 normal controls. The reasons for lack of consistency in results are not clear but may reflect differences in age profiles and composition of study populations.

BMSi was negatively correlated with weight and BMI, but there was no association with height or BMD. Similarly, Sundh *et al.* (21) reported a negative correlation ($r = -0.17$, $P = 0.01$) between BMSi and BMI in a population-based study of 202 women between 75 and 80 years of age, and Rudäng *et al.* (24) reported a weak inverse correlation ($r = -0.14$, $P = 0.04$) between BMSi and weight in a sample of 211 women between 75 and 80 years of age, and no association with height. A high BMI has been linked to high BMD and conventionally been considered a protective factor against fractures, but there has been recent evidence that a substantial proportion of fractures occur in the obese (42, 43). There are insufficient data to explain our finding; hence, further research in this area is warranted.

We did not observe any association between BMSi and BMD. This is consistent with previous studies by Malgo *et al.* (26) and Duarte Sosa *et al.* (27). In the latter study, involving Norwegian women with higher BMD but lower BMSi than Spanish women, there was no correlation found between BMD and BMSi. In contrast, Rudäng *et al.* (24) reported a positive association between BMSi and BMD in a sample of 211 Swedish women (mean age 78 ± 1.1 years). In this study, we had a wider age span and a lower mean age than the population examined by Rudäng *et al.* (24).

Although there is some evidence to suggest that smoking and high alcohol consumption are linked to lower BMD in postmenopausal women and older men, and increased risk of fracture (44, 45), in our study we observed no differences between smokers and nonsmokers and none between participants who consumed more or less than three units of alcohol daily. To the best of our knowledge, there have been no studies exploring the relationship between BMSi and alcohol consumption. So far, the only published study on the association between BMSi and smoking reported no differences between smokers and nonsmokers (24).

To date, there has only been one exercise intervention study that included assessment with the OsteoProbe, where Sundh *et al.* (46) reported changes in BMSi in response to high impact loading. After a 3-month jumping exercise program, these researchers reported an increase in BMSi in the intervention leg compared with the control leg, in 20 healthy and inactive postmenopausal women aged between 51 and 59 years. In our study, we did not detect any differences in mean BMSi for men who were physically active compared with physically inactive men, based on self-reported mobility criteria. Possible effects on physical activity and BMSi readings warrant further research.

Our results did not detect any difference in BMSi for men with and without secondary osteoporosis. To our knowledge, no previous studies have evaluated the association between BMSi and anticonvulsants, SSRIs, ADT, or the presence of hyperparathyroidism, rheumatoid arthritis, or gastrointestinal diseases. However, in a population-based cohort of 211 women aged between 75 and 80 years, Rudäng *et al.* (24) reported no significant BMSi differences between current users and nonusers of glucocorticoids and bisphosphonates. Similarly, Popp *et al.* (25) reported that BMSi was not

associated with long-term use of bisphosphonates in a sample of 153 postmenopausal women. A smaller study conducted by Aasarød *et al.* (47) involving 17 chronic atrophic gastritis patients aged 54 ± 13 years and 41 sex- and age-matched controls showed no differences in BMSi between patients and controls.

Our findings on the association between BMSi and T2DM contrast with other reports of reduced BMSi in patients with T2DM, even after adjustment for potential confounders (29, 30). The reason for the difference in our findings compared with other studies is unclear, and further investigation into this area is warranted.

Furthermore, Popp *et al.* (25) reported no association between BMSi and serum 25OHD. Vitamin D deficiency has been associated with an increased risk of falls and hip fractures (16) and low vitamin D status associated with low BMD in women (17). However, in our study, we did not detect an association between BMSi and serum 25OHD. Our null result could mean that serum 25OHD level has no impact on bone material properties as measured by IMI. However, few men in our study had 25OHD levels below 28 nmol/L, and it is likely that a wider range of vitamin D levels would be needed to detect a relationship.

A strength of this study is that we investigated the associations between BMSi and recognized risk factors for fracture, including fracture history, lifestyle factors, and secondary osteoporosis, in the largest sample and widest age range of men to date. However, small numbers limited our ability to consider subgroups of secondary osteoporosis or bisphosphonate users. Unlike most of the previous studies, this study is population-based and unselected on the basis of disease status. The outcome will thus be relevant for the broader male population. Where possible, we have used objective measures for factors such as anthropometry and BMD; however, we also used some self-reported data, which may have been subject to recall bias. In addition, we investigated men only and acknowledge that the observations may not be generalizable to women or other populations.

In summary, the cross-sectional data presented here indicate that BMSi, as measured by IMI, captures elements of bone fragility such as effect of ageing, prior fracture, and parental hip fracture. Therefore, our data contribute to the evidence that IMI has a potential utility for identifying differences in cortical bone that may predispose men to fracture and lays the foundation for longitudinal studies that could explore how BMSi might add to the predictive ability of BMD for fracture. Nonetheless, more research on how traditional bone parameters and other health and lifestyle factors influence BMSi is warranted.

Acknowledgments

Financial Support: The Geelong Osteoporosis Study was supported by grants from the National Health and Medical Research Council (NHMRC; projects 299831 and 628582, to J.A.P.) and Amgen-GSK OA-ANZBMS (to J.A.P.), but these supporters played no role in the collection or interpretation of data. P.R.-M. is supported by a Deakin University Postgraduate Industry Research Scholarship. K.L.H.-K. is supported by an Alfred Deakin Postdoctoral Research Fellowship. A.D.-P. owns shares of Active Life Scientific, Inc., the manufacturer of the RPI device. M.A.K. and J.A.P. are recipients of grants from the NHMRC and Amgen-GSK OA-ANZBMS.

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Disclosure Summary: The authors have nothing to disclose.

Data Availability: The datasets generated during and/or analyzed during the current study are not publicly available but are available from the corresponding author on reasonable request.

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