μCT scan of a cochlea obtained at μ-VIS X-Ray Imaging Centre, University of Southampton

UNIVERSITY OF Southampton

Investigating immune priming in the auditory system as a cause of variable outcomes after cochlear implantation

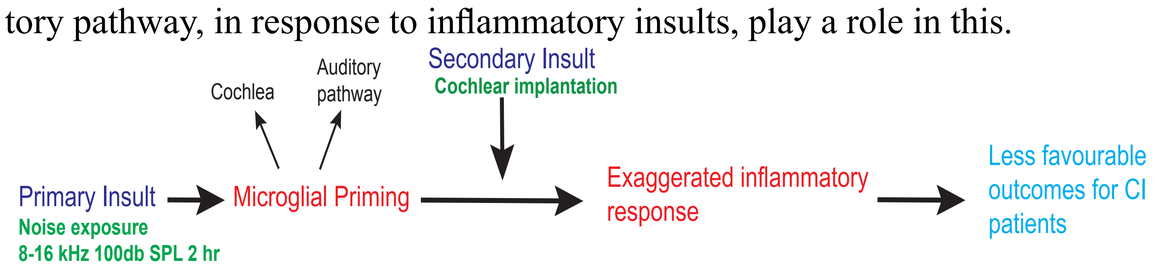
K. Hough[1], A. Sanderson[1], L. Anderson[2], B. Impey[4], C. Verschuur[3], T. A. Newman[4]

[1]Institute of Sound and Vibration Research, Faculty of Engineering and the Environment, University of Southampton, UK; [2]Ear Institute, University College London, UK; [3]Auditory Implant Centre, University of Southampton, UK; [4]Faculty of Medicine, University of Southampton, UK

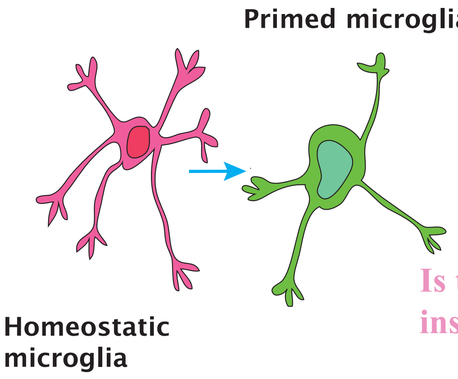
Introduction

Hearing loss affects 466 million people worldwide. People experience hearing loss due to loss of, or damaged hair cells in the cochlea, the inner ear. Hair cells lose their ability to convert mechanical energy from sound waves into neural impulses. Cochlear implants (CIs) are auditory prostheses which replace the function of damaged sensory cells in the cochlea. Despite the huge success of CIs, some individuals do not do as well with their implants and for some people their implants do not work at all.

We hypothesise that the priming of innate immune cells in the cochlea and auditory pathway, in response to inflammatory insults, play a role in this.



What is priming?



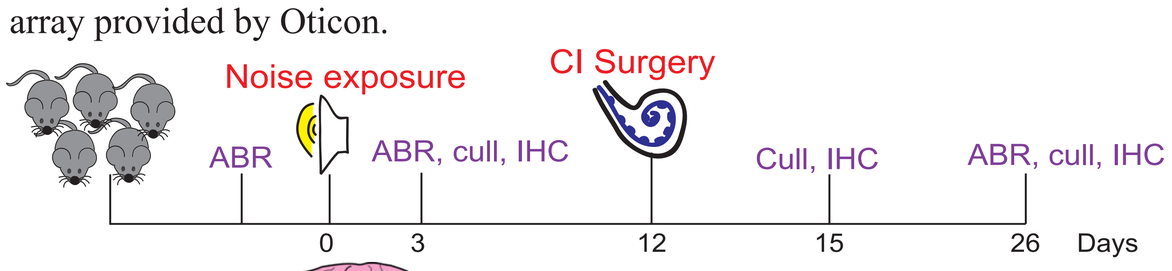
Priming describes a change in phenotype and expression profile whereby microglia and macrophages exhibit an exaggerated inflammatory response to a second stimulus having previously been exposed to an initial 'trigger' stimulus.

Is the variability between individuals after cochlea insults linked to microglial/macrophage priming?

Methods

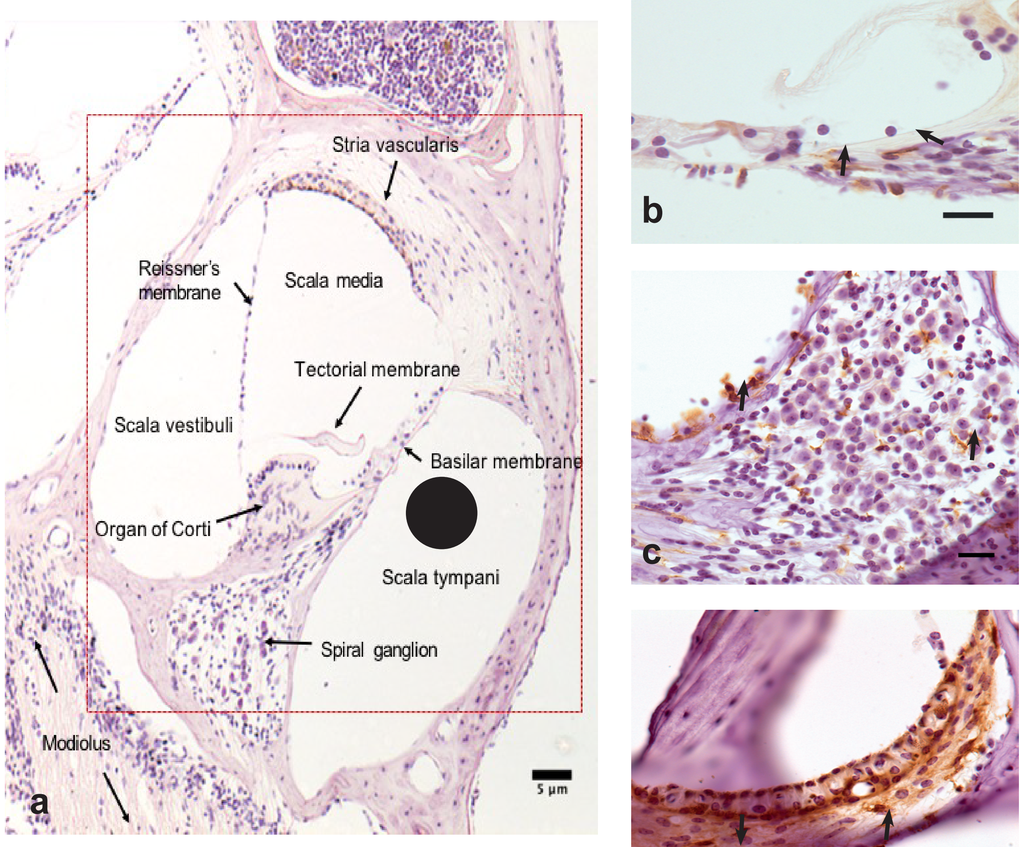
Immunohistochemistry (IHC): The presence and immunoreactivity of inflammatory markers will be measured from tissue collected from the cochlea and central auditory pathway of noise-exposed and control (sham-exposed) CBA mice.
Auditory Brainstem Response (ABR): ABR were recorded using sub-dermal electrodes positioned on the bullae and vertex in response to 50 μs clicks and tone pips at sound levels from 10-80 dB SPL. All stimuli were presented from a pre-calibrated, free-field speaker positioned 45° from midline.
μCT: A single scan will image the cochlea samples complete internal 3D structure at high resolution with the aim to corroborate this information with IHC data, to determine damaged regions of the cochlea after cochlear implantation.
Gene expression: The expression of genes that may be involved in the priming of innate immune cells will be investigated in situ, with high specificity and sensitivity using RNAscope.

Mouse strain: Male CBA mice
Initial insult - Noise exposure: Octave-band noise (8-16 kHz), 100 dB SPL was presented free-field to anaesthetised mice for 2 hours.
Secondary insult - Cochlear implantation**:** Implanted with a functional electrode array provided by Oticon.



From Ear to Brain

Figure 1. Haematoxylin stained mid-modiolar cochlea section of a 22-month-old C57 mouse (a) and Iba1 stained cochlea sections of a 22-month-old C57 mouse counterstained with haematoxylin (b,c,d). C57 mice are a model for age-related hearing loss and have a greater number of Iba1-positive cells of an activated morphology in older mice compared to young.
a. The three scalae that form one cochlea turn and the three main regions of interest within this turn are labelled. A cochlear implant would sit in the scala tympani as shown by the black circle. Scale bar: 5 μm.
b, c, d. Organ of Corti (b), spiral ganglion (c), stria vascularis (d). This figure indicates that there is a population of macrophages/microglia in different regions across the cochlea, which could play a role in the sensory, neuronal and structural functions of the cochlea. Scale bar: 20 μm.



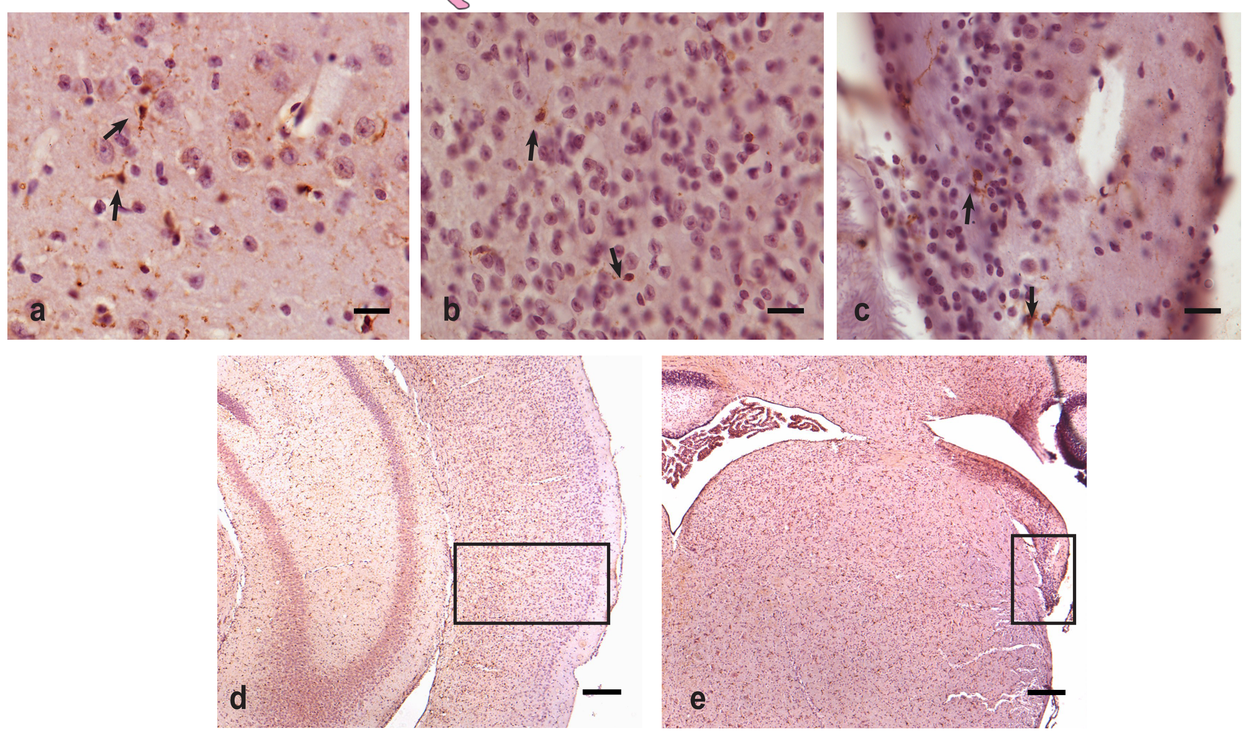


Figure 2. Iba1 stained brain sections counterstained with haematoxylin from a 9-month-old noise-exposed CBA mouse, from regions along the auditory pathway. Iba1-positive cells are indicated with a black arrow. Scale bar: 20 μm.
a, b, c. Brain sections with Iba1-positive cells in the auditory cortex (a), inferior colliculus (b) and cochlear nucleus (c), which indicates there is a population of microglial cells in the main regions along the auditory pathway. We will investigate whether these cells become primed in response to cochlea and systemic inflammatory insults.
d, e. Cortical section with the auditory cortex (d) and cerebellar section with the ventral cochlear nucleus (e) highlighted by a black box.

Figure 3. ABR thresholds from 9-month-old CBA mice before and 3 days post-noise exposure (8-16 kHz 100 db SPL 2 hrs). This figure shows increased thresholds post-noise exposure compared to before, indicating functional damage to hearing function following noise exposure.

Findings and Future Work

- This level of noise exposure causes an increase in hearing threshold 3 days post-injury.
- Iba1-positive cells are expressed across the cochlea and auditory pathway indicating a population of microglia/macrophages that could become primed as a result of a cochlear or systemic insult.
- Changes in phenotype and regional distribution of microglia and macrophages after cochlear insult will be investigated using different antibodies. Whether priming leads to a heightened inflammatory response upon a secondary insult (cochlear implantation) will be determined by measuring cytokine production.

Understanding whether microglia and macrophages become primed after initial insults and the effect on the subsequent inflammatory response, will contribute to determining whether robust control of inflammation will improve hearing outcomes after cochlear implantation.

EPSRC
Engineering and Physical Sciences Research Council

oticon MEDICAL Because sound matters

klh1g13@soton.ac.uk