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# Parametric Imaging of a Trabecular Bone by a Scanning Acoustic Microscope

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#### **Abstract**

A nondestructive method is proposed for bone quality assessment at a resolution of about 200  $\mu m$ . The properties of the bone matrix itself, independent of the mass or porosity of the bone, can be determined using a Scanning Acoustic Microscope (SAM). The high resolution of the SAM makes it possible to image a single trabecula of the biopsed iliac bone sample.

Two microscopic acoustic images of a trabecula are recorded. The first is created by the waves reflected from the surface of the sample. The other is formed by the reflections from the bottom. These images are processed further and two parametric images are calculated. They reflect the distribution of impedance and attenuation in the bone. The method is applied to examine trabeculae in bone samples biopsed from patients with metabolic bone diseases.

#### Introduction

Macroscopic, X-ray methods for bone quality assessment are mainly based on porous bone density measurements. The bone quality is a property that is difficult to define, as it is related to both density and structure of the bone. In recent years, several new ultrasonic diagnostic methods have been developed to examine bones "in vivo" which are based on measurements of the velocity and attenuation of waves penetrating

porous bones. The large interest in these methods is a result of the fact that they provide information not only about the bone density but also about their structure without using ionizing energy [1].

The principal element which determines the bone strength is the trabecular structure of a porous bone. Ultrasonic microscopy makes it possible to assess "in vitro" the quality of bones and to examine their structure [2,3]. At higher frequencies, the acoustic microscope ensures sufficient resolution

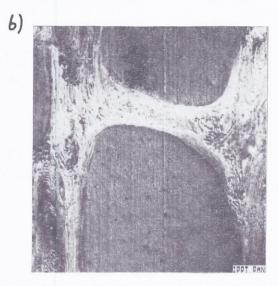
for imaging the internal structure of a single trabecula (Fig. 1). At lower resolutions, the structure of the distribution of trabeculae in a porous bone can be imaged (Fig. 1).

The unique properties of the acoustic microscope make it possible to measure and image such acoustic properties of a

a)

Fig.1 SAM images of a trabecular bone

single trabeculae as the acoustic impedance and attenuation of longitudinal waves in selected areas of a porous bone, in the case, too, of samples from patients who suffer from metabolic bone diseases, such as osteoporosis, osteomalacia and osteoidosis.



a) at 35 MHz - crossection of a bone b) at 200 MHz - a single trabeculea

#### Materials and methods

Bone samples were prepared by Dr. A. Sawicki's team at the Food and Institute. They Nutrition were preparations of a porous pelvic bone presubmerged in methyl methacrylate. Flat, 0.5 mm thick parallelepiped samples were cut off using a wire saw. In order to obtain as smooth a surface as possible for microscopic examination, its surface layer was removed from it beforehand using a microtome. The samples prepared in this way were placed in special clamps. They ensured convenient access of the head and the coupling medium (distilled water) to the surface under examination while keeping the air layer on the other side. A sapphire sample with the impedance z = 44 MRayl

was used as the impedance standard in defining the reference reflection. The size of the reflection (reference signal) was obtained as the mean amplitude of the points of the sapphire surface image (200x200 of such points) while keeping intact all the orientations of the microscope transmitter and receiver, such as applied when bone samples are imaged.

Scanning Acoustic Microscope (SAM), built at the author's laboratory, was used to image and measure. The microscope works at frequencies of 35, 100 and 200 MHz, making it possible to obtain ultrasonic surface and subsurface images. The images are memorized by computer, greatly facilitating their further processing.

Over the incidence angle range up to about 40 degrees, the reflection

coefficient of a longitudinal wave on the water-bone boundary may be taken as constant. A 100 MHz head with a lens with a 20-degree half V-angle was used in the imaging. The application of a lens with a small V-angle allows for the normal wave incidence on the sample surface to be assumed in the model of wave and bone interaction. In subsurface imaging, waves which are weakly focused by the lens do not generate surface waves that would distort measurements, as is the case when lenses with large V-angles are used.

### Imaging of impedance distributions

The brightness of images of flat samples obtained using the scanning acoustic microscope working in the reflection mode, with the microscope focused on the surface being imaged, mainly depends on the reflection coefficient on the water-sample boundary.

The dependence between the bone impedance Zb and the water impedance Zw and that of the reference medium Zr can be represented as:

where Ab(x,y) is the amplitude at the point (x,y) of the bone image and Ar is the amplitude of the reference signal - a reflected wave focused on the surface of the sapphire sample (Zr).

The microscopic trabecula images obtained were processed according to the formula given above and normalized to the maximum impedance, providing impedance distribution images. Then, impedance histograms for particular impedance images were calculated for samples of bones with lesions. The results are shown in Fig.2.

# Imaging of attenuation and impedance distributions for bone samples

The scanning acoustic microscope permits subsurface imaging. A highfrequency ultrasonic pulse incident on the sample being imaged is partly reflected on the water-sample boundary and received by the microscope transducer. Part of the pulse penetrates the sample; it is reflected on the sample-substrate boundary and also detected by the microscope. The pulse transmitted into the sample is partly attenuated as it penetrates the sample. Given the length of the acoustic pulses applied, it is possible to separate the wave reflections from the sample surface from those from the sample bottom in the case of bones which are 0.5 mm thick. The same bone area can be imaged in two ways: using the wave reflected directly from the sample and using the wave which propagates through the sample twice. In the first case, the image can be interpreted as the acoustic impedance distribution of the bone. In the other, changes in the amplitude of the wave propagating through the sample are related to wave attenuation in the bone.

In the attenuation image, it is necessary to account for changes in the amplitude of the wave penetrating the sample, caused by local changes in the sample impedance, and the resulting changes in the reflection coefficient. It was assumed for calculations of images of the reflection coefficient distribution that the coefficient of reflection from the sample bottom is constant and equal to unity. This results from the fact that the sample bottom is loaded only with air the acoustic impedance of which is so small that it can be taken as zero.

Using the method for finding the sample impedance as described in the previous section, the distribution Zk(x,y)

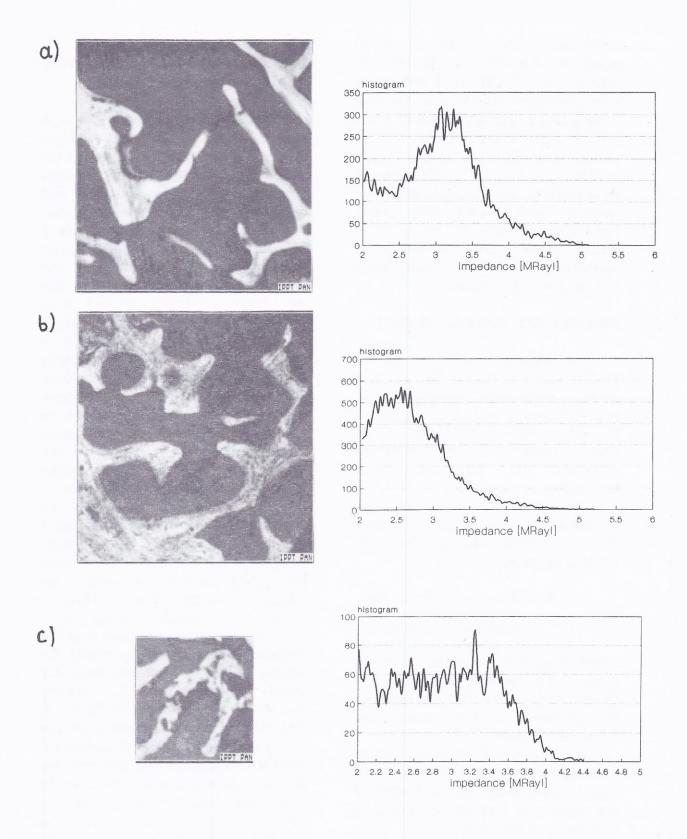


Fig. 2 SAM images and histograms of an impedance distribution for trabecular bone

- a) osteoporosis
- b) osteomalacia
- c) osteoporosis with osteoidosis

determined for the trabecular bone under study.

The amplitude of the signal which provides the image of attenuation in the bone can be represented in the following way:

$$A_{t}(x,y) = A_{0} \times T_{bw} \times T_{wb} \times \exp(-2d\alpha)$$

where Tbw and Twb are the transmission coefficients on the bone-water and water-bone boundaries,  $d\alpha$  is the product of the attenuation coefficient and the sample thickness, and Ao is the reference signal amplitude.

This dependence can be transformed so as to make it possible to calculate the

wave attenuation coefficient (2dα).

On the basis of this train of thought, the bone images obtained for waves focused on the surface and the sample bottom were processed into those of the impedance and attenuation distributions. The images shown in Fig.3 are those of the cross-section of the compact pelvic bone as obtained for waves reflected from the surface, for waves reflected from the sample bottom and processed into sample impedance and attenuation distributions. At the same time, histograms of impedance and attenuation images were found.

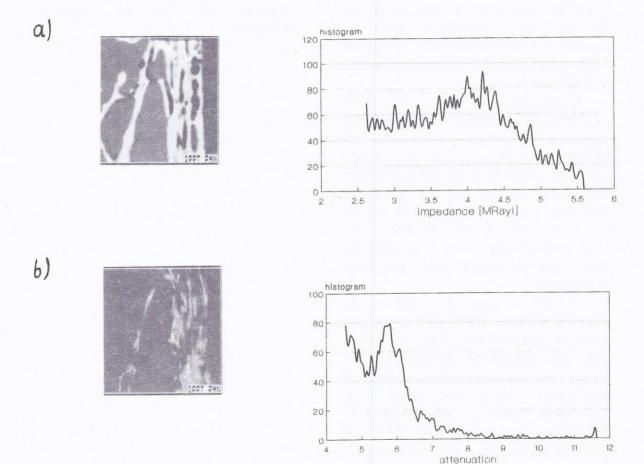


Fig.3 SAM images of a cortical bone a) an impedance image and a histogram of an image b) an attenuation image and a histogram

#### **Initial conclusions**

impedance distributions The (histograms) measured for trabecular bones in the case of samples with different lesions indicate that not only the value of the impedance but also the character of the histograms may be significant factors which distinguish a given disease. The histograms of the trabeculae of bones with osteoporotic lesions are characterized by a sharp maximum evidencing the relatively homogeneous impedance throughout the volume of the trabecula. The position of the histogram maxima changes greatly (2.4 versus 3.1 MRayl); it is probably related to the degree of osteoporosis. In the case of osteomalacia, too, the histogram is characterized by a distinct peak, but it occurs for a lower impedance (2.5 MRayl).

In the case of osteoporosis with osteoidosis, the histograms are almost flat over a wide impedance range (2.6 - 3.5 MRayl), suggesting large impedance variation within a single trabecula. The cortical bone is characterized by a slight histogram peak but for a much higher impedance (about 4.2 MRayl). The impedance images make it possible to select areas with maximum and minimum impedances.

A distinct peak for  $2d\alpha \cong 6$ , corresponding to attenuation of about 52dB/mm can be found. When compared with the impedance distributions the images of the attenuation coefficient distribution suggest that there is a dependence between attenuation and impedance. At the same time, the areas with low attenuation are characterized by high acoustic impedance.

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