The changing landscape for hearing loss therapeutics: novel advances of gene and cell therapies

BY MARCELO RIVOLTA

Recent years have seen advances in hearing loss therapeutics, with novel treatments trialled in humans, and others nearing promising first-in-kind clinical trials.

First successful clinical trials for a specific form of genetic hearing loss

Very exciting news has emerged in our field in recent months. Several different groups based in the US, the UK and China have reported the first successful application of gene therapy to treat a particular form of deafness [1–3]. This has created enormous enthusiasm, as it is the first time scientists and clinicians have successfully repaired a genetic defect that produces deafness. These teams targeted a mutation in a gene named otoferlin (OTOF). Otoferlin is involved in the transmission of the auditory information between the inner hair cell and the neurons in the cochlea. When mutated, the defect leads to a failure in the synaptic connections and the onset of irreversible deafness. Healthy copies of the otoferlin gene are packed into a virus (known as a 'viral vector') and the genetically modified virus is then injected into the cochlea of affected patients. The virus binds onto the surface of the inner hair cells and delivers the healthy gene. This replaces the mutated one, and starts producing a healthy otoferlin protein. Patients have manifested a restoration of their hearing, as measured by improved auditory brainstem thresholds.

This is truly remarkable, as it shows treatment is possible to fix a defective gene and to restore function of patients with hearing loss. It opens up new possibilities and suggests that treatments that repair the damaged molecular make-up of the inner ear are not just feasible but are on the cusp of becoming viable treatment options alongside medical devices.

While highly encouraging and exciting, this technology does not herald the end of hearing loss; this approach has limitations and important questions still remain unanswered. The technology of gene therapy can be applied to monogenic causes of hearing loss that, while accounting for ~35% of all congenital hearing impairments [4], remain a relatively small portion of all cases of hearing loss overall. Each gene (and there are over 150 genes described that lead to deafness when mutated) will have to be targeted by a specific, customised viral vector, meaning a new therapy and new clinical development pathway for each mutation. While it is likely the gene could be delivered to the right target cells, it is not clear if the intervention would suffice to restore function in all mutations. There is also a limit on the gene size a virus can carry as a payload. For otoferlin, this was solved by splitting the gene into two to three different viral vectors but, in some cases, the defective gene may be just too big to be effectively encapsulated into multiple viral particles.

This is without doubt a substantial step forward. However, alternative, more broadly applicable strategies are needed to treat the majority of people who have a hearing loss that is attributable to a large variety of environmental and polygenic factors.

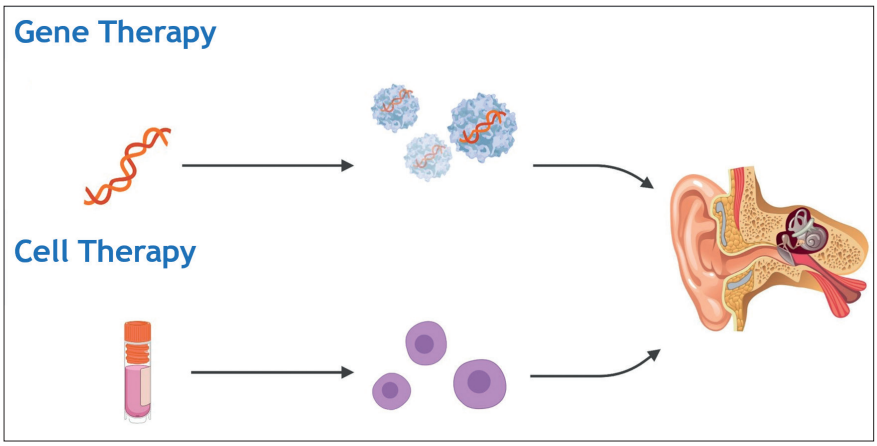

Figure 1: Graphic representation of the differences between gene and cell therapy. In a gene therapy strategy (top), a gene sequence is packed into a viral vector and delivered into the cochlea to correct a genetic defect. On the other hand, cell therapy (bottom) delivers whole functioning cells to replace or restore lost cell populations.

The potential of stem cell-based therapies

While both cell and gene therapies are formally grouped together in what is known as advanced medicinal therapeutics products (ATMPs), they work in a very different way (Figure 1). Gene therapies aim to repair, add or replace a genetic sequence. This works only for patients with a specific known genetic mutation, as the mechanism of action of the gene therapy is directly related to the genetic expression of the sequence the therapy contains. For these therapies to work, the fundamental cells need to have been preserved. Cell-based therapies, on the other hand, regenerate, repair or replace human cells, and have the capability of restoring entire cell populations that may have died and been lost, irrespective of the underlying cause of the cellular loss. Cell therapies do this by using the incredible potential of stem cells. Stem cells are cells that can multiply and expand in a laboratory test tube but are 'undifferentiated', meaning they are a 'white canvas', waiting for instructions to produce a specific cell type. Furthermore, the most useful type of stem cell for therapeutic purposes is called 'pluripotent', as these have the potential to produce virtually any cell type in the human body.

There are two types of pluripotent stem cells: embryonic and induced. Embryonic

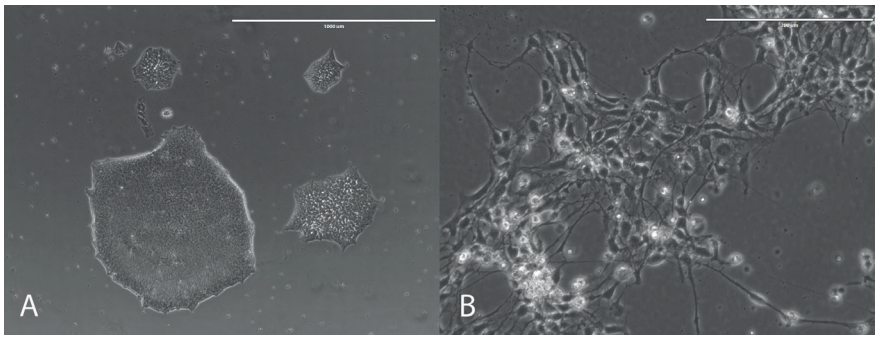

Figure 2: A) Image shows undifferentiated, human pluripotent stem cells in culture. B) Otic neuroprogenitors generated from human pluripotent stem cells by differentiating them using chemical signals that mimic human development.

stem cells are derived from embryos surplus to in vitro fertilisation treatments. Rather than being discarded, they are donated for research and to develop new treatments. These cells are isolated and established as cell lines that can be continuously maintained in culture in the laboratory, negating the need to find new cells each time you want to use them. Induced pluripotent stem cells are similar to embryonic cells in their properties and potential. However, these cells are generated from mature cells (such as white blood cells or skin cells) and artificially 'reprogrammed' into becoming undifferentiated. It is a bit like resetting a clock, turning them back to an early stage and making them competent to receive instructions to become any other cell type.

A key part of the process to turn either of these types of pluripotent cells into a cell therapy is to understand how to direct them down the right track, to make them become the cell type of choice. Both types of cells can go through a process in the lab called 'differentiation', which would mimic in the test tube what would normally happen during human development in vivo. By using chemicals and molecules that modulate the cellular signalling pathways, we can trigger differentiation and turn the stem cells with unlimited potential into a more restricted 'progenitor', a cell still immature but capable only of differentiating into a more specific target lineage (Figure 2). These 'progenitors' can then be transplanted into a patient, allowed to engraft and used to replace the damaged cells. Using restricted progenitor cells, which are committed only to developing into mature cells of a given cell type has a functional advantage and higher safety profile over undifferentiated products.

The advancement of cell therapies for hearing loss restoration

Our work started over 20 years ago in the laboratories of the Centre for Stem Cell Biology at the University of Sheffield. There, we developed the first methods to guide undifferentiated human embryonic stem cells into otic progenitors (an immature inner ear cell, equivalent to those found during development in the cochlea) and then to make them differentiate further into hair cell types and spiral ganglion neurons. We also demonstrated, in an animal model that had lost their cochlear neurons, that when transplanted with otic neuroprogenitors, these could mature into neurons, reestablish the innervation in the cochlea and partially restore hearing by improving auditory brainstem thresholds [5].

Rinri Therapeutics was born in 2018 to translate these initial discoveries into a realistic therapeutic solution. Enormous progress has been achieved since its inception.

We have since adapted the initial methods used to culture and manipulate cells to meet the rigorous standards that are required for clinical use and industrialisation. We have also developed parameters and assays that allow us to define, with confidence, the cells that will be used clinically. The otic neuroprogenitors initially developed in the university lab have now become Rincell-1, a first-in-kind cell-based therapeutic with the potential to treat neural hearing loss. The restoration of lost neurons through Rincell-1 could be used either alongside traditional prosthetic devices or independently in a stand-alone treatment.

Rincell-1 will be applied to treat the loss of spiral ganglion neurons in the cochlea. In an initial phase, we are going to test their application together with a cochlear implant. In this context, the cochlear implant would functionally replace the hair cells while Rincell-1 would restore the lost neurons. We expect Rincell-1 could improve the performance of the implant, but we can also use the implant to measure the success and efficacy of the cell therapy transplant.

It is our ambition to be ready to commence human trials by next year. In our first-in-human study, we are hoping to explore this treatment in two groups of people with hearing loss. Our first cohort will be a group of individuals who have neural hearing loss as a result of ageing. There is an increasingly large body of evidence that suggests that the innervation is the first component to degenerate with the passing of time, and many people over the age of 60 have a substantial loss of neurons and nerve fibres. The second group are those with auditory neuropathy spectrum disorder. Younger than those with presbycusis, the innervation of these patients is largely affected while the hair cells are preserved. This is usually evidenced by the loss of auditory brainstem thresholds but with preservation of otoacoustic emissions and pure tone audiometry.

If successful, these trials would open exciting new possibilities for those with neural hearing loss. Although more work is still needed, other cell types important in hearing loss (like auditory hair cells) could also be generated by redirecting the original population of stem cells to differentiate into the relevant progenitors, using slightly different developmental chemical signals.

Patients with hearing loss have long awaited a solution that would restore their audition. The recent results obtained with the gene therapy trials and the significant advances in the cell therapy space are now giving us reason to be optimistic that therapeutic treatment of hearing loss will become a reality.

References

1. Wang H, Chen Y, Lv J, et al. Bilateral gene therapy in children with autosomal recessive deafness 9: single-arm trial results. *Nat Med* 2024;**30**:1898–1904.
2. Qi J, Tan F, Zhang L, et al. AAV-Mediated Gene Therapy Restores Hearing in Patients with DFNB9 Deafness. *Adv Sci (Weinh)* 2024;**11(11)**:e2306788.
3. Lv J, Wang H, Cheng X, et al. AAV1-hOTOF gene therapy for autosomal recessive deafness 9: a single-arm trial. *The Lancet* 2024;**403(10441)**:2317–25.
4. Petit C, Bonnet C, Safieddine S, et al. Deafness: from genetic architecture to gene therapy. *Nat Rev Genet* 2024;**24(10)**:665-86.
5. Chen W, Jongkamonwiwat N, Abbas L, et al. Restoration of auditory evoked responses by human ES-cell-derived otic progenitors. *Nature* 2012;**490(7419)**:278-82.

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