

doi:10.1017/S0022215116007271

ID: IP231**middle ear mucosal regeneration by nasal mucosal epithelial cell sheets transplantation**Presenting Author: **Yuichiro Yaguchi**Yuichiro Yaguchi¹, Kazuhisa Yamamoto²,
Tsunetaro Morino², Hiromi Kojima²¹*St. Marianna University School of Medicine,*²*Jikei University School of Medicine**Learning Objectives:*

Postoperative regeneration of the middle ear mucosa and pneumatization of the middle ear cavity are of great importance after middle ear surgery. This study developed a new method to transplant autologous nasal mucosal epithelial cell-sheets into the damaged middle ear cavity. The aim of this study was to evaluate postoperative healing after the transplantation of the cell sheets in rabbits. Rabbit nasal mucosal epithelial cell-sheets were fabricated from a temperature-responsive culture dish and transplanted into the damaged middle ear of rabbit, which was surgically created. The healing of middle ears was evaluated with histological methods and computed tomography findings at 8 weeks after transplantation. Functional evaluation was performed by measuring the maximum middle ear total pressure reflecting a trans-mucosal gas exchange function. Two control groups were used: the normal control group and the mucosa-eliminated control group. Transplantation of nasal mucosal epithelial cell-sheets suppressed the bone hyperplasia and the narrowing of pneumatic space in the middle ear cavity more clearly than the mucosa-eliminated control group. The mucosal gas exchange function was also found to be good in the cell sheet-transplanted group. These results suggested that posttransplanted middle ear cavity was not only morphologically but also functionally similar to the normal middle ear cavity. Nasal mucosal epithelial cell-sheet was confirmed to be useful as an effective graft material after middle ear surgery and hopefully become a novel therapy in the future.

doi:10.1017/S0022215116007283

ID: IP232**KGF controls on the epithelial stem/progenitor cell proliferation in external auditory canal**Presenting Author: **Tomomi Yamamoto-fukuda**Tomomi Yamamoto-fukuda¹, Haruo Takahashi²,
Hiromi Kojima¹¹*Department of Otorhinolaryngology Jikei University School of Medicine,* ²*Department of Otolaryngology-Head and Neck Surgery, Department of Translational Medical Science, Nagasaki University Graduate School of Biomedical Sciences**Learning Objectives:*

Introduction: The epidermal basal stem/progenitor cell maintains homeostasis of epidermis under development, self-renewal and differentiation. In many cases of adult basal stem/progenitor cell regulation, the importance of extracellular signals provided by the surrounding cells are well recognized. Keratinocyte growth factor (KGF) is a mesenchymal-cell-derived paracrine growth factor that specifically participate in tissue development as well as wound repair. In this study, we investigated the effects of over-expressed KGF during epithelial cell proliferation and differentiation by using a cell labeling system.

Methods: After anesthetized ICR mouse Flag-hKGF cDNA driven by a CMV14 promoter was transfected into ear skin with electroporation. The ears with empty vector transfection were used as controls. 5-bromo-2'-deoxyuridine (BrdU) and 5-ethynyl-2' deoxyuridine (EdU) were administered at different time points before or after KGF expression vector transfection to identify stem cells or progenitor cells, which are believed to divide slowly or to segregate chromosomes asymmetrically. At 1, 4 and 7 days after vector transfection, 3 mice at each time-point were sacrificed. The paraffin sections were used for H&E and immunohistochemistry for Flag, KGF, BrdU and cytokeratin (CK)14. EdU staining was performed according to the manufacturer's protocol (Life Technologies).

Results: Each plasmid was transfected into the epithelial and subepithelial cells, successfully. After KGF transfection, keratin accumulations were observed 3 of 3 ears at 4 days. BrdU(+)EdU(+) cells (stem/progenitor cell) were detected in the upper layer of thickened CK14 positive epithelium in KGF transfected specimens at 4 days.

Conclusions: These findings indicated that KGF overexpression may possibly increase stem or progenitor cell proliferation and block terminal differentiation, resulting in epithelial hyperplasia and stratification.

doi:10.1017/S0022215116007295

ID: IP233**Middle Ear Mucosal Regeneration by tissue-engineered cell sheets transplantation**Presenting Author: **Kazuhisa Yamamoto**Kazuhisa Yamamoto, Tsunetaro Morino, Takanori Hama,
Yuichiro Yaguchi, Hiromi Kojima*Jikei University School of Medicine**Learning Objectives:*

Introduction: Recurrence of cholesteatoma is mainly caused by poor mucosal regeneration in the middle ear cavity and mastoid cavity. If middle ear mucosa can be preserved and the rapid postoperative regeneration of mucosa on the exposed bone surface can be achieved after middle ear surgery, surgical treatment for otitis media including