# Local ultrasonic wave velocities in articular cartilage measured by micro-Brillouin scattering technique

Brillouin 光散乱法による関節軟骨中の局所的音速評価

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## 1. Introduction

Articular cartilage has a function to absorb impact pressure and to prevent friction between bones. Actually, the articular cartilage is composed of cartilage cells and extracellular matrix (mainly collagen type II and proteoglycan). The proteoglycan in the collagen networks contains water. When the stress is applied, the cartilage releases water and the stress is distributed [1]. The extracellular matrix structure is important to perform normal cartilaginous functions.

For the evaluation of cartilage, MRI and ultrasonic diagnostic systems are used recent years. However, these methods cannot evaluate elastic properties. Nano-indentation is then used to evaluate nonuniformity of the cartilage [2], although this only measures elastic properties in the thickness direction and cannot evaluate the anisotropy.

In this study, we measured wave velocities in the cartilage by using a micro Brillouin scattering. This technique enables the measurements in the small area of the cartilage. Especially, we investigated wave velocity distribution, velocity anisotropy and the effect of water.

#### 2. Material and methods 2.1. Specimen

The articular cartilage specimens were obtained from the distal end of 31-month-old female bovine left femur (**Fig. 1**). We prepared two specimens in the plane of bone axis and medial-lateral direction. One was a dry specimen, which was dried well in the incubator at 37 °C. The other was a fully wet specimen. The specimen thicknesses were approximately 150  $\mu$ m and 200  $\mu$ m, respectively.

# 2.2. Brillouin scattering technique

Brillouin scattering measurements were carried out with a six-pass tandem Fabry-Pérot interferometer using a solid state laser with wave length of 532 nm. The system included an optical microscope for Raman scattering. The actual spot

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diameter of the focused laser beam in the specimen was approximately 10 µm.

The reflection induced  $\Theta A$  scattering geometry is shown in **Fig. 2**. The geometry enables the simultaneous observation of phonons propagating in each direction of wave vectors  $q^{\Theta A}$ and  $q^{180}$  in one measurement [3]. In this study, focusing on the  $q^{\Theta A}$  which is the in-plane phonon wave vector, we measured the longitudinal wave velocities in the cartilage. To evaluate the effect of the water content on the cartilage, we also measured the velocities of specimens under the dry and wet conditions.



Fig. 1 Specimen preparation.



Fig. 2 Reflection induced  $\Theta A$  scattering geometry.

#### 3. Results and discussion

### **3. 1. Wave velocity distribution and anisotropy**

**Figure 3** shows velocity distribution in the dry articular cartilage. Wave propagation direction was parallel to the surface of the subchondral bone. The range of the wave velocity was  $3.36-3.83 \times 10^3$  m/s. This means that the articular cartilage has

complex elastic properties. The deep layer near the subchondral bone tends to show high wave velocities. It is reported that cartilage contains a significant portion of cartilage cells in the deep layer [1]. The high velocity in the deep layer seems to depend on the cartilage cells.

In addition, it is reported that the articular cartilage has different anisotropic character due to the layer [1]. We measured anisotropies of cartilage by rotating the specimen. As shown in **Fig. 4**, the anisotropies of cartilage (difference between maximum and minimum velocity / maximum velocity) were about 2.6 % at position C and 1.2 % at position B. Thus, weak anisotropies were found in the cartilage. The anisotropic character seemed to mainly depend on the orientation of the collagen. The anisotropies of cartilage were weaker than that of trabecula (4.5 %) reported in former studies [4].

# **3. 2.** Wave velocities in the wet and dry specimens

Measured spectra obtained from (a) the dry specimen and (b) the wet specimen at position A are shown in **Fig. 5**. From the dry specimen spectrum, wave velocity was estimated as  $3.58 \times 10^3$  m/s. In the wet specimen, the wave velocity was around  $2.04 \times 10^3$  m/s. In the wet specimen, the velocity was much lower than that in the dry specimen. The results suggest that water content in the cartilage affects the elastic property. Additionally, this wave velocity difference under dry and wet conditions was much larger than that of the artificial isotropic collagen film (velocity difference was about 270 m/s).

#### 4. Summary

Wave velocities in the cartilage were measured by a micro Brillouin scattering technique. Wave velocity distributions in the cartilage were complex. Anisotropies of wave velocity in the specimen were different due to the position in the layer. In addition, the velocity in the dry specimen was much higher than that in the wet specimen, telling that the water content affects elastic properties.

#### References

- K. Fujii, et al., "The biology of the bones and cartilages", Kanehara publication, pp.90-92 (2002) [in Japanese].
- 2. O. Franke, et al., Acta Biomaterialia., **3**, pp. 873-881 (2007).
- 3. J. K. Krüger, et al., J. Phys. D., **31**, pp. 1913-1917 (1998).
- 4. K. Fukui, et al., Proc. Forum Acusticum, pp. 2841-2844 (2011).





Fig. 5 Observed spectra by Brillouin scattering from (a) the dry specimen and (b) the wet specimen.