SU95

Predicting Profiles of Cochlear Pathology from Non-Invasive Physiological Biomarkers

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INTRODUCTION

Temporal bone histology shows that damage to outer hair cells, inner hair cells, the stria vascularis, and the auditory nerve contribute to sensorineural hearing loss. Physiological measures such as otoacoustic emissions, the middle ear muscle reflex, and the auditory brainstem response are used in research and clinically to detect these pathologies and to determine a more precise site of lesion. However, it remains difficult to establish a one-to-one correlation between physiological responses and histological profiles, especially when more than one pathology is present.

In this study, we examined the relationship between histological profiles of cochlear pathology and responses on a battery of physiological tests sensitive to both OHC and non-OHC dysfunctions. Our preliminary results focused primarily on inner and outer hair cell counts across chinchillas with a variety of exposure histories.

METHODS

Animal Models and Exposure Conditions

Experimentally-Induced Hearing Loss		Unexposed
Noise Induced Temporary Threshold Shift (TTS) - Cochlear Synaptopathy N = 6 animals	Noise Induced Permanent Threshold Shift (PTS) - Complex SNHL N = 10 animals	Control N = 6 animals
Carboplatin (CA) Induced Inner Hair Cell Dysfunction N = 7 animals	Gentamicin Induced Outer Hair Cell Dysfunction N = 5 animals	Mystery Chinchillas Older/High-Risk N = 5 animals

Young chinchillas (6 months to 3 years) were used in the exposure groups and the controls. “Mystery chinchillas” were older animals (3 to 6 years) who had been used for other experiments in the lab.

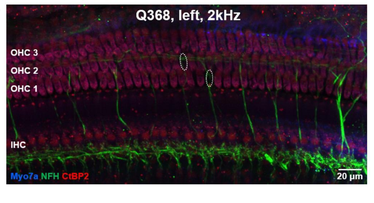
Biomarker Test Battery

ABR Thresholds .5, 1, 2, 4, 8 kHz	**Envelope Following Response Rectangular Amplitude Modulated** 4kHz tone, 223 Hz modulation	**Distortion Product Otoacoustic Emissions**
Wideband Middle-Ear Muscle Reflex	**Click ABR** High level stimulus	**Stimulus Frequency Otoacoustic Emissions**

Physiological assessment with the above battery was performed pre-exposure (see Control Group physiology) and two weeks after exposure. Physiological results shown here include all animals from a given paradigm whether or not histological data was included. Physiological responses are shown for the tests that contributed most to classifying across exposure groups and are normalized to the mean and standard deviation of the pre-exposure results (37 animals).

Cochlear Histology

After all testing was completed, animals were euthanized and perfused. Cochleas were harvested, post-fixed, and decalcified. Cochleas were then bisected through the modiolus, immunolabeled, and dissected into half turns.

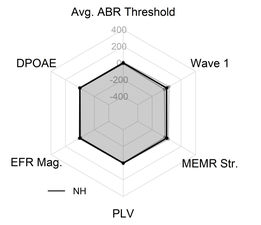


Specimens were labeled for:
- hair cells (Myo7a)
- afferent neurons (NFH)
- hair cell presynaptic ribbons (CtBP2)
- cholinergic efferent neurons (ChAT)
- In a subset, stereocilia (ESPN)

Confocal z-stack images were obtained at .5, 1, 2, 4, and 8 kHz cochlear locations. Hair cells counts were obtained at each of the 5 cochlear locations when dissection and image quality allowed. Hair cell survival is averaged across ears when both ears were available, otherwise, only data from one ear is shown.

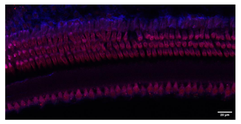
RESULTS

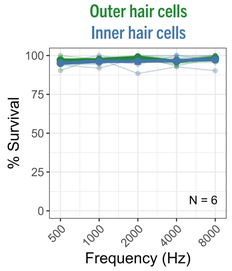
Control



Physiological test results were averaged across the group and visualized as a radar plot with values for each measure normalized to the control (baseline) group.

3 rows of OHCs and a single row of IHCs are visible with 20x confocal microscopy

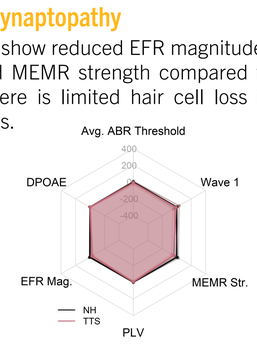


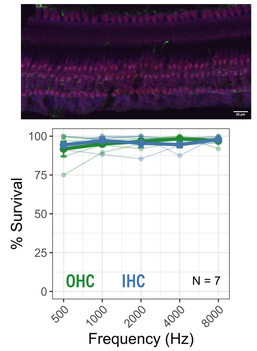


Inner hair cell and outer hair cell survival was quantified. Control/unexposed animals show essentially no hair cell loss.

Cochlear Synaptopathy

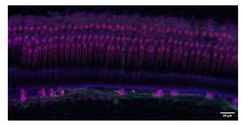
TTS animals show reduced EFR magnitudes and reduced MEMR strength compared to baseline. There is limited hair cell loss in these animals.

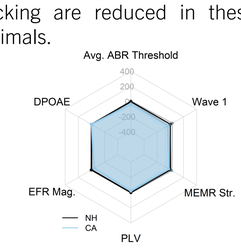


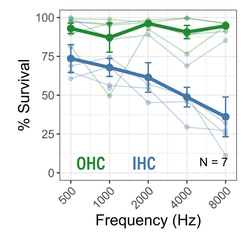


Inner Hair Cell Loss

Carboplatin results in significant IHC loss across the frequency range. Some frequency locations reveal missing OHCs. Both EFR magnitude and phase locking are reduced in these animals.

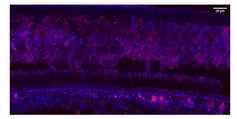


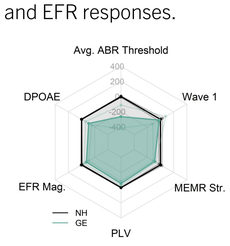


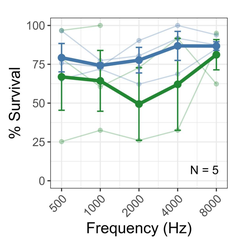


Outer Hair Cell Loss

Gentamicin exposure resulted in both inner and outer hair cell loss, accompanied by reduced ABR thresholds, DPOAEs, and EFR responses.

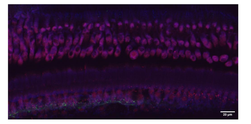


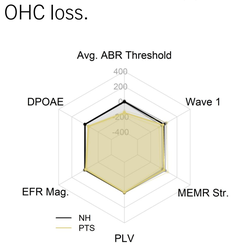


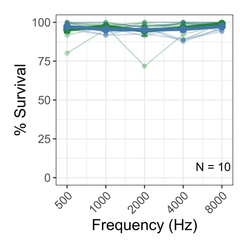


Noise Induced Hearing Loss

NIHL reduced ABR thresholds and DPOAEs as expected. Despite 20-40 dB ABR threshold shifts, only one animals showed appreciable OHC loss.



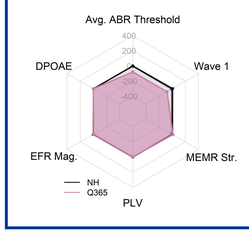


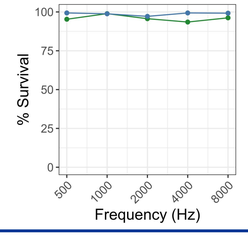


Mystery Chinchillas

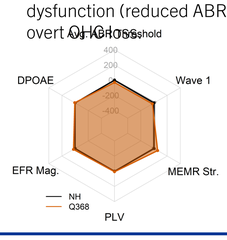
We also explored the relationship between histology and physiology at the individual level in a subset of animals without experimentally-induced hearing loss, but greater risk for sensorineural pathology given their age and noise exposure history.

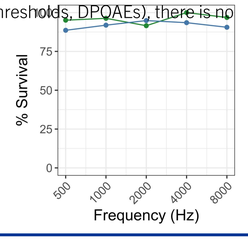
Chinchilla 1: Q365



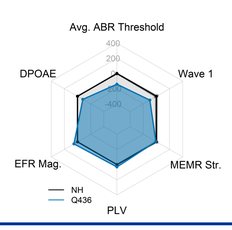


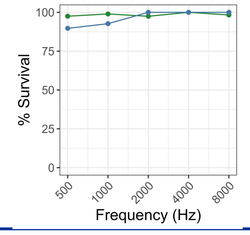
Chinchilla 2: Q368 Despite evidence of OHC dysfunction (reduced ABR thresholds, DPOAEs), there is no overt OHC loss





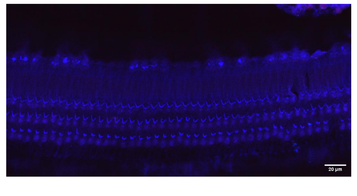
Chinchilla 3: Q436 Despite evidence of OHC dysfunction (reduced ABR thresholds, DPOAEs), there is no overt OHC loss.





FUTURE DIRECTIONS

Future datasets will be analyzed for other features such as synapses, auditory nerve fibers, and stereocilia.



Stereocilia from a carboplatin-exposed animal shows IHC stereocilia disarray in remaining cells.

CONCLUSIONS

- Hair cell counts alone are likely inadequate to account for the full range of pathology possible and more subtle effects such a stereocilia damage.
- These precision biomarkers show promise for non-invasively assessing inner ear health.

REFERENCES

[1] XXX [2] XXXX

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