Int J Thermophys (2015) 36:862–867
DOI 10.1007/s10765-014-1701-6

Bone Composition Diagnostics: Photoacoustics Versus Ultrasound

Lifeng Yang · Bahman Lashkari · Andreas Mandelis · Joel W. Y. Tan

Received: 20 November 2013 / Accepted: 18 July 2014 / Published online: 12 August 2014

Abstract Ultrasound (US) backscatter from bones depends on the mechanical properties and the microstructure of the interrogated bone. On the other hand, photoacoustics (PA) is sensitive to optical properties of tissue and can detect composition variation. Therefore, PA can provide complementary information about bone health and integrity. In this work, a comparative study of US backscattering and PA back-propagating signals from animal trabecular bones was performed. Both methods were applied using a linear frequency modulation chirp and matched filtering. A 2.2 MHz ultrasonic transducer was employed to detect both signals. The use of the frequency domain facilitates spectral analysis. The variation of signals shows that in addition to sensitivity to mineral changes, PA exhibits sensitivity to changes in the organic part of the bone. It is, therefore, concluded that the combination of both modalities can provide complementary detailed information on bone health than either method separately. In addition, comparison of PA and US depthwise images shows the higher penetration of US. Surface scan images exhibit very weak correlation between US and PA which could be caused by the different signal generation origins in mechanical versus optical properties, respectively.

Keywords Bone osteoporosis · Decalcification · Decollagenization · Photoacoustics (PA) · Ultrasound (US) backscatter

L. Yang · B. Lashkari (✉) · A. Mandelis · J. W. Y. Tan
Center for Advanced Diffusion-Wave Technologies (CADIFT), Department of Mechanical and Industrial Engineering, University of Toronto, Toronto, ON M5S 3G8, Canada
e-mail: bahman@mie.utoronto.ca

A. Mandelis
e-mail: mandelis@mie.utoronto.ca

L. Yang · A. Mandelis
School of Optoelectronic Information, University of Electronic Science and Technology of China, Chengdu 610054, People's Republic of China

1 Introduction

Osteoporosis is a major cause of pain and disability in elderly people. X-ray-based measurements of bone mineral density (BMD) represent the gold standard for osteoporosis diagnosis and assessment of fracture risk. Dual-energy X-ray absorptiometry (DXA or DEXA) machines are widely used to diagnose osteoporosis, and assist in making decisions and monitoring progress in treatment [1]. However, there are major issues with the accuracy and safety of DEXA. New evidence reveals that the BMD is not an accurate predictor of bone strength, although it is certainly a major parameter influencing it [2,3]. Several studies now suggest that there is a strong correlation between bone collagenous matrix and bone strength [3].

Recently combined backscatter ultrasound (US) and back-propagating photoacoustic (PA) measurements have been proposed for bone integrity assessment [4–6]. In this study, we used both PA and US modalities to assess the sensitivity of each method not only to the mineral content of bones, but also to the collagen content. The effect of the collagen content on the US signal has been studied before [7]. In the PA field we adapted and extended concepts from the US-bone literature.

The key goal of this study was to investigate US backscattering and PA back-propagating signals from trabecular bone and assess the sensitivity of these modalities to the composition and mechanical properties of bone.

2 Materials and Measurement Protocol

2.1 Bone Specimens

Bone samples were harvested from femur and ischium of three bovines (Angus, Canadian) and cut into a rectangular block. The samples were stored in a refrigerator before being treated or measured and were allowed to equilibrate thermally at room temperature prior to the experiments. The specimens were separated into two groups and treated with different agents to reduce their mineral or collagen contents [7,8] Three specimen were treated with a 50 % buffered solution of ethylenediaminetetraacetic acid (EDTA) (pH = 77) to demineralize the bone Another three specimens were treated with a 5 % solution of sodium liquid hypochlorite to reduce the collagen content Signal detection of every specimen was performed at identical points before and after the treatments. US and PA measurements were performed in distilled water at room temperature. The details of the experimental setup and signal analysis are described elsewhere [5,6]. However, it is important to mention that the laser wavelength employed was 800 nm. The use of the frequency domain (FD) not only facilitated the analysis of spectra but also proved to be helpful in increasing the signal-to-noise ratio (SNR) and extracting small signatures from the signal [9]. The ranges of the frequency measurements for PA and US were 0.3 MHz to 2.6 MHz and 0.3 MHz to 5 MHz, respectively.

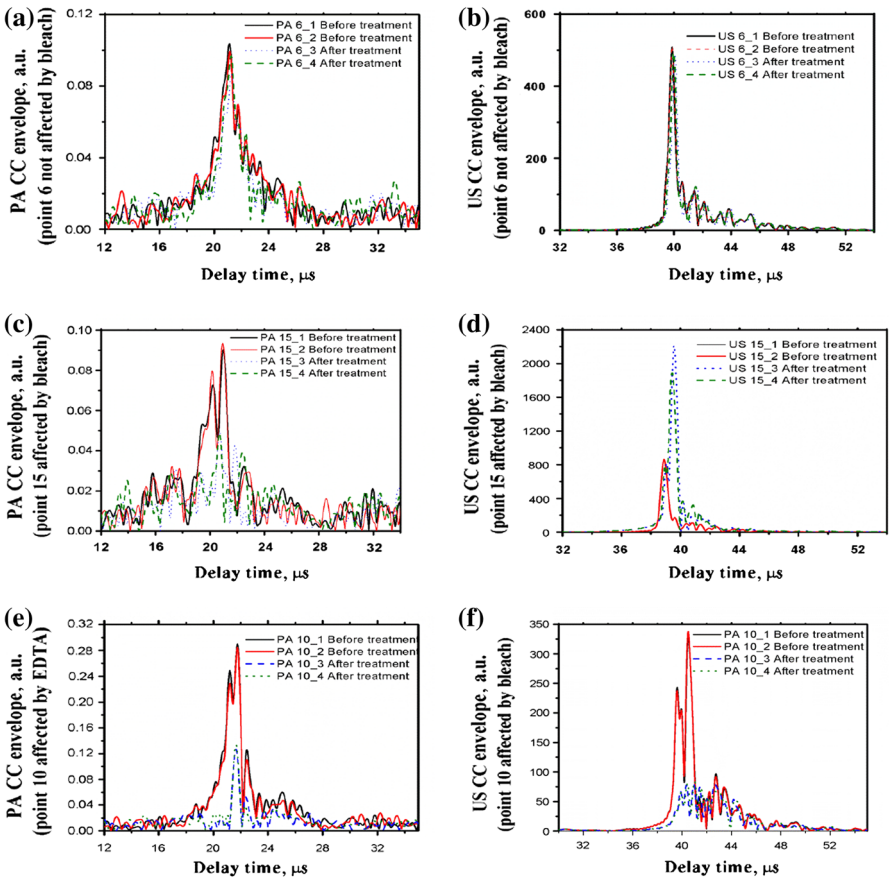

Fig. 1 (a) PA and (b) US cross-correlation signals from a point on the intact part of a bone. (c) PA and (d) US cross-correlation signals before and after decollagenization. (e) PA and (f) US cross-correlation signals before and after demineralization. At each stage measurements at each point were performed at least twice with a 2 h difference in between while the sample remained in water which helps with the reproducibility of the results

2.2 Quantitative Ultrasound (QUS) and Photoacoustic (PA) Measurements

In each sample a landmark was artificially made to distinguish the measurement points and mark the horizontal line below which the sample was immersed in the solution agent. The points above that landmark were not affected by solutions, Fig. 1a, b; and those below were demineralized/decollagenized, Fig. 1c to f. Several points (14 to 16) were measured on each sample. The points on the untreated part of the bone were used as references to reveal the changes in the signal due to factors other than demineralization/decollagenization. The apparent integrated backscattering (AIB) [10,11] was determined by frequency averaging (integrating) the ratio of the power spectrum of the signal (P_b) over the power spectrum of a reference signal (P_r) over the chirp frequency range:

Table 1 Average US and PA AIB changes due to demineralization and decollagenization of bone samples

	Treatment	Treatment (h)	PA AIB change (dB) Average value	US AIB change (dB) Average value
Demineralization	EDTA	5	-6.10 ± 2.18	-6.64 ± 1.52
Decollagenization	NaOCl	3	-6.87 ± 3.55	1.31 ± 1.54

$$AIB = \frac{1}{\Delta f} \int_{\Delta f} 10 \log_{10} \left(\frac{P_b(f)}{P_r(f)} \right) \mathrm{d}f \tag{1}$$

The PA apparent integrated back-propagating signal was calculated using Eq. 1 with P_b being the power spectrum of the signal and P_r being the power spectrum of a reference PA signal. The reference PA signal was obtained from a measurement on a homogeneous absorber. Time gating for both US and PA was performed based on their center frequencies and was six times over their center frequencies. For PA the time gating window was $6/f_{PA} = 414\,\mu s$ and for US it was $6/f_{US} = 226\,\mu s$ [1,12].

3 Results

Figure 1 shows that the PA and US cross-correlation signals have been affected by the treatment while, in the intact part of the bone the signal shape shows very good reproducibility. Several points on every sample were tested, and the averaged changes of US and PA AIB values of each group of the samples are reported in Table 1. The results show that the PA signal is significantly decreased with both treatments while the US signal has decreased significantly with demineralization but it exhibits only a minor increase with decollagenization. The reproducibility of the results was tested with multiple measurements before and after each treatment as shown in Fig. 1.

In another experiment, the PA and US images of a bovine bone sample were generated by raster scanning the sample surface. Fig. 2 shows the sample as well the US and PA images. In addition a micro-computed tomography (μCT) image of the bone is shown for comparison. Here the μCT image has been averaged over $1 \times 1\,mm^2$ on the surface and at a 4 mm depth to generate an image with similar spatial resolution as the US and PA images. Furthermore depthwise PA (Fig. 2e to g) and US (Fig. 2h to j) images of three different cross sections of the bone sample were compared In Fig. 2c to j the laser light impinged on the surface of the sample (left-hand side in Fig. 2e to j). The transmitter and receiving transducers were also located on the same side of the sample and the images were generated by scanning along the three lines shown in Fig. 2a. The higher depth detectivity of US can be seen in Fig. 2h to j

Comparing the three modalities in Fig. 2b to d, there is little correlation between the PA image and the other two. On the other hand, a moderate correlation between US and μCT images can be seen. This can be understood by considering that μCT is only sensitive to the mineral parts and similarly US scattering from mineral parts dominates the US image while the PA image is also sensitive to the organic parts of bone. The

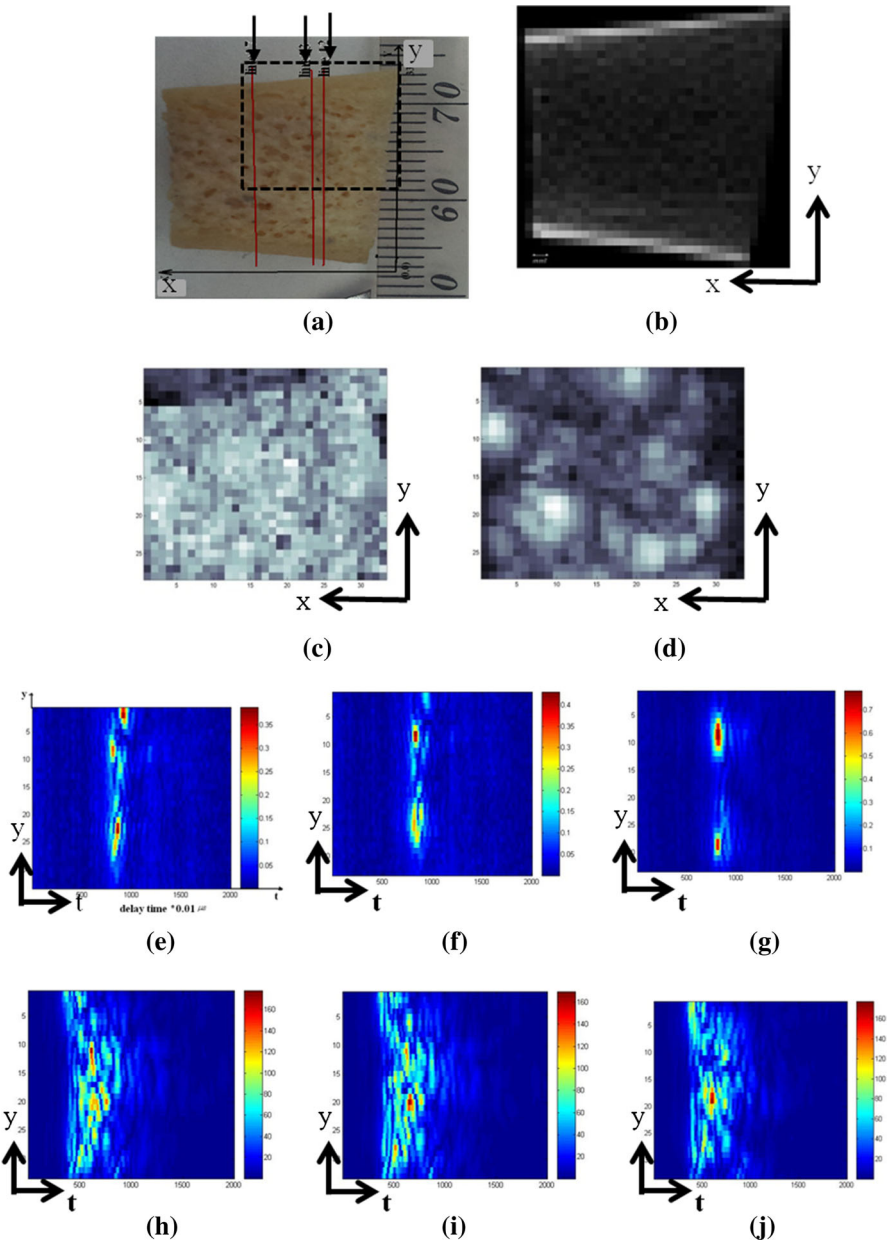

Fig. 2 (a) Photograph of the bone sample; (b) μCT image pixel averaged to attain the same spatial resolution as the US and PA images (~1 mm); (c) US and (d) PA images of the sample generated by raster scanning; (e), (f), and (g) PA images of the three sections in the sample marked with *arrows* in image (a). US images of the same three sections are shown in (h), (i), and (j)

differences and similarities of PA and US modalities and the relative sensitivity of PA to the composition of bone (based on its spectral response) require more investigation

4 Conclusions

Early osteoporosis usually manifests itself as a reduction of bone mass; however, bone health and strength are also connected to the collagen content of bones which cannot be detected by traditional DEXA measurements. Here it has been shown that both US and PA transient probing and imaging can detect the variation in BMD; however, the two modalities respond differently to the artificial collagen content variation generated by the decollagenization solution (NaOCl). This difference can be used to detect variations in the organic components of bone and are also noticeable in the surface scans. In addition, the PA results are more superficial compared with the US signals. Therefore, the two modalities are complementary in determining the composition of bone tissue and combined they may increase the chances of early diagnosis of osteoporosis. It should be added that there are obvious challenges in translating the results of this paper to the clinical diagnosis of osteoporosis, as the presence of blood and marrow as well as skin overlayer interfere with the bone signal. However, the ability of the PA modality to differentiate between the main chromophores using hyperspectral imaging provides an immense advantage that will facilitate the transition of *in vitro* research to clinical application, and will be the objective of a future study.

Acknowledgments The authors gratefully acknowledge the support of the Canada Council for the Arts through a Killam Fellowship to AM and of the Natural Science & Engineering Research Council (NSERC) for a Discovery Grant to AM The guidance and recommendations of Drs. M. D. Grynpas and T. L. Willett, Mount Sinai Hospital, are greatly appreciated. The authors acknowledge the help of Professor J. E. Davis and Dr. E. Ajami (IBBME, University of Toronto) in performing the μCT of the bone samples.

References

1. B. Edelmann-Schäfer, L.D. Berthold, H. Stracke, P.M. Lührmann, M. Neuhäuser-Berthold, Ultrasound Med. Biol. **37**, 29 (2011)
2. J.P. Karjalainen, J. Toyras, O. Riekkinen, M. Hakulinen, J.S. Jurvelin, Ultrasound Med. Biol. **35**, 1376 (2009)
3. M.E. Launey, M.J. Buehler, R.O. Ritchie, Annu. Rev. Mater. Res. **40**, 25 (2010)
4. B. Lashkari, A. Mandelis, Proc. SPIE **8207**, 82076K (2012)
5. B. Lashkari, A. Mandelis, Proc. SPIE **8565**, 85656I (2013)
6. B. Lashkari, A. Mandelis, J. Biomed. Opt. **19**, 036015 (2014)
7. B.K. Hoffmeister, S.A. Whitten, S.C. Kaste, J.Y. Rho, Osteoporos. Int. **13**, 26 (2002)
8. H. Ehrlich, P.G. Koutsoukos, K.D. Demadis, O.S. Pokrovsky, Micron **40**, 169 (2009)
9. B. Lashkari, A. Manbachi, A. Mandelis, R.S.C. Cobbold, J. Acoust. Soc. Am. **132**, EL222 (2012)
10. K. Wear, IEEE Trans. Ultrason. Ferroelectr. Freq. Control **47**, 777 (2000)
11. K.I. Lee, M.J. Choi, J. Acoust. Soc. Am. **131**, EL67 (2012)
12. B.K. Hoffmeister, D.P. Johnson, J.A. Janeski, D.A. Keedy, B.W. Steinert, A.M. Viano, S.C. Kaste, IEEE Trans. Ultrason. Ferroelectr. Freq. Control **55**, 1442 (2008)