Regeneration of Hair Cells of Mammalian Inner Ear

Introduction
1. Sensorineural hearing loss resulting from noise, infection, ototoxin, or age-induced cochlear hair cell damage is the most common form of hearing loss seen today.
2. Vestibular disorders are also common, with 30% of all Americans having experienced episodes of dizziness by the age of 65.
3. As the population ages, hearing and balance disorders caused by inner hair cell loss are expected to increase.
4. Consequently, developing methods to replace lost receptor cells through transplantation or regeneration is an active area of research.

History of improvement
1. Postembryonic proliferation and differentiation of hair cells has been investigated in cold-blooded vertebrates.
2. Both birds and mammals were believed to have their full adult complement of hair cells at birth.
3. The study of the avian inner ear.
4. Electronic earing aid.
5. Digital hearing aid.
6. Biological approach: injection of exogenous agents, such as genetically engineered viral vectors, progenitor or stem cells to replace the damaged cells.

The avian inner ear
1. The avian cochlea, or “basilar papilla”, is curvilinear.
2. Tall hair cells (THC) and short hair cells (SHC): inner hair cells and outer hair cells.
3. Vestibular sensory epithelium consists of supporting cells and two types of hair cells. Type I: pear shaped and enclosed by nerve calyx. Type II: cylindrical and have multiple button type nerve endings.
4. Morphologic recovery: in adult quail THC numbers were near normal by 60 days after acoustic insult, but SHC recovery had a slower time course.
5. Physiologic recovery: ototoxic aminoglycosides.
6. Precursor for the new hair cells: sensory epithelial supporting cells.
7. Trigger mechanism:
   a. The mechanical loss of hair cells may reverse an inhibitory influence on
      progenitor cells, allowing them to reenter the cell cycle and begin epithelial
      recovery
   b. Paracrine mechanism, whereby damaged hair cells, scavenging phagocytic
      cells, or loss of receptor neural activity cause the release of a mitogenic
      substance
8. Hair cell regeneration in the mammal: cells much lower and the proliferative time
    course much slower

**Stem cell overview**
1. A stem cell is characterized by the capacity to self-renew and the ability to
   differentiate asymmetrically to form cell types other than its own
2. Embryonic stem cell: pluripotent
3. Tissue-specific, end organ or adult stem cells: retain the capacity to self-renew
   and to produce different daughter cells: multipotent
4. The observations that end organ stem cells mature into cell types that do not
   typically exhibit regeneration, such as neurons, led to therapeutic applications
5. Avian supporting cell

**Potential use of stem cells in cochlear repair**
1. It would be more practical to use stem cells that are isolated from the end organ in
   which they reside than to use stem cells from another source
2. Embryonic stem cell lines: tumorigenic
3. An adult stem cell: more appropriate
4. Neural stem cell: may be the therapeutic stem cell of choice due to the close
   association of the inner ear and the brain
5. Ito et al (2001): injected aggregates of hippocampal stem cells, or neurospheres,
   into the cochlea of rats
6. Characteristics of the ideal therapeutic stem cell:
   a. a robust capacity to migrate to the site of lesion
   b. must respond to local environmental cues and differentiate into the
      appropriate cell types
   c. must retain the ability to recover the loss of function
7. Possible complication: generation of ectopic hair cells
Fate of neural stem cells grafted into injured inner ears of mice

1. Juichi Ito et al. 2003
2. Fetal mouse NSCs expressing green fluorescence were used as transplants. NSC spheres were obtained from the neuroepithelium of the dorsal telencephalon of embryo
3. A neomycin solution was injected from the left posterior semicircular canal to the recipient animals
4. A small hole was then made in the lateral wall of the cochlear second turn for NSC aggregates
5. The cell fates of grafted NSCs were determined by immunohistochemistry for MAP2 (marker for neuronal cells), GFAP (marker for several types of glial cells), nestin (marker for immature neural cells including NSCs and immature supporting cells), or myosin VIIa (a specific marker for inner hair cells)
6. Injury of sensory epithelia caused by aminoglycoside toxicity may promote integration of grafted NSCs into the sensory epithelia of the inner ear
7. Expression of myosin VIIa was observed in grafted NSCs in the vestibular epithelia, indicating that NSCs have the potential to differentiate into cells characteristic of inner hair cells
8. The number of grafted NSCs in cochlear sensory regions was very limited. The potential of NSCs to differentiate into cochlear hair cells remain controversial

Generation of hair cells by stepwise differentiation of stem cells

1. Huawei Li, Stefan Hellar et al
2. Pool cells from the utricle, the only part of the ear known to retain a limited mitotic capacity in adult mammals
3. Sphere formation was enhanced when cultures were treated with epidermal growth factor (EGF) and insulin-like growth factor-1 (IGF-1), two known vestibular growth factors
4. After two weeks, the sphere cells began to show expression of MyoVIIa and Brn3.1, two diagnostic gene for mature hair cells
5. Inject sphere cells into the otic vesicle of developing chick embryos: murine cells in the bird cochlea
6. How to control the behavior of precursor cells once in the body?
7. How to control the relative orientation of the new hair cells within the plane of the sensory epithelium? Randomly oriented hair cells would relay conflicting information to the brain, submitting the individual to a constant state of vertigo
Gene therapy

1. Wei-Qiang Gao et al. 2002
2. Hair cells are produced in the sensory epithelium of the cultured adult rat utricular maculae, via adenovirus–mediated overexpression of Hath 1, a human atonal homolog
3. Adult rat utricular macula treated with gentamicin, and subsequently infect the tissue using the ad-Hath-1-EGFP virus: the infected cells were myosin VIIa positive
Reference: