# Anisotropy of ultrasonically induced electric potentials in bovine cortical bone

ウシ皮質骨における超音波誘導電位の異方性

Tsukasa Nakamura<sup>1‡</sup>, Mineaki Takata<sup>1</sup>, Itsuki Michimoto<sup>1</sup>, Tomoya Oda<sup>1</sup>, Shinji Takayanagi<sup>1</sup>, and Mami Matsukawa<sup>1</sup> (<sup>1</sup>Doshisha Univ.) 中村 司<sup>1‡</sup>,高田 峰聖<sup>1</sup>,道本 樹<sup>1</sup>,小田 智也<sup>1</sup>,高柳 真司<sup>1</sup>,松川 真美<sup>1</sup> (<sup>1</sup>同志社大)

## 1. Introduction

Low-intensity pulsed ultrasound (LIPU) has attracted attention as a technique for healing bone fractures. This technique typically uses pulsed ultrasound from 1.5 to 3 MHz and shortens the healing time of bone fractures. It is known that the ultrasound stimulates bone cells and promotes the healing. However, the initial mechanism of the physical stimulation of ultrasound and cell response in bone remain unclear [1].

One possible mechanism is the piezoelectricity caused by the crystal structure of collagen and hydroxyapatite in bone. In 1953, Fukada and Yasuda have reported that the mechanical stress at low frequencies induces electrical potentials in bone [2]. Osteocytes might be stimulated by the electrical potentials or the mechanical stress. However, a few studies have investigated the piezoelectricity of bone in the MHz range. Then, we fabricated ultrasound transducers using bones as piezoelectric materials and confirmed very weak piezoelectricity in the MHz range [3].

Cortical bone is mainly composed of collagen and hydroxyapatite. They are highly orientated in the bone axial direction and cause strong anisotropy [4]. The purpose of this study is to observe the effects of bone anisotropy on the ultrasonically induced electrical potentials.

## 2. Material and Methods

Figure 1 shows the preparation method of the bone samples. Three types of cortical cylindrical bone sample (diameter; 10.5 - 11.0 mm, thickness;  $5.00 \pm 0.01$  mm) were extracted from the anterior part of the mid-femoral shaft of a 29 month-old bovine. Then, they were processed into the cylindrical samples with electrodes on the inside and the outside surfaces. The axes of the samples were along radial, tangential, and bone axis directions. Using these cylindrical bone samples as piezoelectric materials, we fabricated bone transducers for ultrasound reception.

-----

In the experiments, a PVDF focus transducer (diameter, 20 mm; focal length, 40 mm; custom made by Toray) was used as a transmitter. The transmitter and the bone receiver were set to cross at right angles in degassed water as shown in Fig. 2. In this setup, a function generator (33250A; Agilent Technologies) generated a short pulse (main frequency; 760 kHz) which was amplified to 70 Vp-p by a bipolar power supply (HAS 4101; NF). The ultrasonic pulse (sound pressure at the measurement point; 7.4 kPa p-p) was irradiated to the side of bone transducer so that the induced potentials were generated. The received signal was amplified 40 dB by a pre-amplifier (BX-31A; NF) and observed by an oscilloscope (DPO3054; Tektronix). The bone transducer was rotated at each 10 degree to check the anisotropy.



Fig. 1 Preparation of the bone transducers.



Fig. 2 Experimental system.

### 3. Results and Discussion

Figure 3 shows observed waveforms measured by the bone transducer of the tangential sample. Here, radial, tangential, and axial directions are defined as 1, 2, and 3 axes, respectively. It has been reported that the piezoelectric constants of bone at low frequencies are  $d_{11}$ =0.18 pC/N,  $d_{22} \le 0.01$  pC/N and  $d_{33}$ =0.15 pC/N [5]. The piezoelectric properties in this study also correspond to  $d_{11}$ ,  $d_{22}$  and  $d_{33}$ , and the results at high frequencies show the polarity change due to the anisotropy.

Next, we measured the waves by rotating the bone transducer at each 10 degrees. Figures 4 (a) and (b) show relationships between the amplitudes of induced electrical potentials and ultrasound irradiation directions in the bone transducers of the radial and tangential samples. The amplitudes showed maximum around 45, 135, 225 and 315 degrees, whereas they showed minimum in the radial  $(d_{11})$ , tangential  $(d_{22})$  and axial directions  $(d_{33})$ as in the previous low frequency study [5]. In addition, the polarity of the wave front changed every 90 degrees. Makino et. al. have irradiated longitudinal ultrasound to the front surface of circular bone samples (normal directions of the plates; radial, tangential, axial, 45° directions from tangential-axial plane and 45° directions from radial-axial plane). They have reported that receiving sensitivities of off axis bone transducers were higher than those of on axis bone transducers  $(d_{11}, d_{22} \text{ and } d_{33}).$ 

Figure 4 (c) shows relationships between the amplitudes of induced electrical potentials and ultrasound irradiation directions measured by the bone transducer of the axial sample. Even if the ultrasound propagation direction was changed, the amplitudes of the induced electrical potentials stayed almost constant. Yamato et. al. have reported almost uniaxial character of cortical bone and the isotropic tangential-radial plane [4]. The crystal structure of collagen, which contributes to the piezoelectricity of bone, is considered to be hexagonal and aligns in the bone axis direction. However, from these data, it could not be judged whether crystal structure of collagen was hexagonal or not.

#### 4. Conclusion

In this study, we investigated relationship between the induced electrical potentials and ultrasound propagation directions using cylindrical bone samples. The polarity and the amplitudes of the induced electrical potentials changed depending on the ultrasound propagation direction.



Fig. 3 Observed waveforms by the tangential sample.



(c) Axial sample. Fig. 4 Relationships between induced electrical potentials and ultrasound irradiation directions.

#### References

- 1. Padilla, Ultrasonics. 54, pp. 1125-1145 (2014).
- 2. Fukada et al., J. Phys. Soc. Jpn. 12, pp. 1158-1162 (1957).
- 3. Okino et al., Appl. Phys. Lett. 103, 103701 (2013).
- 4. Yamato et al., Calcif. Tissue Int. 82, pp. 162-169 (2008).
- 5. Anderson et al. Nature. 227, p. 491 (1970).