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ORIGINAL ARTICLE

Relationship between bone mineral density and trabecular bone pattern in postmenopausal osteoporotic Brazilian women

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Abstract
Objectives The aim of this study is to evaluate the relative efficacy of the mandibular trabecular bone in detecting osteoporotic-associated bone changes in Brazilian women.
Materials and methods Seventy-three digital panoramic radiographs of healthy and osteoporotic postmenopausal women were used. Regions of interest were selected in the mandibular angle and body of the panoramic images, which were analyzed for fractal dimension (FD) and pixel intensity (PI). Statistical analysis evaluated the distribution pattern, compared normal with osteoporotic patients and the right with the left mandible, and assessed the relationship between FD and PI. The significance level was set at 0.05.
Results FD and PI values were significantly different ($p<0.05$) between normal and osteoporotic patients for the right and left mandibular angle and for the left mandibular body. There was no significant difference ($p>0.05$) between the right and left mandibular angle for FD and PI, but a significant difference ($p<0.05$) was observed on the mandibular body for PI values. FD and PI values presented positive correlation.
Conclusions The mandibular trabecular bone was effective in detecting osteoporotic changes, with the exception of the right mandibular body.
Clinical relevance Mandibular trabecular bone shows a promising attempt of accurate detection of osteoporotic changes in some regions of the jaws. Fractal analysis and pixel intensity had strong positive correlation.

Keywords Osteoporosis · Densitometry · Panoramic radiography · Radiographic image enhancement

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Introduction

Osteoporosis is a skeletal disorder characterized by low bone mass and microarchitectural deterioration of bone tissue. It results from diminished organic bone matrix (mostly collagen protein and mucopolysaccharides) rather than from bone calcium. This disorder leads to enhanced bone fragility, with a consequent increase in fracture risk. It is estimated that 200 million people worldwide have osteoporosis [1]. According to the Brazilian Institute of Geography and Statistics, the total Brazilian population is expected to be at 260 million by 2050, of which about 30 % will be older than 65 years of age [2]. Considering that old age is a risk factor for osteoporosis, an elevation in the number of those

affected with this disease is expected. Caucasian females are at highest risk of developing osteoporosis. A higher quality of life is correlated with a lower risk of osteoporosis. The major complication of the disease is the increased risk of fractures, which may spontaneously occur in routine activities or because of low-intensity trauma [3]. The overall rate of mortality is approximately 20 % in the first 12 months after a hip fracture, and the costs to the public health system are significant [4, 5]. In Brazil, there currently are 28 million toothless people. Although the strongest risk factors of tooth loss are dental caries and periodontal disease [6], this may also be related to bone loss [7].

Dual energy X-ray absorptiometry (DXA) is the most typical and accurate method to measure bone mineral density (BMD) [8]. Starting in 1960, the radiographic image of the mandibular bone has been evaluated for its efficacy in the diagnosis of osteoporosis. Since then, several studies have been done on radiographic findings of osteoporosis in the oral cavity [9–13]. Since its introduction into the general practice of dentistry, panoramic radiography has become a popular and valuable diagnostic tool [9]. Digital radiographs are an increasingly popular option in the clinic. Such images are composed of pixels with a specific numerical value for each one. Two important methods of evaluating digital images are fractal dimension (FD) and pixel intensity (PI) analyses. FD is an objective measurement of the complexity of the bone tissue architecture, describing shapes and structural patterns [14]. PI is a grayscale measure, ranging from 0 (black) to 256 (white) in an 8-bit digital image [10].

Because the panoramic radiography is an exam more common and affordable than DXA, and it shows the entire maxillomandibular region on a single film, its application in the early detection of low bone mass would bring the significant benefit of early treatment to those afflicted with osteoporosis. The aim of this paper was to evaluate the relative efficacy of mandibular trabecular bone in detecting osteoporotic-associated bone changes in panoramic radiographs of postmenopausal Brazilian women.

Materials and methods

The experimental protocol was approved by the Human Research Ethics Committee (protocol # 2007.1.1086.58.6). Seventy-three digital panoramic images at 265 dpi were selected from a clinical digital archive of patients referred for dental treatment. The images were acquired using the Veraviewepocs 2D (Morita, Kyoto, Japan) unit, a digital X-ray system, by the same radiology technician. A chin rest positions the anterior teeth in the focal layer, and a head stabilizer centers the patient in the unit. The direct automatic exposure function was disabled and all exposures were taken at 65 kVp, 6 mA, and 7.4 s. The eligibility criteria for selecting images were females at postmenopausal stage with a bone densitometry report of the lower spine and hips classified as either normal (BMD *T* score >−1.0) or osteoporotic (BMD *T* score <−2.5), in accordance with the World Health Organization classifications. Within the group of selected images, any images of females with systemic diseases that could influence bone mineral density, such as secondary osteoporosis, poorly controlled thyrotoxicosis, primary hyperparathyroidism, malabsorption, liver disease, or alcoholism, were not included in the study. The subjects of each image ranged in age from 45 to 70 years. Exclusion criteria were incomplete bone density measurements, inadequate radiographic material, presence of condensing osteitis, osteosclerosis, or local destructive lesions of the mandible. Standardized regions of interest (ROI) of 230× 130 pixels were selected on the right and left mandibular body and angle. The selection of the ROIs on the mandibular body was below the apex of the canine, right anteriorly to the mental foramen. On the mandibular angle, the ROIs were below the mandibular canal, posteriorly to the molar region, so as to prevent interference of masticatory stress in the study. Since patients have different anatomical features, keeping the same image resolution and ROI size was of great importance (Fig. 1).

FD and PI were measured in all ROIs using ImageJ 1.44o, a public domain software developed by the National Institutes of Health (Bethesda, MD, USA). To calculate FD of the trabecular bone structure, the ROIs were submitted to a sequence of procedures as follow: blurring, subtraction, addition, binarization, erosion, dilatation, inversion, skeletonization, and superimposition (Fig. 2) [15]. As the first step of this sequence, the blurring of the image attenuates high frequency signal (Gaussian Blur). The subtraction of the blurred image from the original ROI and the addition of 128 to each pixel location result in an image in which most variation in brightness is due to the presence of trabeculae versus marrow spaces. The binarization stores each pixel as a single bit (black or white). The erosion followed by dilatation avoids the generation of indeterminate values for structures occurring along the edges

Fig. 1 ROI locations in the mandible

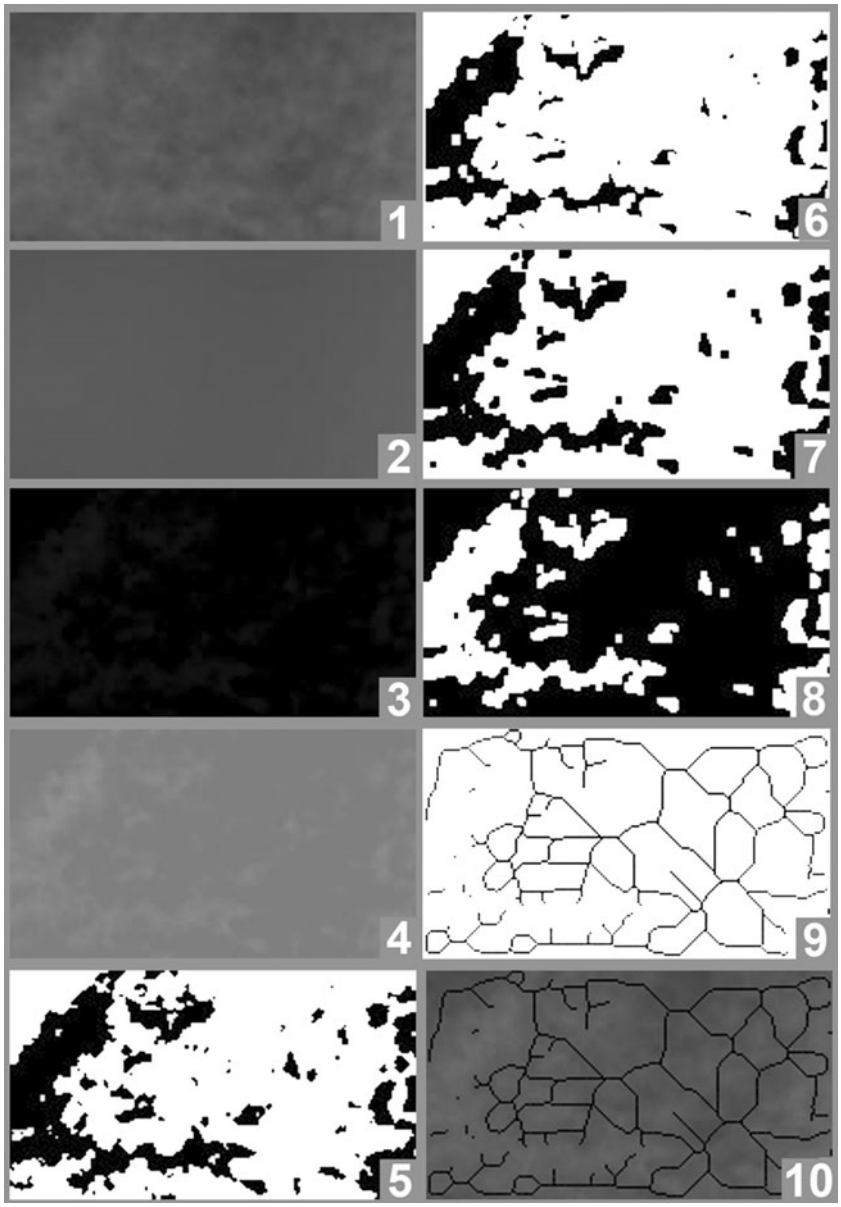

Fig. 2 Resulting images after digital processing: *1* original ROI, *2* blurring, *3* subtraction, *4* addition, *5* binarization, *6* erosion, *7* dilatation, *8* inversion, *9* skeletonization, *10* superimposition

of the image. The inversion makes the trabeculae black, and the skeletonization leaves only the central line of pixels to remain. Lastly, the superimposition of the skeletonized image on the original ROI visually demonstrates that the outline image corresponds to the original trabeculae. FD was calculated for both binary and skeletonized images by the box-counting method (Minkowski–Bouligand dimension). All data were recorded on a Microsoft Excel for Mac 2011 spreadsheet. The Shapiro–Wilk test evaluated the distribution pattern of the data. Based on the type of data distribution, Student's *t* test, Wilcoxon signed-rank, and Mann–Whitney *U* tests compared the normal with the osteoporotic group, and the right with the left mandible. Pearson's correlation was used to determine the relationship between FD and PI values. The significance level was set at 5 % (α=0.05).

Results

Within the 73 selected panoramic images, 38 (52 %) images were of normal patients and 35 (48 %) were of osteoporotic patients. The normal patients presented *T* score of −1.0 or higher at the lower spine and hips. The osteoporotic patients presented at least two sites with *T* score of −2.5 or lower.

FD values were significantly different ($p<0.05$) between normal and osteoporotic groups for the right and left mandibular angle and left mandibular body. On the right side, FD values of the mandibular body did not differ

significantly ($p>0.05$) between normal and osteoporotic groups. Right and left mandibular angle and body did not differ significantly ($p>0.05$) for both patient groups (Table 1).

PI values were significantly different ($p<0.05$) between normal and osteoporotic groups for the right and left mandibular angle and left mandibular body. On the right side, PI values of the mandibular body did not differ significantly ($p>0.05$) between normal and osteoporotic groups. Right and left mandibular angle did not differ significantly ($p>0.05$) from each other. A significant difference between right and left mandibular body was observed ($p<0.05$) in both patient groups (Table 2). FD values were positively and strongly correlated with PI values in all regions and groups (r from 0.84 to 0.92) (Fig. 3).

Discussion

The diagnosis of osteoporosis is made by measuring the BMD. DXA is the most widely used technique for bone measurements, since it is considered to be able to provide an accurate estimation of bone mineral density in adults. There are several bone sites in the body to perform DXA scans. The spine or femur must be one of them in the evaluation of bone density [16]. In the present study, evaluated sites were the lumbar spine and hips.

Assessment of trabecular bone pattern is an important factor in the analysis of mandibular bone quality for surgical planning, implant placement, orthodontic movements, and prosthetic support. The mandible is a single bone, which belongs to the axial skeleton. The effect of masticatory forces on mandibular trabeculae is still inconsistent. Panoramic radiography has been widely used in screening and epidemiological studies because of the convenience of their use. Besides, this technique can offer a dose advantage over large numbers of intraoral radiographs [17].

Table 1 Means (± standard deviation) for fractal dimension values of different mandibular sites based on patient groups

	Normal	Osteoporotic
Mandibular angle[a]		
Right	1.41 (0.08) aA	1.36 (0.10) aB
Left	1.40 (0.07) aA	1.35 (0.11) aB
Mandibular body[b]		
Right	1.40 (0.08) aA	1.38 (0.08) aA
Left	1.42 (0.05) aA	1.37 (0.08) aB

[a] Right and left mandible with different lowercase letters differed by paired t test. Normal and osteoporotic groups with different capital letters differ by unpaired t test ($p<0.05$)

[b] Right and left mandible with different lowercase letters differed by Wilcoxon test. Normal and osteoporotic groups with different capital letters differ by the Mann–Whitney test ($p<0.05$)

Table 2 Means (± standard deviation) for pixel intensity values of different mandibular sites based on patient groups

	Normal	Osteoporotic
Mandibular angle		
Right	7.06 (1.74) aA	6.25 (1.74) aB
Left	6.81 (1.88) aA	5.96 (2.00) aB
Mandibular body		
Right	6.75 (1.28) aA	6.19 (1.69) aA
Left	7.21 (0.95) bA	6.59 (1.43) bB

Right and left mandible with different lowercase letters differed by paired t test. Normal and osteoporotic groups with different capital letters differed by unpaired t test ($p<0.05$)

FD is a value that indicates how completely a fractal appears to fill space. Several authors have reported that structures with high FD have greater structural complexity. Lower FD values are indicative of simpler structures [14]. FD measurements of healthy bone taken from computed tomography images range from 1.70 to 1.80 [18]. In the present study, the mean values of FD for normal and osteoporotic groups taken from digital panoramic radiographs were 1.40 and 1.36, respectively. Such values indicate lower complexity of the mandibular trabecular bone pattern of the osteoporotic group in the sample studied.

Yasar and Akgünlü studied 48 postmenopausal women and found no significant difference in FD between healthy and osteoporotic patients in their study [11]. However, they calculated FD from direct digital periapical radiographs, which present much higher spatial resolution than panoramic radiographs. FD can only be reliably compared when using radiographs at the same spatial resolution [19]. Law et al. made use of fractal analysis to evaluate digitized dental radiographs for signs of osteoporosis [20]. The osteoporotic group presented higher FD than the normal group, which is in disagreement with the results found in our study. These divergent findings may be related to the age range of the sample and the acquisition mode. Law et al. used women from 22 to 85 years old as control group [20]. Our study included only postmenopausal women with ages ranging from 45 to 70. The age range affects the standardization of FD values, since hormonal changes are dissimilar between young and elderly patients. Besides, the present study used direct digital panoramic images at 256 dpi. There is a scarcity of literature concerning fractal analysis with regard to the reliability of digitized images.

In 2003, a group of researchers from five European centers started a project known as OSTEODENT. The aim of this project was to identify the most valid and effective radiographic index, or combination of radiographic and clinical indices, for the diagnosis of osteoporosis in female

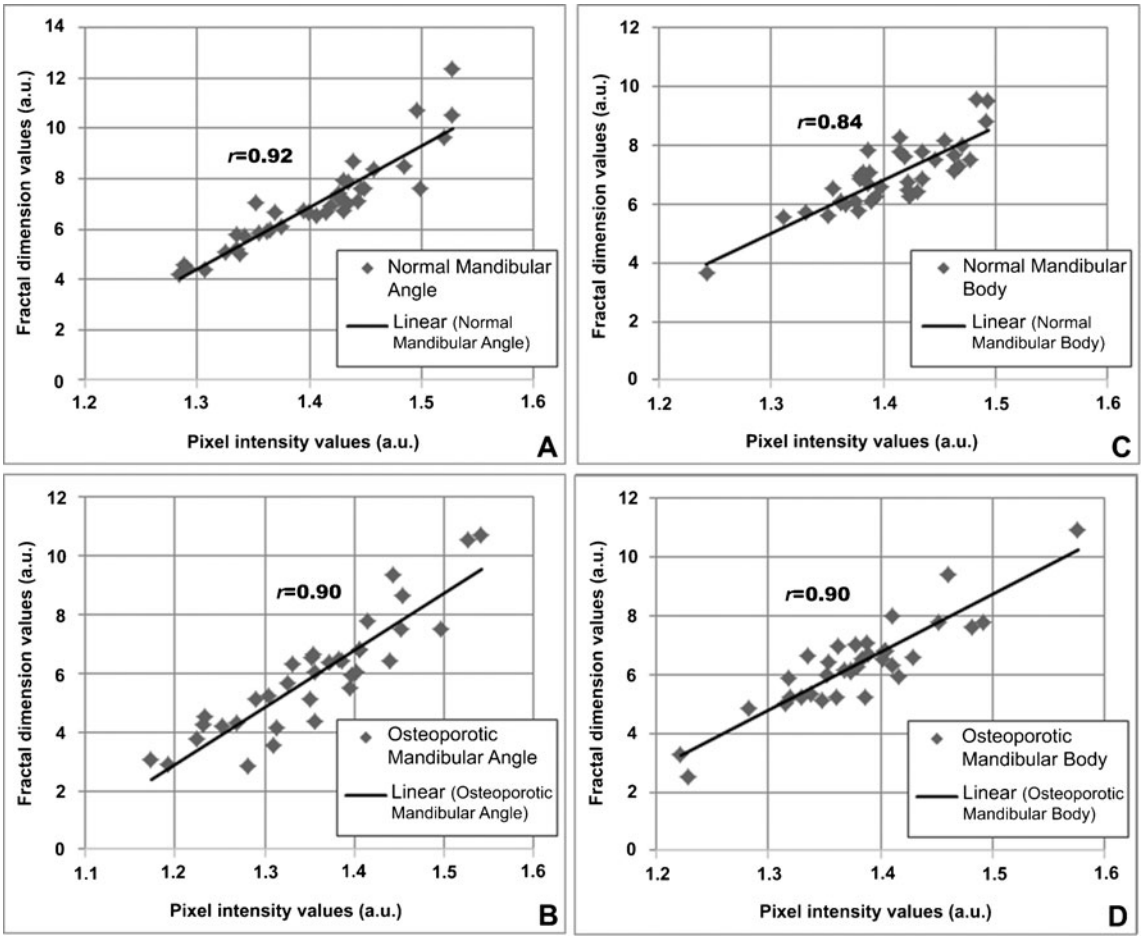

Fig. 3 Relationship between FD and PI values in normal (**a**) and osteoporotic (**b**) mandibular angle, and normal (**c**) and osteoporotic (**d**) mandibular body

patients in the age range 45–70 years who have undergone radiography for usual dental reasons. Over the years, they have shown that the Osteoporosis Index of Risk (OSIRIS)—age, weight, current estrogen therapy, and history of low trauma fracture—has higher diagnostic validity than the measurement of the mandibular cortical width (MCW) on panoramic radiographs [21]. However, patients with MCW thinner than 3 mm should be referred for osteoporosis investigation [22]. More recently, in 2010, the OSTEODENT index, which is the result of the OSIRIS, was showed to have value in prediction of hip fracture risk [23]. Even before the OSTEODENT project, the MCW showed to be directly associated with skeletal bone mass [24], including young adult men [25]. Nevertheless, Ferreira Leite et al. demonstrated that there was no apparent association between vertebral fragility fractures and the appearance of mandibular cortex in elderly men [26]. In a study, which correlated FD from periapical radiographs and panoramic radiographs with MCW and morphology, Bollen et al. stated that FD values increase as BMD decreases [27]. However, in the image processing for fractal analysis, the images were not skeletonized, making the analysis more susceptible to interferences. Image skeletonization emphasizes the geometrical and topological properties of shape by reducing foreground regions of a binary image to a skeletal remnant. Additionally, Van der Stelt and Geraets have stated that any step of the digital image processing can influence the final radiographic image [28].

Feltrin et al. studied the lumbar vertebral cancellous bone architecture and stated that FD of trabecular bone should be considered as a supplement to BMD evaluation in the assessment of osteoporosis [29]. Tosoni et al. found no significant correlation between FD values of three groups (normal, osteopenic, and osteoporotic) of postmenopausal women, differing from our results [10]. This may possibly be due to the size and location of ROIs and the inclusion criteria. Tosoni et al. selected three ROIs of different sizes on the right side of the mandible in panoramic radiographs of healthy, osteopenic, and osteoporotic women [10]. In this study, two groups were evaluated: normal and osteoporotic. The non-inclusion of osteopenic patients aimed to eliminate subtle differences between the studied groups, focusing in

the osteoporosis detection. All ROIs had 230×130 pixels and were located in the right and left mandibular angle and body, away from masticatory stress.

In digital imaging, a pixel is a single point. It is the smallest unit of picture that can be represented or controlled. The intensity of each pixel is variable. In a radiographic image (grayscale), the value of each pixel carries only intensity information, varying from black at the weakest intensity to white at the strongest [30]. Hedstrom et al. found a positive correlation between PI in digital periapical radiographs of the mandible and the BMD of the heel [31]. Unlike Law et al. [20], who analyzed scan lines to determine PI, we analyzed scan areas. However, both studies showed that the mean PI value for the osteoporotic group was lower than for the normal group. Moreover, in Southard and Southard's study, a comparison of digitized radiographic alveolar features between 20- and 70-year-old women allowed the affirmation that a measurable reduction in the magnitude of the PI occurs with age [32]. Tosoni et al. evaluated the relative efficacy of PI analysis in detecting osteoporotic-associated bone density changes [10]. As in our study, the normal group had significantly higher PI than the osteoporotic group. Such results could be expected because of the relatively high penetrability of the X-ray photons into osteoporotic bone. This produces a more radiolucent image with consequent lower PI. It could also be observed that the PI in the right and left mandibular body were significantly different both for normal and osteoporotic groups, which may suggest anatomical variations or sensitivity of this analysis in detecting unilateral chewing. Possible positioning errors should be considered, since the panoramic radiography technique presents a relatively narrow image layer in the anterior region [33]. Although this study had an unbiased random selection of individuals, the authors believe that the sample size might not have been enough to avoid sampling process error. The use of a larger sample could have eliminated such difference.

Examining structural parameters in the assessment of bone quality, Matsubara et al. suggested that PI is a useful tool in the evaluation of bone quality in radiographic images [34]. Their methodology made use of bone specimens, and therefore, there was no soft tissue interference. The presence of soft tissue increases the X-ray attenuation, leading to a decrease of the signal received by the image receptor, and consequent higher PI values. However, similarly to Matsubara et al. [34], our results were not expected to be influenced by soft tissues, since all of the images were made binary and skeletonized as previously described [15]. Moreover, race is an important determinant of BMD, which for instance is consistently higher in Black women [35]. The present study assessed the Brazilian population, which is well known for its extensive miscegenation, as a result of five centuries of interethnic crosses among Europeans, Africans, and Amerindians. Miscegenation reduces the average genetic distance between different population clusters. This results in biological variation, with polygenic human phenotypes, and makes the study of individual phenotypic differences (genetic traits) even more difficult [36]. Limitation in the selection of the ROIs on panoramic radiographs should be considered due to anatomic variation among patients. Although complex and not widespread, both the FD and PI are methods of evaluating digital images available for the dental practitioner, since some of the imaging software applications are of free download. "Dentists have a role to play in the detection and referral of patients at high risk of osteoporosis" [22].

In conclusion, according to the results found in this research and within the limitations of the methodology used, the mandibular trabecular bone was effective in detecting osteoporotic changes in postmenopausal Brazilian women, with the exception of the right side of the mandibular body. The methods of fractal analysis and pixel intensity had similar ability in the analysis of the mandibular trabecular bone with strong positive correlation. Further studies about fractal analysis are necessary to better characterize osteoporotic-associated factors, since it is a contradictory issue.

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Conflict of interest The authors declare that they have no conflict of interest.

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