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Welcome to the ESUCB2007

On behalf of the French-German network on ultrasound assessment of bone strength from the tissue level to the organ level I welcome you to the 2nd European Symposium on Ultrasonic Characterization of Bone, ESUCB2007. This 2-day symposium is open to all researchers interested in ultrasonic characterization of bone. Ample time between and after the scientific sessions will be reserved to promote informal discussions among scientists and young investigators. The meeting will address advances in basic science (propagation models, backscattering, numerical simulations, guided waves), instrumentation and signal analysis (acoustic microscopy, transverse and axial transmission, imaging techniques) and applications (small animals, osteoporosis and other diseases).

The first symposium was held in Paris at University Pierre et Marie Curie, March 6-7, 2006. The meeting was attended by researchers and students from 10 countries. Fifty scientific presentations pointed out many innovative research directions, such as theoretical and computational tissue and wave propagation models, novel instrumentation, as well as micro- and macroscopic acoustic measurements. Moreover, the meeting has resulted in new collaborations between groups from Europe, Japan, and North America.

This year 30 oral and 10 poster presentations from 11 countries will again present the state-of-the-art on ultrasonic assessment of bone.

The ESUCB2007 will be held in the historical Medical Faculty at Martin Luther University of Halle-Wittenberg, situated in the center of the city of Halle. The local organizing committee wishes all participants a fruitful congress and an enjoyable stay in Halle.
ESUCB2007 Scientific Committee

Local Organizer
Kay Raum (Germany)

International Scientific Committee
Pascal Laugier (France)
Frédéric Padilla (France)
Claus-C. Glüer (Germany)
Emmanuel Bossy (France)
Frederic Patat (France)
Keith A. Wear (USA)
Jonathan J. Kaufman (USA)
Petro Moilanen (Finland)
Mami Matsukawa (Japan)

Scientific Program Information

Plenary Speaker
Devakar Epari: “Mechanobiology and Musculoskeletal Regeneration”
Mechanobiology Group, Musculoskeletal Research Center Berlin, Charité, Germany

Oral Presentations
Contributed Oral Presentations have a gross duration of 20 min that breaks down to 15 min for presentation and 5 min for discussion. One major intention of the meeting is to promote discussions among the conference participants. Therefore the presenting authors are asked to strictly stay within the 15-min presentation time.

Plenary and Keynote Presentations have a gross duration of 30 min that breaks down to 25 min for presentation and 5 min for discussion.

Poster Presentations
Poster sessions are scheduled during the afternoon coffee breaks (50 min). Poster panels are 180 cm in height and 100 cm wide. All posters can be mounted during the entire time of the conference (Thursday morning until Friday afternoon). In order to ensure a maximum of interaction, the presenting authors are requested to discuss their work during their scheduled presentation time, but can use the poster panels for discussions at any other time.

Social Program Information

Thursday - Barbeque
After the scientific sessions on Thursday we invite you to meet with your friends and colleagues in one of the most beautiful beer gardens of the city.

Enjoy a typical German barbeque with either local wine from the Saale-Unstrut area or with world famous German beer, while you will have a spectacular view of the ancient castle Giebichenstein and the river Saale from the top of a porphyry cliff.
Friday – Get Together Reception

We encourage you to stay with us after the scientific sessions on Friday. Participants of the ESUCB2007 and members of the DGOOC Network on Musculoskeletal Biomechanics (MSB-NET) often look at bone from a different point of view. The evening will provide a unique atmosphere to get new ideas or even establish new collaborations. And it is free for all participants.

Network Meetings

French-German Network "Ultrasound Assessment of Bone Strength from the Tissue Level to the Organ Level"

The network meeting is scheduled for Saturday morning and is open to ESUCB2007 participants. After the scientific presentations the current research status will be summarized, problems as well as future research directions will be discussed. One of the major goals of the meeting is to identify potential new network partners for future concerted research activities.

DGOOC Network on Musculoskeletal Biomechanics (MSB-NET)

The network meeting is scheduled for Saturday morning and is open to the participating members of the MSB-NET laboratories. However, researchers interested in the field are welcome to attend the meeting. The meeting language will be German.

Internet Access

A WLAN access will be available in the lecture hall and in the foyer in front of the lecture hall. You will find the access information in your information package. If you don’t bring your own WLAN laptop, you can use one of our provided PC’s.
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http://www.q-bam.de

Martin Luther University of Halle-Wittenberg
http://www.uni-halle.de
Program Timetable

Thursday, July 19th, 2007:

9:00 am  Opening
9:15 am  Welcome Note: French-German Network
9:30 am  Plenary Talk
10:00 am Coffee Break
10:30 am Session I - Ultrasound Propagation Models
         noon Lunch Break
2:00 pm  Session II - Characterization of Cancellous Bone: Simulation Studies
3:20 pm  Coffee Break / Poster Session
4:10 pm  Session III - Small Animal Models
5:50 pm  Lab Visit
7:00 pm  Barbeque - Bergschenke

Friday, July 20th, 2007:

9:00 am  Keynote Lecture
9:30 am  Session IV - Novel Instrumentation
10:30 am Coffee Break
11:00 am Session V - Numerical & Experimental Assessment of Bone
         noon Lunch Break
2:00 pm  Session VI - Scanning Acoustic Microscopy
3:40 pm  Coffee Break / Poster Session
4:30 pm  Session VII - Applications
5:50 pm  Lab Visit
7:00 pm  Get Together Reception - (ESUCB2007 participants & MSB-NET)

Saturday, July 21st, 2007:

9:00 - noon Meeting of the French-German Network "Ultrasound Assessment of Bone Strength from the Tissue Level to the Organ Level" (open to ESUCB2007 participants)
9:00 - noon Meeting of the DGOOC Network on Musculoskeletal Biomechanics (MSB-NET)
Scientific Program
Thursday, July 19th, 2007:

9:00 am Opening
Kay Raum

9:15 am Welcome Note: French-German Network
Pascal Laugier

9:30 am Plenary Talk
Mechanobiology and Musculoskeletal Regeneration
D. Epari

10:00 am Coffee Break

10:30 am Session I - Ultrasound Propagation Models
Chair: Emmanuel Bossy

10:30 am 3D simulation study of the properties of slow and fast wave modes versus bone volume fraction and structural anisotropy in human trabecular bone
G. Haïat, F. Padilla, P. Laugier

10:50 am Dispersion of guided waves in bone-mimicking plates with microstructural effects
M. Vavva, V. Protopappas, L. Gergidis, A. Charalambopoulos, D. Fotiadis, D. Polyzos

11:10 am Stochastic simulation of the axial transmission technique based on a multi-scale model of bone material properties

11:30 am Modeling of velocity dispersion in trabecular bone: effect of multiple scattering and of viscous absorption
G. Haïat, F. Padilla, S. Lonne, A. Lhémery, P. Laugier, S. Naili

noon Lunch Break

2:00 pm Session II - Characterization of Cancellous Bone: Simulation Studies
Chair: Jonathan J. Kaufman

2:00 pm A computationally efficient finite element model for ultrasound propagation through trabecular bone
J. Vanderoost, L. Goossens, S.V.N. Jaecques, S. Boonen, J. D'Hooge, W. Lauriks, G. Van der Perre

2:20 pm Is Biot theory adequate to model sound wave propagation in cancellous bone?
N. Sebaa, M. Naas, L. Goossens, Z. Fellah, G. Van der Perre, S. Boonen, W. Lauriks, C. Depollier

2:40 pm Two-phase macroscopic modelling of wave propagation in cancellous bone
M. Pakula, F. Padilla, M. Kaczmarek, P. Laugier

3:00 pm The contribution of density fluctuations in ultrasound scattering in cancellous bone
A. Elsariti, T. Evans
### Thursday, July 19th, 2007:

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<th>Time</th>
<th>Session</th>
<th>Title</th>
<th>Authors</th>
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<tr>
<td>3:20 pm</td>
<td>Coffee Break / Poster Session</td>
<td>Development of a simplified trabecular model and ultrasound simulation in cancellous bone</td>
<td>A. Hosokawa</td>
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<tr>
<td>3:20 pm</td>
<td>Coffee Break / Poster Session</td>
<td>Simulation of the US propagation in 3D cortical bone porous networks: is the apparent velocity dependant on signal processing?</td>
<td>M. Talmant, Q. Grimal, C. Baron, P. Laugier</td>
</tr>
<tr>
<td>3:20 pm</td>
<td>Coffee Break / Poster Session</td>
<td>Finite element model of the lamellar osteonal structure</td>
<td>A. Gerisch, T. Hofmann, K. Raum</td>
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<tr>
<td>3:20 pm</td>
<td>Coffee Break / Poster Session</td>
<td>Modelling of ultrasonic wave propagation through trabecular bone using a finite element model</td>
<td>L. Goossens, J. Vanderooost, S.V.N. Jacques, S. Boonen, J. D'Hooge, W. Lauriks, G. Van der Perre</td>
</tr>
<tr>
<td>3:20 pm</td>
<td>Coffee Break / Poster Session</td>
<td>How does a gradient of material properties due to endosteal resorption affect wave propagation in the axial transmission technique?</td>
<td>G. Haïat, S. Naili, Q. Grimal, C. Desceliers, C. Soize</td>
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### 4:10 pm  Session III - Small Animal Models

**Chair:** Mami Matsukawa

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<th>Title</th>
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<td>4:10 pm</td>
<td>Session III - Small Animal Models</td>
<td>Bone elastic phenotyping of small animal models</td>
<td>K. Raum, T. Hofmann, I. Leguerney, A. Saïed, F. Peyrin, L. Vico, P. Laugier</td>
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<tr>
<td>4:30 pm</td>
<td>Session III - Small Animal Models</td>
<td>Assessment of bone structure and elasticity under the influence of leptin deficiency in C57BL/6 mice using high resolution scanning acoustic microscopy</td>
<td>J. Marr, K. Ruschke, K. Raum</td>
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<tr>
<td>4:50 pm</td>
<td>Session III - Small Animal Models</td>
<td>Ultrasonic mechanical characterization of bone formed at the interface of two different grafting materials</td>
<td>H. Schneider, D. Busenlechner, G. Fugger, M. Schulz, S. Tangl, K. Raum</td>
</tr>
<tr>
<td>5:30 pm</td>
<td>Session III - Small Animal Models</td>
<td>Monitoring of elastic properties during bone regeneration</td>
<td>M. Schulz, J. Brandt, K. Brehme, K. Raum</td>
</tr>
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### 5:50 pm  Lab Visit

### 7:00 pm  Barbeque - Bergschenke
Friday, July 20th, 2007:

9:00 am  Keynote Lecture
First data of hip fracture discrimination using the Femur Ultrasound Scanner (FEMUS)
R. Barkmann, S. Dencks, P. Laugier, F. Padilla, C.C. Glüer

9:30 am  Session IV - Novel Instrumentation
Chair: Guillaume Haïat

9:30 am  Separation and characterization of superimposed waves at the proximal femur
S. Dencks, R. Barkmann, F. Padilla, P. Laugier, G. Schmitz, C.C. Glüer

9:50 am  Effects of crystallites orientation on the longitudinal wave velocities in bovine cortical bone
M. Matsukawa, Y. Yaoi, H. Mizukawa, Y. Yamato, K. Yamamoto, K. Yamazaki, A. Nagano

10:10 am  A new system for ultrasonic assessment of the forearm
J. Kaufman, G. Luo, R.S. Siffert

10:30 am  Coffee Break

11:00 am  Session V - Numerical & Experimental Assessment of Bone
Chair: Reinhard Barkmann

11:00 am  Attenuation through trabecular bone: a comparison between simulated values obtained using finite-difference time-domain 3D simulations and experimental values measured on the same samples
E. Bossy, P. Laugier, F. Padilla

11:20 am  Experimental study on the fast wave velocity and structural anisotropy in the cancellous bone
K. Mizuno, M. Matsukawa, T. Otani, M. Takada, I. Mano, T. Tsujimoto

11:40 am  SAM data is used to derive the continuum level elastic properties of cortical bone
Q. Grimal, K. Raum, A. Gerisch, P. Laugier

noon  Lunch Break

2:00 pm  Session VI - Scanning Acoustic Microscopy
Chair: Pascal Laugier

2:00 pm  Characterization of bone samples by ultrasound holography at about 100 MHz and general aspects concerning the mechanical properties of porous anisotropic inhomogeneous materials including bone replacements
H. Hartmann, U. Cobet, W. Grill

2:20 pm  Measurement of wave velocity in bovine cortical bone by μ-Brillouin scattering method
M. Sakamoto, M. Matsukawa, Y. Yamato, K. Yamamoto, K. Yamazaki, A. Nagano

2:40 pm  The elastic and structural properties of human femoral cortical bone at tissue level using high resolution acoustic measurements
S. Lakshmanan, A. Bodí, K. Raum

3:00 pm  Assessment of bone remodeling using high resolution acoustic microscopy
F. Rupin, A. Saïed, K. Raum, L. Vico, V. David, P. Laugier

3:20 pm  Confocal and non-confocal acoustic transmission microscopy of tissue-engineered cartilage constructs
M. von Buttlar, E. Twardowski, W. Grill
**Friday, July 20th, 2007:**

**3:40 pm  Coffee Break / Poster Session**

P6  A combination of mechanical and ultrasonic methods is used to assess the anisotropic elasticity and strength of in vitro cortical bone samples
Q. Grimal, S. Haupert, D. Mitton, L. Vastel, B. Raphaël, P. Laugier

P7  Precision control and seasonal variations in quantitative ultrasound of calcaneus, tibia and radius

P8  Assessment of skeletal age in children and adolescents using radius sonography in comparison to standardised radiography

P9  Short-term reproducibility of in vivo guided wave measurements
V. Kilappa, P. Moilanen, J. Timonen, S. Cheng

P10  Elastic changes in cartilage and subchondral bone due to primary arthrosis
S. Leicht, K. Raum

**4:30 pm  Session VII - Applications**

Chair: Frédéric Padilla

4:30 pm  Ultrasonic assessment of trabecular structure in proximal tibia
A. Tatarinov, P. Saparin

4:50 pm  Guided wave ultrasound in the assessment of tibial bone in elderly females
M. Määttä, P. Moilanen, P. Nicholson, J. Timonen, S. Cheng, T. Jämsä

5:10 pm  Beneficial effects of Tai Chi on women’s skeletal status assessed by quantitative ultrasound (QUS) at the hand phalanges: the cross-sectional and one-year follow-up study
M. Bolanowski, W. Pluskiewicz

5:30 pm  Quantitative ultrasound, a reliable measurement for bone changes assessment during two months bedrest: gender, control, exercise groups comparison
M. Defontaine, M.N. Eddin, D. Felsenberg, J. Rittweger

5:50 pm  Lab Visit

7:00 pm  Get Together Reception - (ESUCB2007 participants & MSB-NET)
Saturday, July 21st, 2007:

9:00 – noon  Meeting of the French-German Network "Ultrasound Assessment of Bone Strength from the Tissue Level to the Organ Level" (open to ESUCB2007 participants)

9:00 - noon  Meeting of the DGOOC Network on Musculoskeletal Biomechanics (MSB-NET)
Abstracts
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Mechanobiology and Musculoskeletal Regeneration

Devakar Epari

Mechanobiology Group, Musculoskeletal Research Center Berlin, Charité, Germany

e-mail: devakara.epari@charite.de
Quantitative ultrasonic techniques are clinically used to estimate bone mechanical properties. Two longitudinal wave modes may be observed in trabecular bone according to bone volume fraction (BV/TV), structural anisotropy and direction of propagation relatively to the direction of main trabecular alignment (MTA). Both wave modes are predicted by the Biot’s theory. In some circumstances, the overlapping of both waves in the time domain is such that only a single pulse seems to be transmitted. Analytical predictions are unable to establish reliable relationships between structural anisotropy, BV/TV and the behavior of both wave modes. The aim of this work was to investigate the conditions on bone volume fraction and structural anisotropy under which the fast and slow waves can be separated. Our approach uses 3D finite-difference time-domain simulations coupled with 34 real human femoral trabecular microstructures. The structural anisotropy (MTA, degree of anisotropy (DA)) of each sample was determined using a dedicated 3D image processing method. The influence of BV/TV on both waves was studied using an image processing algorithm modifying all initial 3D microstructures. A heuristic signal processing method was developed in the frequency domain aiming at determining when both wave modes overlap in time and when they can be distinguished. Both wave modes generally overlap in time when the MTA is perpendicular to the direction of propagation. When MTA is parallel to the direction of propagation, both separated wave modes can be observed for samples with high values of DA and BV/TV. Moreover, our data show that the BV/TV under which non overlapping wave modes are observed is negatively correlated with the DA ($r^2=0.53$). Our results show that simultaneous conditions on structural anisotropy and BV/TV have to be fulfilled to be able to separate both wave modes.
The use of guided waves has drawn significant interest in the ultrasonic characterization of bone. However, the microstructure of bone plays an important role in guided wave propagation since it induces both geometrical and material dispersion. The objective of the present work is to determine the velocity dispersion curves of the guided modes that propagate in isotropic bone-mimicking plates with microstructural effects. The analysis is performed in the context of a simple gradient elastic theory, which can be considered as the simplest case of the general elastic theory proposed by Mindlin. We assume that the plate’s faces are free from stresses and double stresses. The characteristic frequency equations corresponding to symmetric and antisymmetric modes are obtained and solved numerically. By comparing with the theoretical dispersion curves derived from an isotropic plate in the classical elastic case, we show that microstructure has a significant effect on mode dispersion, and therefore should be taken into consideration in experimental and simulation studies.

This work was supported by research grant #PENED 2003“ 03ED140.
Stochastic simulation of the axial transmission technique based on a multi-scale model of bone material properties

Quentin Grimal a, Kay Raum b, Christophe Desceliers c, Christian Soize c, Guillaume Haïat d, Salah Naili d, Maryline Talmant a, Pascal Laugier a

a Laboratoire d’Imagerie Paramétrique, CNRS, UPMC-Paris 6, France
b Q-BAM Group, Martin Luther University of Halle, Germany
c LAM, UMLV, France
d B2OA, Univ. Paris 12, France
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The identification of the geometrical and material properties of bone structures in QUS could be performed within the framework of statistical inverse problems. This would enable to use more information contained in the acoustic signals compared to what is used currently and would also provide robust inversion algorithms. Setting a statistical inversion framework for QUS is a challenging task. The first step is to build a stochastic model of the QUS acoustic problem. Such model includes (i) a representation of the bone material properties and geometry using probability density functions (pdf); (ii) the formulation and simulation of the acoustic propagation problem. In this paper, we investigate the direct stochastic problem for the axial transmission technique. The bone thickness, porosity and impedance of the bone matrix are taken as random parameters. The pdf for the thickness is constructed based on in vitro scanner data. The pdf for porosity and impedance are constructed based on spatial distributions of impedance obtained with acoustic microscopy (50 MHz, 23 micron resolution). The elastic properties of bone required for the simulation are derived from porosity and impedance values upon using a finite element homogenization technique. The direct stochastic problem is solved with a Monte Carlo simulation. For each realization of the random parameters, the wave propagation problem is solved with a finite difference scheme. The results show the dependence of the first arriving signal (FAS) on the model parameters. The comparison of the simulated FAS dispersion to the dispersion of in vitro measurements is used to test and discuss the developed stochastic model of the axial transmission technique.
Modeling of velocity dispersion in trabecular bone: effect of multiple scattering and of viscous absorption

Guillaume Haïat a, Frédéric Padilla b, Sébastien Lonne c, Alain Lhémery c, Pascal Laugier b, Salah Naili a

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b Laboratoire d’Imagerie Paramétrique, CNRS, UPMC-Paris 6, France
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Bone quantitative ultrasound (QUS) techniques play an increasing role in fracture risk prediction. However, the physical interaction between trabecular bone (TB) and ultrasound remains unclear. In particular, the origin of negative velocity dispersion measured in TB is still a matter of debate. The aim of this paper is to propose a model predicting the frequency dependence of phase velocity. This study also aims at estimating the effect of multiple scattering phenomena by comparing the results with another model assuming independent scattering. A 2D homogenization model, initially developed in the context of composite materials, and accounting for multiple scattering effects is employed to calculate the dependence of phase velocity on frequency, bone volume fraction (BV/TV) and trabecular thickness (TbTh). A simple biphasic description of TB in which the trabeculae are assumed to be identical and parallel cylinders is considered. Moreover, this model takes into account multiple scattering phenomena as well as viscosity of the marrow. In addition, the results obtained with a model assuming independent scattering which uses the Faran model was compared to the multiple scattering model. The values of velocity dispersion at 500 kHz and its dependence to bone properties predicted with the multiple scattering model are in good agreement with experimental results obtained by Wear in 2D phantoms mimicking TB. The modified Faran model is also in good agreement for low values of TbTh and BV/TV. Moreover, for typical TB parameters, the multiple scattering models predict negative velocity dispersion, although the values of velocity dispersion are slightly underestimated compared to experimental results. Moreover, including viscous properties of the marrow weakly modifies the results. This simple geometrical description of TB allows to derive a physical explanation for negative values of dispersion and to assess the influence of multiple scattering phenomena.
A computationally efficient finite element model for ultrasound propagation through trabecular bone

Jef Vanderoost a, Liesbet Goossens a, Siegfried V.N. Jaecques b, Steven Boonen c, Jan D’Hooge d, Walter Lauriks e, Georges Van der Perre f

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c K.U.Leuven, Centre for Metabolic Bone Disease, Belgium
d K.U.Leuven, Imaging and Cardiovascular Dynamics, Belgium
e K.U.Leuven, Acoustics and Thermal Physics, Belgium
f K.U.Leuven, Biomechanics and Engineering Design, Belgium

Metabolic bone diseases such as osteoporosis diminish the strength of bones by affecting the structure of porous trabecular bone. By gaining insight into the propagation of ultrasound waves through trabecular bone, new clinical analysis tools can be developed to detect structural changes. The complex microstructure of trabecular bone is widely investigated through the use of FE-models based on µCT-scans (voxel-based models). We propose an alternative simplified FE-model based on a structural skeleton (skeleton-based model). The bone microstructure is reduced to its mechanical essence, composed of rods and plates or trabecula. This model is used to estimate the stiffness or strength of trabecular bone as well as to simulate the propagation of ultrasound waves. Since only the solid bone phase is modeled, only the propagation of the fast wave is calculated. Through skeletonisation, voxel-based models of trabecular bone samples are simplified to complex structures of rods and plates. An optimized procedure with special care for boundary effects is used. By relating the skeleton to the original structure each rod and plate is characterized with local structural parameters (thickness, area moments of inertia). These characteristics are implemented into a FE-model consisting of beam and shell-elements. To validate the equivalence of both models in modeling US propagation, the SOS of 17 samples of calcanean trabecular bone (BIOMED I project) is calculated using the voxel-based and the skeleton-based finite element model. Each cubic sample is tested in the three principal directions, adding up to 51 equivalent SOS values for each model. Preliminary results show a linear correlation ($R^2=0.7$) between the SOS-values of both models with an almost 500-fold reduction in required CPU time. After further optimization and validation tests the skeleton-model will be used for parametric analysis of the effect of the bone structure on the propagation of ultrasound waves.
Is Biot theory adequate to model sound wave propagation in cancellous bone?

Naima Sebaa, Mouna Naas, Liesbet Goossens, Zine Fellah, George Van der Perre, Steven Boonen, Walter Lauriks, Claude Depollier

K.U.Leuven, Belgium

*e-mail: naima.sebaa@fys.kuleuven.be*

During the last decade, to improve the ultrasound techniques for the diagnosis of osteoporosis, several models were developed to facilitate the understanding of the sound propagation through cancellous bone. More specifically, the motivations of these investigations were the dependence of ultrasound velocity and their attenuation upon physical parameters of the structure of bones such as density, porosity, elastic moduli. The Biot's theory of poroelasticity is one of the most realistic model to describe the sound propagation in porous media. It takes into account different couplings between fluid and solid structure: i) inertial coupling, modelled by the dynamical tortuosity of the medium, ii) viscous coupling (due to the viscosity of the fluid) and iii) elastic coupling (due to the reciprocity principle of fluid-structure interactions). Its most important success is the prediction of two compressional waves which travel in the porous medium at different velocities. The fast wave is the motion where the fluid filling the pore space and the solid matrix are locked together through viscous or/and inertial couplings: fluid and solid move in phase. Conversely the slow wave describes the relative motion between the fluid and the matrix. In porous media, attenuation results from different processes: i) relative motion between viscous fluid and solid, ii) scattering by inhomogeneities and interfaces of porous medium sample. The first process appears mainly in the damping of the slow wave. In the second process, because of the conversion modes process, an additional attenuation results when the wave is scattered by an interface. This is due to the fact that the attenuation of the two kinds of waves are quite different: roughly speaking, the attenuation is governed by the slow wave. In this communication we show that some confusions may appear in conclusions of authors about Biot theory. While in vitro studies on slices of cancellous bone confirm that the Biot's theory predictions are in good agreement with the experimental results, it does not accurately predict attenuation and the absence of slow wave in vivo situations. Simulations of sound waves propagation based on the time domain model reveal that a sealed surfaces sample of porous media behaves almost as a nonporous solid transmitting only ordinary compressional wave. This confirms the importance of interface hydraulic condition on the generation of slow wave: in such media, relative motions between an incompressible fluid and the solid structure are impossible. About the excess of attenuation of fast wave, we show that the scattering process i) increases the path length of waves and ii) over-damps the fast wave through the conversion modes process. We present results of numerical simulations and experimental results which show that Biot theory is an appropriate model to describe the sound wave propagation in cancellous bone.
Two-phase macroscopic modelling of wave propagation in cancellous bone

Michal Pakula a, Frederic Padilla a, Mariusz Kaczmarek b, Pascal Laugier a

a Laboratoire d’Imagerie Parametrique, CNRS, UPMC-Paris 6, France
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The study is focused on modelling of wave propagation in cancellous bones using two-phase macroscopic Biot’s theory. Almost all required input mechanical and structural parameters for 31 pure trabecular bone specimens (human femurs) were measured individually and then were used to calculate attenuation coefficient and phase velocity. Then frequency dependent wave parameters predicted by the model were compared with the results of ultrasonic tests performed on the same specimens. To compare the predictions to measurements done using an insertion method, within our theoretical framework, additional interactions of the plane harmonic wave with the slab of cancellous bone treated as a saturated porous material was considered. The reflection coefficient and transmission coefficients (for fast and slow wave) at the normal wave incidence were calculated and their contribution in the global attenuation was evaluated. The most important finding is that the values predicted for the wave attenuation accordingly to the Biot’s model (without corrections for reflection at the boundaries) were two orders of magnitude lower than measured values. Considered wave interaction exhibited significant contribution of the fluid/bone and bone/fluid boundaries on the global attenuation coefficients. The corrected values of attenuation coefficient are of the same order of magnitude compared to measured values. Moreover, the theoretical results are also consistent with the higher attenuation of fast wave (compared to that of the slow wave) that is usually observed in experiments. Considering the amplitude ratio of simulated time domain signals of both longitudinal waves (accordingly to the Biot’s model with boundary corrections), one can conclude that they are of the same order of magnitude compared to measured values. However, an analysis in the frequency domain shows that the frequency content of the simulated pulses of the fast and slow wave differs from that observed in the experiments. The most probable source of discrepancy in the frequency dependence of attenuation and phase velocity comes from the basic assumption of Biot’s theory, which was introduced for wavelengths much higher than the typical dimension of heterogeneities and therefore does not account for important physical interactions such as scattering.
The contribution of density fluctuations in ultrasound scattering in cancellous bone

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An understanding of the interaction between acoustic waves and cancellous bone is needed in order to realize the full clinical potential of ultrasonic bone measurements. Scattering is likely to be of central importance but has received little attention to date. Few theoretical approaches have been described to explain scattering of ultrasound from bone.

As Nicholson et al (2000) mentioned that one of their limitations was to account for density fluctuations. In this study, a scattering model based on velocity and density fluctuations in a binary mixture (marrow fat and cortical matrix) was used to estimate the ultrasonic attenuation in cancellous bone as a function of volume fraction. Predicted attenuation and backscatter coefficient were obtained for a range of porosities and scatterer size. At 600 kHz and for different scatterer size the effect of velocity and density fluctuations in the predicted attenuation was approximately 60 % higher than velocity fluctuations.
Development of a simplified trabecular model and ultrasound simulation in cancellous bone

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The objective of this study is to develop a three-dimensional (3D) cancellous bone model without using the real bone data for numerical simulations of ultrasound propagation. Trabecular frame in cancellous bone has numerous pore spaces with various sizes and shapes. Assuming that the complicated pore structure was made up of more numerous spherical pores, in this study, the trabecular frame was simplified to easily make a cancellous bone model. In the simplified model, spherical pores with normally distributed sizes were more regularly arranged than in the real bone. The trabecular-oriented model was made by adjusting the structural parameter values, and the fast and slow wave propagations through cancellous bone, which can be experimentally observed in the direction parallel to the trabecular orientation, were numerically simulated in 3D using a viscoelastic finite-difference time-domain (FDTD) method. The changes in the propagation properties, the wave amplitude and speed, with model reconstructed from a 3D x-ray micro-computed tomography (µCT) image of bovine femoral cancellous bone. Moreover, the fast and slow wave simulations using the realistic µCT model were also performed to compare with the simulated results obtained using the simplified model. The validity of the simplified model was investigated with these comparisons.
Simulation of the US propagation in 3D cortical bone porous networks: is the apparent velocity dependant on signal processing?

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The propagation of ultrasonic waves in cortical bone involves interactions with a porous network which may result in dispersion and attenuation. As a consequence, the procedure used to retrieve the wave velocity should be considered carefully. The purpose of the present work is to elucidate the dependence on signal processing of the apparent velocity in cortical bone in the range [500 kHz – 10 MHz]. A time domain finite difference code is used to compute the US propagation of longitudinal and shear waves, transverse to, and along the bone axis, in typical cortical bone porous networks. The 3D networks, which were obtained with micro-CT synchrotron radiation in a previous study, essentially consist of resorption cavities and Haversian pores. We will present the apparent group velocity and phase velocity calculated for each simulated signal with various classical techniques.
A recent study of single osteon lamellae by 900-MHz scanning acoustic microscopy (SAM), hyperspectral Raman imaging and nanoindentation (Hofmann et al., 2006) supported the hypothesis that the majority of the lamellae in a single secondary osteon have no great differences in the degree of mineralization or elasticity, but distinct alternating orientations. This causes (i) a characteristic bimodal lamellar pattern in high resolution acoustic images and (ii) the elastic tissue anisotropy of the tissue compound. The lamellar pattern is believed to represent the anisotropic properties of an asymmetric twisted plywood structure composed of transverse isotropic mineralized collagen fibrils.

In order to confirm this hypothesis, a finite element model was developed, in which the osteon is considered to consist of a central Haversian canal filled with an incompressible fluid and surrounding sets of lamellar units. Each lamellar unit was further subdivided in five sublayers, whereas the orientation of the symmetry axis between adjacent sublayers was shifted clockwise by an angle of 30 degrees. A sublayer consists of one to ten layers of parallel oriented mineralized collagen fibrils (thickness: 0.2 μm, transverse isotropic elasticity: $E_1 = 24$ GPa, $E_2 = 27$ GPa, $G_{12} = 9$ GPa).

Results: A variation of the sublayer thicknesses results in either isotropic or anisotropic tissue compound properties. By changing the individual layer thicknesses various degrees of anisotropy could be produced. The best agreement with the lamellar pattern obtained in 900-MHz SAM images as well as with the anisotropic elastic coefficients measured at the tissue level (50-MHz ultrasound) was obtained be choosing an asymmetric lamellar unit.

Reference:

Modelling of ultrasonic wave propagation through trabecular bone using a finite element model

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Osteoporosis is a degenerative bone disease associated with biochemical and hormonal changes in the ageing body, resulting in a net decrease in bone mass. Early detection of this disease is very important for adequate treatment and fracture prevention. So far the DEXA technique which determines bone mineral density (BMD) is the most used diagnostic tool. Recently ultrasound (US) has been extensively used as alternative. Many aspects of the propagation of ultrasonic waves through biological tissues remain unknown. Today only statistical correlations exist between US parameters and bone strength. This approach yields interesting conclusions but with limited physical insight. Studying the interaction of ultrasonic waves with trabecular bone using a simulation technique can improve the knowledge and interpretation of ultrasonic parameters related to the diagnosis of osteoporosis. The goal of this study is to investigate whether finite element modeling (FEM) can be successfully applied to simulate ultrasonic wave propagation through trabecular bone samples. FEM requires a 3D structural representation of the trabecular bone sample. These 3D structures are created using microfocus computed tomography (microCT). To simulate wave propagation, a time-dependent displacement is applied at one side of the sample. The output is the displacement of the elements at the opposite side. MicroCT images of trabecular bone samples from the BIOMED1 project were used with a voxel size of 40 µm. The average dimensions of the bone cubes were 4x4x4 mm³. All elements in the model were given isotropic material properties with a Young’s modulus (E) of 20 GPa, Poisson coefficient 0.3 and density 1.962 g/cm³. Starting from this FE model some parameters were calculated: Speed of Sound (SOS), apparent Young’s modulus and BV/TV. It was found that SOS correlates better with the ratio of E and BV/TV than with E or BV/TV separately for some frequencies of the input signal.
How does a gradient of material properties due to endosteal resorption affect wave propagation in the axial transmission technique?

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The axial transmission technique is now widely used in the clinic for cortical bone assessment. However, ultrasonic propagation in this complex medium remains unclear, in particular because of the heterogeneous nature of cortical bone. Owing to different pathologies such as osteoporosis, endosteal parts of cortical bone undergo a resorption of the bone structure, leading to an increased porosity and to a transformation into trabecular bone. It creates a spatial gradient of material properties at the organ level. The aim of this work is to evaluate the effect of a spatial gradient of elastic modulus on the ultrasonic response of the bone structure. Therefore, a finite element method developed in the time domain accounts for the interaction of a transient elastic wave with the bone structure using a 2D model, which consists of a layer of transverse isotropic solid sandwiched between two semi-infinite fluid media. The model couples the acoustic propagation in both fluid media with the elastodynamic response of the solid. The conditions of continuity in normal stress and in displacement are used for the fluid-structure interaction. The effect of a spatial gradient of elastic modulus i) along the entire structure and ii) over half of the bone width, on the ultrasonic response as measured by a multi-element receiver is investigated. The amplitude and time-of-flight of the first arriving signal are evaluated as a function of the spatial gradient by keeping the mean value of both elastic properties constant. Our results show that the gradient of material properties plays a more important role when the thickness to wavelength ratio is high (h/λ > 1) than when it is low (h/λ < 0.25). This work gives further insight on the understanding of the interaction between ultrasound and the bone structure, which may lead to significant improvements of bone assessment.
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Bone elastic phenotyping of small animal models

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200-MHz scanning acoustic microscopy (SAM) and synchrotron radiation μCT (SR-μCT) were used to assess microstructural parameters, acoustic impedance $Z$ and tissue degree of mineralization of bone ($DMB$) in site-matched regions of interest in femoral bone of two inbred strains. Transverse femoral sections taken from 5 C57BL/6J@Ico (B6) and 5 C3H/HeJ@Ico (C3H) mice (5.5-months old) were explored. Mass density $\rho$, elastic coefficient $c_{11}$ and Young’s modulus $E_1$ were locally derived in the distal epiphysis, distal metaphysis for trabecular bone and mid-diaphysis for cortical bone using a rule-of-mixture model. Structural parameter estimations obtained from X-ray tomographic and acoustic images were almost identical. Both strains had the same bone diameter, but the C3H mice had greater cortical thickness and smaller cancellous diameter than did B6 mice. The average $DMB$ and impedance values were in the range between 1.13 and 1.33 g cm$^{-3}$ and 5.8 and 7.8 Mrayl, respectively. All tissue parameters were lower in B6 mice than in C3H mice. However, interstrain differences of $DMB$ were much less (up to 3.8 %) than differences of $Z$ (up to 13.2 %). $c_{11}$ was highly correlated with $Z$ ($R^2 = 0.995$), but not with $DMB$ ($R^2 = 0.13$).

SAM and SR-μCT fulfill the requirement for a simultaneous evaluation of cortical bone microstructure and material properties at the tissue level. However, SAM provides a quantitative estimate of elastic properties at the tissue level that cannot be captured by SR-μCT. The strong differences in the measured acoustic impedances among the two inbred strains indicate that the impedance is a good parameter to detect genetic variations of the skeletal phenotype in small animal models.
Assessment of bone structure and elasticity under the influence of leptin deficiency in C57BL/6 mice using high resolution scanning acoustic microscopy

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We investigated the influence of leptin deficiency on the elastic and structural bone properties of leptin deficient (ob/ob) and wildtype (wt) C57BL/6 mice using 200-MHz time-resolved scanning acoustic microscopy. Transverse femur sections from mice sacrifices at ages of 3, 6, 9, and 12 months were explored. The acoustic impedance values in the epiphysis ($Z_{\text{epi}} = 8.36 \pm 0.5 \text{ Mrayl}$) were generally lower than in the cortical tissue of the mid-diaphysis ($Z_{\text{dia}} = 8.74 \pm 0.41 \text{ Mrayl}$). While in wt mice the impedance was progressively increasing up to the 9-month group, no significant difference was observed between 3, 6, and 9 month old ob/ob mice. However, in the 12-month group the impedance values in both strains were equalized. These findings suggest that bone formation and maturation is decelerated in ob/ob mice, but a reduced bone turnover compensates for genetically induced difference of tissue elasticity in mature mice.
Ultrasonic mechanical characterization of bone formed at the interface of two different grafting materials

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It is a prerequisite for a successful placement of dental implants, that there is a sufficient amount of bone tissue available that lends mechanical stability to the device. To increase bone quality and quantity a multitude of substitute materials has been used. Their usefulness has already been empirically demonstrated. The purpose of this study was to characterize and measure the elasticity of two grafting materials and the different types of tissues that are involved in their osseointegration with scanning acoustic microscopy (SAM). Five mini-pigs were subjected to a bilateral sinus elevation procedure. One side was augmented with an injectable nanocrystalline hydroxyapatite, while contralaterally particulate deproteinized bovine bone was employed. After 6 weeks the animals were sacrificed. Tissue blocks containing the augmented area were removed from the sinus, fixed with buffered formalin and imbedded in acrylic glass. SAM measurements were performed with a 50MHz transducer on a three axis scanning time resolved microscope. Resulting impedance maps were masked with manually selected regions of interest to derive the distribution of acoustic impedance for the different bone tissue types. Strong and significant differences in measured acoustic impedance were only found between nanocrystalline hydroxyapatite (5.4 ± 1.5 Mrayl) and deproteinized bovine bone (8.5 ± 2.1 Mrayl). The treatment groups showed similar results for old bone tissue (6.3 ± 0.9 Mrayl) that was already present before surgical intervention. It was possible to demonstrate that after 6 weeks of osseointegration, nanocrystalline hydroxyapatite possesses mechanical properties that differ dramatically from deproteinized bovine bone. However the quality of the bone tissue that is newly formed near the interface region, exhibits no such differences.
Transosseous application of Low-Intensity Ultrasound on healing at tendon graft -bone interface in rabbits results in altered gene expression levels

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Introduction:

The purpose of this study is to investigate the effect of transosseous low-intensity pulsed ultrasound (LiUS) during ligamentization process on the healing at tendon graft-bone interface in rabbits, by examining the expression levels of TGFb1, biglycan and collagen I using semi-quantitive RT-PCR.

Materials and Methods:

The anterior cruciate ligament in both knees of 36 New Zealand white rabbits were excised and replaced with the long digital extensor. Custom-made ultrasound transducer was implanted onto the bone fragment and along the surface of the bone tunnel. The LiUS-treated right knees received 200-µsec bursts of 1 MHz sine waves with pulse repetition rates of 1 KHz and average intensity of 30 mW/cm2, for 20 minutes daily (study group), while the left knee received no LiUS (control group). Semi-quantitative RT-PCR was performed from RNA samples representing both study and control groups at 1, 2, 5, 7, 9, 12, 14 and 21 days, using specific primers.

Outcome:

Analysis of the RT-PCR products showed statistically significant up-regulation of biglycan and collagen-encoding genes in the study group compared to the control group. In addition, TGFb1 gene expression exhibits a bimodal profile. In the study group, we observed statistically significant down-regulation of its mRNA levels from day 1 until day 7 compared to the control group (~70% in day 7). From day 9 until day 21, TGFb1 gradually restored its mRNA levels to the mRNA levels of the control group (day 12). Subsequently, we observed a gradual up-regulation compared to the control group after day 14 (~30% in day 21).

Conclusions:

Transosseous LiUS treatment affects the expression levels of significant genes and possibly enhances the healing rate of the tendon graft-bone interface in rabbits.
Monitoring of elastic properties during bone regeneration

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The bone regeneration occurs after fractures, arthrodeses, osteotomies and bone grafting operations. It occurs in several steps like fraction, granulation, modeling and remodeling.

The aim of this study was the characterization and quantification of the elastic properties for new modelled bone near graft material after substitution at 5 different time periods using scanning acoustic microscopy (SAM).

Bilateral distal femur defect procedure was applied to 66 female New Zealand white rabbits. On both sides were augmented with different substitutes. One side was filled only with an injectable nanocrystalline hydroxyapatite paste and other side was filled using the same graft mixed with particulate autogeneic or allogeneic bone. After 2, 4, 6, 8 and 12 weeks the animals were sacrificed. Tissue blocks containing the augmented area were removed from the distal femur, fixed with buffered formalin and imbedded in PMMA. Data acquisition was performed using a 50MHz transducer. Recorded were c-scans in time resolved mode. Acoustic impedances were analyzed on distal femur samples which included only bone graft substitute. Regions of interest were hand made selected from impedance maps to derive the distribution of different bone tissue types. Significant differences in acoustic impedance were found between new formed bone over the whole time range of 12 weeks from 5.0±0.2 Mrayl at 2 weeks to 6.0±0.4 Mrayl at 12 weeks.

This study allowed the assessment of the dynamic behaviour of the elastic properties of new modelled bone over treatment time.
New Trends in Scanning Acoustic Microscopy

Dr. Peter Czurratis,¹ Dr. Klaus Krämer²

The uniqueness of acoustic microscopy lies in the ability to image the interaction of acoustic waves with the elastic properties of a specimen with microscopic resolution. A lens with good focusing properties on axis can be used for both transmitting and receiving the signal, and an image is formed by scanning the lens mechanically over the specimen. For routine use 2 GHz is the highest practical frequency, which offers a resolution of about 0.3 µm = \lambda/2. The information is contained in the way that the acoustic wave is reflected from the specimen. In higher stiffness specimens, including most metals, semiconductors and ceramics, a dominant role in the contrast can be played by Rayleigh waves in the surface. If the specimen has a surface layer, then the propagation of the Rayleigh waves is sensitive to the perturbing action of the layer. If the specimen is anisotropic, then there will be dependence on the orientation of the surface and the direction of propagation in it. If there are surface cracks or boundaries, then there will be strong contrast when they scatter the Rayleigh waves. In many applications of acoustic microscopy, the microscope is used to image the interior of an opaque material. In such cases somewhat lower frequencies 10-100 MHz are used to achieve greater penetration. Uses of this type often include examination of electronic packaging materials to ensure integrity, especially in high value-added applications.

Acoustic microscopy can be used for example to examine solder in side-brazed dual-inline packages and glass hermetic seals in ceramic quad flat pack packages, plastic quad flat pack packages, die attach layers, interconnect integrity on thin-film and high density organic substrates, biological samples (cells, bones e.g.) and the mechanical properties of packaging materials. Composites can also be inspected in this way, and the confocal properties of acoustic microscopy can be exploited to give enhanced depth discrimination and hence to examine individual interfaces, density variations, material inhomogeneity one at a time using new KSI scan modes and image technics. There are many situations in which a thin coating is applied to the surface of a material, and in which it would be useful to be able to measure the elastic properties of the coating layer. Even more desirable in some applications would be knowledge of stresses in the layer. It is now becoming possible to make measurements by quantitative acoustic microscopy that will yield this kind of information. Accurate elastic measurements by acoustic microscopy were pioneered using cylindrical lenses which brought the acoustic beam to a focus along a line. This meant that Rayleigh waves were excited in the surface of the material in one azimuthal direction in the surface only, so that even if the sample was anisotropic only one Rayleigh velocity would be involved. Line-focus-beam lenses give particularly well defined oscillations in the variation of signal with defocus—the so-called V(2) curve—and by suitable Fourier analysis surface wave velocities can be determined from the period of the oscillations with an accuracy which can exceed 0.1 %. The attenuation of surface waves can also be measured.
Friday, July 20th, 2007:

9:00 am Keynote Lecture

**First data of hip fracture discrimination using the Femur Ultrasound Scanner (FEMUS)**

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Recently, we presented correlations between Quantitative Ultrasound (QUS) and Dual-X-ray Absorptiometry (DXA) both measured at the proximal femur in vivo. Now, we extended the study by including 10 patients with recent hip fractures. We tested if these correlations are similar in the patient group and in the control group. Additionally, we investigated the power of femur QUS for fracture discrimination in comparison with DXA.

Controls without hip fractures were recruited from an ongoing population based study. Patients recruited from a clinical database had suffered an osteoporotic hip fracture within the last half year. All participants were measured using DXA and the FEMUS scanner. QUS transmission signals through the centre of the greater trochanter were evaluated and QUS variables speed of sound (SOS) and broadband ultrasound attenuation (BUA) were calculated. SOS was adjusted for variations in hip width using arrival times of signals reflected from both the anterior and posterior surfaces of the hip. Correlations of QUS variables with trochanteric BMD were calculated. The ability of discriminating between patients and controls was calculated for QUS and DXA using logistic regression analysis and expressed as standardized odds ratios per 1 SD decrease of the population variance, adjusted for age (confidence intervals in brackets).

Both SOS and BUA correlated significantly (BUA in volunteers only as trend) with trochanteric BMD in both groups (controls: $R^2=0.53$, $p<0.05$ for SOS; $R^2=0.31$, $p=0.09$ for BUA; patients: $R^2=0.60$, $p<0.01$ for SOS, $R^2=0.69$, $p<0.01$ for BUA; pooled: $R^2=0.66$, $p<0.0001$ for SOS, $R^2=0.54$, $p<0.001$ for BUA). Odds ratios for fracture discrimination were 3.9 (2.0-7.6) for trochanteric BMD, 7.8 (2.9-20.7) for total BMD, 8.7 (3.3-22.8) for SOS and 2.2 (1.3-3.7) for BUA. Weight and height did not contribute significantly to any of these associations.
Separation and characterization of superimposed waves at the proximal femur

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For QUS measurements at the proximal femur we recently introduced a model-based signal analysis to separate superimposed waves [1]. In this study, we compare results for BUA and SOS obtained from separated signal components with those of conventional algorithms. Signal component separation was successfully applied to ex-vivo (35 specimens, 74 ± 11 years) and to in-vivo (19 patients, 75 ± 8 years) measurements. In both cases, we observed two distinct waves. In the trochanter the group delays of the first and the second wave were strongly correlated (ex-vivo: R²=0.93, r<0.0001; in-vivo: R²=0.84, r<0.0001) whereas the correlations of the centre frequencies, which are equivalent to BUA, were moderate to weak (ex-vivo: R²=0.66, r<0.0001; in-vivo: R²=0.44, r<0.001). BUA was higher for the first wave, which is in agreement with Biot’s theory for fast and slow dilatational waves in porous media. For the total hip region of the ex-vivo measurements the characterization of the dominant second wave resulted in similar correlations of BMD with SOS and BUA (R²BMD-SOS=0.71, r<0.0001; R²BMD-BUA=0.69, r<0.0001) while conventional algorithms lead to a lower correlation of BMD with BUA (R²=0.61) compared to BMD with SOS (R²=0.81)[2]. Combining SOS and BUA of the second wave, the correlation with BMD becomes substantially stronger (R²=0.88, r<0.0001). Our results demonstrate that QUS parameters are sensitive to signal superposition. Further studies should investigate which additional information on bone properties are comprised in the distinct waves.

For the ultrasonic evaluation of bone, it is important to understand the anisotropy and complicated distribution of elastic properties with both micro- and macroscopic points of view. We focused on the body of femoral bovine cortical bones and reported the velocity distribution of longitudinal waves propagating in three directions (axial, tangential and radial) using cubic specimens. We have observed clear dependence of velocity anisotropy on the bone structure (plexiform or Haversian) and pointed two different types of velocity anisotropy in the plexiform specimens, which have similar structures and densities.\(^1\)

In this study, then, we have investigated the distribution of longitudinal wave velocity in the axial direction, using ring shaped cortical bone specimens and self-made PVDF transducers with diameter of 3 mm. The velocity distribution strongly depended on the measured position in the ring specimen, which is also dependent of bone structure. The result was in agreement with our former velocity distribution data obtained by cubic specimens. We have next focused on the effect of hydroxyapatite (HAp) crystallites on the axial velocities. The HAp crystal has a hexagonal symmetry, whose orientation can be experimentally measured by the X-ray diffraction. By adjusting the ultrasonic and X-ray measured points on the same specimen surface, we have clarified the relation between crystallites orientation and longitudinal wave velocities in the axial direction. Here, the crystallites orientation at each measured point was obtained from the averaged X-ray data of both front and reverse sides of the ring specimen. The wave velocities in the area of plexiform structure clearly depended on the degree of crystallites orientation (\(R^2 = 0.525, p < 0.001\)), however, the effect of crystallites orientation is not clear in the Haversian area.

A new system for ultrasonic assessment of the forearm

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The overall objective of this study was to develop an ultrasonic method for estimating bone mineral density (BMD) at the forearm. Estimation of BMD is an important component in diagnosis and managing osteoporosis. The use of BMD is based on the well-established thesis that bone strength is strongly related to the amount of bone material present and that a stronger bone in a given individual is associated generally with a lower fracture risk. Radiological densitometry (e.g., DXA), which measures the BMD at a given site is currently the accepted indicator of fracture risk. Because of its expense, inconvenience, and associated x-ray exposure, ultrasound has been proposed as an alternative to DXA. This study used both empirical ultrasound measurements and data generated in computer simulations. In vitro measurements were conducted on a set of 6 plastic rods, 7 plastic tubes, and 15 human radii. A 3.5 MHz source and receiver in a through-transmission configuration were used within a water tank, between which the plastic or bone samples were placed. The received waveform was stored for subsequent processing. A set of ultrasound simulations of analogous models was also carried out using a commercial software package (Wave2000 Pro, CyberLogic, Inc., NY). The ultrasound data obtained from both the bench-top experiments and the computer simulations were processed to obtain an ultrasound parameter known as net time delay (NTD), associated with each sample. Results obtained show extremely close correspondence between the simulated and empirical results, validating the use of the software for further development. Both sets of data demonstrated high correlation with cortical thickness (bone mass). Moreover, clinical data obtained using a novel ultrasonic array system demonstrated close correspondence with the in vitro and simulated data. This study suggests that ultrasonic transmission measurements at the forearm may be a useful, simple, and radiation-free alternative to DXA for assessing bone mass and fracture risk.
Attenuation through trabecular bone: a comparison between simulated values obtained using finite-difference time-domain 3D simulations and experimental values measured on the same samples.

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The present study compares values of attenuation and BUA values through trabecular bone obtained by numerical simulation and experimentally. The comparison is done between results obtained in the exact same samples for simulation and experiments. The simulations were performed using three-dimensional (3-D) high-resolution micro-computed tomography reconstructions of a cohort of human femoral specimens that were previously investigated experimentally using a through-transmission technique. The simulation code (SimSonic) uses a finite-difference time-domain (FDTD) algorithm to compute ultrasound propagation into both fluid and solid structures. The version of the code that was used did not account for dissipative mechanisms. When comparing the predicted frequency-dependent attenuation coefficients to the experimental ones, a strong correlation was found between simulated and experimental values both for the attenuation and slope of the frequency dependent attenuation coefficient (a linear regression yielded \( \text{nBUA}_{\text{experiment}} = 1.12 \times \text{nBUA}_{\text{simulation}} + 1.6 \text{ dB} \times \text{(cm} \times \text{MHz}^{-1}) \), \( R^2 = 0.83 \). Both simulated and experimental nBUA exhibited the same dependence on bone volume fraction. However, it was found that the attenuation systematically underestimated in the simulation compared to the experimental results. Under assumption that the absence of absorption losses in the simulations is responsible for the difference between simulation and measurements, the results suggest that scattering is probably the main contribution to attenuation for the specimens with a low bone volume fraction; and that other attenuation mechanisms such as absorption may be significant in the specimens of highest density.
Experimental study on the fast wave velocity and structural anisotropy in the cancellous bone

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Cancellous bone is comprised of the trabecular frame and soft tissue, and considered as anisotropic due to the complex structure. It is known that the ultrasonic longitudinal wave which passed through the cancellous bone is separated into two waves, the fast and the slow waves [1]. The fast wave mainly reflects the characteristics of trabecular frame. The fast wave velocity therefore increases with the bone volume fraction. The velocity of the fast wave is influenced not only by the bone volume fraction but also the anisotropy of cancellous bone. In this study, then, the relationship between the velocity of the fast wave and the anisotropy of cancellous bone is experimentally investigated. We used an ultrasonic pulse method [1] to measure the wave velocity in the cancellous bone. By rotating the cylindrical cancellous bone specimen obtained from the head of bovine femur, the velocity changes due to the rotating angle have been observed. In addition to the ultrasonic evaluation, the structural anisotropy of each specimen was measured by X-ray micro CT (Hitachi). The trabecular alignment of each specimen was then evaluated by “3D-Bon” (Ratoc) using the CT data. Here we have introduced “Bone alignment index (BA)” which is the averaged angle between the direction of wave propagation and trabecular alignment. The ultrasonic and CT results showed that the fast wave velocity depended on the structural anisotropy, especially on the trabecular alignment. The fast wave velocities were always high in the small BA region, though the degree of velocity anisotropy (velocity changes as a function of the incident angle to the specimen) depended on the bone volume fraction.

SAM data is used to derive the continuum level elastic properties of cortical bone

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The aim of this project is to elucidate the relationships between the microscopic properties of bone and its elasticity at the continuum scale, or millimetre scale, or mesoscale. We propose a method to estimate the mesoscale properties of cortical bone based on a spatial distribution of acoustic properties obtained with SAM (50 MHz, 23 micron resolution). The procedure used to compute the mesoscopic stiffness tensor involves i) the segmentation of the pores to obtain a realistic model of the porosity; ii) the construction of a field of anisotropic elastic coefficients at the microscopic scale which reflects the heterogeneity of the bone matrix; iii) finite element computations of mesoscopic homogenized properties. The computed mesoscopic properties compare well with available experimental data. It appears that the tissue anisotropy at the microscopic level has a major impact on the mesoscopic anisotropy.
Characterization of bone samples by ultrasound holography at about 100 MHz and general aspects concerning the mechanical properties of porous anisotropic inhomogeneous materials including bone replacements

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Point source and detection ultrasound holography has been applied to bovine and human bone samples. The method is based on defocused phase sensitive scanning acoustic microscopy (PSAM) in transmission. Conversion of the phase images into appropriate data matrices and subsequent data processing yielded the angle dependent ultrasound velocities for the anisotropic samples. The results match conventional direction resolved measurements of the sound velocity in bone. However, the presented technique requires only small specimens. The demonstrated results are obtained for sample sizes of $5 \times 5 \times 0.5$ mm³. General features of porous anisotropic and inhomogeneous materials and anomalies of the Poisson Ratio observed for porous sintered Titanium samples are discussed also to highlight the special elastic properties of porous inhomogeneous and anisotropic materials of which human bone is a natural occurring example.
Measurement of wave velocity in bovine cortical bone by µ-Brillouin scattering method

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Cortical bone tissue is the main element to carry loads and transmit stresses in the skeletal structure. The material properties of this tissue strongly depend on the micro-macroscopic structures, from mineral crystallites and collagen fibers to the orthotropic lamellae or Haversian structures. The conventional ultrasonic wave evaluation induces the information of the multi-scale elastic properties in the large area where the ultrasonic waves passed through. In order to understand the microscopic “bone quality” without the effect of macroscopic structures, we then tried the measurement of elastic anisotropy of minute area using µ-Brillouin scattering technique. Brillouin scattering is a non-destructive and non-contact measurement technique. Making use of the focused laser beam and scattered light from the thermal phonons in thin layer sample, it enables to measure wave velocities which propagate in the different directions of the layer. Additionally, µ-Brillouin scattering technique can measure the minute area with diameter of 10µm and confirm the irradiation area by a microscope. However, the samples for Brillouin scattering measurement should be transparent. We have then tried to obtain very thin sample with thickness around 30µm from the bovine femur cortical bone. We have fabricated two types of samples from radial-tangential and axial-radial surfaces of the plexiform bone. After the optimization of the light scattering geometry, we have succeeded in measuring longitudinal wave velocities. The measured wave velocities were around 48-50×100 m/s in the tangential direction, whereas the measured wave velocities in the axial direction were around 50-52×100 m/s. In spite of the small difference, wave velocities in the axial direction were faster than those in radial direction, which indicated the anisotropic elasticity in the minute area.
The elastic and structural properties of human femoral cortical bone at tissue level using high resolution acoustic measurements

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In this study the spatial distribution of elastic properties of human femoral cortical bone was analyzed at the tissue level using high resolution cylindrical ultrasonic scanning microscopy. This scanning procedure allows to measure the angular dependence of the acoustic impedance of cylindrically shaped samples (diameter: 4.4 mm) with a single measurement. The measurements were performed with a 50-MHz transducer that provided a lateral resolution of 23 µm. The procedure maps the surface reflection amplitude of cylindrically shaped tissue sections. From the angular reflection the independent elastic coefficients were derived using the assumptions of transverse isotropy and continuum micro-mechanical model constraints. This method was applied to study the spatial distribution of elastic properties of human cortical bone at 14 spaced locations (from 20% to 85% of femur length) along its length, four anatomical positions as well as 10 sections (from endosteum to periosteum) along the radial direction. The ranges of elastic constants were: $C_{33}$: 26.8 - 34.9 GPa; $C_{11}$: 21.0 - 24.5 GPa; $C_{12}$: 8.6 - 11.1 GPa, $C_{13}$: 9.0 - 11.6 GPa, $C_{44}$: 5.4 - 7.8 GPa. The coefficients of correlation $R^2$ between the individual elastic coefficients were between 0.04 and 0.99.
The remodeling process enables bones to be adapted to their mechanical environment by modifying both architecture and tissue properties of the skeleton. An imbalance of this process can increase the fracture risk. Scanning acoustic microscopy (SAM) provides high resolution mapping of acoustic impedance (Z) related to stiffness and consequently could be an interesting tool to investigate changes in tissue elastic properties resulting from mechanical loading. In this study, trabecular bone specimens were extracted from bovine sternum and ulna. From each bone, the samples were divided in three groups. The C group was fixed post-extraction, the NL and L groups were kept viable in a culture cell over 3 weeks. Samples of L group underwent dynamic stress while NL samples were left free from loading. Then all the samples were prepared for further investigation. After histological measurements, they were explored with SAM. For each specimen, Z distribution was obtained. The mean value and standard deviation (SD) were average over each group and were compared to histology. Z varied with anatomical location and was 25% higher in the ulna C group compared to sternum. This could be the consequence of the local differences in mineral and collagen organization exhibited naturally between the two bones. The sternum responded more significantly than ulna and displayed a 25% increase in L group compared to C group. The SD of Z distributions was 50% higher for the loaded group, due probably to the heterogeneity of the sternum tissue response. In conclusion, SAM reflects very local changes in bone intrinsic material properties as a consequence of mechanically induced remodelling process.
Confocal and non-confocal acoustic transmission microscopy of tissue-engineered cartilage constructs

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Acoustic transmission microscopy at a frequency of about 100 MHz was employed for the characterisation of tissue-engineered disc-shaped cartilage transplants. In the confocal mode imaging was performed in an axially aligned plane, yielding results similar to that of a B-scan. In the non-confocal mode, the transducers were focused on the upper and lower surfaces of the disc-shaped transplant giving results similar to shadowing but with additional information on the time-of-flight. From the images in phase contrast local variations of the sound velocity can be determined and respective variations of the mechanical properties can be imaged even with 3-dimensional resolution. Respective tomographic images are demonstrated for test samples including structured mixed tissues.
A combination of mechanical and ultrasonic methods is used to assess the anisotropic elasticity and strength of in vitro cortical bone samples

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We present a strategy for the measurement of both anisotropic elastic properties and strength on the same in vitro bone samples. The method was applied to 88 human femur samples. Young’s modulus and strength are obtained with a three-point bending test. Two original ultrasonic setups adapted to the specific shape of plate-like samples used in three-point bending are introduced for the measurement of elastic constants. In the axial direction of the sample, the bar velocity is measured with a transmission setup using 60 kHz transducers; in the transverse direction, the bulk wave velocity is measured with a double transmission setup using a 20 MHz-focused transducer. The elastic coefficients calculated from the measured velocities are in the range of those reported in the literature obtained with other techniques. We will present a comparison of the sample properties (acoustical and mechanical) obtained with the different techniques.
Objective: To assess the dependence of quantitative ultrasound (QUS) indices speed of sound (SOS) and broadband ultrasound attenuation (BUA) at different sites on temperature and season we measured these indices monthly, from January to July 2006. Methods: 10 healthy volunteers were included. QUS was performed on calcaneus (Sahara, Hologic), on tibia and on radius (Omnisense 7000P, Sunlight) at four consecutive times after the start, at 0, 20, 40, and 60 min from the start in an examination room. The room temperature was kept constant. Skin temperature was monitored. BUA and SOS were evaluated. Results: There was a significant inverse association between the skin temperature at the calcaneus and SOS at the beginning of the measurements (R= -0.47; p<0.01); at 60 min correlation was R= -0.21 (p<0.01). There was no significant correlation between temperature and SOS at the radius (R= 0.17) and tibia (R= 0.09). Standard deviation of the parameters BUA and SOS in repeated measurements at the same time was higher on cold days than on warm days. Conclusion: SOS depends inversely on the skin temperature. QUS should measured after 60 min of rest in the examination room. There was no significant seasonal variation in the QUS indices. But, the variation was higher in colder days. The precision was best at the calcaneus.
Assessment of skeletal age in children and adolescents using radius sonography in comparison to standardised radiography

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Purpose: The assessment of skeletal development in children is important. The standard is to study left hand radiograms based on the Gräulich&Pyle (G&P) method. Aim of our study was to compare the accuracy of a new ultrasound method with the G&P method. Method: 70 consecutive evaluated patients (calendary age 6–17y; 34f, 36m) who received a X-ray of the left hand were evaluated. Ultrasound of the same hand was performed immediately using the BonAge system (Sunlight Med. Ltd., Israel). This system evaluates the relationship between the velocity of the wave and growth using gender- and ethnicity-based algorithms. X-rays were analyzed and bone age scores based on the G&P were evaluated. Results: In 65 patients BonAge measurement was successful. The correlation between BonAge and G&P was 0.82. The averaged accuracy (i.e. absolute difference in years between GP reading and BA ultrasonic results) was calculated. Results were similar for boys or girls – for the whole left hand 1.0+/−0.8 and for the distal radius 0.8+/−0.7. The difference between BonAge and CA was the same as the difference between GP and CA, i.e. 1.4 years. Conclusion: The BonAge device demonstrates the ability to produce an accurate assessment of bone age using an ultrasound method.
Short-term reproducibility of in vivo guided wave measurements

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The first arriving signal (FAS) as measured by a low-frequency (200kHz) axial transmission device is known to be a guided wave, and its velocity (V1) is influenced by both the geometry and material properties of cortical bone. An inexperienced operator of the current prototype may produce relatively high reproducibility errors (up to 5%) in V1. Such high errors explain most of the observed variability in V1. In this study we focused on investigating the performance of the device by carefully executing measurements on the antero-medial mid-shaft tibia of children (n=84) and adults (n=161). Co-localized volumetric cortical bone mineral density (CBMD) and cortical thickness (CTh) were also assessed by peripheral quantitative computed tomography (pQCT). Preliminary analysis of the data showed that for children the rms coefficient of variation (RMS CV) was 1.3%, whereas for adults it was higher, 2%. V1 was significantly correlated with CBMD (r=0.56 and r=0.33 for children and adults, respectively; p<0.001) and with CTh (r=0.26 and r=0.17, respectively; p<0.05). On the other hand, ranges in all recorded variables were slightly narrower for the children, thus the weaker correlations obtained for adults were mostly due to poorer precision. Soft tissue thickness and its variation were bigger in adults than in children, and these two factors are known to be the most significant sources of measurement uncertainty. Although the bidirectional method was used to account for the variation in soft tissue thickness, it did not completely eliminate the error. Future development of the device will focus on optimization of its mechanical performance.
Elastic changes in cartilage and subchondral bone due to primary arthrosis

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This study aimed at assessing elastic changes of cartilage and subchondral bone relating to primary osteoarthrosis. Samples were obtained from 28 human individuals during alloplastic implant surgery. Sagittal sections were explored in hyperosmolar (2.5 molar) saline solution at 50 MHz at 25°C using a scanning acoustic microscope (SAM). Cartilage and bone impedance distributions were evaluated as a function of the distance to the cartilage-bone interface. The degree of cartilage degeneration was derived by histological and histochemical analysis using the Mankin Score. The mean impedance value in cartilage was 2.12 ± 0.02 Mrayl. The well-known layered cartilage structure was revealed by different impedance values in most samples. Generally values were higher close to the bone interface and decreased continuously towards the cartilage surface. Higher grades show a loss of the layered structure and a rather uneven surface with large height differences. The content of Proteoglycans as a histochemical sign for cartilage degeneration was associated with increasing impedance values for cartilage and subchondral bone. The mean impedance value in subchondral bone was 6.28 ± 0.54 Mrayl. A significant increase of the impedance within the first 150 µm relative to cartilage-bone interface was observed in 65.5 % of the investigated sections. We hypothesize that the impedance increase close to the bone cartilage boundary is an indicator for subchondral sclerosis.
Ultrasonic assessment of trabecular structure in proximal tibia

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Proximal tibia presented access to the medial tibia plateau and a possibility to monitor status of load-bearing bone during osteoporosis or in microgravity. This site is composed mainly of the highly structured porous trabecular bone, where loss of bone mass is tightly bound with changes of the structure. 25 proximal tibiae bone specimens with broad range of the trabecular bone mineral density (BMD) were examined. Region of interest was located 17 mm below the tibial plateau. Ultrasonic parameters measured by the surface transmission on 40 mm acoustic base orientated in transverse plane included:
- Ratio of signal intensities at 100 and 300 kHz (R_{100/300}) that was considered as a derivative of the frequency slope of attenuation in the low frequency band;
- Integral intensity of broadband ultrasonic response as a measure of attenuation;
- Arrival times of signal front and of slower wave packets.

The ultrasonic parameters were correlated with Structural Measures of Complexity and BMD calculated from pQCT slices, and histomorphometric parameters, all obtained from the same location. The highest correlations were found between parameter R_{100/300} and those structural parameters, which are responsible on the scattering properties: Trabecular Network Index (r=0.89), Index of Global Ensemble (r=0.88) and size of maximal connected marrow area (r=-0.86). Good correlation between R_{100/300} and BMD (r=0.86) is explained by tight relation of the BMD with trabecular architecture; meanwhile correlation of R_{100/300} with trabecular thickness was rather poorer (r=0.53-0.64) showing the secondary contribution of bone mass itself. The study demonstrated potential of low frequency ultrasonic measurements in the surface transmission mode for acquiring information about subcortical trabecular structure in the tibia epiphysis.

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Guided wave ultrasound in the assessment of tibial bone in elderly females

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Axial transmission technique is found to be suitable for measurements on cortical bone. Lamb waves propagate throughout the cortical layer of bone making them appealing e.g. for assessing osteoporosis. In this study we focussed on the first arriving signal which at low frequencies includes a contribution from the S0 Lamb mode. Subjects were 98 elderly women (78-82 years). A low-frequency (200 kHz) quantitative ultrasonic (QUS) axial transmission prototype was used to measure ultrasound velocity (V1) in the mid-tibia. Femoral shaft diameter (FSD) and medial and lateral femoral shaft cortical thicknesses (FSC) were measured from digital radiographs below the minor trochanter (FSD1 and FSC1) and at the femoral shaft 2% of the subject’s height distally from the minor trochanter (FSD2 and FSC2). Bone mineral density (BMD) was measured at the hip and at lumbar vertebrae L1-L4 using dual energy x-ray absorptiometry. Significant but weak correlations were found between lateral FSC1 and V1, and between medial FSC2 and V1. Highly significant but still moderate correlations were found between V1 and hip BMD. Both L1 and L2 BMD also correlated significantly with V1. The observed correlations, although low, between V1 and femoral cortical thickness parameters suggest that tibial ultrasound can to some extent predict femoral properties. The method is comparable to other QUS methods in terms of characterizing hip and lumbar spine BMD and it might thus be suitable for skeletal status assessment. Improving the reproducibility of V1 measurements from the present prototype level is expected to further enhance the capability of V1 in bone characterization.
Beneficial effects of Tai Chi on women’s skeletal status assessed by quantitative ultrasound (QUS) at the hand phalanges: the cross-sectional and one-year follow-up study

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Aim of the study was to assess the skeletal status in women exercising Tai Chi. At baseline a group of 115 women (55.9±10.2 y) was compared with age-matched control group of 1030 women, and one-year follow-up comprised 46 subjects. Skeletal status was assessed using QUS measurements at hand phalanges. Results at baseline: amplitude-dependent speed of sound (Ad-SoS) had greater values in a whole group of exercising subjects than in controls (2012±77 vs. 1981±81 m/s, p=0.00009). In premenopausal women, Ad-SoS did not differ in comparison with controls, but in postmenopausal women did (1946±60 vs. 1975±59 m/s). There was correlation between Ad-SoS and exercise duration in the whole group and in postmenopausal women (r=0.24, p=0.01, and r=0.29, p=0.01, respectively). Age affected Ad-SoS significantly less in exercising women than in controls (r=-0.64 and -0.74, respectively). A stepwise multiple regression analysis of Ad-SoS regressed on age, duration of postmenopause, duration and intensity of exercises, and body size showed that age had negative influence and duration of the use of Tai Chi exercises remained a protective factor (regression equation: Ad-SoS (m/s) = 2183 m/s – 0.43 x age (y.) + 0.32 x duration of exercises (y.) (R=0.52, SEE=51.5, p<0.00001). Results at follow-up: the increase of Ad-SoS from 1995±80 to 2012±75 m/s (p=0.001) and Z-score from 0.673±1.054 to 1.053±1.178 (p=0.0005) were observed in subjects studied. These changes revealed statistically significant positive correlations with age of the exercising subjects (r=0.39, p=0.007; and r=0.35, p=0.018, respectively). The increase in Ad-SoS exceeded the value of the LSC in 19 subjects (41%), and in pre- and postmenopausal women in 2 (22%) and 17 (46%), respectively. Conclusion: due to its beneficial effect on skeletal status, Tai Chi exercises may be recommended in the prevention of postmenopausal osteoporosis, especially in older individuals.
Astronauts bone loss is a major concern on long duration space flights. It is now commonly accepted that skeletal unloading leads to increased bone remodeling with an imbalance between the increasing bone resorption process and the quasi-normal bone formation process. During the last 4 decades, several papers reported bone losses at different skeletal sites and particularly at the Calcaneus, during either space missions or bedrest simulated microgravity studies. Several resistance exercises and drug therapy have been developed and applied as countermeasures to prevent from bone loss. In this context, we developed a dry Quantitative Ultrasound (QUS) Imaging device dedicated to heel and wrist bone. It provides two parametric images of attenuation (BUA) and velocity (SOS), with precision errors of 2% and 0.3%, respectively. Two 60 days Bedrest studies were organised in Benjamin Franklin Universitätsklinikum (Berlin, BBR study), and in MEDES (Toulouse, WISE study). 16 healthy male volunteers, and 24 healthy female volunteers, were included in the BBR and the WISE studies, respectively. Three QUS measurements were performed with foot repositioning. Several measurement sessions were planned: 1 before Bedrest, every 15 days during the Bedrest, and after 1, 3, 6 and 12 months. The subjects were divided in 2 groups: Control and Exercise. The exercise consisting in training the lower limbs and feet. We clearly exhibit different bone modification between these two groups, either for BBR or WISE experiments. Bone loss was higher in the male group (-6%) compared to the female group (-3.5%). All of them recovered nearly their initial BUA or SOS values after 1 year.
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