Buenos Aires – 5 to 9 September, 2016
Acoustics for the 21st Century…

PROCEEDINGS of the 22nd International Congress on Acoustics

Biomedical Acoustics: Paper ICA2016-630

Advances in cortical bone assessment using ultrasonic resonances and guided waves

Jean-Gabriel Minonzio[(a)], Quentin Vallet, Nicolas Bochud, Quentin Grimal, Pascal Laugier

[(a)] Sorbonne Universités, UPMC Univ Paris 06, CNRS, INSERM, Laboratoire d'Imagerie Biomédicale (LIB), Paris, France jean-gabriel.minonzio@upmc.fr

Abstract

Assessment of bone mechanical properties is an important clinical issue. Recently, progresses achieved in quantitative ultrasound have stimulated a renewed interest for basic and clinical bone studies. Resonant ultrasonic spectroscopy has been adapted for the full characterization of the anisotropic stiffness of small specimens of highly damping materials such as cortical bone. In RUS, the elastic properties are estimated by solving an inverse problem by fitting model predicted frequencies to the measured resonant frequencies. We have introduced a Bayesian approach in which an a priori knowledge of the exact pairing between measured and predicted resonant frequencies is not necessary, the optimal pairing being determined in the course of the optimization process. RUS is prone to provide answers to questions that remain open regarding the determinants of cortical bone elastic properties. On the other hand, guided waves measured in axial transmission, have been proposed for the *in vivo* investigation of appendicular bones (tibia, radius) which are relatively accessible to measurements. We have developed a specific data processing and an inverse problem solving scheme using genetic algorithms to overcome the challenges caused by surrounding soft tissues and by the complex structure of the cortical waveguide. Our procedure allows estimating cortical bone biomarkers such as cortical thickness, stiffness and porosity.

Keywords: cortical bone, ultrasound, guided waves, Resonant ultrasonic spectroscopy (RUS), axial transmission (AT)

Advances in cortical bone assessment using ultrasonic resonances and guided waves

1 Introduction

Measurement of bone mineral density (BMD) using Dual-energy X-ray absorptiometry (DXA) has become the gold standard for the diagnosis of osteoporosis and fracture risk assessment. Adequate evidence indicates that these instruments have only modest predictive value for fractures and for guiding decisions about which patients to treat. For example, more than 50% of fragility fractures are sustained by women who are classified as being non-osteoporotic by DXA-derived BMD measurements [1]. This reveals that BMD-unrelated determinants of bone strength are not captured by this modality. It is known that conditions such as physical exercise, aging, bone turnover, specific diseases or therapeutic agents can affect independently bone composition (i.e., mineralization), microstructure (e.g., porosity) and material properties (e.g., stiffness, toughness). In osteoporotic patients, long bones have a thinner and more porous cortex [2].

A typical case that attracted special attention recently is that of mineralization. The relevance of bone mineralization as a factor contributing to bone strength and stiffness is generally acknowledged. In the context of bisphosphonate-based treatment, material property changes (i.e. increase in mineralization) are expected. However, very high mineralization may result in brittle bone. To date controversy remains on the question of maximum treatment duration to avoid these detrimental effects (i.e., the occurrence of atypical femoral fractures suspected to result from long-term use of biphosphonate). In addition, it is not clear how such changes could be measured *in vivo* to help an eventual decision about treatment discontinuation, in case a negative impact of enhanced mineralization can be verified and linked to measurable bone properties.

The non-invasive assessment of bone material properties in patients, particularly independent assessment of porosity and mineralization, remains unattainable to date. One promising approach to bridge this gap is based on quantitative ultrasound (QUS) [3]. An overview of recently developed axial transmission techniques is proposed in the second part of this paper.

In parallel, we directed part of our work towards gaining *ex vivo* a detailed knowledge of cortical bone material characteristics. One motivation is to provide guidance for the interpretation and the optimization of *in vivo* cortical bone QUS assessment. A second motivation is to contribute to fundamental knowledge of mechanical properties of bone, particularly by investigating the relation of anisotropic elastic stiffness to bone composition and microstructure. Elastic properties of bone are nowadays widely used in fundamental studies, in conjunction with numerical models (e.g. finite element modeling), to investigate the structure-function relationships and in clinical applications to predict fracture risk or to monitor fracture healing. A novel emerging quantitative ultrasound technology (namely, resonant ultrasound spectroscopy RUS) has emerged to non-invasively investigate the anisotropic elastic properties of bone. We

have followed a systematic approach to the goal of developing this technique to show how it can help in characterizing the elastic properties *ex vivo*. This will be presented in the third part of the paper.

2 Axial transmission (AT)

The last decade has seen the emergence of ultrasound axial transmission (AT) techniques to assess cortical bone [3]. Cortical bone is best accessible at the radius or tibia.

A key feature is the development and testing of a new (AT) device that is not limited to the first arriving signal as in typical clinical procedures [4-9]. The new protocol is based on the waveguide concept and includes measurements of all guided modes that are transmitted in the cortical waveguide and their corresponding dispersion spectrum [4-6]. Mode dispersion curves depend on the structure and material properties of the waveguide. Fitting an appropriate waveguide model to the experimental data allows one to retrieve the waveguide characteristics [7-9]. This measurement includes virtually more comprehensive information on cortical bone, such as cortical thickness, porosity and anisotropic elastic stiffness, compared to conventional QUS or X-ray measurements. Particularly, speculating on the different impact vascular porosity and mineral crystal deposit may have on the anisotropy, e.g. of elasticity, the potential to assess anisotropy should be carefully investigated in the perspective of the differentiation of alterations of our bones resulting from changes in porosity or mineralization.

Validation of guided wave axial transmission technique (GW-AT) measurements was achieved through extensive basic *ex vivo* measurements [7]. Measurements were obtained with a multi-emitter and multi-receiver ultrasound transducer following a bidirectional principle, to achieve measurements of guided waves propagating in cortical bone with an automatic compensation for the overlying soft tissues [6]. The probe (Vermon, Tours, France), aligned with the bone axis (Figure 1), consists of a 24 element receiving array placed between two arrays of 5 transmitters each. A custom-made electronic device (Althaïs, Tours, France) was used to transmit ultrasonic pulses (centre frequency: 1 MHz, -6dB frequency bandwidth: 800 kHz) and to record the received ultrasound signals. An ensemble of 240 rf signals corresponding to all possible pairs of transmitter/receiver elements were digitized (12 bits, 20 MHz, 1024 samples) and time averaged (16 times). The experimental dispersion curves are extracted using a 2-D spatio-temporal Fourier transform combined with a singular value decomposition denoising step, to overcome the limitations of the classical 2-D Fourier algorithm. The method has been extensively described in our previous publications [4-9].

Figure 1: *Ex vivo* (left) and *in vivo* (right) measurement with the custom made multi-element probe

Identification of the waveguide material and structural characteristics was achieved by fitting a waveguide model to the experimental dispersion branches. A 2-D non-absorbing transverse isotropic free plate model, i.e., the Lamb model, was considered here [7, 9].

An *ex vivo* study by our group showed that a multimodal axial transmission approach can yield simultaneous estimates of waveguide thickness and stiffness [7]. Prior to the measurements on bone, the method has been validated on cortical bone-mimicking phantoms. The repeatability and the trueness of the estimated parameters on bone-mimicking phantoms were found around a few percent. Estimation of cortical thickness on bone samples was in good agreement with cortical thickness derived from high-resolution peripheral quantitative computed tomography data analysis of the samples (Figure 2).

Such a model-based approach typically requires a complex inverse procedure for pairing the incomplete experimental (colour points in Figure 2) data with the model (continuous and dotted lines in Figure 2). In this first ex-vivo study, the problem was solved by grouping the incomplete data into user-defined experimental trajectories, where each trajectory was associated to a specific Lamb branch.

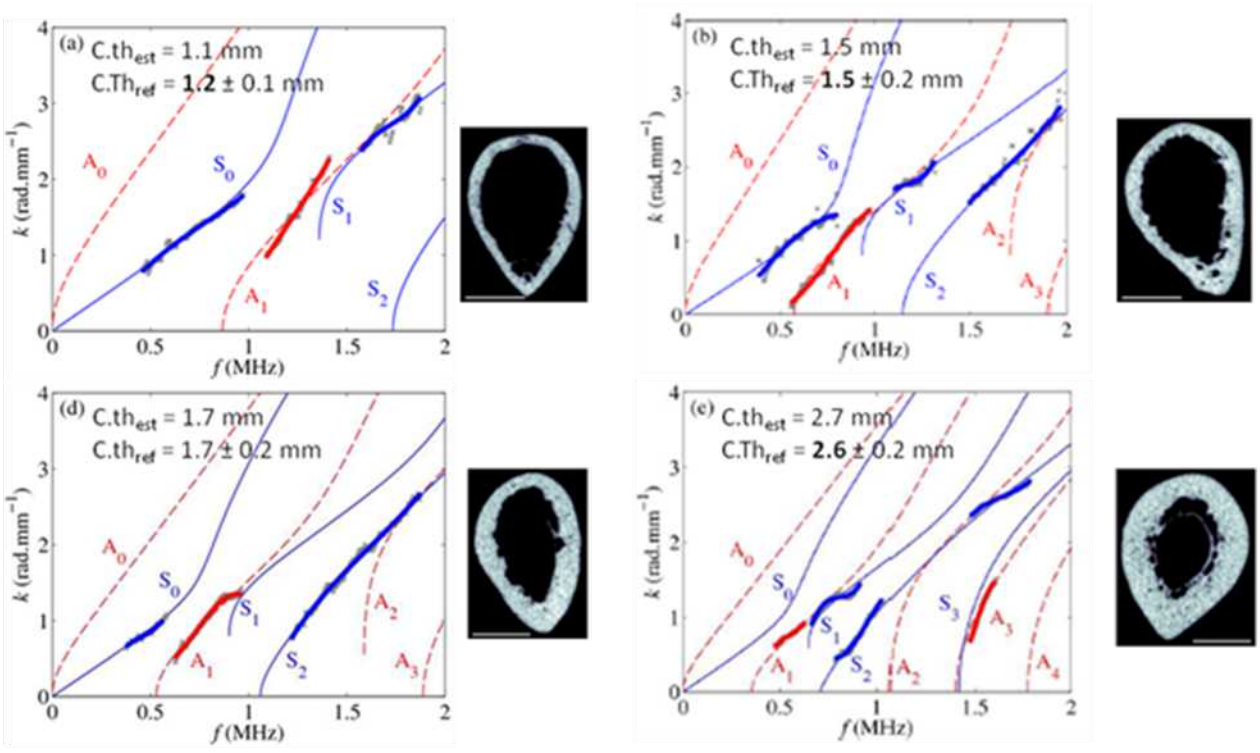

Figure 2: Dispersion curves (i.e., wave number k vs. frequency f) of guided modes propagating in cortical bone (A/ antisymetric modes and S symetric modes). Superposition of ex vivo measurements (points) and the best fit model computed (lines) for 4 human radii (Ct.Th=1.2 mm; 2.5 mm; 1.7 mm; 2.6 mm). Comparison of estimated thickness with the actual thickness derived from peripheral computed tomography (small side panels) shows good agreement.

Nonetheless, this prior assignment is far from trivial in the case of in vivo measurements. To overcome this difficulty, we have developed a global search approach, which enables us to estimate the properties of cortical bone, avoiding any prior knowledge on the experimental trajectories. This can be achieved by including an additional model parameter, which represents the number of theoretical Lamb branches that are necessary to fit the experimental trajectories. The model parameters (Cortical thickness, apparent porosity, number of Lamb branches) are found by a global search algorithm that minimizes the discrepancy between the measured and numerically predicted dispersion spectra. Genetic algorithms are used as search algorithms due to their capability of finding a global solution where the cost functional has multiple local minima.

This approach has been successfully tested on 14 healthy subjects [9]. The inversion procedure based on the 2-D transverse isotropic non absorbing free plate model allowed a combined estimation of cortical thickness and porosity (the main determinant of cortical stiffness). The study was completed with measurements on 5 ex vivo human radii. The cortical thickness values were validated by comparison with site-matched estimates derived from X-ray high-resolution peripheral quantitative computed tomography (Figure 3). Results showed an excellent agreement between both measurements (R^2 = 0.91, p < 0:05, RMSE = 0.19 mm). This preliminary study demonstrates the potential of bidirectional axial transmission for the *in vivo* assessment of bone strength-related factors, such as cortical thickness and porosity, which cannot easily be captured by X-ray densitometry techniques.

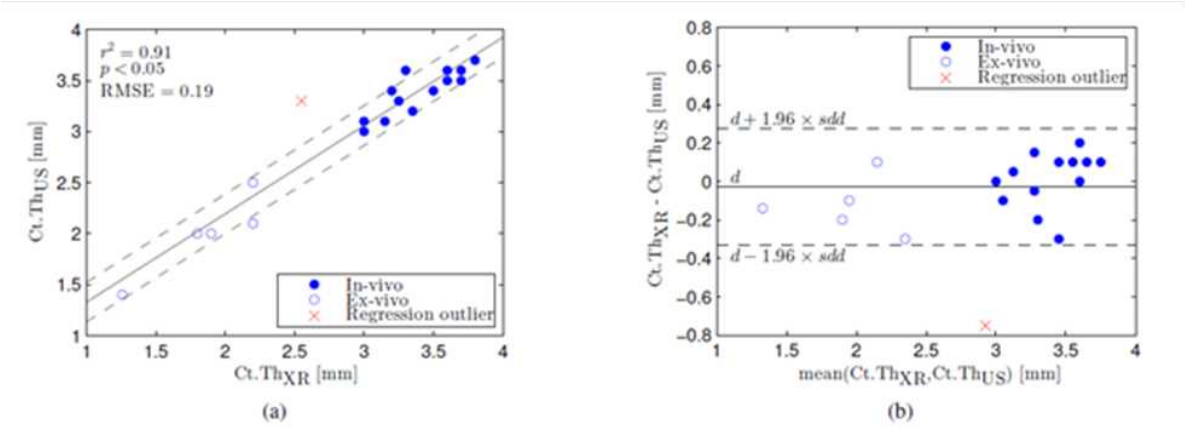

Figure 3: Linear regression (a) and Bland & Altman plot (b) between ultrasound-estimated cortical thickness and the reference value measured with HR-pQCT.

The *in vivo* multimodal response of human radius has been measured for the first time using GW-AT technique and a consistent estimation of cortical thickness has been performed by making use of a fully automatic inversion procedure. A good agreement has been found with reference values derived from site matched HR-pQCT and no bias has been noticed between US-based estimates and reference values. First estimates of cortical porosity have also been obtained from those measurements but they still have to be validated by comparison to micro-computed tomography measurements.

3 Resonant Ultrasonic Spectroscopy (RUS)

The insufficiently understood and still poorly documented variability of cortical bone elastic properties and their relation to composition and structure at different scales are currently investigated in our group using resonant ultrasound spectroscopy (RUS).

RUS is an accurate measurement method in which the full stiffness tensor of a material is assessed from the free resonant frequencies of a small sample. In RUS, the free vibration resonant frequencies of a material specimen of simple shape are measured and the stiffness coefficients adjusted until model-predicted frequencies match the experimental results. This inverse approach gives accurate elastic parameters because the resonant frequencies, which are entirely determined by elasticity, specimen dimensions and material mass density, can be measured with high accuracy. However, high viscoelastic damping (such as in cortical bone) causes the resonant peaks to overlap and therefore complicate the measurement of the resonant frequencies and the inverse identification of material properties. For that reason, RUS has been known to be fully applicable only to low damping materials. RUS has been used successfully for the first time on bone specimens by our group [10-13].

Spectrum measurement using shear transducers combined with dedicated signal processing is employed to retrieve the resonant frequencies despite overlapping [4-7]. Briefly, the specimens are placed with slight contact between a pair of shear ultrasonic transducers (V154RM, Panametrics, Waltham, MA), one acting as an emitter and the second one as a receiver (Figure 4). The frequency response is recorded using a vectorial network analyzer (Bode 100, Omicron Electronics GmbH, Klaus, Austria) and a broadband charge amplifier (HQA-15 M-10 T, Femto Messtechnik GmbH, Berlin, Germany). The frequency band of analysis is tuned for each specimen so as to contain the ~20–30 first mechanical resonant frequencies (typically 150–500 kHZ). Six successive measurements on each specimen are completed, with intermediate rotation of the specimens by a small angle between each measurement to vary the relative amplitudes of the excited resonant modes in order to maximize the number of detectable resonant frequencies.

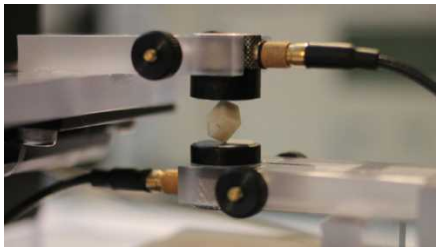

Figure 4: Photograph of the bone specimen inserted between the two transducers.

The elastic properties of the specimen are estimated by solving an inverse problem which consists of the comparison of the measured resonant frequencies to model predicted frequencies. The model is that of a vibrating elastic specimen with stress-free boundary

conditions, whose resonant frequencies are efficiently obtained with the Rayleigh–Ritz method. The geometry, mass density and stiffness of the specimen entirely determine the frequencies, so that the stiffness can be adjusted until the best match between the model and the experiment has been reached.

An issue of the inverse approach is that the measured frequencies are not easily paired with their predicted counterpart, leading to ambiguities in the definition of the cost function. In the past, this issue has been overcome through trial-and-error methods requiring the experimentalist to find the correct pairing, or through involved experimental methods measuring the shapes of the normal vibration modes in addition to their frequencies. A probabilistic (Bayesian) formulation of the inverse problem, tackling the problem of correctly pairing the measured and predicted frequencies, has been proposed. We showed, through this Bayesian formulation, that the inverse problem can be solved automatically and without requiring additions to the usual experimental setup. The pairing of measured and predicted frequencies is considered unknown, and the joint posterior probability distribution of pairing and stiffness is sampled using Markov chain Monte Carlo sampling. Introduction of prior information through Bayesian formulation reduces ambiguities. The method has been tested on calibration materials and then on a set of 51 tibia specimens [13]. The determined diagonal elastic constants (in GPa) (C_{11} = 20.3±0.6 ; C_{22}= 20.2± 0.6 ; C_{33} = 31.7±0.8 ; C_{44} = 6.38±0.02 ; C_{55} = 6.32±0.03 ; C_{66} = 4.80±0.02) were found in good agreement with concurrent reference sound velocity measurements performed in the principal directions of one specimen (C_{11}=21.6±0.7 ; C_{22} = 21.4±0.7 ; C_{33}= 31.3±1.0 ; C_{44} = 6.5±0.3 ; C_{55}=6.5±0.3 ; C_{66} = 4.8±0.2) [13].

Bone specimens harvested from the tibia of 19 donors were measured with RUS. The stiffness tensor with the assumption of transverse isotropy was retrieved for each specimen. We found that all the stiffness coefficients are correlated to each other (R^2 > 0.71) and that density variations determine a large part of stiffness variations (R^2 > 0.79 for all coefficients except for C_{13} for which R^2 = 0.52) (Figure 5).

Previous work on femur bone [14] supports the idea that the variations of elasticity with density are mostly explained by the variations of porosity, i.e., that the inter-specimen variations of the elasticity of the extracellular matrix are less important that inter-specimen variations of porosity [15].

We tested this idea on the data of [13]. Density values were converted to porosity values by assuming a fixed density for pores on the one hand and matrix on the other hand. This was done for the 51 tibia specimens. Then the porosity was fed into a micromechanical model [14] which predicts the apparent specimen stiffness (Figure 5, red curve). This model assumes that the elastic stiffness constants of the matrix are fixed and the values considered were (in GPa) C_{11} = 18.7, C_{33} = 31, C_{13} = 10.1, C_{44} = 6.98, C_{66} = 4.93. We found that the prediction of apparent stiffness with the physical micromechanical model is very satisfactory and is interestingly close to the linear fit of the experimental data.

These strong relationships between porosity and anisotropic apparent stiffness, which was already pointed out in [13] and which is confirmed in the above-presented results, are of major importance for the processing of in vivo measurements with AT. These results pave the way for in vivo determination of porosity using apparent stiffness data.

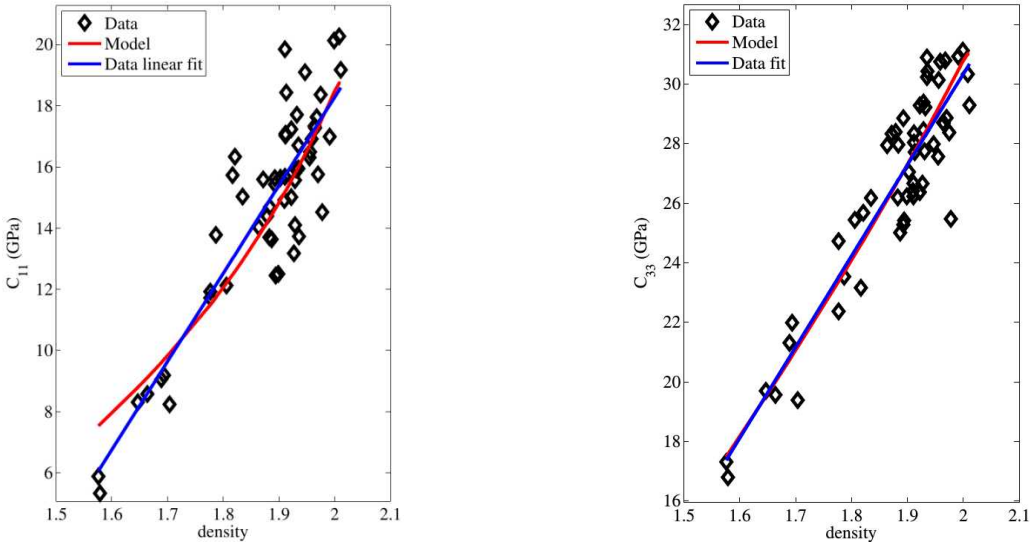

Figure 5. RUS data from human tibia specimens. Stiffness coefficients C11 (left) and C33 (right) versus density. Prediction of the micromechanical model (red) and linear regression (blue).

4 Conclusion

The newly developed QUS methods, namely AT and RUS, are tailored to address 1/ the limitations of conventional mechanical testing in the assessment of the anisotropic elastic stiffness of cortical bone specimens, and 2/ the limitations of DXA approaches with the goal to yield improved performance both for risk assessment and in the context of monitoring treatment effects.

Acknowledgments

This research was supported by the Fondation pour la Recherche Médicale through project number FRM DBS201311228444, by the Agence Nationale pour la Recherche under Grant No. ANR-13-BS09-0006-01 and Grant No. ANR-14-CE35-0030-01 and by the spin-off company AZALEE.

References

[1] E. S. Siris, Y.-T. Chen, T. A. Abbott, E. Barrett-Connor, P. D. Miller, L. E. Wehren, and M. L. Berger, “Bone mineral density thresholds for pharmacological intervention to prevent fractures.” Arch Intern Med **164**, 1108–1112 (2004).

[2] R. Zebaze, A. Ghasem-Zadeh, A. Bohte, S. Iuliano-Burns, M. Mirams, R. Ian Price, E. J. Mackie, E. Seeman., “Intracortical remodelling and porosity in the distal radius and post-mortem femurs of women: a cross-sectional study,” The Lancet **375**, 1729 – 1736 (2010).

[3] P. Moilanen, “Ultrasonic guided waves in bone,” IEEE Trans. Ultrason. Ferroelectr. Freq. Control **55**, 1277-1286 (2008).

[4] J. G. Minonzio, M. Talmant, P. Laugier, “Guided wave phase velocity measurement using multi-emitter and multi-receiver arrays in the axial transmission configuration,” J Acoust Soc Am **127**, 2913-2919 (2010).

[5] J. G. Minonzio, J. Foiret, M. Talmant, and P. Laugier, "Impact of attenuation on guided mode wavenumber measurement in axial transmission on bone mimicking plates," J. Acoust. Soc. Am. **130**, 3574–3582 (2011).

[6] L. Moreau, J. G. Minonzio, J. Foiret, E. Bossy, M. Talmant, and P. Laugier, "Accurate measurement of guided modes in a plate using a bidirectional approach," J. Acoust. Soc. Am. **135**, EL15-EL21 (2014).

[7] J. Foiret, J. G. Minonzio, C. Chappard, M. Talmant, and P. Laugier, "Combined estimation of thickness and velocities using ultrasound guided waves: A Pioneering study on in vitro cortical bone samples," IEEE Trans. Ultrason. Ferroelectr. Freq. Control. **61**, 1478-1488 (2014).

[8] J. G. Minonzio, J. Foiret, P. Moilanen, J. Pirhonen, Z. Zhao, M. Talmant, J. Timonen and P. Laugier; “A free plate model can predict guided modes propagating in tubular bone-mimicking phantoms,” J. Acoust. Soc. Am. **137**, EL98-EL104 (2015).

[9] Q. Vallet, N. Bochud, C. Chappard, J.-G. Minonzio, and P. Laugier, "In-vivo cortical bone characterization using guided waves measured by axial transmission," IEEE Trans. Ultrason. Ferroelectr. Freq. Control. (accepted)

[10] S. Bernard, Q. Grimal and P. Laugier, Accurate measurement of corical bone elasticity tensor with resonant ultrasound spectroscopy;” J Mech Behav Biomed Mater **18**, 12–19 (2013).

[11] S. Bernard, Q. Grimal and P. Laugier, “Resonant ultrasound spectroscopy for viscoelastic characterization of anisotropic attenuative solid materials,” J Acoust Soc Am **135**, 2601–2613 (2014).

[12] S. Bernard, G. Marrelec, P. Laugier and Q. Grimal, “Bayesian normal modes identification and estimation of elastic coefficients in resonant ultrasound spectroscopy,” Inverse Problem **31**, 65010 (2015).

[13] S. Bernard, J. Schneider, P. Varga, P. Laugier, K. Raum and Q. Grimal, “Elasticity–density and viscoelasticity–density relationships at the tibia mid-diaphysis assessed from resonant ultrasound spectroscopy measurements,” Biomechanics and modeling in mechanobiology **15**, 97-109 (2016).

[14] M. Granke, Q. Grimal, A. Saïed, P. Nauleau, F. Peyrin, P. Laugier, “Change in porosity is the major determinant of the variation of cortical bone elasticity at the millimeter scale in aged women,” Bone **49**, 1020-1026 (2011).

[15] W. J. Parnell, M. B. Vu, Q. Grimal, S. Naili, “Analytical methods to determine the effective mesoscopic and macroscopic elastic properties of cortical bone,” Biomech Model Mechanobiol **11**, 883-901 (2012).