J Nutr Health Aging. 2021;25(1):71-76

ASSOCIATION BETWEEN BONE MINERAL DENSITY AND NUTRITIONAL STATUS, BODY COMPOSITION AND BONE METABOLISM IN OLDER ADULTS

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Abstract: *Purpose:* To identify an association between bone mineral density (BMD) and nutritional status, body composition and bone metabolism in older patients. *Methods:* Cross-sectional study, involving older adults, with osteopenia/osteoporosis and with normal BMD. The mineral density of the lumbar spine from L1 to L4 and the proximal region of the femur was assessed using dual energy X-ray absorptiometry. Biochemical analyzes were performed of 25(OH)-D, calcium and parathormone. Weight, knee height, and abdominal (AC), mid-upper arm (MUAC) and calf (CC) circumferences were measured. The percentage of body fat (%BF) and Fat-Free Mass (FFM) were quantified by electrical bioimpedance analysis. The Body Mass Index (BMI) was calculated. The statistical analysis used bivariate and multivariate, parametric and/or non-parametric tests, and was considered significant when $p < 0.05$. *Results:* Of the total 51 older adults assessed, 30 of them (58.8%) were diagnosed with osteopenia/osteoporosis. Body weight ($p = 0.001$), BMI ($p = 0.001$), % BF ($p = 0.030$) and serum concentrations of 25(OH)-D ($p = 0.003$) were higher in the group without changes in BMD. BMI and serum levels of 25(OH)-D demonstrated a positive correlation with the BMD of all bone compartments and the AC displayed a positive correlation with the lumbar vertebrae. In the logistic regression models, adjusted for sex and age, the BMI and the serum concentration of 25(OH)-D were presented as a protective factor against osteopenia/osteoporosis. *Conclusions:* Higher body weight, BMI, AC and %BF, and sufficient serum levels of vitamin D, were shown to be promoters of BMD.

Key words: Aging, body composition, bone diseases, bone density.

Introduction

Aging is a complex phenomenon, during which hormonal and metabolic changes are observed, which cause changes in body composition, with reduced muscle mass, greater weight gain and a loss of bone mass, the latter predisposing to a greater risk of fractures and bone diseases (1, 2).

Osteoporosis is a skeletal disorder often observed in the older population, especially women, characterized by impaired bone microarchitecture that causes a reduction in bone mineral density (BMD), predisposing to an increased risk of falls and routinely, the occurrence of fractures, thereby compromising the quality of life and increasing morbidity and mortality in older patients (3).

The measurement of BMD is used to assess the bone density of different bone compartments. According to the World Health Organization (WHO) the reference standard for diagnosing bone diseases is the assessment of BMD at the femoral neck and the lumbar spine, due to the fact that these areas present the highest fracture rates, with significant morbidity and mortality rates (4).

Body composition measurements (muscle tissue and adipose tissue) have presented controversial associations in different studies regarding their contribution to the adequate maintenance of BMD (5-7). For some researchers, excess weight is related as a protective factor against developing osteoporosis (7, 8). Despite the contradictions observed, there seems to be a consensus amongst authors that adequate nutritional status is also a protective factor for maintaining bone health. The preservation of muscle mass is proposed as the main protective factor against metabolic bone diseases, since the mechanical and hormonal effects promoted by muscles stimulate the activity of bone tissue cells and promote the maintenance of adequate bone density (6, 7). Low weight or weight loss, on the other hand, have been characterized as a predictor of low BMD, therefore presenting a greater occurrence of fractures in older adults (3, 8, 9).

Given the above, the objective of this study was to identify the association between anthropometric indicators of nutritional status and bone mineral density in older adults.

Methods

This was an observational cross-sectional study with a non-probabilistic sample, composed of individuals aged 60 years or older, admitted to the Clinic for Nutrition and Metabolism in Aging (LANME) in the Faculdade de Nutrição at the Universidade Federal de Alagoas. Were included people aged between 60 and 80 years, who were not taking hormonal, calcium and vitamin D supplements, and had no previous diagnosis of metabolic diseases, known to compromise calcium homeostasis, vitamin D and parathormone, such as liver

Received May 8, 2020
Accepted for publication June 1, 2020
Published online August 10, 2020, http://10.1007/s12603-020-1452-y

disease, chronic kidney disease, and primary or secondary hyperparathyroidism.

The bone mineral density of each participant was assessed using the dual energy x-ray absorptiometry test (DEXA). The BMD of the L1-L4 vertebrae and the proximal region of the femur (neck, greater trochanter and Ward triangle) were taken using the dual-energy x-ray densitometry device (DXA), according to the Hughes protocol (10).

The BMD values (g/cm^2) were standardized as T-score and Z-score values, values based on the variability of the BMD standard deviation (SD) measured in patients, using the BMD mean of the reference population as a comparison. The T-score was used to diagnose osteopenia and osteoporosis according to criteria established by WHO (11).

All measurements were performed in triplicate, using the mean values of each. The participants were weighed, wearing light clothing and no ornaments, on a digital calibrated G-TECH® scale, with a sensitivity of 50g and a maximum capacity of 150 kg.

Considering the structural changes inherent to the aging process, it was decided to use the height estimated by measuring the knee height, which was performed with an inelastic tape and the measurement obtained was applied in a predictive equation proposed by Chumlea and cols (1985) (12). With the data of weight (kg) and estimated height (m), the Body Mass Index (BMI) was calculated through the ratio between weight and square height, adopting the classification proposed by Lipschitz (1994) (13).

The abdominal circumference (AC) was measured with an inelastic tape, positioned at the height of the navel. The cutoff points established by the Education Program - Adult Treatment Panel III (NCEP-ATP III) were adopted, which consider a circumference ≥ 88 cm for women and ≥ 102 cm for men borderline values for cardiovascular events (14).

The mid-upper arm circumference (MUAC) and calf circumference (CC) were measured according to the techniques proposed by Lohman et al (1988), using inelastic fiberglass tape. A measurement of the tricipital skinfold (TSF) was also performed on the non-dominant arm using a Cescorf Innovare® adipometer, with a capacity of 100 mm and a sensitivity of 1 mm, following the criteria of Lohman et al (1988). The adequacy of the measurements was verified according to established protocols, using the 75thpercentile for the respective age and sex as a reference (15). Muscular reserve was based on calculating the arm muscle circumference (AMC), which was performed according to the equation proposed by Gurney and Jelliffe (16).

The body composition of each participant, percentage of body fat (% BF) and Fat-Free Mass (FFM), was determined with a bio-electrical impedance analysis (BIA) using a Sanny® tetrapolar device. On the day before the assessment, participants received a set of instructions so that the results would suffer no interference, such as: no strong physical exercise the day before, having a good night's sleep, nochange in their eating habits the day before the assessment, no alcoholic beverages for at least 8 hours, to drink regular amounts of water and maintain a 4-hour fasting period before the assessment.At the time of the assessment, participants were instructed to urinate before undertaking the examination and to remove their shoes, socks and any ornaments made of electrically conductive material. They were then placed in the supine position on a non-conductive surface with arms and legs separated at an angle of 30º. Asepsis was performed with cotton wool and alcohol on the right hand and foot where electrodes were then placed, at least 5 cm apart, according to the evaluation protocol suggested by Lukaski (17) and the manufacturer of the device used.

In this study, serum concentrations of calcium, 25(OH)-D, and parathormone (PTH) were assessed as markers of bone metabolism. Calcium was measured using the selective electrode technique with automatic correction for pH variation. The reference values used were 8.4-10.2 mg/dL. The 25(OH)-D was measured using the quantitative determination method, based on the principle of chemiluminescence (CLIA). The reference values used were: deficient (serum levels <20 ng/mL; insufficient (serum levels between 20 and 30 ng/mL) and sufficient (levels ≥ 30 ng/mL). The PTH was calculated using the chemiluminescence method and the reference values adopted were between 4.0 - 58 pg/mL Participants were also requested to undergo a 12-hour fast period in order to perform biochemical tests the following day.

The collected data were first organized into Excel® 10.0, and were later transferred to the SPSS Statistical Program 23.0®. To verify the distribution of variables, the Kolmogorov-Smirnov test was performed. Continuous variables were expressed as mean ± standard deviation or median ± interquartile range, respecting the distribution of variables. Categorical variables were presented by simple and absolute frequency.

Association between the categorical variables was tested using Pearson's X^2 tests or Fisher's exact test. The difference between two means was identified using the Student's t test or the Mann Whitney. The analysis of variance (ANOVA) was performed to identify the difference between three mean values. To assess the existence of a linear correlation between the anthropometric measures, body composition, bone metabolism markers and BMD, Pearson's Correlation test was performed for the normal distribution of variables and Spearman's correlation test for the non-normal distribution of variables. Measures for the coefficients were also adopted in order to measure the degree of the relationship between the variables. Thus, r <0.4 (weak correlation); r ≥ 0.4 and <0.6 (moderate correlation); r ≥ 0.6 (strong correlation). Finally, a logistic regression model was used to assess continuous variables. The level of significance was set at p <0.05.

All participants signed the informed consent forms, and the study was approved by the Research Ethics Committee at Universidade Federal de Alagoas, under nº. 432.659/2013.

Table 1
Comparison of the mean values of the anthropometric variables, body composition measurements and bone metabolism markers in the group of older adults diagnosed with osteopenia/osteoporosis and the group of healthy older adults, 2019

	BMD normal		BMD osteopenia/osteoporosis		CI	p
	Mean/Median	SD/IQR	Mean/Median	SD/IQR		
Weight (Kg)	78.12	14.1	64.54	10.62	6.62 — 20.54	0.001*
AC (cm)	97.91	12.55	89.84	92.5	1,87 — 14,26	0.012*
MUAC (cm)	31.82	3.33	30,49	3.54	- 2.14 — 4.81	0.429*
CC (cm)	37.22	4.93	36.58	3.63	- 3.27 — 4.55	0.736*
TSF (mm)	22.75	10.36	26.00	8.93	- 12.17 — 5.67	0.455*
AMC (cm)	26.09	3.54	25.14	6.23	- 4.5 — 6.40	0.717*
BF (%)	43.08	6.33	38.64	5.09	0.,45 — 8.43	0.030*
FFM (%)	57.11	6.26	61.51	4.88	- 829 — - 0.51	0.028*
BMI (Kg/m²)	28.63	4.51	24.13	3.66	2.18 — 6.80	0.001*
Ca (mg/dL)	9.60	0.51	9.59	0.47	- 0.26 — 0.29	0.916*
25(OH)D (μg/ dL)	40.38	12.77	31.51	6.74	3.33 — 14.39	0.003 *
PTH (pg/mL)	40.00	23.50	43.00	16.50	-	0.667#

BMD Bone Mineral Density; AC abdominal circumference; MUAC mid-upper arm circumference; AMC arm muscle circumference; CC calf circumference; TSF tricipital skinfold; AMC arm muscle circumference; BF (%) = Percentage of body fat; FFM (%) = Percentage of fat-free mass; BMI Body Mass Index; 25(OH)-D = Vitamin D; CI = Confidence Interval - 95%; IQR = Interquartile Range (p25 - p75); * Student t test; # Mann-Whitneytest; p value <0.05

Results

The older adults were divided into two groups, according to their bone health status: Group with a normal BMD, (21 older adults), and group diagnosed with ospeopenia/osteoporosis (30 older adults). Among the evaluated, female gender was the majority, mainly in the group diagnosed with changes in BMD, 80%, and without changes in BMD, 61.9% were women. The mean age of the participants in the whole sample was 66.9 ± 5.12 years, with no difference between groups (p> 0.05). Sufficient circulating levels of 25-hydroxyvitamin D was found in 85.7% and 43,3% of the older adults with and without change in BMD, respectively.

It was observed that the highest number of eutrophic or overweight older adults belonged to the group with a normal BMD (p = 0.03). When comparing the mean values of the anthropometric measurements, body composition and bone metabolism markers, it was observed that weight (p = 0.001); AC (p = 0.012); % of body fat (p = 0.030); BMI (p = 0.001) and 25(OH)-D (p = 0.003) presented higher mean values in the group of older adults presenting with a normal bone mineral density. The group with osteopenia/osteoporosis however, presented a higher mean value of% FFM (p = 0.028) (Table 1).

Serum concentrations of 25(OH)-D and BMI presented a moderately positive correlation with the BMD of all bone compartments and the AC presented a moderately positive correlation with all the assessed lumbar vertebrae (Table 2).

Analyzing the BMD of the older adults according to the classification of their nutritional status, it was observed that the mean values of the bone compartments were significantly higher in the overweight group (Table 3).

A logistic regression analysis was performed to identify the association between the indicators of nutritional status and the markers of bone metabolism with an impaired bone health status, adjusted for age and/or sex. The initial model included BMI, TSF, % BF, AC, MUAC, AMC, CP,% FFM, Calcium, PTH and serum concentrations of 25(OH)-D. With 95% confidence, it was observed that when adjusted only for sex or for sex and age, an increase of each 1 kg/m2 of BMI reduced the chance of older adults presenting with inadequate BMD maintenance by approximately 26% (adjusted OR = 0.743/adjusted OR = 0.742) (Table 6). Similarly, when adjusted only for sex or for sex and age, for an increase of each 1μg/dL of vitamin D, there was a 9% reduction in the probability of an individual presenting BMD impairment (adjusted OR = 0.905 / adjusted OR = 0.903) (Table 4).

Discussion

In this study, being overweight, an increased AC and a high percentage of body fat were associated with normal BMD, suggesting that these variables are involved in maintaining bone health. This finding could be justified according to the principle of Wolff's law that defends the idea that this tissue is capable of adapting to the mechanical stresses caused by body weight that result in bone deformation, which in turn may trigger a signal transduction cascade that culminates in the activation of cells that promote bone mass maintenance, such as osteoblasts (18).

Table 2
Correlation between bone mineral density, anthropometric variables, body composition measurements, bone metabolism markers, 2019

	Bone Compartment Assessed					
	L1	**L2**	**L3**	**L4**	**R Femur**	**L Femur**
BMI (Kg/m²)	r= 0.571	r= 0.572	r= 0.511	r= 0.510	r= 0.342	r= 0.367
	p= 0.000	p= 0.000	p= 0.000	p= 0.000	p= 0.015	p= 0.008
CC (cm)	r= 0.286	r= 0.164	r= 0.157	r= 0.080	r= - 0.017	r= - 0.021
	p=0.209	p= 0.477	p= 0.497	p= 0.729	p= 0.943	p= 0.930
MUAC (cm)	r= - 0.026	r= - 0.067	r= 0.025	r= 0.222	r= - 0.134	r= - 0.158
	p= 0.915	p= 0.784	p= 0.920	p= 0.361	p= 0.595	p= 0.519
AMC (cm)	r= - 0.240	r= - 0.165	r= - 0.089	r= - 0.118	r= - 0.141	r= - 0.164
	p= 0.323	p= 0.498	p= 0.716	p= 0.631	p= 0.576	p= 0.502
AC (cm)	r= 0.560	r= 0.551	r= 0.506	r= 0.553	r= 0.195	r= 0.176
	p= 0.000	p= 0.000	p= 0.000	p=0.000	p= 0.180	p= 0.220
TSF (mm)	r= 0.001	r= - 0.110	r= - 0.129	r= - 0.112	r= - 0.415	r= -0.313
	p= 0.997	p= 0.634	p= 0.578	p= 0.630	p= 0.069	p= 0.167
BF (%)	r= 0.124	r= 0.096	r= 0.114	r= 0.183	r= 0.358	r= 0.269
	p= 0.483	p= 0.590	p= 0.520	p= 0.301	p= 0.041	p= 0.124
FFM (%)	r= - 0. 143	r= - 0.111	r= - 0.123	r= -0. 196	r= - 0.344	r= - 0.251
	p= 0.419	p= 0. 533	p= 0.487	p= 0.267	p= 0.050	p=0.152
25(OH)-D (ng/mL)	r= 0.555	r= 0.485	r= 0.415	r= 0.522	r= 0.392	r= 0.437
	p= 0.000	p=0.000	p= 0.002	p=0.000	p=0.005	p=0.001
Serum Calcium (mg/dL)	r= - 0.115	r= - 0.143	r= - 0.229	r= - 0.212	r= - 0.131	r= - 0.098
	p=0.421	p= 0.316	p= 0.106	p= 0.135	p= 0.365	p= 0.495
PTH (pg/mL)#	r= 0.005	r= 0.037	r= 0.000	r= - 0.048	r= - 0.114	r= - 0.073
	p= 0.970	p= 0.796	p= 0.998	p= 0.739	p= 0.430	p= 0.611

CC calf circumference; MUAC mid-upper arm circumference; AMC arm muscle circumference; AC abdominal circumference;; TSF tricipital skinfold; AMC arm muscle circumference; BF (%) = Percentage of body fat; FFM (%) = Percentage of fat-free mass; BMI Body Mass Index; 25(OH)-D = Vitamin D; R Femur= Right Femur; L Femur= Left Femur; r – Pearson correlation; # - Spearman correlation; P value< 0.05

Increased AC and risk of fractures have been documented in the literature, being observed a negative effect of visceral adiposity on bone health, increased the risk of fracture (19, 20).

A prospective cohort conducted in ten European countries, reported results similar to the present study, in which the highest BMI and the highest body weight were inversely related to a lower BMD and the consequent lower risk of vertebral fractures (21). Prieto-Alhambra et al (2012) observed that obesity, defined by the highest BMI, in males participating in the study, was associated with a significantly reduced risk of fracture in the lumbar spine, due to the maintenance of bone mass (22).

Androgen hormones are associated with body weight for the maintenance of BMD (23). A higher number of adipocytes causes a higher level of sex steroids with a consequent reduction in bone remodeling due to the anti-resorptive and anabolic effects (24). Osteoclasts in women have specific estrogen receptors, called estrogen alfa - ERα - which, when recognized by osteoclastic cells, inhibit the differentiation and activity of these cells and stimulate bone synthesis and mineralization, by intensifying the differentiation of new osteoblasts and the inhibition of calcium mobilization (23). In men, the same occurs due to the action of testosterone and to a lesser extent of androsterone (25).

The adipose tissue currently recognized as an endocrine organ, in addition to offering substrate for the synthesis of sex hormones, important for the maintenance of BMD, secretes adipokines responsible for stimulating the activity of cells that promote bone density (26). Adiponectin and leptin, hormones that are secreted by adipose tissue, promote BMD, mainly acting to regulate bone turnover by stimulating the proliferation and differentiation of osteoblasts (24).

Published data demonstrate that in obese individuals, there is less incidence of hip and spine fractures, due to the cushioning

Table 3
Comparison between the mean values of the bone compartments assessed with the classifications of the nutritional status of the older patients according to bone health status, 2019

	Low Weight (n=10)		Eutrophy (n=21)		Overweight (n=20)		F	P*
	Mean	± SD	Mean	± SD	Mean	± SD		
L1	0.871[c]	0.082	1.023	0.209	1.334[a]	0.203	6.474	0.003
L2	0.924[c]	0.148	1.052	0.194	1.168[a]	0.218	5.338	0.008
L3	0.967[c]	0.116	1.118	0.238	1.223[a]	0.239	4.522	0.016
L4	1.003[c]	0.107	1.153	0.259	1.255[a]	0.284	3.43	0.041
R Femur	0.799[c]	0.132	0.897	0.149	0.938[a]	0.131	3.321	0.045
L Femur	0.788[c]	0.133	0.903	0.148	0.925[a]	0.123	3.55	0.037
Total	0.843[c]	0.139	0.964	0.159	1.057[a]	0.119	7.788	0.001

a. ≠ Low weight; b. ≠ Eutrophy; c. ≠ Overweight; SD = standard deviation; p value <0.05; * ANOVA test; Post-hoc Bonferroni

Table 4
Logistic regression models for the association between bone health compartment with anthropometric indicators of nutritional status and bone metabolism markers, 2019

	B	OR adjusted	IC	Standard Error	Z (Wald)	p
*Model 1**						
BMI	-0.297	0.743	0.603 - 0.916	0.107	7.729	0.005
25(OH)-D	-0.099	0.905	0.822 - 0.997	0.049	4.111	0.043
*Model2***						
BMI	-0.299	0.742	0.601 - 0.915	0.107	7.74	0.005
25(OH)-D	-0.102	0.903	0.822 - 0.993	0.048	4.46	0.035

* Model adjusted for sex and age; ** Model adjusted for sex; 25(OH)-D = Vitamin D; CI = Confidence Interval - 95%; OR = OddsRatio; p value <0.05

promoted by adipose tissue. One possible explanation is that obese older adults tend to have a greater accumulation of fat on the hip and abdominal region, than in the upper and lower limbs, therefore, fractures are less likely to occur in these locations (21, 26).

We also investigated the serum concentrations of vitamin D in the participants, in order to identify the relationship between this bone metabolism marker and body composition and bone density. We identified that the group with the highest serum concentration of vitamin D was that of individuals without osteopenia or osteoporosis and the participants diagnosed with excess weight. It is known that vitamin D deficiency decreases intestinal calcium absorption, leads to secondary hyperparathyroidism and accelerates bone loss, thereby increasing the risk of falls and fractures (10, 27, 28), facts that justify the need to investigate this marker. In addition, we compared the PTH concentrations of participants diagnosed with and without osteopenia or osteoporosis and found the opposite behavior to that observed with vitamin D, i.e., participants diagnosed with the disease have higher serum concentrations of this marker. However, there was no significant difference between the groups studied.

The analysis of PTH concentrations in association with the analysis of vitamin D is a sensitive method for assessing the behavior of bone metabolism, due to the regulation exerted by PTH on the metabolism of vitamin D and in controlling the mobilization of calcium reserves (27). In a study that investigated the correlations between serum concentrations of vitamin D, PTH and BMD in Korean individuals aged> 50 years and that reported results similar to ours, serum concentrations of 25(OH)-D demonstrated positive and moderate correlations with the BMD of vertebrae L1 to L2 and the femur, and inverse correlations with PTH (28).

Low body weight or weight loss has been associated as a predictor of low BMD and a higher risk of fracture in older adults. This reinforces the recommendation that reducing body weight is not an appropriate therapy for maintaining BMD in older adults, due to the fact that weight loss contributes to the development of sarcopenia, which associated with low bone mineralization, significantly compromises the functional capacity of these individuals, thereby decreasing the quality of life (3, 8, 21). A study conducted with older adults to

determine the associations of weight loss with bone strength and microarchitecture observed that weight loss in men towards the end of life is associated with lower peripheral bone strength and BMD (29).

Eutrophy and adequate muscle mass are viewed as protectors against osteoporosis, particularly in men, while adipose mass provides greater protection for women. Satisfactory nutritional status is important for adequate BMD, considering that the bone compartments of individuals with adequate body weight do not suffer stress. This is due to the absence of a mechanical load exerted by the body weight, in addition to the use of body reserves, such as the mobilization of the body calcium deposited in bone tissue that promotes resorption, culminating in bone fragility with a significant increase in the risk of fractures (6, 7).

One limitation of this study is the lack of investigation into the practice and frequency of physical exercises by the participants, considering that regular physical activity promotes the necessary mechanical stimulus for the proper maintenance of BMD.

Conclusion

Our findings have reinforced the hypothesis that normal and higher levels of BMI, body composition measurements indicative of greater fat reserves, such as waist circumference, the proportion of body fat and sufficient serum concentrations of vitamin D, promote adequate maintenance of BMD in the lumbar spine and femoral neck, and consequently, are protective factors against the appearance of bone diseases. We emphasize that maintaining the body fat reserve is related to the maintenance of bone mineral density in older adults.

Acknowledgements: This research was financed by the Fundação de Amparo a Pesquisa de Alagoas in partnership with the State Health Secretariat of Alagoas and the Brazilian Ministry of Health with resources for Research for the Integrated Health System and, in part, it was also financed by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Fiance Code 001.

Conflicts of interest: The authors declare that there is no conflict of interest.

Ethical standards: The study was submitted to the Research Ethics Committee at the Universidade Federal de Alagoas and was approved under Protocol Nº 432.659/2016. The patients who were eligible to participate in the study signed an informed consent form before participating in the study.

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