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▶ **To cite this version:**

Jean-Philippe Berteau, Martine Pithioux, Patrick Chabrand, Philippe Lasaygues. Children bone elastic properties characterization: an ultrasonic issue ?. 10ème Congrès Français d'Acoustique, Apr 2010, Lyon, France. hal-00534635

HAL Id: hal-00534635
https://hal.science/hal-00534635

Submitted on 10 Nov 2010

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10ème **C**ongrès **F**rançais d'**A**coustique

Lyon, 12-16 Avril 2010

Children Bone Elastic Properties Characterization: An Ultrasonic Issue ?

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Bone is a composed structure including cortical bone, trabecular bone and bone marrow, but organization of these composites changes during ageing ; indeed, differences can be shown from childhood to old age. But these changes in childhood are not clearly shown. For example, certain studies compare bone density (Bone Mineral Density : BMD) of children, like in Chron disease, but not the stiffness, strength or elastic modulus. Some have chosen specific bone, close to cancellers cells or cadaver fragments. The aim of this study was to determine the elastic properties of the children bone. We have used surgery waste (bone transplantation) from long bone (fibula : essentially composed of cortical bone) of children without metabolic or mineralization disturbances. A specific ultrasound frame able to process very small sample (36 mm high, 11 mm tall, and 0.2 mm thickness) was used to evaluate velocity grading along bone axis. They are moved with 0.1 mm accuracy in x and y axis. For Z axis, azimuth got 0.01° of accuracy (Fig 1). Nominal frequency can increase from 3 to 10 MHz. Our first results (two samples) show longitudinal (vL) and transversal (vT) velocity of propagation (values in m.s-1) We used the same method as used for isotropic transverse bovine bone frame. In the case of orthotropic we can right 12 elements of rigidity matrix. Comparisons were made between juvenile specimens and mature specimens, and the first result is that the wave velocities, and the associated elastic modulus, are then lower in children than adult around 30 percent less for 11 years old child.

1 Introduction

Bone is a composed structure including cortical bone, trabecular bone and bone marrow. They are categorized in different groups (long bone as tibia, flat bone as scapula, sesamoïd as patella) showing specific mineral and tissue organization (as a composite). But organization of these composites changes during ageing ; indeed, differences can be shown from childhood to old age.

Ageing adversely affects the elastic and ultimate properties of human cortical bone, lot of studies show differences between normal bone and osteoporotic adult for stiffness. In ageing bone, there is modification in the elastic properties of the material. But these changes in childhood are not clearly shown.

In paediatric, the diagnosis of bone diseases, the estimation of fracture risk and treatment decision are provided without specific knowledge of bone mechanical properties.

Most of the time, the bone mineralization density distribution (BMDD) provides important information about the effect of metabolic bone diseases and therapeutic interventions on the mineralization of the bone material. It exists remarkably small inter-individual variation in BMDD, suggesting that even small deviations have biological relevance. Indeed, low matrix mineralization was found in osteoporosis and mild primary hyperparathyroidism while a shift towards higher mineralization was found in osteogenesis imperfecta. .

The capacity of bone to resist to mechanical forces and fractures depends not only on the quantity of bone tissue but also on its quality ; indeed bone matrix is a two-phase system in which the mineral phase provides the stiffness and the collagen fibers provide the ductility and ability to absorb energy (i.e., the toughness).

For example, certain studies compare bone density (Bone Mineral Density : BMD) of children, like in Chron disease, but not the stiffness, strength or elastic modulus[1, 2]. During childhood there is a specific process from cartilage to bone with difference for each bone group. Few studies consider mechanical characteristic of this process[3, 4]. Some have chosen specific bone, close to cancellers cells, or cadaver fragments[5, 6].

Consequently, there is a lack of reference which led us to analyse children bone's in histomorphometry, BMDD (Bone Mineral Density Distribution) and macroscopic mechanical properties by ultrasonic method and three points bending system. The aim of this study is to give an average value of mechanical properties of cortical part of children bone, using histomorphometric BMDD data ; macroscopic values and correlation with clinical informations (weight, height, age...) In our study, we have used surgery waste (bone transplantation). These samples come from long bone (fibula) and are essentially composed of cortical bone. In order to find young modulus and certain parts of stiffness matrix, we have made a pre study to evaluate our protocole and the impact of water bench on collagen fibers before comparing ultrasonic data to three points bending evaluation.

2 Method

2.1 Population

The study population comprised Caucasian subjects aged from 7 to 13 years (female, male) who got surgery using bone transplant for various orthopedic conditions, such as lower limb deformities, scoliosis, clubfeet and other problems that require corrective surgery. We have kept bone wastes and rank it in differents age, sex and kind of bone group . Litterature show difference in different group and in different conditions [7, 4, 8, 9, 10] resumed in FIG 1. All subjects were ambulatory, had normal renal function and had no evidence of any metabolic bone disease. None was immobilized prior to surgery or received medications known to affect bone metabolism. This cohort is part of paediatric population from the Timone hospital of Marseille.

criterion	inclusion	exclusion
ETHNOS	CA	A / AS / HIS
SEX	XX / XY	GENETIC DISORDER
CLINICAL PATHOLOGY		METABOLIC DISEASE / RENAL DYSFUNCTION/ DIABETES / ASTHMA
CHRONIC OR FREQUENT USE OF MEDICINE		CORTICOID INHALED STEROID ANTIEPILEPTIC DRUGS GROWTH HORMONES
BEDDRIDEN	NOT BE	WAS FOR SHORT OR LONG PERIOD
PUBERTY		UNASCERTAINABLE
CALCIUM DAILY AMOUNT	MORE THAN 1000mg	LESS THAN 1000mg

FIGURE 1 – population criterion

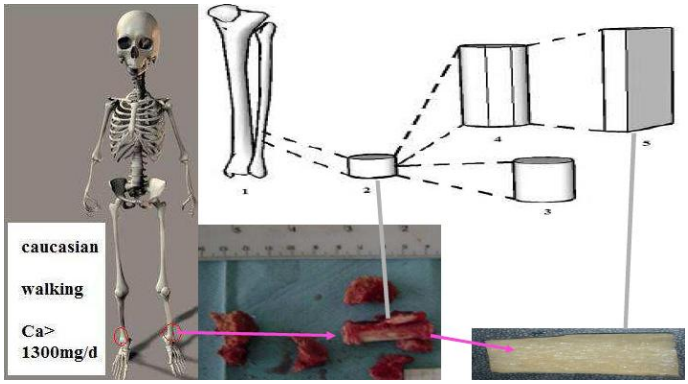


FIGURE 2 – fibula samples

2.2 Ultrasonic method

The physical principle of the technique is based on velocity mesurement from the surface of a sectioned bone area, it induces some values wich can provide some part of the matrix rigidity. Ultrasound technics is employed to determine speed of sound (transverse and longitudinal velocity) through bone samples, it necessits specific ultrasound scanner.

The signal receipt from the first to the second transducer were converted into values of the acoustic impedance using a time-of-flight (TOF) based defocus correction and impedance calibration. The pulse signal is measured as a function of the transducer-sample-transducer distance which led us to determine the value of sample's thickness. For each transducer-sample transducer-distance, the TOF and the corresponding normalized intensity were determined.

We have developed a specific ultrasound frame (Fig.3), which is able to process very small sample.

Fresh fibula bone samples were prepared using a low speed diamond saw (Isomet 1000, Buehler ; Lake Bluff, IL, USA) in order to obtain plane and parallel surfaces.

The samples prepared for ultrasonic measurements were completely immersed in a temperature-controlled tank filled with distilled, degassed water at 25° C. After size fitting, fibula was separated in three part, three bone samples trending to look like parallelepiped were suspended to a mechanical stage and placed at the center of the measuring system. The sections were frozen less than 5 times before experimentation to preserve their mechanical properties.

They are moved with 0.1 mm accuracy in x and y axis. For Z axis, azimuth got 0.01° of accuracy. Nominal frequency can increase from 3 to 10 MHz.

We have evaluated bone velocity using the difference of sound path duration and Snell Descartes law (1)(2)(3), which led us to an expression of the bone velocity. First, we record the longitudinal velocity and after rotation we can record the transversal velocity. A, B, B' geometrical dot shown on Fig 2. i : incidence angle and r : refraction angle.

First step was to evaluate our bench with known material and with adult cortical bone. Close to the reference values, these results allow us to evaluate children cortical bone.

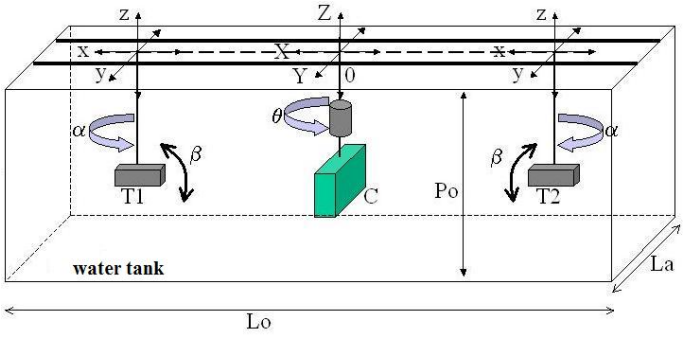


FIGURE 3 – watertank

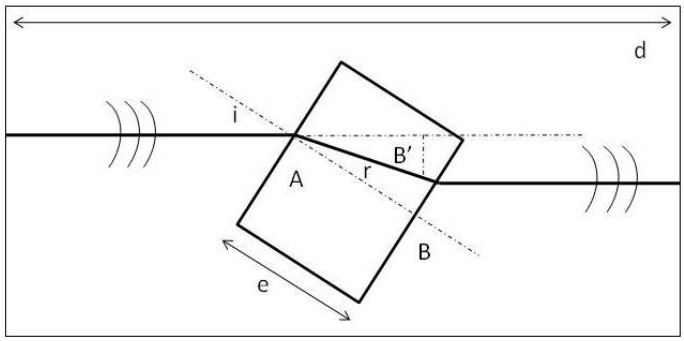


FIGURE 4 – sample position

$$\Delta t = t_{AB'} - t_{AB'} = \frac{AB'}{c_w} - \frac{AB}{c_b} \quad (1)$$

$$n = \frac{c_w}{c_b} = \frac{sin\ i}{sin\ r} \quad (2)$$

$$c_b = \frac{c_w}{\sqrt{1 + \frac{c_w \Delta t}{x} \times \left(\frac{c_w \Delta t}{x} - 2cos_1\right)}} \quad (3)$$

2.3 Protocol evaluation

Due to the small size of the sample, we are bound to optimize bone's sample. In first, we have to evaluate the impact of water immersion on collagen fibers. That's why, a biochemical analysis of collagen cross links before and after ultrasound test indicate us if we can use ultrasound method as a safe test. Two grammes bones' samples, five animals(ewe, chiken, rabbit, calf) and two humans (children long bone and old female vertebrae), were analysed. If there is no impact of us we can use directly us evaluation, if not we should cut 2 grammes before water immersion.

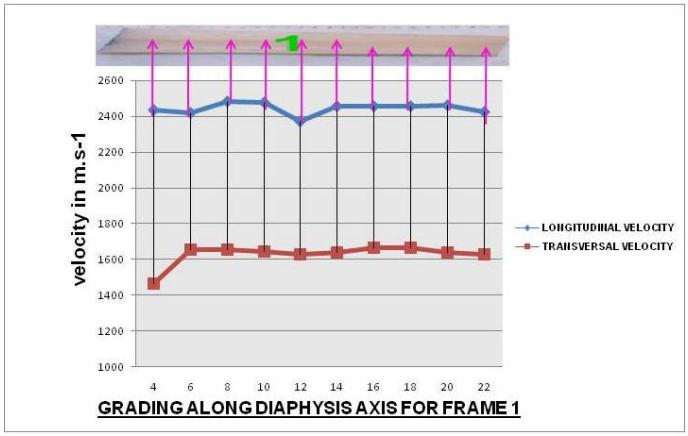


FIGURE 5 – results 1

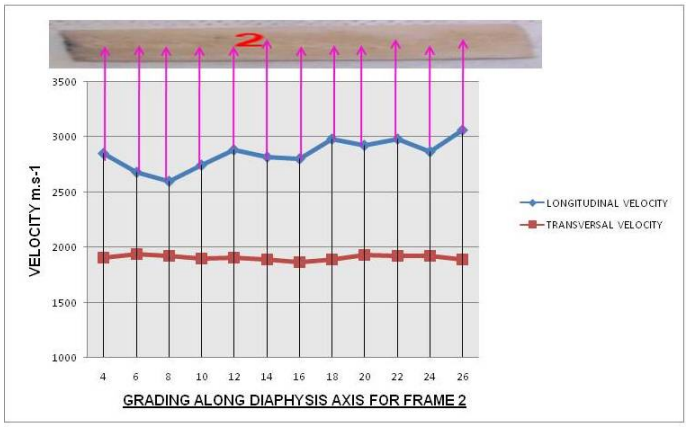


FIGURE 6 – results 2

3 Results

Our first results (two faces of two fibula samples) show longitudinal (CL) and transversal (CT) velocity of propagation (Figure 5,6 and 7). We used the same method as used for isotropic transverse bovine bone frame [11]. The first results show around 2500 m.s-1 for first face and around 3000 m.s-1 for the second. The third face was not adapted for ultrasonic method due to a non parallelipedic form. Results are equal for the first bne (eleven years old girl) and the second (twelve years old boy). The poisson's ration is around 0.3 in each case.Biochemical evaluation is in progress, no value are available right now.

Longitudinal wave velocity in adult bone (m/s)	Longitudinal wave velocity in adult bone with our ultrasonic scanner (m/s)	Longitudinal wave velocity in child bone with our ultrasonic scanner (m/s)
2700 – 3800 (Katz et al, 1984) 3550 – 4180 (Yoon et Katz, 1976)	3400 - 3600	**2500 - 3000**

FIGURE 7 – results values

4 Discussion

The aim of this study was to determine the elastic properties of the children bone. For this purpose, an ultrasonic method, using a mechanical scanner with which linear and sectorial scanning can be carried out, was implemented to determine acoustical parameters such as the wave velocities and poisson's ratio. These results show different spatial distribution of elastic characteristics per bone side, like it was observed in adult long bone using classical mechanical tests [12] and by ultrasonic technique [13]. This study is the first step of a more general work. Comparisons were made between juvenile specimens and mature specimens, and the first result is that the wave velocities, and the associated elastic modulus, are then lower in children than adult. In order to find young modulus and certain parts of stiffness matrix, we have made an ultrasonic evaluation of bone celerity as a first step ; but aqueous atmosphere used for the *in vitro* ultrasonic method of characterization can be the purpose of some differences in compare to mechanical test. To reach our goal, the first step will be to delineate acoustic parameters (velocity and attenuation) of children bone and try to describe its anisotropy. The second step will be to correlate our ultrasonic result with mechanical results and we could balance these results with data from histological processing. In order to validate our protocole of investigation, the impact of water immersion on collagen is in progress.

5 Acknowledgement

The authors thank to the Paediatric Orthopaedic Surgery Department (La Timone Children Hospital, Marseille, France) and of the Inserm research team (U831, University of Lyon, Lyon, France) for helping them in the preparation of the bone's sample.

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