AAV Gene Therapy for noise induced hearing loss using cerebrospinal fluid as route of delivery

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BACKGROUND

- Noise induced hearing loss (NIHL) is one of the most prevalent disabilities for which there is no effective therapeutic treatment.
- IHC-SGN synapses must be maintained or regenerated to improve hearing after auditory insult.
- Delivery of effective compounds to the target cells is the main challenge for clinical translation.
- AAV9 can be utilized to effectively transduce cells in the CNS—we hypothesize we can utilize AAV9 to deliver therapeutic compounds to IHC of the cochlea.

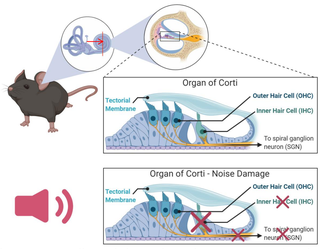


Figure 1. Auditory injury via noise exposure leads to metabolic and/or mechanical changes in inner hair cells (IHC) and spiral ganglion neuron (SGN) degeneration. Figure created with Biorender.com.

METHODS

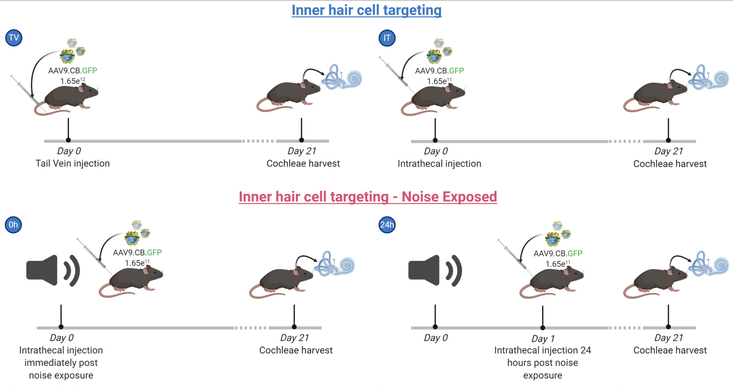


Figure 2. Experimental design to determine optimal AAV9 injection route and to analyze AAV9 transduction of IHC 0 and 24 hours post noise injury. Figure created with Biorender.com.

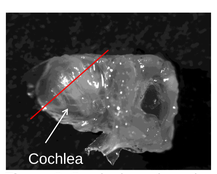


Figure 3. Sectioning orientation.

Tissue Processing:

- Cochleae were fixed in 4% PFA for 2 hours then decalcified in 120mM EDTA in PBS for 3 days.
- Cochleae were embedded in OCT and sectioned at 14µm on cryostat in the orientation indicated.

CONCLUSIONS

- TV injected AAV9.CB.GFP reaches the cochlea but does not transduce IHC.
- IT injected AAV9.CB.GFP can transduce IHC before and up to 24 hours post noise injury.
- At 24 hours post noise injury, less IHC are transduced and expression is lower than in 0 hours post injury injected animals, though IHC are not physically damaged by the noise exposure protocol utilized.
- AAV9 can be utilized as a drug delivery system to administer therapeutic compounds to the IHC pre and post noise injury.
- Ongoing experiments: analysis of vector targeting utilizing a hair cell specific promotor (AAV9.Myo7A.GFP); IT injections 48 hours post noise injury; analysis of IHC targeting via intratympanic injections.

RESULTS

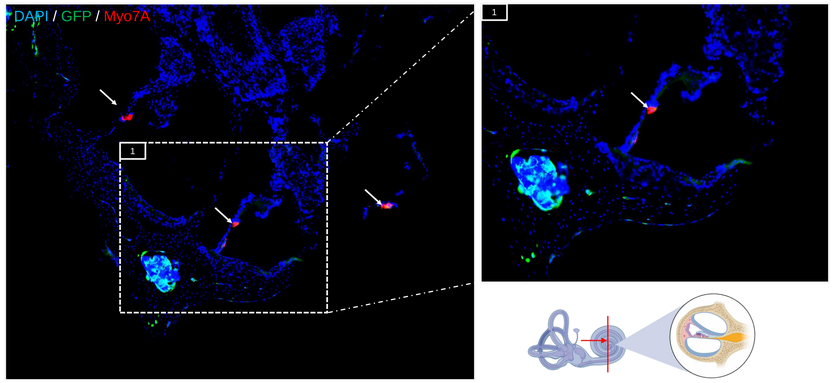
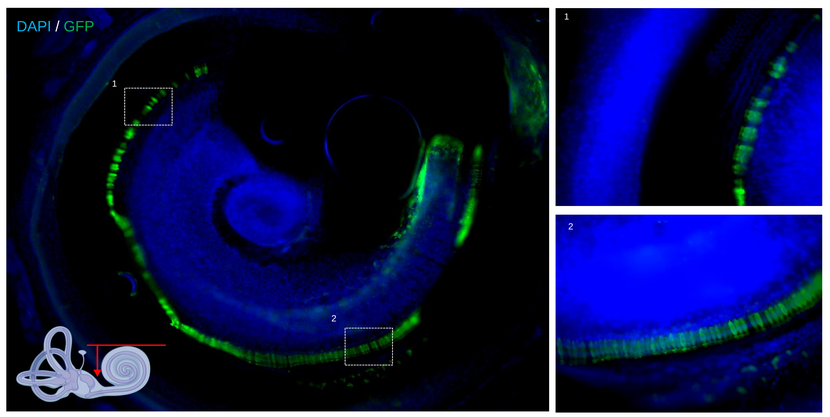


Figure 4. IHC Transduction—Tail Vein AAV9.CB.GFP Injection. Transduction of IHC was not observed with tail vein injection in wild type mice. Myo7A was used to detect hair cells (red). Non-transduced hair cells are indicated by white arrows.



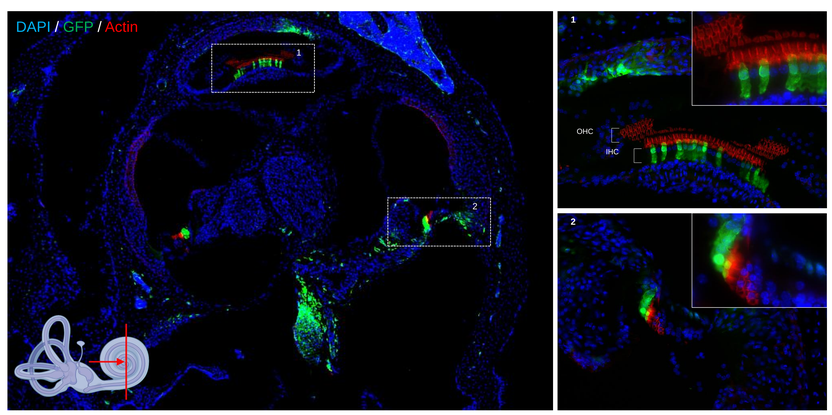


Figure 5. IHC transduction—Intrathecal AAV9.CB.GFP Injection. High transduction of IHC (green) is observed with intrathecal injection in wild type mice. Outer hair cells are detected with actin (red).

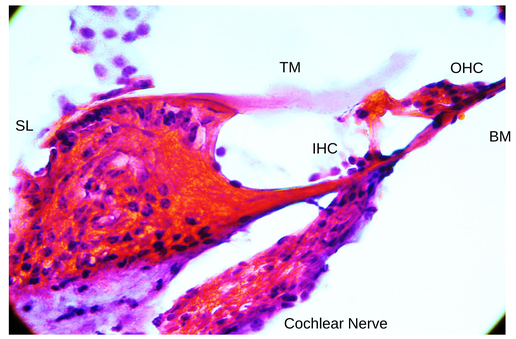


Figure 6. Noise damaged mouse Organ of Corti. Spiral limbus (SL), inner hair cells (IHC), outer hair cells (OHC), basilar membrane (BM), tectorial membrane (TM). No structural damage is observed post noise injury.

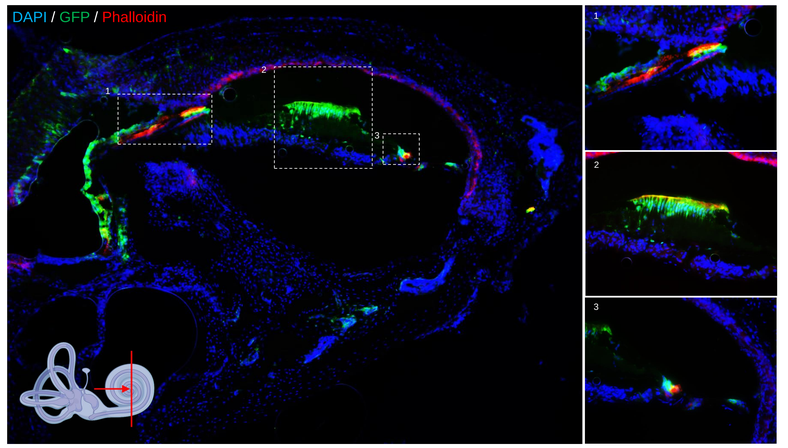


Figure 7. IHC transduction 0 hours post noise injury. Wild type mice were IT injected immediately after noise exposure. High transduction of IHC is observed (green). Phalloidin was used to detect actin/outer hair cells (red).

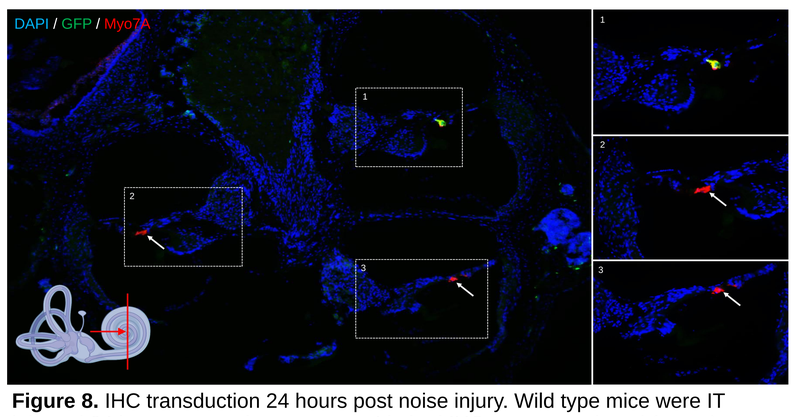


Figure 8. IHC transduction 24 hours post noise injury. Wild type mice were IT injected 24 hours post noise exposure. Limited IHC were transduced (green)—non transduced hair cells are indicated by white arrows. Myo7A was used to detect hair cells (red).