

# Study on photoacoustic properties of bovine cortical bone

## ウシ皮質骨の光音響特性に関する検討

Yoshihiko Maekawa<sup>‡</sup>, Keita Yano, Taishi Hattori, and Mami Matsukawa<sup>1</sup> (<sup>1</sup>Doshisha Univ.)  
前川 慶彦<sup>‡</sup>, 矢能 啓太, 服部 大志, 松川 真美 (<sup>1</sup>同志社大)

### 1. Introduction

Diabetic patients have higher risks of fractures than healthy people and the evaluation of their bone strength is important. It is known that bone is primarily composed of hydroxyapatite and collagen<sup>[1]</sup>. The National Institutes of Health (NIH) consensus development panel has noted that both bone mineral density (BMD) and bone quality contribute to the bone strength<sup>[2]</sup>. Bone quality has many factors such as bone elasticity and microcracks. However, glycation due to diabetic diseases change properties of collagen in bone, which cannot be evaluated by X-ray techniques. Saito et al. have pointed out abnormal crosslinks of collagen in bone due to diabetes and aging<sup>[3]</sup>. One example of the abnormal crosslinks is known as advanced glycation end products (AGEs). These crosslinks are thought to affect the elastic properties of bone. For these reasons, investigation of collagen properties are important for bone quality assessment.

One technique to physically evaluate collagen properties in bone is the photoacoustic (PA) method, which is the energy conversion from light to sound in the material. Lashkari et al. reported characteristic changes of PA signal due to the collagen content in bone<sup>[4]</sup>, however, the evaluation of diabetic bone by the PA method has not been studied yet.

In this study, we experimentally measured the PA signals from bone specimens with and without glycation, to check the possibility of diabetes evaluation.

### 2. Material and Methods

#### 2.1 Bone specimen preparation

The first step of our study was to obtain 2D PA images from bone. One cortical bone specimen ( $7 \times 7 \times 0.5 \text{ mm}^3$ ) was fabricated from a femur of a 46-month-old bovine.

The next step is to evaluate the effect of glycation on photoacoustic properties of bone. Two cortical bone specimens ( $7 \times 3.5 \times 0.5 \text{ mm}^3$ ) were obtained from a femur of a 46-month-old bovine. These specimens were divided into glycated and reference specimens. All specimens were kept in an incubator at 37°C for 14 days in solutions (Normal saline (reference) or Normal Saline with D-(-)-Ribose for glycation (glycated)).

### 2.2 Ultrasonic measurements

#### 2.2.1 2D PA images

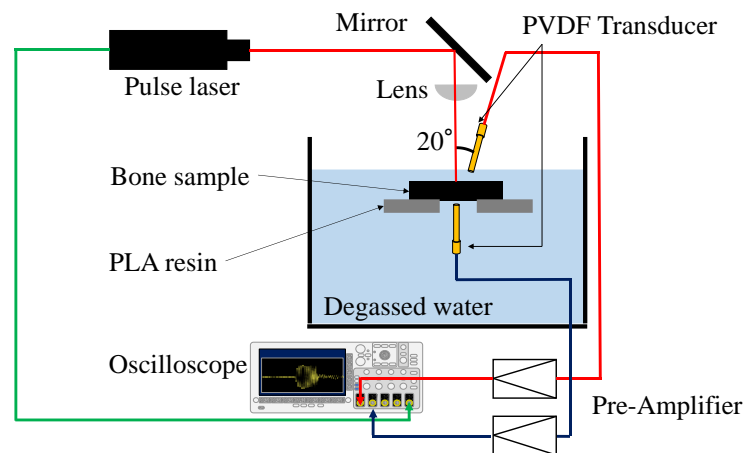


Fig.1 Experimental system.

Figure 1 shows the experimental system. We used a nanosecond pulsed laser (1064 nm, Cobolt Tor<sup>TM</sup> XS, HÜBNER PHOTONICS, pulse width 1.2 ns, beam diameter at the focal point 80  $\mu\text{m}$ ). A PVDF handmade transducer was placed above or under the bone specimen to measure generated waves. After amplification (SA430F5, NF Co.), the waveforms were observed with an oscilloscope (DPO7254C, Tektronix Co.).

Figure 3 (a) represents the area ( $1.3 \times 1.5 \text{ mm}^2$ ) scanned for the image. Some markers (black ink) and small holes were included in the area. We scanned this area with a step of 100  $\mu\text{m}$ , and analyzed the waves observed from the transducer above the bone. A two-dimensional image was obtained by measuring the peak to peak values of photoacoustic waves.

#### 2.2.2 Diabetic bone measurement

Using the nanosecond pulsed laser, we measured generated waves at 20 positions of each specimen by a PVDF transducer. We observed waveform by amplifying 66 dB (above the bone) or 46 dB (under the bone).

### 3. Results and Discussion

Figure 2(a) shows observed waveforms generated at the surface of the specimens. A strong first peak (A) was observed at the time of light

irradiation. It may be due to the pyroelectric effect<sup>[5]</sup>. Another peaks (B) seemed to be due to photoacoustic effect. Figure 2 (b) shows enlarged photoacoustic waves.

Figure 3 (b) presents scanned image of peak to peak values of photoacoustic waves. Here, the black ink area absorbs light and showed the large amplitudes. We also confirmed the needle holes. In addition, amplitude changes were observed between the area of the burr holes and its surrounding area.

Figure 4 (a) shows observed waveforms when the transducer was placed under the bone. We also confirmed waves similar to Fig. 2. Figure 4 (b) shows the enlarged photoacoustic waves. The photoacoustic waves were smaller than the first peak(A).

Figure 5 (a) shows normalized amplitudes of photoacoustic waves measured above the bone. Figure 5 (b) shows normalized amplitudes of photoacoustic waves measured under the bone. The amplitudes of the photoacoustic waves seemed larger in the reference specimen. In addition, the wave amplitude of the glycated specimens seemed smaller, especially when the waves were observed under the specimen. This result may indicate that the glycated specimen has larger attenuation inside the bone.

The present results come from the first trial of photoacoustic evaluation of glycated bones. The changes of collagen may be evaluated by the photoacoustic measurements as changes of wave properties.

#### 4. Conclusion

In this study, we investigated the effect of glycation on photoacoustic properties of bone. The generated waves seemed to depend on the glycation. However, more detailed investigations may be required, in addition to the study of bone pyroelectricity.

#### Acknowledgement

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#### References

- 1.M. Saito, et al, *Calcif. Tissue Int*, pp. 160-168, 79, (2006).
- 2.NIH, *JAMA*, 285, pp.785-795 (2001).
- 3.M. Saito, et al., *Osteoporosis Int.*, Vol.17, pp.1514-1523 (2006).
- 4.B. Lashkari et al, *Quant Imag Med Surg.*, Vol.20, 076016, (2015).
- 5.D. Khare et al, *Biomaterials*, Vol.258, 120280, (2020).

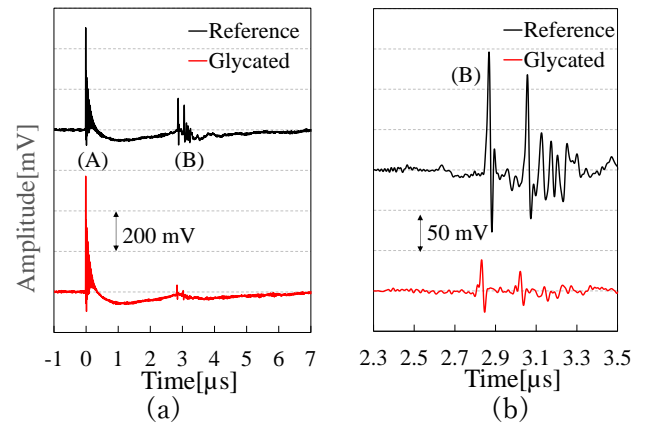


Fig.2 Waveforms observed above the bone.

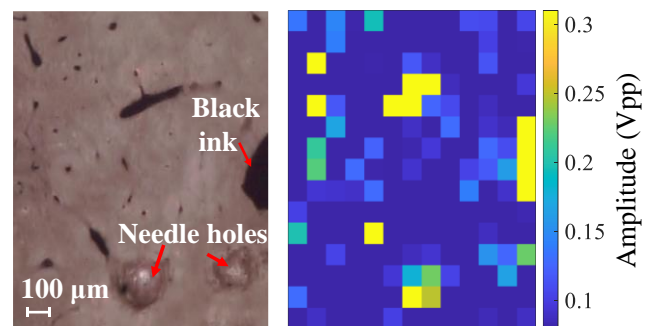


Fig.3 (a) scanned area of the bone, and (b) photoacoustic images of the bone.

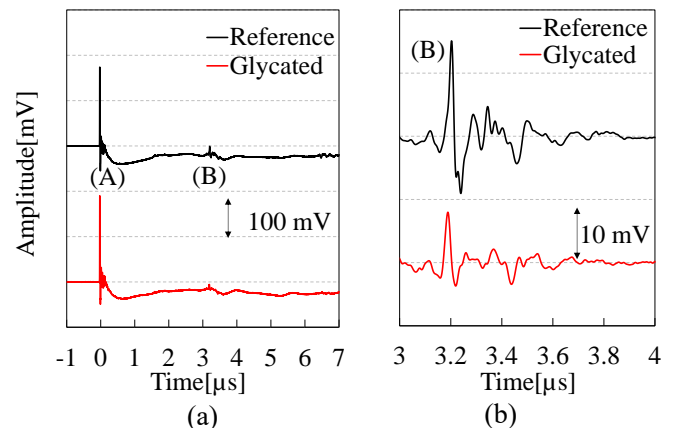


Fig.4 Waveforms observed under the bone.

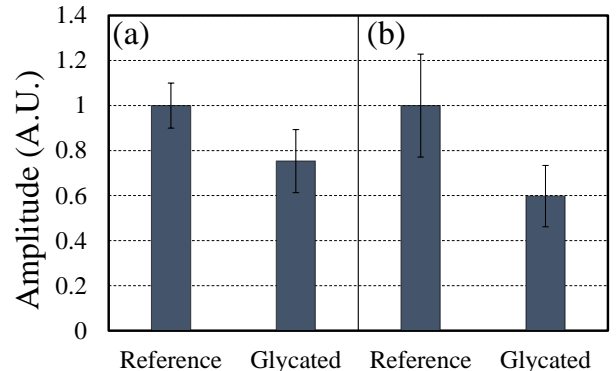


Fig.5 Normalized average amplitude of observed waves (a) above the bone, and (b) under the bone.