*Introduction*: This research project seeks to identify genetic pathways predisposing to cholesteatoma. Familial clustering of cholesteatoma has been observed in East Anglia (Prinsley 2009). DNA sequencing has advanced so that whole exome sequencing of affected and unaffected individuals is now feasible.

*Methods*: A database of East Anglian families with cholesteatoma forms the core recruitment group for this study. However, the British Society of Otology (BSO) network could help identify other families. Pedigree charts and blood/saliva samples will be obtained from affected families for DNA extraction.

In the second stage, exome sequencing will be coupled to a linkage analysis in the families in which cholesteatoma is segregating. In conjunction with the pedigree mapping, we will have an opportunity to identify genetic polymorphisms predisposing to formation of cholesteatoma, and by using multiple affected families, to identify recurrent pathways or genes identified through this methodology.

*Results*: A research team of clinicians and scientists has been assembled and a systematic literature review has been carried out. Data extracted from the literature review will be used to identify pathways to focus on during the filtering steps to identify variants of interest that co- segregate with the disease phenotype. Funding has been secured from the Royal College of Surgeons of England and from the Rosetrees Foundation. The project will be adopted on to the NIHR Portfolio subject to Research Ethics Approval. The whole exome sequencing and analysis will be performed at The Genome Analysis Centre in Norwich.

*Conclusions*: A project has been created to identify genetic causes of cholesteatoma.

By selecting the right families, the project has potential to yield information that may widen our understanding of the disease pathophysiology.

doi:10.1017/S0022215116004126

## Genetics in Otology (R831)

### ID: 831.2

### Gene expression profiling reveals expression of tumor-relevant

Presenting Author: **Johannes Greiner**

Johannes Greiner, Janine Müller, Jörg Ebmeyer, Holger Sudhoff

*Klinikum Bielefeld Mitte*

*Learning Objectives*: Cholesteatoma is a destructive, potentially life-threatening lesion of the middle ear Cholestatoma tissue expresses tumor markers SERPINB3 and SERPINB4 Oncogenes like Lipocalin 2 are upregulated in Cholestatoma tissue, while tumor suppressor gene are downregulated.

*Introduction*: Cholesteatoma is a gradually expanding destructive epithelial lesion within the middle ear, which leads to extensive tissue destruction in the temporal bone followed by conductive and sensorineural hearing loss and facial nerve palsy. To develop new treatment strategies, gaining further insights into the complex gene regulation and signaling underlying the formation and progression of cholesteatoma are mandatory.

*Methods*: Gene expression profiling of cholesteatomas and regular external auditory skin from 17 patients via full genome micro-arrays containing 19,596 human genes followed by validation using real time PCR analysis.

*Results*: Full genome micro-arrays showed significantly increased expression of 811 genes in cholesteatoma tissue compared to regular external auditory skin, while 334 were found to be downregulated. Next to matrix metalloproteases MMP9, MMP10 and MMP12, the anti-apoptotic genes BCL2L1 and A20 were upregulated in cholesteatoma tissue. Providing a further linkage to tumorigenic tissue, expression of the tumor markers SERPINB3 and SERPINB4 as well as the oncogene Lipocalin 2 was increased in cholesteatoma tissue in comparison to external auditory skin. Accordingly, downregulation of the cell adhesion molecule cadherin 18 as well as the tumor suppressor gene inhibitor ID4 was observed in cholesteatoma tissue. Linking the characteristic expression of tumor-relevant genes in cholesteatoma to inflammation, the inflammation-related calcium binding protein S100A7A was found to be highly upregulated.

*Conclusions*: The Expression profile of cholesteatoma was found to be similar to a tumorigenic and chronically inflamed tissue, giving new insights in the complex biology of cholesteatoma.

doi:10.1017/S0022215116004138

## Genetics in Otology (R831)

### ID: 831.3

### Molecular pathology of cochlear gap junction in GJB2 associated hearing loss

Presenting Author: **Kazusaku Kamiya**

Kazusaku Kamiya, Ichiro Fukunaga, Katsuhisa Ikeda

*Juntendo University Faculty of Medicine*

*Learning Objectives*:

*Introduction*: Hereditary deafness affects about 1 in 2000 children and GJB2 gene mutation is most frequent cause for this disease. GJB2 encodes connexin (Cx) 26, a component in cochlear gap junction. We recently demonstrated that the drastic disruption of gap junction plaque (GJP) macromolecular complex composed of Cx26 and Cx30 are critical pathogenesis starting before hearing onset (Kamiya *et al.,* 2014, *J Clin Invest* 124, 1598–1607). To develop the effective therapy for GJB2 associated hearing loss, restoration of gap junction plaque (GJP) macromolecular complex using virus vectors or multipotent stem cells such as induced prulipotent stem (iPS) cells and mescenchimal stem cell (MSC) are expected to rescue the hearing function of GJB2 related hearing loss.

*Methods*: Mouse induced pluripotent stem cells (iPS) were used for generation of Cx26-expressing cells with proper gap junction plaque between the cells. Adeno associate virus (AAV) were used for the GJB2 gene transfer and restoration of GJP.

*Results*: By diffentiation of iPS cells, we generated the Cx26-expressiong cells with large gap junction plaque as cochlear cells. Cochlear delivery of Gjb2 using AAV significantly improved the auditory responses and development of the cochlear structure of Cx26f/fP0Cre mice (Iizuka, *Hum Mol Genet*, 2015, 24(13):3651–61).

*Conclusions*: Using cell therapy or gene therapy to restore hearing in the mouse models of Gjb2-related deafness may lead to the development of effective therapies for human hereditary deafness.

doi:10.1017/S002221511600414X

## Genetics in Otology (R831)

### ID: 831.4

### Is Cholesteatoma heritable and how can we find the genes involved?

Presenting Author: **Mahmood Bhutta**

Mahmood Bhutta

*Royal National Throat Nose and Ear Hospital*

*Learning Objectives*: To review evidence that cholesteatoma is heritable, and to discuss methods that can be used to ascertain genetic pathways involved.

Abstract is for the round table on "Genetics in otology"

The aetiology of cholesteatoma remains elusive. Those with a history of chronic mucosal disease are susceptible, but only a few such individuals will develop cholesteatoma. What evidence is there that cholesteatoma is a heritable disorder, and what methods can we use to elucidate genetic susceptibility?

I will discuss evidence from a recent systematic review of the heritability and genetics of cholesteatoma. This evidence includes reports of familial clustering of disease, and family history in the Kibutz population of Israel. Presence of disease in certain syndromes, in particular congenital malformation syndromes of the head or ear, also suggests genetic pathways are perturbed in cholesteatoma, and that a relatively small number of loci may be relevant.

I will introduce the other speakers for this session, and outline epidemiological and laboratory methods that can be exploited to further research molecular and genetic pathways involved in cholesteatoma. I will discuss how the discovery of such pathways could lead to potential clinical benefit.

doi:10.1017/S0022215116004151

## Free Papers (F832)

### ID: 832.1

### Anatomical understanding in canal wall down mastoidectomy using a medical image processing system – simulation and education of ear surgery

Presenting Author: **Kazunori Nishizaki**

Kazunori Nishizaki[1], Yuko Kataoka[2], Yorihisa Orita[2], Shin Kariya[2]

*[1]OKayama University, [2]Okayama University*

*Learning Objectives:* To understand surgical anatomy of the temporal bone using a medical imaging processing system.

*Introduction*: Although canal wall down mastoidectomy still plays an important role in treatment of cholesteatoma, the chance of performing this procedure appears to be decreasing by appropriate intervention for ear diseases that develop cholestatoma. The decreasing chance to master this technique should be compensated by other methods. As one of the alternatives we introduce a simulation and education method of ear surgery using a medical image processing system.

*Methods*: Sagittal 2 and 3 dimensional reconstructive images (DRI) of the temporal bone CT scan are made for this purpose using a three-dimensional image analysis system volume analyzer (SYNAPSE VINCENT, Fuji Film Co, Tokyo, Japan).

*Results*: Sagittal 3DRIs introduced here show, in the order from lateral to medial, the antrum cavity, the prominence of the lateral semicircular canal, the incus body, the malleus head, the bridge being formed, the second genu of the facial nerve canal, the bridge resected at the level of the malleus neck, the mastoid segment of the facial nerve canal, the completely resected bridge, the lateral semicircular canal, and the completely opened facial recess. These images also show that the lateral wall of the attic has anterior-posterior and superior-inferior slants. 2DRIs parallel to the lateral wall of the attic show that the resection of the bridge parallel to the lateral wall is safe without risk of injury to the ossicles, the facial nerve, and the inner ear. However, sagittal 2 and 3DRIs should be evaluated for each patient due to individual differences in the temporal bone anatomy and bone structural changes affected by the disease.

*Discussion and Conclusions*: Since ear surgery usually progresses from lateral to medial, sagittal 2 and 3DRIs from lateral to medial simulate ear surgery including canal wall down mastoidectomy. Medical imaging processing systems are a useful and inexpensive tool to