# Cochlear Damage, Hippocampal Injury and Behavioral Assessment in a Rat Model of Blast Wave Exposure

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## Introduction

• Blast wave exposure is a major cause of hearing loss and tinnitus in military personnel

• In addition to damaging the inner ear, blast wave exposure has the potential to induce traumatic brain injury (TBI)

## Rationale

• Previous reports have identified the hippocampus, one of two regions in the adult brain where neurogenesis occurs, as particularly vulnerable to TBI

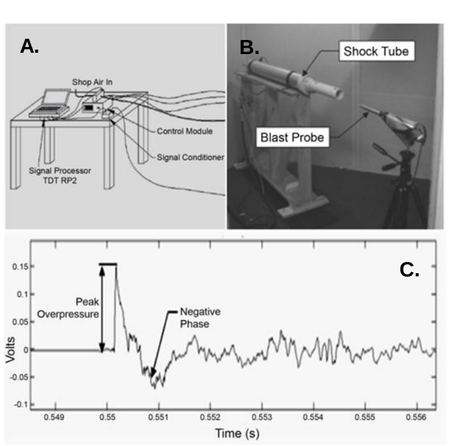
• At present, little is known about how blast exposure affects hippocampal neurogenesis– a process strongly associated with memory function

## Approach

• We exposed adult rats to repeated blast waves (3; ~187 dB pSPL, ~45 kPa) and assessed the extent of damage in the cochlea and hippocampus at various times after exposure (7 & 42 days post-blast)

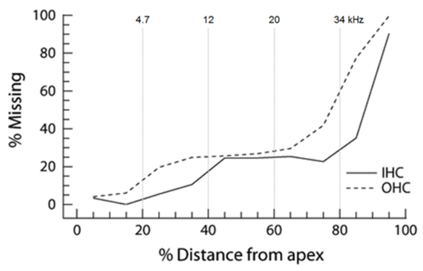
### Blast Wave Generator

• Shock tube (PVC pipe & outer protective metal shield) sealed at one end by a thin metal diaphragm (Fig. 1B). Once the chamber was charged with compressed air (A), a solenoid-driven arrow inside the chamber pierced the diaphragm and triggered the primary blast wave. High-pressure probe (B) measured peak overpressure of the primary blast wave (C).

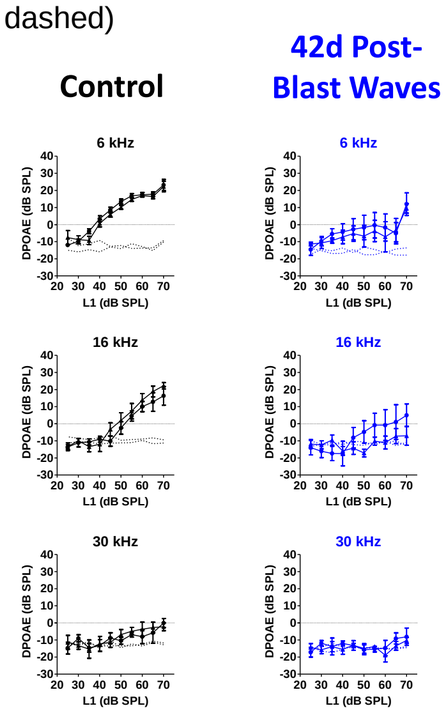



## COCHLEA: Methods & Results

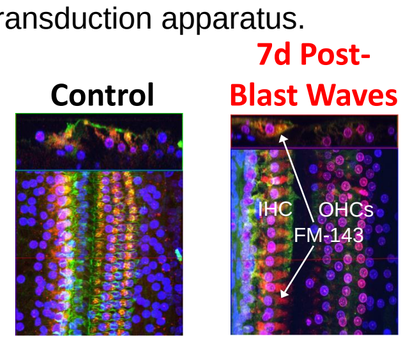
• Rats (n=4) were sacrificed **42 days** post-blast exposure, and a mean **cochleogram** was constructed to determine the amount of inner (IHC) and outer hair cell (OHC) loss. Cochlear position was related to frequency using a cochlear frequency-place map.




• Prior to sacrifice, **distortion product otoacoustic emissions** (DPOAE ; f2/f1=1.2, L1=L2+10dB) were assessed in both ears at low (6 kHz), mid (16 kHz) and high (30 kHz) frequencies (left ear- circle; right ear- triangle; noise floor- dashed)




• **FM-143**, a small styryl dye that labels hair cells by passing through the mechano-electrical transduction channels in the stereocilia, was used to monitor the functional status of the transduction apparatus.




FM-143(red) was taken up by IHCs & OHCs in control tissue, but only by IHCs post-blast; findings consistent with damage to the OHC stereocilia.

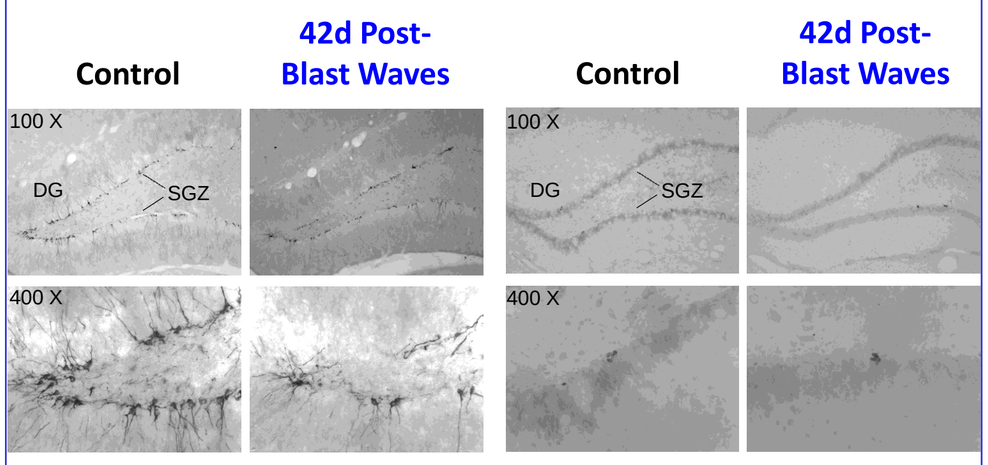
## HIPPOCAMPUS: Methods & Results

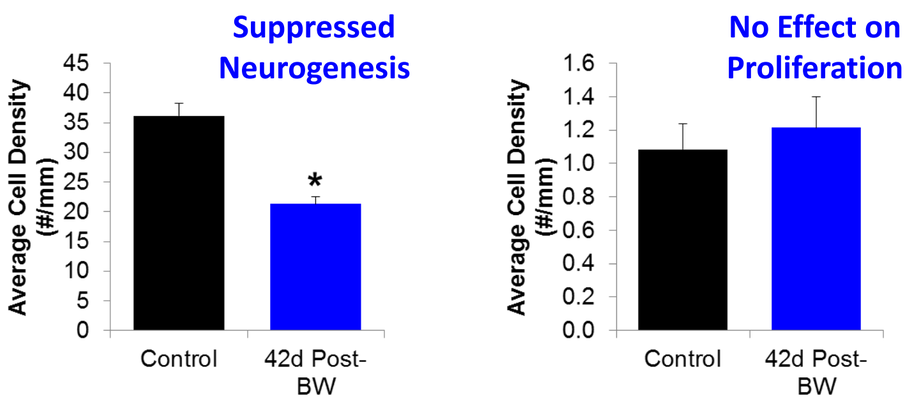
### Doublecortin (DCX) Immunolabeling: Neurogenesis

• **DCX** is a microtubule-associated protein that identifies neuronal precursor cells
• Brain harvested after intracardial perfusion, and then cryosectioned (coronal) for immunolabeling
• Treatment-blinded counts of DCX-positive cells along the measured length of the subgranular cell zone (SGZ) in the dentate gyrus were used to calculate the average cell density (#/mm)

### Ki67 Immunolabeling: Proliferation

• **Ki67** is a mitosis marker that identifies proliferating cells
• Brain harvested after intracardial perfusion, and then cryosectioned (coronal) for immunolabeling
• Treatment-blinded counts of Ki67-positive cells along the measured length of the subgranular cell zone (SGZ) in the dentate gyrus were used to calculate the average cell density (#/mm)

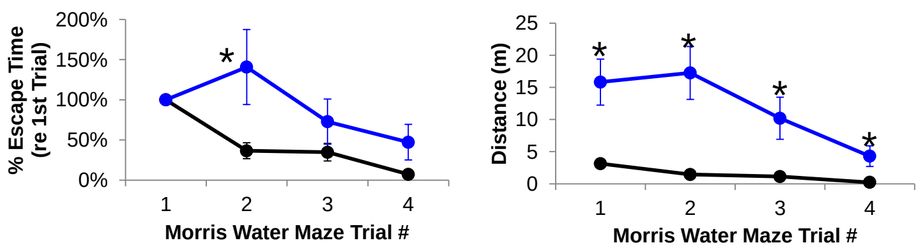



## BEHAVIORAL ASSESSMENT: Methods & Results

### Morris Water Maze: Spatial Learning and Memory

• Rat placed in small, darkened pool of water which contained a hidden platform slightly below water surface
• Four successive trials were performed with same platform location
• Video camera recorded the time taken for rat to swim to the hidden platform, and total distance traveled
• Compared to **controls**, **blast wave exposed rats** showed limited performance improvements over successive trials, indicative of impaired spatial learning and memory




## Discussion

### COCHLEA

• The majority of hair cells survived repeated blast wave exposure, but the DPOAE were significantly reduced, indicative of impaired outer hair cell (OHC) function.

• Additional experiments using FM-143 staining revealed that although OHCs survived blast wave exposure they had permanent damage to their stereocilia transduction apparatus which likely contributed to their impaired function.

### HIPPOCAMPUS

• Neurogenesis (DCX labeling), but not cell proliferation (Ki67 labeling), was significantly reduced 42 days after repeated exposure to primary blast waves

• These findings could represent a blast-induced shift of differentiation from a neuronal lineage toward a glial lineage, or alternatively, a decreased survivability of immature neurons

• Ultimately, our findings confirm an earlier report of down-regulated gene expression related to neurogenesis after primary blast wave exposure

## Conclusion

• Suppressed hippocampal neurogenesis in the weeks following blast exposure may contribute to the cognitive deficits common in military personnel exposed to blast trauma

• We are continuing to investigate whether the well-established paradigms for tinnitus assessment in animal models will need to be modified to accommodate the additional TBI associated with blast-induced hearing loss

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