CONTRAST-ENHANCED MICRO-COMPUTED TOMOGRAPHY IMAGING OF THE RODENT AND HUMAN INNER EAR

Annalisa De Paolis (1), Luis Cardoso (1)

1. City University of New York, Biomedical Engineering, NY, NY, USA

Introduction

Imaging of the inner ear is critical to better understand changes on hearing function due to pathologies such as hearing loss. Synchrotron radiation-based µCT imaging provides high resolution and contrast, allowing researchers to visualize soft tissue structures in the inner ear [1, 2]. Unfortunately, access to a synchrotron-based imaging system is often limited. Laboratory benchtop µCT imaging systems can provide high-resolution images of hard tissues, but not soft tissues. In this study we tested the ability of a µCT system and Iodine Potassium Iodine (IKI) staining to provide improved images of both soft and hard tissues in the intact murine and human cochleae.

Methods

A human temporal bone was explanted right after death from a donor (female, 62 yo) and the bullae of a Wistar rat (14 do) were harvested after death. Fixation was carried out in 10% buffered formaldehyde for 1 week and the inner ear was isolated [3]. Both samples were stained with 50% IKI/Buffer saline solution (0.1 Normal/0.05 M, Fisher Scientific, USA) and µCT-scanned (1172, SkyScan, Belgium) 1, 2, 3, 5 and 7 days after incubation in IKI to determine the time required for the contrast agent to diffuse and reach equilibrium within the tissues. Scanning was performed at 100 kV and 100 µA, using a 0.5 mm thick aluminum filter. Images of the inner ear were acquired at 3-µm and 2.1-µm resolution for the human and the rat cochleae respectively. Figure 1A–B shows cross and axial sections of the human cochlea. Scala vestibuli, SV and the scala tympani, ST, auditory nerve, spiral ligament, oval and round window membranes, as well as basilar and tectorial membranes were visible in the scans. In Figure 1C and in the right panel is a 3D reconstruction of the human ear displaying outer, middle and inner ear. In Figure 1D, a 3D reconstruction of the SV and ST and scala media, SM is shown. In Figure 1E the finite element mesh of the 3D volume representing SV and ST is also shown and in Figure 1F the regions corresponding to the oval and round window membranes are highlighted. Figure 2 show the cross sectional area of the rodent ear after 5 days of incubation in the IKI solution. SV, ST, SM as well as auditory nerve, spiral ligament, basilar memebrane, tectorial membrane and Reissner's membrare were clearly recognizable.

Results

Cochlea bone microarchitecture and soft tissues morphology were shown in great detail when samples were treated with IKI and imaged with µCT at high-resolution (Figure 1). The shapes of Reissner's, auditory nerve, basilar and tectorial membranes and the contour of the Organ of Corti (OC) can be extracted using this technique. However, no feature of the OC such as hair, pillar and phalangeal cells can be clearly observed. Five days incubation was considered the most appropriate for a successful imaging of the tissues in the human and rat cochlea

Discussion

In this study we tested the ability of IKI to provide increased image contrast when compared to the X-ray absorption of soft tissues in the human and rodent inner ear. The presented data demonstrated that a contrast-enhanced µCT imaging approach can be used for the 3D imaging of soft tissues in the human and rodent inner ears to investigate 3D changes of soft tissues due to hearing pathologies in human and the rodent inner ear samples.

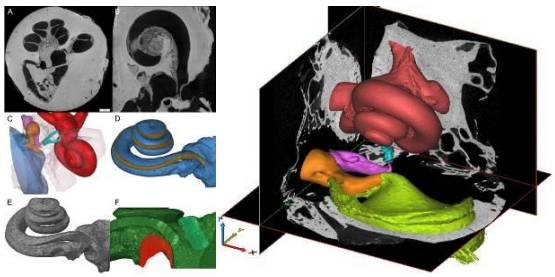

Figure 1: High-resolution 3D reconstruction of Human Ear from microCT images.

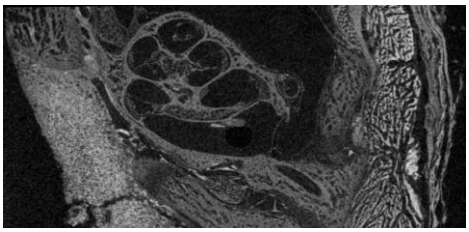

Figure 2: High-resolution contrast-enhanced microCT images of a rodent ear.

References

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