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Tissue analysis of electrically sealed vessels using an ultrasonic microscope

超音波顕微鏡による血管シーリング時の組織変性評価

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1. Introduction

Acoustic analysis of various biological tissues and understanding of correlation between changes in acoustic parameters and tissue structure are greatly important, because acoustical properties of tissues could provide another insight to conventional histopathological analysis. Rough and average acoustic properties of biological tissues have been measured, however, understanding of changes in acoustic parameters associated with pathological or degenerative changes are still not of satisfactory.

Electric bipolar vessel sealing systems are widely used in various kind of surgeries for dissecting tissues without bleeding. Stable and reliable sealing performances are required when major blood vessels are divided with these devices. Thus, we have been evaluating vessel sealing performances of them with conventional histopathological analysis of the sealed specimens. In this study, the possibility of new analysis method which could provide additional information of degenerative condition of vessel by ultrasound microscopy was tested.

2. Methods

Bilateral carotid arteries were harvested from 5 castrated male domestic pigs of 100 kg in body weight under general anesthesia and were stored at -20°C until sealing analysis. Enseal PTC system (Ethicon Endo-sugery, Japan) was used for vessel sealing. Total of 9 sealing was performed with varied activation time from 0 (clamping 8 seconds only) to 8 second with increment by one second. Sealed vessel specimens were fixed with 10% formalin solution and embedded in paraffin blocks. Three consecutive sections were prepared from all paraffin blocks, two with 4 μm thicknesses for hematoxylin-eosin (HE) or Heidenhain's AZAN trichrome staining, and the other with 10 μm thickness for ultrasound microscopy. AZAN staining was used to detect thermal degeneration if tissues as indicated in the report by Kinoshita et al.[1]. Distribution of speed of sound of the sections was calculated with a modified type of AMS-50SI (Honda Elec. Inc., Japan), acquiring echo signals in the domain of 2.4 mm × 2.4 mm by two-dimensional operation of the transducer of 80 MHz center frequency by 300 × 300 points[2,3,4]. Experimental protocol was reviewed and approved with the internal review board.

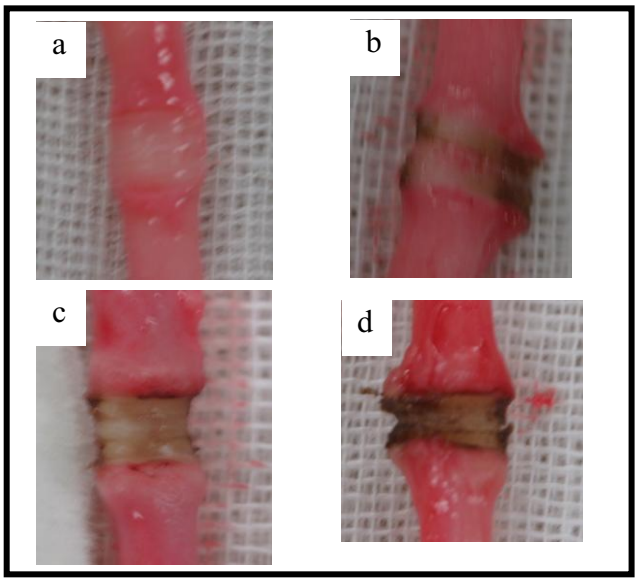

Fig. 1: General appearances of sealed site on the vessel specimens with a) 0, b) 2, c) 4, and d) 6 seconds sealing activation.

3. Results

General appearances of sealed sites of the vessel specimens with sealing activation for 0, 2, 4, and 6 seconds are shown in Fig. 1. The deformity of the sealing site increased as the sealing activation time increased. Histological findings of the specimens with conventional tissue staining and ultrasound microscopy are shown in Figure 2 (0 second sealing activation; only clamping) and Figure 3 (6 seconds sealing activation).

Conventional histopathological analysis of the specimen without sealing activation showed normal tissue structure, and inner, medium, and outer layer of the vessel were clearly distinguishable. Especially, medium layer of the

vessel homogeneously stained red and blue with HE and AZAN staining, respectively (Fig. 2a and 2b). Layer structures of the vessel were similarly distinguishable with ultrasound microscopy showing speed of sound with 1405-1515 m/s for inner layer, 1640-1823 m/s for medium layer, and 1831-1982 m/s for outer layer (Fig. 2c). The speed of sound in adipose tissue accompanied by the vessel showed 1434-1506 m/s.

In contrast, vessel walls are somehow compressed and apparently coapted at the sealing site of the specimen underwent 6 seconds sealing activation (Fig. 3). Both inner and outer layer of the vessel were fused with medium layer, and the layer structure of it became ambiguous (Fig. 3a). Furthermore, outer half of the vessel wall stained red with AZAN staining at the same site, indicating thermal degeneration of the tissue (Fig. 3b). Distribution of the speed of sound calculated at the same site showed more heterogeneity compared with the histological finding with AZAN staining, ranging from 1616 to1993 m/s (Fig. 3c).

Similar findings were obtained from histopathological and ultrasonic analyses of the vessels underwent sealing activation for 1-5, 7 and 8 seconds (data not shown in here. None of the parameter mentioned above showed linear correlation with the activation time.

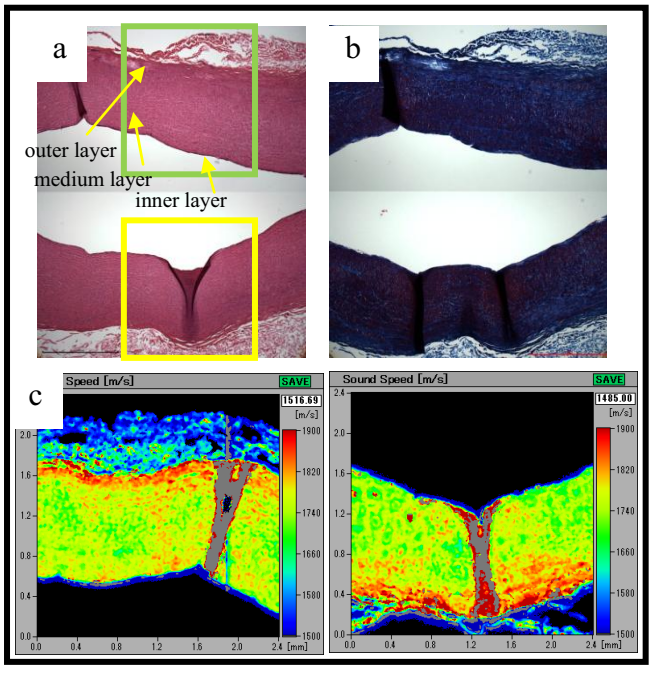

Fig. 2: Histopathological and ultrasound microscopy of the vessel with 0 second sealing activation (a: HE staining, b: AZAN staining, c: speed of sound).

4. Discussions

Major physiological difference between the specimens shown in Fig. 2 and that in Fig. 3 was heating up of the tissue by energization. Thus, clamping by the jaws of the sealer (0 second sealing activation) itself would not exert any influence on tissue structure. In contrast, combination of compression and heating up the vessel walls led to sealed and thermal degeneration of the tissue indicated in Fig. 3b.

Distribution of the speed of sound on the specimen underwent 6 seconds sealing activation showed much more heterogeneity compared with the conventional histological findings, suggesting it could associate with sealing strength of the vessels.

5. Conclusions

We evaluated performances of electric bipolar vessel sealing system with animal tissues and conventional histopathological analysis. Ultrasound microscopy of deformed vessel was suggested to provide additional information for the sealing strength of the vessels compared with the common histopathological analyses. Further study is required to elucidate its usefulness in this field.

References

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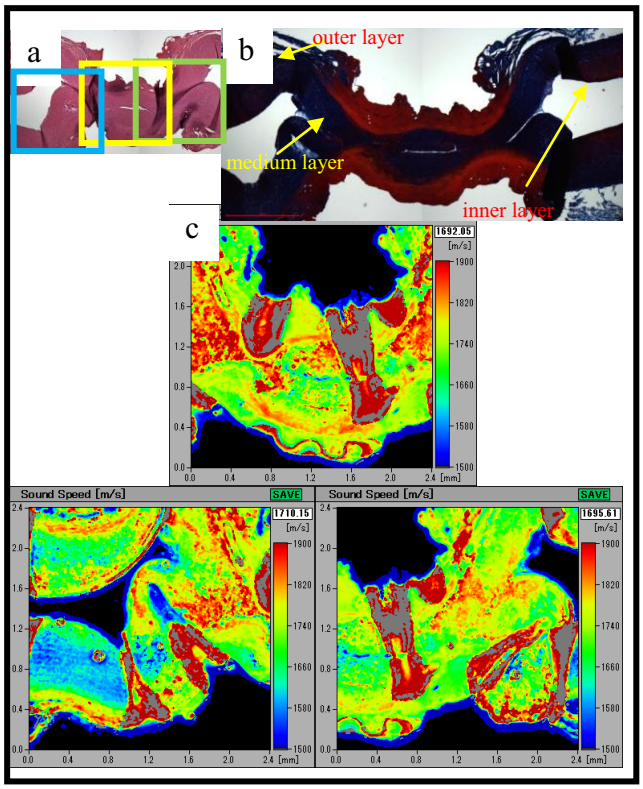

Fig. 3: Hisitopathological and ultrasound micoroscopy of the vessel with 6 seconds sealing activation (a: HE staining, b: AZAN staining, c: speed of sound).