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In vitro ultrasonic and mechanic characterization of the modulus of elasticity of children cortical bone



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ABSTRACT

The assessment of elastic properties in children's cortical bone is a major challenge for biomechanical engineering community, more widely for health care professionals. Even with classical clinical modalities such as X-ray tomography, MRI, and/or echography, inappropriate diagnosis can result from the lack of reference values for children bone. This study provides values for elastic properties of cortical bone in children using ultrasonic and mechanical measurements, and compares them with adult values. 18 fibula samples from 8 children (5-16 years old, mean age 10.6 years old ±4.4) were compared to 16 fibula samples from 3 elderly adults (more than 65 years old). First, the dynamic modulus of elasticity (E_{dyn}) and Poisson's ratio (v) are evaluated via an ultrasonic method. Second, the static modulus of elasticity (E_{sta}) is estimated from a 3-point microbending test. The mean values of longitudinal and transverse wave velocities measured at 10 MHz for the children's samples are respectively 3.2 mm/µs (±0.5) and $1.8 \text{ mm/}\mu s$ (±0.1); for the elderly adults' samples, velocities are respectively $3.5 \text{ mm/}\mu s$ (±0.2) and 1.9 mm/ μ s (±0.09). The mean $E_{\rm dyn}$ and the mean $E_{\rm sta}$ for the children's samples are respectively 15.5 GPa (±3.4) and 9.1 GPa (±3.5); for the elderly adults' samples, they are respectively 16.7 GPa (± 1.9) and 5.8 GPa (± 2.1) . $E_{\rm dyn}$, ν and $E_{\rm sta}$ are in the same range for children's and elderly adults' bone without any parametric statistical difference; a ranking correlation between E_{dyn} and E_{sta} is shown for the first time.

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1. Introduction

Cortical bone is an organic structure with mineral comprises approximately 80% of the human skeleton. Pathologies impacting human cortical bone quality include osteoporosis [1] in adults and osteopenia [2], Crohńs disease [3] or osteopetrosis [4] in children. In addition to its cost in terms of health, European estimates predict that the direct cost of osteoporotic fracture will reach 76.7 billion ϵ by 2050 due to demographic changes, notably the ageing of European populations [5]. A low bone mass in childhood is now recognized as a high risk factor for osteoporosis in later life [6] and authors consider the assessment of bone mineral status in children as a priority [7,8]. The Bone Mineral Density (BMD), which is one of the most gold standard parameters to assess mineral status, requires in the first intention, the use of the dual energy X-ray absorptiometry (DXA). The reference study [9] concerns only 7–17-year-old children, and was conducted with a Hologic DXA

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scanner (Hologic Inc., Waltham, MA, USA). However, the pediatric evaluation of the BMD raises problems of interpretation when the size of the bone varies related to the statural age of the child. The BMD is not correlated with the bone micro-architecture, and several studies have shown the ability of ultrasound measurement to assess the quantity and the quality of the explored bone area (elasticity and structure) [10,11].

There is a tremendous lack of data on young bone strength and mechanical behaviors: several papers [12–14] report age-dependence for ultrasonic axial transmission data but, to our knowledge, the elastic properties of cortical bone in children have been quantitatively investigated by only two *in vitro* mechanical studies [15,16], both using destructive tests on dry samples. In both cases, the experimental values for bone in children support the theoretical optimization hypothesis [17] of an increasing bone modulus of elasticity from neonate values to adult values, which is currently used in pediatric computational methods. It has been developed in accidentology [18] but it has been invalidated by [19] for young child. All this would suggest the likelihood of lower ultrasonic wave velocities and modulus of elasticity in children compared to adults. Yet the findings of a recent study performed by our team

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on rib cortical bone from teenagers with scoliosis [20] do not support that hypothesis. Although our study concerned pathological bone, our conclusion was that the *in vitro* ultrasonic wave velocities and the Young Modulus E values were close to the elderly adult values found in the current literature. The lack of reference concerning normative pediatric ultrasonic wave velocities and elastic properties of children's prevents the medical community from using the diagnosis devices based on analytic model of ultrasound scattering dedicated to adult's population (Quantitative ultrasound and echography). Consequently, the data collection and the development of relevant models of bone growth is a critical need to investigate an effective device of diagnosis and to meet the needs expressed by the medical community.

The aim of this study was to obtain ultrasonic wave velocities, dynamic and static modulus of elasticity, and Poisson's ratio for cortical bone samples from children, and then to compare these results with elderly adult cortical bone samples. Our two-stage study proceeded first by performing experimental ultrasound measurements to assess ultrasonic wave velocities, dynamic modulus of elasticity ($E_{\rm dyn}$) and Poisson's ratio (ν) and second, via 3-point microbending tests, to assess static modulus of elasticity ($E_{\rm sta}$).

2. Materials and methods

Fig. 1 is a diagrammatic representation of the method used.

2.1. Samples

In accordance with the stipulations of the French ethical committee, we studied cortical bone samples from Caucasian patients (5-16 years old, mean age 10.6 years old ±4.4) of the University Hospital in Marseille who required auto transplant surgery. Surgical waste bone, largely consisting of cortical bone from fibula diaphysis was studied; the selected population was composed of walking children not on drugs disturbing their bone metabolism. All auto transplant samples were excised from a non-pathological location in the fibula 5 cm above the ankle (Fig. 1). The elderly adult bone fibula samples were extracted from the same location as for the children's samples, but from cadavers (+65 Years old) at Inserm U1033 and UMR-T 9406 Ifsttar/UCBL (Lyon, France) bone bank. Samples for study were obtained by cutting the waste fragments in parallelepipeds (plane and parallel surfaces) using a low-speed diamond saw (Isomet 1000, Buehler; Lake Bluff, IL, USA). A total of 18 cortical bone parallelepipeds extracted from 8 children fibula samples were obtained and measured with a digital caliper. Dimensions of all the samples are in Table 1. Because of small dimensions of the samples (15-50 mm long, 4-10 mm wide,

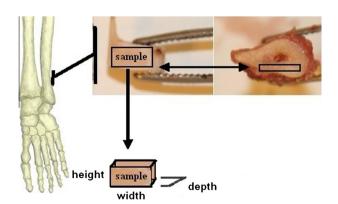


Fig. 1. Samples preparation: the waste fragment from auto transplant which was selected to prepare one of the children's cortical bone samples tested.

0.5–3 mm thick) the US protocol only tested the transversal axis (perpendicularly to the length–width plane) of bone samples.

Sixteen elderly adult (+65 years old) bone samples excised from cadavers were similarly prepared. Each sample was stored at $-20\,^{\circ}\text{C}$ in phosphate buffered saline with less than 5 freezing cycles.

2.2. Ultrasonic measurements

We used an ultrasonic protocol specifically developed, to process small and thin samples, and which has been validated on standard materials (plexiglass) [22] and on animal and human adult bones. The ultrasonic bench [21,22] used consisted of a main arm carrying two linear stages. Each linear stage was carrying the end-rod transducer, and was moved linearly with increments of hundredths of millimeters. Angular scanning is carried out by rotating either the main arm or the object holder. The transducers can also be positioned and oriented precisely; such precision allows for both linear and sectorial scanning.

The parallelepiped bone sample to be tested was placed in the presumed geometrical center of the bench so that the maximum distance between the transducers and the center was 30 mm. The surrounding fluid medium was water at an average temperature of 18°, the variations of temperature were taking into account using Marczak's equation [23].

The thickness of the samples estimated by caliper measurements ranged from 1.5 to 3 mm, consequently a nominal frequency of 10 MHz was chosen, making the wavelength in water ten times greater than the sample thickness.

Two focused broadband transducers at $10\,\mathrm{MHz}$, $(5\,\mathrm{mm}$ diameter, $6\,\mathrm{dB}$ -bandwidth ranging from 9 to $13\,\mathrm{MHz}$; Imasonic, Besançon, France), facing each other with their axes aligned, were used to scan along transverse parallel directions through the sample (perpendicular to the bone axis). The focal area of the transducer was 3×3 mm with a focus set at $30\,\mathrm{mm}$. Only propagation processes were taken into account and the ultrasonic wave attenuation was assumed to be weak. Firstly, thickness e was calculated for each sample using a pulse-echo technique [21]. The wave reached the interface, also assumed to be plane at this scale, perpendicularly.

Time of flight (TOF) of the propagating waves was determined in the reflection mode, from the left (respectively right) transducer to the left (respectively right) interface of the sample. The TOF measured between transducers without sample was used as reference. All the thicknesses thus obtained were compared to caliper measurements.

Then the longitudinal and transverse velocities were determined in the transmission mode by rotating the bone around the clamp axis so as to change the incidence of the acoustic beam. The emitter was fixed and the receiver could be moved laterally.

We define θc as the critical angle such that θc = arcsin C_{water}/V_l . When the incident angle $\theta i < \theta c$ we can observe the longitudinal waves and when $\theta i > \theta c$ we can see the transverse ones.

The velocities V_l (respectively V_t) of the longitudinal (respectively transverse) waves in the samples were determined in the transmission mode using the following equation:

$$V_{l,t} = \frac{C_{\text{water}}}{\sqrt{1 + C_{\text{water}} \frac{\Delta t}{e} \left(C_{\text{water}} \frac{\Delta t}{e} - 2 \cos \theta i \right)}}$$
(1)

 C_{water} is the ultrasonic wave velocity in water, measured without bone sample, e the thickness of the sample. For every angle, Δt is obtained by cross-correlation between the first signal going through the zero stage with the sample, and the reference signal measured earlier without the sample (Fig. 2).

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