The estimation of structural anisotropy of trabecular and cortical bone tissues based on ultrasonic velocity and attenuation

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Velocity of ultrasound and broad-band ultrasound attenuation (BUA) were measured in three perpendicular directions in anisotropic samples of cancellous and cortical bones. The aim was to study the influence of microarchitecture of bone tissue on ultrasound propagation in bone.

The pulse transmission method (1 MHz and 3.5 MHz) and the Fourier analysis of transmitted pulses were applied. The dependence of both ultrasonic parameters on the orientation and the correlation between the degree of the anisotropy and bone density were stated. In cancellous bone samples, both velocity and BUA had extreme value in the direction of trabeculae alingment. In cortical bone, maximal velocity was in the axial direction, while BUA in this direction was minimal. The influence of the anisotropy of bone structure, both cancellous and cortical, on ultrasonic parameters is discussed.

Key words: structural anisotropy, trabecular and cortical bones, ultrasonic velocity, attenuation

1. Introduction

Quantitative ultrasound techniques have been introduced into the clinical practice at the beginning of the 90-ties as alternative methods for the assessment of skeletal status. The method consist in the measurements of velocity and attenuation of ultrasonic impulses passing through heel bone [1], [2]. The basis of the method is the correlation between ultrasonic parameters and bone density though an influence of bone structure is always stressed [3]–[7]. Information on the relationship between ultrasonic parameters and bone microarchitecture may significantly improve the ability of ultrasound to assess fracture risk. However, it is not fully understood which parameters of bone structure are sensitive to ultrasounds and to what extent.

There were numerous attempts to analyze the relationship between ultrasonic parameters and the microstructure of bone as well as the influence of anisotropy of

bone architecture on ultrasonic parameters [5], [7], [8]–[20]. Results demonstrated a close dependence of ultrasound velocity on the direction of trabeculae alignment in cancellous bone [9], [15], [16] and on orientation of ultrasonic beam with respect to main anatomical axes in cortical tissue [13], [14]. Attenuation of ultrasound in bone is also dependent on the structure of bone, especially in highly anisotropic cancellous bone samples [5], [11], [15], [17], [18], [20] though the dependence of attenuation on the direction of propagation in cortical tissue has been also reported [14]. However, the influence of orientation of trabeculae in cancellous bone and bone lammelae in cortical bone on ultrasound propagation remains still an unresolved question.

The aim of this study was to measure ultrasound velocity and attenuation in three perpendicular directions in anisotropic samples of cancellous and cortical bone tissues in order to give an insight into the influence of the bone microarchitecture on ultrasound propagation in bone.

2. Material and methods

Thirty four rectangular samples of cancellous bone from bovine and human vertebrae and femora and seven samples of cortical bone from bovine femoral shaft were used in the study. The size of samples was 8–23 mm for cancellous bone and 9.5–18 mm for cortical bone. The edges were aligned along main anatomical axes. In cancellous samples, the basic alignment of trabeculae was established under microscope and the edge consistent with this direction was denoted as the *x*-axis. The second edge with minimal number of trabeculae was accepted as the *z*-axis and the third edge of the rectangular sample – as the *y*-axis (figure 1). In cortical samples

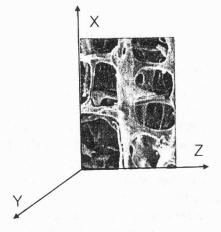


Fig. 1. Orientation of axes in bone samples; the central part of an osteoporotic human vertebra is used as the example

from bovine femoral shaft, the x-axis was in longitudinal, the y-axis in tangential, and the z-axis in radial directions.

The apparent density of each sample was determined from the wet weight divided by the volume as determined by the overall physical dimensions of the specimen.

The ultrasonic measurements were performed in a thru-transmission immersion setup following the method applied in diagnostic devices [1]. Two ultrasonic nonfocused transducers (transmitter and receiver) were mounted coaxially in a tank filled with the degassed water, at a room temperature (figure 2a). Transducers with the central frequency of 0.95 MHz and the diameter of 20 mm were used for porous samples of cancellous bone, while for cortical samples, transducers with the central frequency of 3.5 MHz and the diameter of 14 mm were mounted. The sample was placed between the transducers in such a way that one axis was positioned along the axis of an ultrasonic beam. The distance between transducers was 140 mm. Transducers were connected to a computer-controlled pulse generator and an analogue-digital circuit digitizing the signal at a frequency of 14 MHz. First 20 µs of the received signal were analyzed.

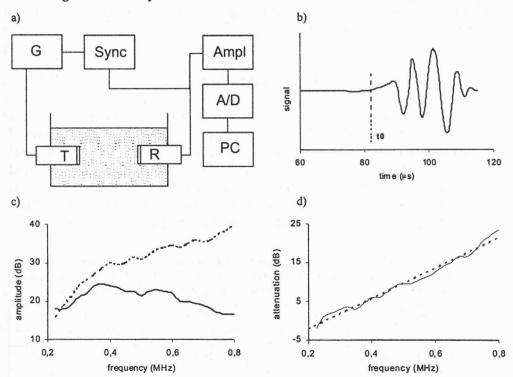


Fig. 2. (a) Experimental setup: T, R – transmitter, receiver; G – pulse generator; Ampl – amplifier; Sync – synchronization; A/D – digitizer; PC – computer; (b) ultrasonic pulse, t_0 – time of the first zero-crossing; (c) amplitude spectra of the signal transmitted through the bone (solid line) and of the reference signal (dashed line); (d) the difference between both spectra, the slope of the fitted line is BUA

Two transmitted signals were compared: one obtained with the bone immersed between transducers and a reference signal obtained without the bone. Velocity of ultrasound was calculated using the transit time of the signal obtained by determining the first zero-crossing for the waveform of pulse (figure 2b). In order to obtain the attenuation, the frequency spectra of the signals were calculated by using a standard Fourier transform algorithm (figure 2c). The slope of the difference spectrum (figure 2d) normalized to unit length of a sample provides the broad-band ultrasound attenuation (BUA). The spectra were calculated over the frequency range from 0.2 to 0.7 MHz for cancellous bone, and from 3.5 to 4.5 MHz for cortical bone. In some bone samples, dimensions in the direction perpendicular to ultrasonic beam were less than the diameter of a transducer, in such a case BUA measurements were performed only in one or two directions. The series of six measurements was made for each sample with repositioning after each measurement. Coefficient of variation in the series, defined as CV = standard deviation /mean value, was calculated in order to estimate the precision of the ultrasonic measurements in a sample.

Coefficients of correlation between bone density and ultrasonic parameters as well as significance of differences between groups of samples were calculated using standard procedures in Statistica for Windows.

3. Results and discussion

Results of measurements of ultrasonic parameters in bone samples are given in the table. Samples were divided into three groups: I – cancellous bone samples of the density $< 1.18 \text{ g/cm}^3$, II – cancellous bone samples of the density $\ge 1.18 \text{ g/cm}^3$ and III – cortical bone samples of the density $> 1.9 \text{ g/cm}^3$. Velocity of ultrasound and broadband ultrasound attenuation depend on the orientation and type of a tissue. In each group, maximal value of ultrasound velocity was found in the x-direction, i.e. in the direction of a general alignment of compressive trabeculae in can-

Table. Velocity of ultrasound (V) and broad-band ultrasound attenuation (BUA) in low-density trabecular bone samples (I), high-density trabecular bone samples (II), and cortical bone samples (III) in three perpendicular directions; mean values and standard deviations are given

No. of group	Density (g/cm ³)	V_x (m/s)	V_y (m/s)	V_z (m/s)	BUA_x (dB/cmMHz)	BUA_y (dB/cmMHz)	BUA_z (dB/cmMHz)
I	1.128	2451	1864	1794	30.3	25.0	24.1
	0.036	103	143	143	4.6	5.3	6.8
II	1.227	2562	2307	2109	33.0	32.2	32.7
1	0.038	202	240	120	3.9	3.5	3.7
III	2.019	4265	3643	3298	6.2	11.3	12.1
	0.024	116	82	45	0.7	3.3	2.6

cellous bone samples and in the axial direction in cortical samples. BUA in cancellous bone depends on the direction only in low-density group and is the highest in the *x*-direction. In cortical bone, BUA is maximal in radial direction.

The correlation between ultrasonic parameters and bone apparent density is shown in figures 3 and 4. Each point is a mean value from six measurements. The coefficient of variation in a single bone sample did not exceed 0.7% for the velocity and 4.3% for BUA measurements, and was less than variation between samples in the group (table).

The points for the x- and z-directions are given; points for the y-direction are omitted for transparency inasmuch as the differences between ultrasonic parameters for the y- and z-directions are insignificant, except for the velocity in cortical samples (table). There is a high correlation between the apparent density of cancellous bone and the velocity of ultrasound in the z-direction perpendicular to the alignment of main compressive trabeculae (R = 0.968). In the x-direction, parallel to these trabeculae, the correlation is lower (R = 0.720) and the velocity changes slower with density than in the z-direction. This is probably due to a change of trabecular architecture of cancellous tissue during bone loss. The loss of tissue is not isotropic, trabeculae perpendicular to the direction of main compressive trabeculae disappear first [21], [22].

The attenuation of ultrasound in cancellous bone depends on the orientation only in low-density samples. In dense cancellous bone samples, the dispersion of BUA values in the x-direction is so large that it is difficult to find any regularity. A similar result is reported by Rho et al. [17]. They found that in dense cancellous bone,

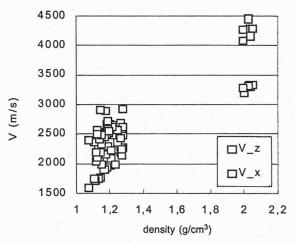


Fig. 3. Velocity of ultrasound (V) versus apparent density of cancellous (density $< 1.3 \text{ g/cm}^3$) and cortical (density $> 1.9 \text{ g/cm}^3$) bone samples in two perpendicular directions. In cancellous samples: x – the direction of basic alignment of trabeculae, z – the direction of the minimal number of trabeculae. In cortical samples: x – the axial direction, z – the radial direction

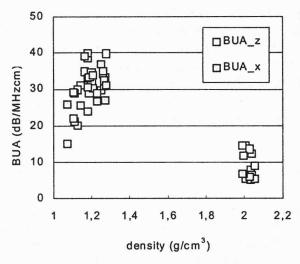


Fig. 4. Broad-band ultrasound attenuation (BUA) versus apparent density of cancellous (density < 1.3 g/cm³) and cortical (density >1.9 g/cm³) bone samples in two perpendicular directions. Denotations as in figure 3

anisotropy of ultrasound attenuation was slight and correlation between BUA and density was insignificant. In another study on samples from vertebra and epiphyses of long bones, BUA in the direction of compressive trabeculae was significantly higher than in the perpendicular direction [11], [15]. However, arrangement of trabeculae in those samples was extremely regular; moreover, an apparent density was rather low.

A relationship between the density of bone and ultrasonic parameters was usually reported as linear [4], [10], [17], [18]. However, a non-linear relationship between BUA and the density of human and bovine cancellous bone samples was also established [6], [12]. Serpe and Rho [19] showed a linear regression between velocity and density of bone over the entire range of densities from low-density cancellous bone to dense cortical bone. For BUA and apparent density in the same samples they reported the second-order polynomial regression model. However, their measurements on low-density cancellous samples showed linear correlation with BUA. Similarly, in the present study, the maximal and direction-independent values of BUA were found in dense cancellous samples. BUA in cancellous bone is, to a large extent, related to scattering on bone trabeculae. Orientation of the trabeculae is one of the parameters determining the structure of the tissue. The other parameters, i.e. size and shape of intertrabecular spaces, thickness and connectivity of trabeculae, influence the scattering and may cause the dispersion of BUA values in dense tissue overshadowing an influence of orientation. Qualitative and quantitative correlation between the parameters mentioned-above and the propagation of ultrasound is still an unanswerable question.

In each cortical sample, BUA is significantly lower than in cancellous bone (figure 2). In contrast to cancellous samples, where maximal BUA and maximal velocity are in the same direction in nearly all samples, in cortical samples the directions of maximal values of BUA and velocity are mutually perpendicular. This difference is due to the fact that cortical and cancellous bones are two different types of material. Cancellous bone is a highly porous structure with more or less regular arrangement of pores of diameter in a range of 10^{-1} mm, or even larger. Cortical bone is a dense tissue formed from lamellae and little pores of the size of about 10^{-2} – 10^{-3} mm arranged in structures aligned parallel to a long axis of bone [14]. In the cortical tissue, attenuation may be almost entirely due to absorption, since the structure is relatively homogeneous and continuous for the waves used in this study (wavelength in the range of 1 mm). However, higher values of BUA in the radial direction in comparison with those in the axial direction may be due to scattering and reflection of ultrasound on the borders of radially aligned layers of bone lammelae.

From the study performed we conclude that ultrasonic parameters in bone are influenced significantly by the spatial arrangement of bone structure both of cancellous and of cortical tissues. The results of this research prove that ultrasonic techniques have considerable potential for noninvasive assessment of bone structure complementing existing densitometry techniques.

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