

Poster presentation

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Microarray analysis of gene expression during auditory hair cell regeneration in zebrafish (*Danio rerio*)

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Background

Fishes are capable of regenerating sensory hair cells in the inner ear after overexposure to sound [1]. However, the time course and gene expression of auditory hair cell regeneration has not been established for zebrafish (*Danio rerio*). This study establishes a time course of cell proliferation associated with hair cell regeneration in the zebrafish inner ear and examines gene expression patterns exhibited during this process.

Materials and methods

Adult zebrafish were exposed to a 100 Hz pure tone at 179 dB re 1 μ Pa RMS for 36 hours, and then allowed to recover for 0 to 14 days before morphological analysis. Saccular hair cells were quantified through visualization of stereocilia stained by phalloidin at 0, 2, 7, and 14 days post-sound exposure. Cell proliferation was quantified through BrdU labeling at 0, 1, 2, 3, 7 and 10 days after the end of sound exposure. Groups of 18–20 fish were allowed to survive 0, 2 or 4 days post-sound exposure at which time the whole ears from each group were pooled and RNA extracted. Fluorescent cRNA from each treatment sample was hybridized to three duplicate Agilent Zebrafish oligonucleotide arrays (4 × 44 K, 60-mer oligonucleotides).

Results

Immediately following noise exposure, zebrafish saccules exhibited significant hair cell loss in the caudal region. Hair cell counts increased over the course of the experi-

ment, reaching pretreatment levels by 14 days post-noise exposure. Cell proliferation peaked two days post-noise exposure in the caudal region, and to a lesser extent in the rostral region. Microarray analysis showed that a number of genes are significantly regulated in the sound-exposed zebrafish inner ear on days 2 and 4 following sound exposure (see Figure 1). Some of the most highly regulated genes included those encoding major histocompatibility complex, growth hormone, and cytoskeletal organization proteins.

Conclusion

The zebrafish ear is a good model for examining genes involved during the process of hair cell regeneration. A large number of genes were either up- or down-regulated at 2- and 4-days post-sound exposure. Putative biological processes for some of these genes have been identified, but more work will be needed to determine their function in the zebrafish inner ear.

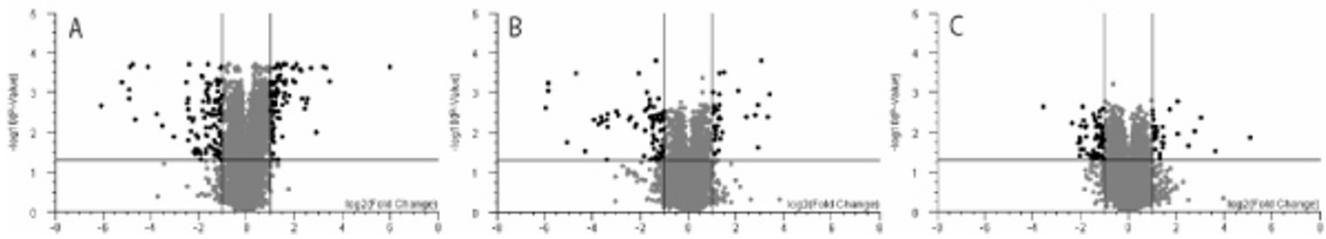


Figure 1

Volcano plots of normalized median intensities showing differential expression between **A.** controls and 2 days post-sound (188 genes), **B.** controls and 4 days post-sound (106 genes), and **C.** 2 and 4 days post-sound (91 genes). Black dots represent differentially expressed genes with a significance cut-off p-value of 0.05 and fold change value of ≥ 2 .

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References

1. Smith ME, Coffin AB, Miller D, Popper AN: **Anatomical and functional recovery in the goldfish (*Carrasius auratus*) ear following noise exposure.** *J Exp Biol* 2006, **209**:4193-4202.

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