Hearing related disorders affect millions of people worldwide. Most people develop hearing related disorders through excessive exposure to loud noise in recreational and/or occupational settings. The mechanisms underlying noise induced hearing disorders remains poorly understood. Excessive production of reactive oxygen species (ROS) in auditory nuclei is implicated as a major contributor of noise hearing loss. On the other hand, loud noise leads to diminished gene expression of tyrosine hydroxylase, the rate limiting enzyme in the production of dopamine. Thus, because ROS can modulate neuronal processes, including synaptic dopamine release, we hypothesise that excessive noise exposure will lead to the production of ROS which in turn will attenuate dopamine levels in auditory nuclei particularly, the inferior colliculus (IC).

### Experimental Design and Methods

<table>
<thead>
<tr>
<th>4-hour noise exposure</th>
<th>Hydrogen peroxide assay</th>
</tr>
</thead>
<tbody>
<tr>
<td>Getting sound box ready for 24 hours before exposure</td>
<td>Sacrifice immediately after exposure or wait sacrifice</td>
</tr>
</tbody>
</table>

**Animals**
- Adult Sprague Dawley were used in this study according to the International Animal Care and Use Committee (IACUC) of the University of Northern Colorado.
- Animals were separated into three groups: Control group (not exposed to noise), 4-hour noise exposed /same day sacrificed group, and 4-hour noise exposed /sacrificed 24 hr later group.

**Sound exposure**
- Animals were exposed to sound of 10 kHz at 118 decibels (dB) at 1/3 octave band.
- Exposures were done for four hours.

**Analysis of Sample**
- The animals from both control and test groups were sacrificed according to laboratory protocols.
- The inferior colliculus (IC) was extracted from the brain and protein extraction done. Protein fraction was analyzed for the presence of the ROS, hydrogen peroxide.

### Results

**Analysis of hydrogen peroxide in control rat**

- **Figure 1.** The schematic describing the process of noise exposure. The sound bath was prepared to deliver 10 KHz at 118 decibels (dB) at 1/3 octave band. After which rats will be exposed to the sound for 4 hours. The same-day group was sacrificed immediately after sound exposure and the 24-hour group was sacrificed 24 hours after sound exposure. The IC was extracted from rats, protein extraction done followed by hydrogen peroxide assay.

- **Figure 2.** The proposed mechanism that is being tested in this study. Excessive noise exposure produced changes in the concentration of the ROS, hydrogen peroxide, in the IC of the rat brain. The IC is the main hub of dopamine. Therefore we believe that dopamine levels will be attenuated in response to changes in ROS ultimately resulting in hearing loss.

- **Figure 3.** Determination of hydrogen peroxide in control rat. The results shown are for the left and right IC hydrogen peroxide levels in the control rat. $P^2$ values were 0.9839 and 0.9795 respectively.

- **Figure 4.** Determination of hydrogen peroxide in 4-hour sound exposed rat. The results shown are for the left IC and right IC hydrogen peroxide levels in the same-day exposed rat. $P^2$ values were 0.9013 and 0.8958 respectively.

- **Figure 5.** Determination of hydrogen peroxide in 24-hour sound exposed rat. The results shown are for the left IC and right IC hydrogen peroxide levels in the 24-hour post exposure rat. $P^2$ values were 0.9612 and 0.9777 respectively.

### Conclusions
- Volumes of hydrogen peroxide in left and right IC.
- 4-hour sound exposed rat was 0.21 ml and 0.64 ml respectively.
- 24-hour sound exposed rat was 0.13 ml and 0.11 ml respectively.
- Control rat was 0.68 ml and 0.52 ml respectively.
- There was a general decrease in peroxide levels between control and sound-exposed rats. With the 24-hour post exposure group showing a significantly lower volume of peroxide in the IC than the same-day group.
- Hydrogen peroxide is a signaling molecule that has been shown to modulate synaptic dopamine release through calcium channels.
- Contrary to our hypothesis, this preliminary finding shows that excessive noise exposure causes the level of the ROS, hydrogen peroxide, to decrease while we expected an increase.
- However further work has to be done to add to the numbers of rats in the three experimental groups and to assess the effect on dopamine in the sound exposed rats versus control rats.

### Acknowledgement
We would like to thank the University of Northern Colorado for the financial support for this research project.