

# Determination of Relationships between the Ultrasound Velocity and the Physical Properties of Bovine Cortical Bone Femur

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## ABSTRACT

**Background:** Accurate measurements of physical characteristics of bone are essential for diagnosis, assessment of change following treatment, and therefore, indirectly, for evaluation of new forms of therapy. This is particularly true of osteoporosis and aging skeleton, in which fractures occur easily. **Methods:** In this study an ultrasonic system was set-up and calibrated on Plexiglas tubes of variable thickness then used to detect the cortical bone thickness change in calf and bovine adult femurs. Lamb waves have been generated and detected using a pair of Piezoelectric point transducers (transmitter and receiver) operating at 60 kHz in contact with the surface of the bone. **Results:** A link has been established between the ultrasound velocity and the bone thickness. On the other hand, the density variation has been also investigated by the simulation of the bone decalcification chemically. The results show that the velocity is very sensitive to both thickness and density, its value reduces as the cortical bone thickness and density decrease. **Conclusion:** This technique might be considered in the attendance of certain bone diseases expressing itself by gradual change in physical properties. *Iran. Biomed. J. 11 (3): 193-198, 2007*

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## INTRODUCTION

The methods used at present to estimate the quality of the bone through the measure of the cortical thickness are the radiographic densitometry and the photon absorpsiometry [1, 2] However, none of them can permit the determination of the mechanical properties of the bone. The ultrasound techniques have much advanced for some years to try to respond to this need. Among investigators having used ultrasonic techniques in bone [3-6], Rich *et al.* [3] suggested that bone thickness or mass could be estimated from ultrasonic measurement. Abendschein and Hyatt [4] reported that ultrasonic velocity may provide a valuable non-destructive tool for more precise determination of the physical characteristics of bone, such as, healing and pathology of bone. Singh [5], Tatarinov *et al.* [6] and Lin *et al.* [7] developed a non-destructive technique for quantitative cortical bone thickness measurements. For this reason, we turned to

measurement of the speed of sound in bone, which depends on not only its density but also its elasticity according to the following equation [8]:

$$E = \rho C^2 \quad (1)$$

In fact, the module of elasticity is low for the sick bones, especially by osteoporosis compared to the one of the normal bone. Osteoporosis affects not only the bone mineral density, but also the internal microstructure of bone characterized by its non-isotropic and inhomogeneous nature and consequently causing reduced bone strength and increased fracture risk. Recently, another diagnosis method based on ultrasonic Lamb waves has been used to measure the cortical bone thickness to predict the osteoporosis disease [9, 10]. In the present work, a pair of transducers composed from Piezoelectric (PZT) transducers and fixed to point wave guide has been used to make measurements even in a localized regions. The ultrasound wave guides are used in various medical applications,

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especially for speed and attenuation measurements in the tissues at low frequency [11] and also in non-destructive testing of materials [12]. Like our previous works [13, 14] where the technique of Lamb wave has been set-up and validated on Plexiglas tubes of variable thickness. When a pair of transducers is used to excite and receive  $A_0$  mode Lamb wave in tubes, any change in thickness can be detected by the change in the Lamb wave velocity due to the dispersive nature of the  $A_0$  mode. With this technique, we have measured Plexiglas tubes thickness with an accuracy of 5%. The developed method was then applied to measure the thickness change of calf and bovine femurs from the phase velocity of the  $A_0$  mode. As the osteoporosis does not only make the bone thickness weak but also change its mechanical properties too, we turned our investigation to study the variations of the density with the ultrasound velocity. The change of the bone density has been obtained by the simulation of the bone decalcification chemically. This decalcification has been obtained by dipping the bone in a 10%-hydrochloric acid solution. A mineral component of the bone will be chemically dissolved in the solution causing a decalcification and therefore a density decreasing. We noticed a decrease of 48% of the ultrasound velocity for 23% decrease of density.

Lamb waves are elastic waves which propagate as a combination of longitudinal and transverse wave in plates. The velocities of all Lamb waves are dispersive in any plate of thickness,  $2d$ , for a particular angular frequency; there will be a finite number of real roots of Rayleigh-Lamb equation given by equation 2.

$$\frac{\tan \omega d \sqrt{(c^2 - c_2^2)/c_2^2 c^2}}{\tan \omega d \sqrt{(c^2 - c_1^2)/c_1^2 c^2}} + 4 \left( \frac{\sqrt{((c/c_1)^2 - 1)((c/c_2)^2 - 1)}}{[2 - (c/c_2)^2]} \right)^{\pm 1} = 0 \quad (2)$$

The phase velocities of Lamb waves as a function of the frequency-thickness product may be obtained by solving the previous equations [15]:

The +ve and -ve signs relate to symmetric and anti-symmetric Lamb waves, respectively,  $c$  is the phase velocity and  $c_1$  and  $c_2$  are the bulk longitudinal and shear wave velocities, respectively. The positive and negative real roots of equation (2) correspond to propagating harmonic waves in the +ve and -ve  $x$  directions, respectively, while the imaginary or complex roots relate to non-propagating spatially varying vibrations.

The dispersion curves of Figure 1, plotted from the equation 2, represent the phase velocities as a function of frequency-thickness product ( $fd = f \times d$ )

for each mode. Inspection of this dispersion curves shows that all Lamb modes except the lowest order symmetric ( $S_0$ ) and anti-symmetric ( $A_0$ ) modes, have lower cut-off frequencies. This particularly makes

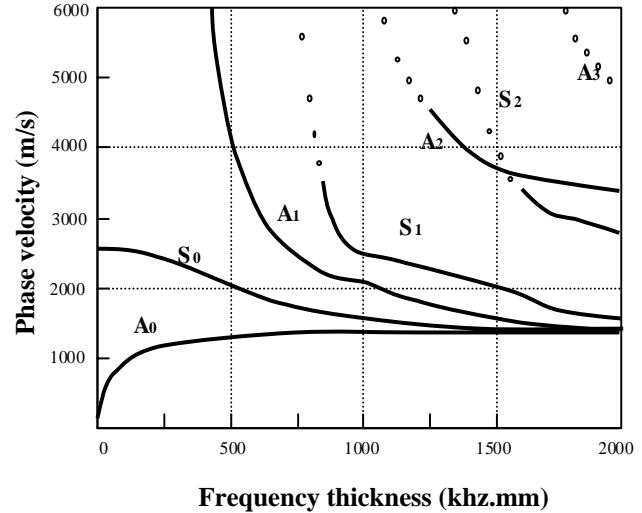


Fig. 1. Dispersion curves of Lamb waves in Plexiglas plate.

the modes  $A_0$  and  $S_0$  very interesting because they are the only modes of propagation existing for the product which corresponds to our experimental conditions. One can notice that the phase velocity of the mode  $S_0$  is practically constant if  $f.d < 0.2 \text{ MHz} \times \text{mm}$ . On the other hand, the phase velocity of the mode  $A_0$  is highly dispersive in that region with phase velocity approaching very small values as  $f.d \rightarrow 0$ . As  $f.d$  gets larger, both modes converge to the Rayleigh surface wave. The lack of cut-off frequency for these two modes can be exploited particularly for the smaller values.

## MATERIALS AND METHODS

**Bones preparation.** In order to calibrate our system, we have carried out the measures on 5 Plexiglas tubes of 300mm length, and 200 mm large with different thickness respectively of 20 mm, 15 mm, 12 mm, 9 mm and 2 mm. On each tube, straight lines joining the two ends have been drawn in the centre to overcome the reflections on edges. The lines were divided into measuring sites of 10 mm distances. For the animal sample, two calf bones femurs of 257 mm length, 320 mm of respective diameter 36 mm and 34 mm and also one bovine femur of 412 mm length and with 41 mm diameter in the centre of diaphysis were obtained fresh from a

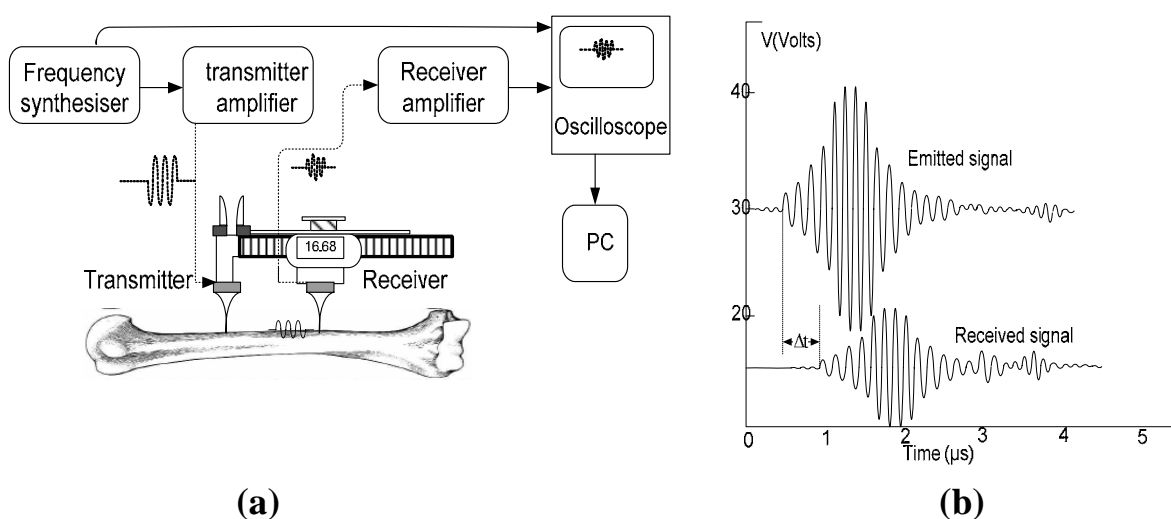


Fig. 2. (a) Measurement principle; (b) The emitted and received signals.

local slaughterhouse. The calf femurs were selected for their dimensions and their mechanical features close to those of the human femur [16]. The bovine femur was used for the purpose of comparison. All bones have been cleaned from soft tissues to be used for study and were then preserved in a fridge at 5°C before the start of the experiments in order to minimize degradation. As for the Plexiglas tubes, straight lines joining the two epiphysis have been drawn on each bone. After doing the measures of ultrasound speed at room temperature, a longitudinal cut has been fulfilled on the femurs in order to measure the cortical thickness along the diaphysis with a micrometer.

**Ultrasonic techniques.** A schematic diagram of the hardware is given in Figure 2a. The principal components are the low frequency PZT transducers vibrating at 60 kHz, prolonged by an exponential metallic wave guide and assuring a contact point with the sample. The Nicholson and Mc Dicken works [17] have shown that in the low frequency range (some Kilohertz), the loss of transmission energy between the wave guide and the ultrasound transducer is much less important than in the high frequency range (some Megahertz). These sensors allow one to get a point source-point receiver system which can fulfill short distance  $\Delta d$  (10 mm) measurements. This distance has been selected in order to permit local measures corresponding to a relatively constant thickness of the bone's cortical layer. Realizing that the relative error increases as soon as the distance among transducers decreases, 10 mm has been considered as a minimal distance

for an acceptable error.

The pulse generator transmits a programmable number of sinusoidal pulses at a resonant frequency of the transducers. This signal amplified at 30 Volts RMS (root mean square) is afterwards applied to the emitting transducer coupled with the sample. The signal received from the transducer receiver which can be moved from a micrometer along the straight line on the bone axis, is first amplified and then filtered in order to obtain a good signal to noise ratio. The time measured  $\Delta t$  is the time flight (Fig. 2b) corresponding to the distance covered in the sample between the emitted and the received signal, respectively the first and second points distance which is given by the digital oscilloscope. Finally, the speed of the wave is simply given by the formula:  $v = \Delta d / \Delta t$ .

## RESULTS

**The measured velocities in sensitive region of  $A_0$  mode.** One of the problems that we often meet with Lamb waves is the coexistence of several modes in the tubes, which makes the measures difficult. According to the material, the only excitation of that  $A_0$  and  $S_0$  modes imposes a lower frequency-thickness product as it is shown in Figure 1 for the case of Plexiglas plate. Applying a normal traction of the emitter and the receiver on one surface of the plate, the  $A_0$  mode is selected over the  $S_0$  mode [12]. In the low  $f \cdot d$  region, this mode is very sensitive to the thickness of the tubes.

As it is shown in Figure 3, the variation of the

ultrasound Lamb wave velocity as functions of the bones thickness follows the profile of the  $A_0$  mode in the sensitive region. These curves were plotted to confirm that the measurement are in the dispersive region of the  $A_0$  mode. As the frequency is fixed to 60 kHz, then the variation of the velocity will depend only on the thickness of the material. One can also notice that the calf bone velocity variation is higher than the bovine ones due to the difference in the mechanical characteristic.

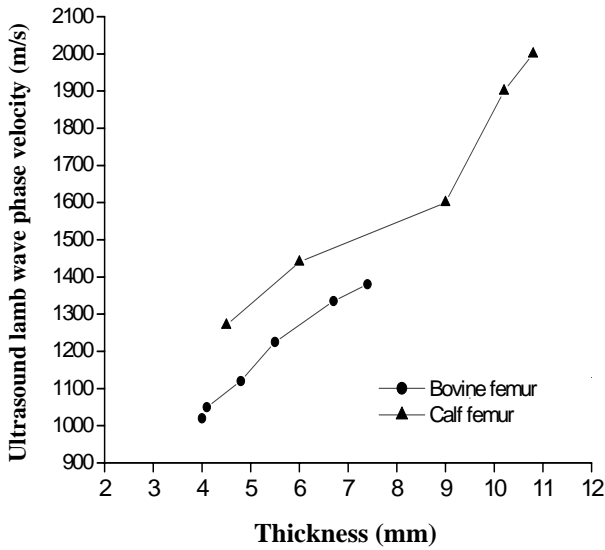


Fig. 3. Variation of ultrasound Lamb wave phase velocity in  $A_0$  mode as function of bone thickness.

**The measures of ultrasound velocities in calf and bovine bone femurs.** While plotting the profiles of the ultrasound velocity and the cortical thickness as a function of the positions of the measuring points along the diaphysis (Figs. 4 and 5), we can notice a very good correlation between these two parameters. The velocity measured is weak at the epiphysis extremities of the bones where the cortical thickness is weak, while it is more important at the centre of the diaphysis where the cortical thickness is maximal. The maximum thickness variation between the extremity and the centre of the diaphysis measured by micrometer was 9.4 mm for bovine femur and 8.2 mm for the calf femur. In our experiment we have taken 10 measurements sites, 5 sites on both sides from the centre of the diaphysis.

The results show that the ultrasound velocity change is very sensitive to the cortical thickness change. An average of 360 m/s change (from 1380 m/s to 1020 m/s) from the measured velocities was obtained for a variation of 3.4 mm (from 7.4 mm to

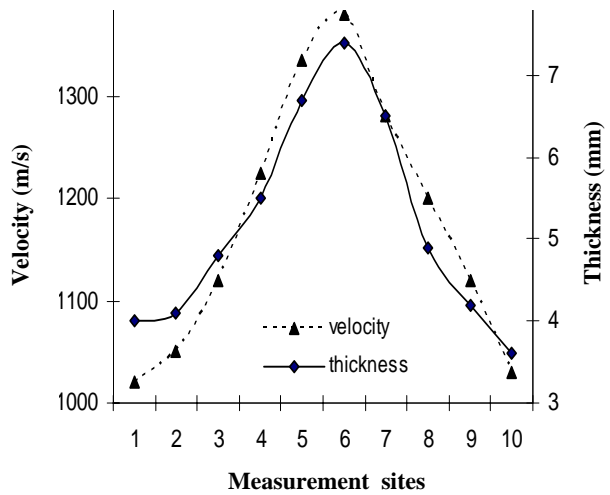


Fig. 4. Velocity and thickness profile for bovine femur bone vs. measurement sites.

4 mm) for the bovine bone, corresponding to 26% velocity change for 46% thickness variation. However for the calf femur, 36% velocity change was obtained for 56.5% thickness variation. To recapitulate, these results give an average of 106 m/s velocity variation for 1mm thickness change for bovine femur, whereas for calf femur, an average of 115.5 m/s velocity variation for 1mm thickness change. This relation between the two parameters represents the sensitivity of the system to detect the variation change in the cortical bone thicknesses.

The errors in ultrasound phase velocity measurement results from the uncertainty of time delay and distance measurements. Since the time delay measured from the digital oscilloscope is very accurate by signal averaging, the resulting error in time delay measurements is in the order of

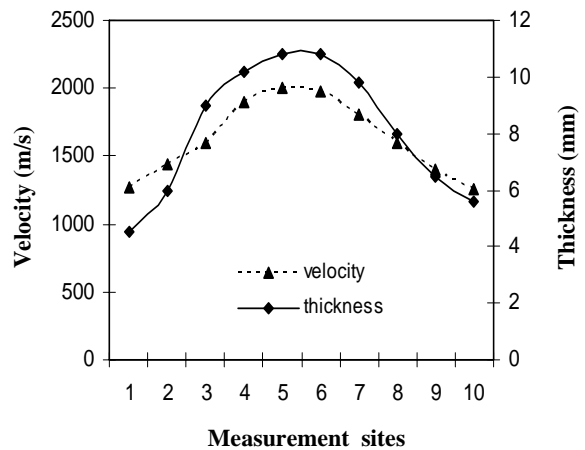


Fig. 4. Velocity and density of bovine femur bone vs. dipping time in the hydrochloric acid solution.

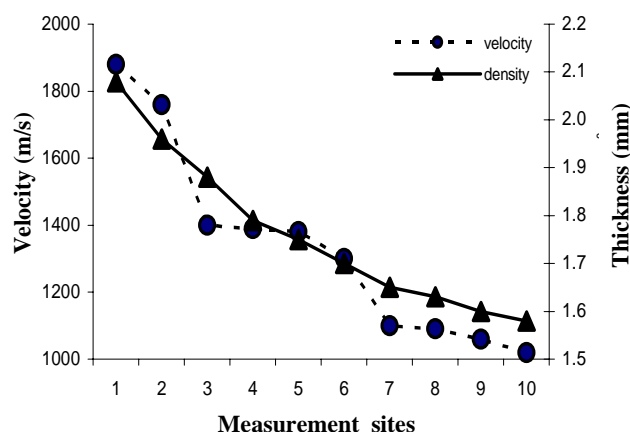
$2 \times 10^{-4}$  %, assuming that the digital oscilloscope has a high resolution time interval measurement. Thus, the accuracy of the phase velocity measurements is determined mostly by the emitter to receiver distance measurement. However, the transducer distance is mounted on mechanical micrometer stages for accurate lateral motion. By this method, the uncertainty in determining distance between two receiver positions for phase velocity measurement is reduced about 0.5 mm. The resulting maximum error in phase velocity measurement can be estimated to be of the order of 5% for distance measurement of 10mm and this error decrease as soon as the distance between the transducers gets larger. This means that it is possible to detect at least 0.5mm thickness change which represents a good precision for our application.

**Ultrasound velocity related to density measurement.** The simulation of decalcification has been fulfilled by dipping sections of bovine femur bone in a 10% hydrochloric acid solution. More time the bone is left inside the solution more it loses its mineral component. This chemical process causing a decalcification and therefore a density decreasing may simulate the osteoporosis disease as the latter causes the bone to have a low bone mass, thus, fragile and more likely to break.

The measures of ultrasound velocity in the bovine femur bone have been fulfilled in a centre site of the diaphysis during the decalcification process for each hour. The measures of the density have been done by taking a sample subjected to the same decalcification process; these measures were followed in parallel to the velocity measures. The curves representing the ultrasonic velocity and density as a function of time process are shown in the Figure 6. During a 10-hour process, the velocity decreases from 1956 m/s to 1008 m/s, which represent 48% decreases from the initial value. As for the density, it passes from 2.07 to 1.58 g/cm<sup>3</sup>, which represents 23% loss from the initial value.

## DISCUSSION

In this study, we have shown that there is a strong correlation between the measured velocities and the profile of the cortical thickness of the bone. With this technique, we are capable of measuring the thickness with an accuracy of 5%. The measurements errors are acceptable and the fact that they have been carried out in the dispersive region of



**Fig. 6.** Velocity and density of bovine femur bone vs. dipping time in the hydrochloric acid solution.

the A<sub>0</sub> mode, this has increased the sensitivity of the measured velocity as it was shown in figure 3. The results are in accordance with those found in the bibliography.

As the osteoporosis disease does not only make the bone thickness weak but also changes its mechanical properties too, we turned our investigation to study the variations of the density with the ultrasound velocity. The results obtained from the simulation of decalcification in figure 6 bring out again that ultrasonic velocity decreasing depends to the density loss and also to the bone elasticity. This is true because more the bone is decalcified more it became soft and flexible. This study suggests that this technique might be envisaged in the attendance of certain bone diseases expressing itself by a graduate decreasing of the cortical thickness and changing their mechanical properties.

The technique might be feasible to make measurement *in-vivo*, the effect of soft tissue on velocity measurement is automatically cancelled as long as its thickness is constant over the measured site [18]. The technique mainly aims at measurement on long bone such tibia, since at the medial face; the soft tissue is rather thin, with constant thickness over a large site.

The procedures of measures using ultrasonic PZT transducers for the characterisation of the materials become inoperative in the case of structures of narrow and irregular shapes. That is why some methods said of non contact or of punctual contact take a significant expansion for the generation and detecting the acoustic signals in the material for testing. However their sensitivity is very weak compared to the conventional systems. For all these

reasons, a system of characterisation of the bone by the point-source point-receiver has been developed for the measures of ultrasound velocity: Measures which demand less preparation of the samples and can be fulfilled without coupling in very localised regions. Compared to other techniques like spectroscopic or differential measures, the method suggested in this study, though very simple permits to make correct measures of the phase velocity which give some mechanical and physical characteristic of the examined biological element.

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