# Verification of stability of depth assessment in threedimensional acoustic impedance analysis

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## 1. Introduction

Conventional acoustic impedance evaluation of biological tissues using scanning acoutic microscopy has focused on the relationship between the impedance and the surface information, i.e., the two-dimensional tissue structure of the target tissue. However, in order to correspond to clinical ultrasound diagnosis, the acountic impedance analysis that reflects the three-dimensional structure of the tissue interior is necessary. In this study, we calculated three-dimensional acoustic impedance using ultrahigh-frequency ultrasound by signal separation based on the autoregressive (AR) model, and evaluated the relationship between the internal structure of biological tissue and its acoustic properties.

### 2. Materials and Methods

### 2.1 Data Acquisition

The back muscles were removed from 8week-old non-obese type 2 diabetic rats (SDT/Jcl), and short-axis sections cut orthogonally to the muscle fibers were used for evaluation. A scanning acoustic microscopy (modified AMS-50S, Honda Electronics) incorporating a 250 MHz ZnO transducer (Fraunhofer IBMT) with a spatial resolution of 7  $\mu$ m in the aperture direction at the focal point was used to measure the sample on a 50  $\mu$ m thick polystyrene film dish (HPS-3805, Honda Electronics). The sample was scanned twodimensionally (1,200  $\mu$ m × 1,200  $\mu$ m) from the underside of the dish and the echo data were collected at a sampling frequency of 2 GHz with 8 bit quantization.

### **2.2 Calculation Principle**<sup>[1]</sup>

Under the measurement conditions in this study, the acoustic impedance  $Z_{target}$  of the evaluation target can be calculated by the following equation using the acoustic impedance  $Z_{ref}$  of the known reference material (purified water) and the acoustic impedance  $Z_{sub}$  of the substrate (polystyrene film dish).

$$Z_{target} = \frac{1 - \frac{S_{target}}{S_{ref}} \cdot \frac{Z_{sub} - Z_{ref}}{Z_{sub} + Z_{ref}}}{1 + \frac{S_{target}}{S_{ref}} \cdot \frac{Z_{sub} - Z_{ref}}{Z_{sub} + Z_{ref}}} Z_{sub} \quad (1)$$

The acoustic impedance of purified water and the substrate (polystyrene) were set to  $1.50 \times 10^6$  and  $2.37 \times 10^6$  kg/m<sup>2</sup>/s, respectively, based on the results of the preliminary evaluation. *S* represents the signal intensity of each signal at a given frequency, and *S*<sub>ref</sub> and *S*<sub>target</sub> are the reflected signals from the purified water and the measurement sample, respectively.

### 2.3 Acoustic Impedance Evaluation

In all  $300 \times 300$  observation points in the acquired 3D RF echo data, the signals were separated by applying the sixth-order AR model set in the preliminary study <sup>[2]</sup> to remove a large number of interfering components in the signals to be evaluated. The original signal and four signals whose bandwidths were limited to -30, -25, -20, and -15 dB of the peak frequency were evaluated as input signals, and the results were compared, taking into consideration the reduction of specific frequency components that occur in very high-frequency ultrasonic measurements. Of the six separated echo components, the signal of the first component was used for analysis. The A-mode signal of the first component at each measurement point was upsampled by a factor of 10, and the acoustic impedance was calculated using the amplitude envelope after logarithmic compression. Acoustic impedance maps were created at each time (= depth) every 50 ps based on the time when the echo amplitude was the largest.



Fig. 1 Signal separation by AR model.

## 3. Results

Figures 2 shows the acoustic impedance at each depth before (a-1) and after (b-1~4) signal separation. Red lines show the signal determined to be the target (biological tissue), blue lines are the amplitude of the signal from the reflector used as a reference, and black lines are the acoustic impedance calculated from both signals of up-sampled first component and reflector using equation (1). The acoustic impedance without waveform separation (a-1) and with the frequency domain set to -30 dB (b-1) and -25 dB (b-2) at the time of separation shows unstable values in the deep region. On the other hand, the values of (b-3) with a frequency range of -20 dB and (b-4) with a frequency range of -15 dB are about 1.5 Mrayl at all depths, which are evaluated as values that are consistent with general biological tissues. Figure 3 shows the results of Fig. 2(a-1) and (b-3) as two-dimensional acoustic impedance maps at each depth. Although the upper and lower rows differ in the depth in which the acoustic impedance of the muscle tissue is strongly characterized, Figure 3(b), which shows waveform separation in the -20 dB frequency band, is considered more valid considering the structural differences between the surrounding tissue and the muscle tissue and the literature values for each tissue.

## 4. Conclusions

The acoustic impedance of rat back muscles were evaluated from the echoes at each depth and confirmed the relationship between the internal structure of the biological tissue and its acoustic properties. We confirmed that the evaluation was more robust than the conventional method and effective for understanding the tissue properties in the depth direction.

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## References

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Fig. 2 Target and reference signals and acoustic impedance in each frequency domain.



Fig. 3 Acoustic impedance maps for each depth before signal separation (a) and in the -20 dB frequency domain after signal separation (b).

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