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Novel algorithm detecting trabecular termini in μCT and MRI images

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Abstract

In this paper, a novel algorithm detecting trabecular termini in three-dimensional images of trabecular bone is introduced. The algorithm is applied to the analysis of μCT and MRI images of distal radius trabecular bone samples. In μCT images, the volume of the trabecular termini constitutes at most 2.1% of the bone volume fraction BV/TV and is typically smaller than 1% of BV/TV. Isolated trabeculae are not observed in the interior of the trabecular bone samples. Trabecular bone structure assessed with μCT appears thus highly optimized. The volume and the number of the trabecular termini do not correlate with BV/TV. These quantities do not correlate also with apparent Young's modulus of the samples. In contrast in MRI images, segmented with the dual reference limit method, the volume of the trabecular termini and the volume of isolated parts constitute even up to 14% of the apparent bone volume fraction App.BV/TV. For MRI images, the volume of the trabecular termini increases significantly with decreasing App.BV/TV. The volume and the number of the trabecular termini, derived from MRI images do not correlate with Young's modulus. There is also no correlation between the number and the volume of the trabecular termini, estimated from MRI and μCT images. The volume of the trabecular termini is overestimated 15 times in MRI images. App.BV/TV correlates strongly with BV/TV. Young's modulus derived from MRI images correlates strongly with Young's modulus found for μCT data. It is shown that the diagnostic significance of latter result is highly limited.

Keywords: Trabecular termini; Finite elements; MRI; Microtomography

Introduction

The problem of optimizing the process of diagnosing osteoporosis remains a challenge for trabecular bone research. The quantity of primary interest for diagnosis is the risk of fracture, which could not be however straightforwardly assessed because of structural complexity of trabecular bone and limitations of imaging methods. Multiple procedures have been proposed to quantify the risk of fracture, among which measurement of BMD is a standard clinical approach. Diagnosis based on BMD only is however incomplete. It is well established that there are subjects suffering osteoporotic fractures in spite of apparently adequate BMD value, while others, with inadequate BMD do not sustain fractures. Thus, it has been claimed that BMD measurement should be supplemented with architectural information to improve diagnosis.

Histomorphometric studies have shown that trabecular number and trabecular thickness decrease in osteoporosis, what in turn leads to increased trabecular separation [8]. Apart from these changes, there is another factor, contributing to the stiffness of trabecular network and affected by osteoporosis, namely trabecular connectivity, assessed with number of methods, like node-strut analysis [9], Euler number [3,7] or star volume of the marrow space [15].

Promising approach to the node-strut analysis has recently been demonstrated in a paper of Aaron et al. [1], who have shown that the number of trabecular termini is significantly higher in fracture subjects than in matched for bone density nonfracture ones. In that study, thick (300 μm) slices of trabecular bone were stained on both sides and then real (i.e. unstained) trabecular termini were manually

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counted. In other studies, node-strut analysis has been applied to thin (typically of the order of 10 μm) sections obtained in vitro (e.g. Ref. [2,4]) or to low-resolution in vivo 2D images (e.g. Ref. [2]), however, without distinction between real and artificial trabecular termini (which distinction is in these circumstances often impossible). The node-strut analysis of 3D in vitro (μCT) and in vivo (MRI) images has also been reported [10]. Except of the study of Aaron et al. [1], counting trabecular termini is not straightforward and is based on conversion of the original gray-level images to binary ones, followed by skeletonization. The final result (i.e. the number of trabecular termini) is in fact quite sensitive to the parameters of skeletonization algorithm [10]. On the other hand, manual counting is difficult to apply if the thickness of the analyzed samples is too large.

Motivated by difficulties encountered when other algorithms are used, a new algorithm for counting trabecular termini is developed. In the algorithm, the skeletonization step is omitted. The algorithm is applied to the analysis of μCT and MRI 3D images of trabecular bone. The number of trabecular termini, found for μCT data is in excellent agreement with the previous reports.

Finally, the results of μFE analysis of μCT and MRI data are presented. Because the number of trabecular termini was found to be higher for fracture subjects, than for nonfracture ones, one may expect correlation between this parameter and trabecular bone apparent elastic constants. This problem, to the best of the author's knowledge has not been addressed in any previous study.

Materials and methods

Materials

3D data

Fifteen radius bone cubes (side length 12 mm) from the distal radius were obtained from 15 individuals (8 men, 7 women) aged 49 to 92 years. The samples were harvested about 10 mm from the endplate of the radius and scanned with a μCT-20 scanner (Scanco Medical, Zurich, Switzerland) with a voxel size of 34 μm in all three spatial dimensions.

For magnetic resonance experiments, the samples were defatted and immersed in 0.3 vol.% gadolinium-DTPA-doped water solution. Prior to MR image acquisition, samples were fixed in a plastic box of the size of human wrist. The box was filled with doped water. Samples were oriented with the main direction of trabeculae (cranio-caudial direction) aligned with the static magnetic field to reproduce conditions of in vivo wrist measurement.

The images were acquired with a receive-only wrist coil (Medical Advances, Milwaukee, WI) on a GE SIGNA 1.5 T echo-speed system (General Electric Medical Systems, Waukesha, WI). High-resolution MR images were obtained with slice thickness of 300 μm. The reconstructed spatial resolution was equal to $156 \times 156\ \mu m^2$.

The same material and data were used in the previous studies of Laib et al. [5] and Pothuaud et al. [10], in which further details, concerning the measurement protocols and material preparation are described. In these studies, structural and topological parameters of trabecular bone were measured.

MRI images were segmented into bone and marrow phase, using the method of dual reference limit [6]. This thresholding method is based on assuming a two phase model for the segmented structure. Then mean signal intensity I_A of the whole field of view is measured together with mean intensities of dark I_B (bone) and light I_M (marrow) phases. Apparent bone volume fraction App.BV/TV is derived from the equation:

$$I_A = \text{App.BV/TV} \cdot I_B + (1 - \text{App.BV/TV}) \cdot I_M$$

2D data

Sixteen lumbar vertebral body L_3 specimens were taken from 8 young (mean age of 27 years, range 25 ÷ 33 years) and 8 elder men (mean age 75 years, range 70 ÷ 77 years) without metabolic bone disease and no known vertebral fractures. The vertebral bodies were sectioned perpendicularly to the vertical axis of the vertebra to a thickness of approximately 3 mm, cleaned with a water jet, dehydrated, placed in acetone and next embedded in methylmethacrylate. Embedded specimens were ground to a uniform thickness of 200 μm and then rinsed in chloroform to remove any remaining debris.

For each section obtained from a middle part of the vertebral body, images were acquired using a low-magnification digital camera. The images were composed of 512×512 pixels (pixel size 30 μm × 30 μm). In all cases, full 256 grey values were retained. The interfaces between trabecular bone and bone marrow were identified on the basis of brightness histograms.

Methods

The backbone-finding algorithm

The backbone-finding algorithm was applied to binary images of trabecular structures. The analyzed regions of interest (ROIs) were of cuboid shape. The backbone is defined as this part of the structure, which determines its mechanical properties [14]. According to this definition, trabecular termini (as well as isolated trabeculae) do not belong to the trabecular backbone, because the loads applied to their network-connected ends could not be transferred to the rest of the network. The algorithm consists of the following parts:

Step A—Generation of self-avoiding paths, connecting the seed point with the opposite face of ROI. Selecting a seed point defined as any point (pixel or voxel) which belongs