

## SUMMARY OF THE INVENTION

To study the neuronal networks in behaving animals, the University of Oregon Institute of Neuroscience (ION) acquired a Thorlabs 2-photon mesoscope. This mesoscope allows for the recording of activity of over 10,000 neurons simultaneously at single-cell resolution with an acquisition rate of ~3 Hz over much of one hemisphere of the cerebral cortex of a mouse while they perform complex tasks in real-time. These tasks can involve seeing, hearing, smelling, movement, etc., even in a virtual environment.

During the auditory tests, the inventors detected unwanted high-frequency noise from the mesoscope. This noise is also detected by the mice performing auditory tasks and interferes with the data collection task being performed. Using a microphone and an oscilloscope, the inventors discovered the source of the noise to be the resonant scanners and the tubes that connect the light paths.

The inventors discovered a solution to reduce the noise level of the high-frequency sound, making use of 3D-Printers and software that enable complex designing and manufacturing of devices for optimizing scientific instrumentations. The inventors 3D-Scanned and designed a baffle having both solid and closed-cell honeycomb lattice. After installing the baffle, the high-frequency (12.5 kHz) noise went from 60 dB without the baffle down to an average of 10 dB with the baffle installed. The mice are not able to hear the high-frequency sound if the noise is below 16 dB at this sound frequency, based on published, species-specific auditory brainstem response threshold curves. Therefore, adding the baffle resulted in the mice not being able to hear the sound and also made a significant improvement in the data collection for both generic experiments where the mouse is awake and will experience stress as a result of the sound, and auditory-task-based research where precise control over the auditory environment is critical.

The sound pressure level was measured with a Bruel and Kjaer 4938 1/4" pressure-field microphone (see attached) calibrated with a Bruel and Kjaer Type 4231 Sound Calibrator, with the microphone signal amplified through a Bruel and Kjaer 1708 sound conditioner.

The sound measurements were recorded in fast fourier transform mode (FFT) on a Tektronix TDS 1002 oscilloscope. Each division on the oscilloscope display corresponded to an explicit, calibrated sound pressure level increment in decibels (dB). The output from the sound conditioner to the oscilloscope was sent by BNC cable.

A Creaform HandyScan Black Elite 3D-Scanner was used to create a digital CAD of the existing setup of the Thorlabs Mesoscope. Live capture of the scan data was performed using VX Elements software and the same software was used to convert the live scan to a CAD file of .STL (stereolithography) file format. Using the scanned CAD as a reference, the team designed a baffle to custom fit onto the mesoscope

resonant scanner and tubes using Autodesk Fusion-360 software. After designing the baffles, the CAD file of the designed baffle was imported into Materialise Magics software and the lattice design having solid walls followed by honeycomb latticing was designed using this software. The latticed files were saved in .STL file format. In addition, using Materialise Magics, a manual fixing operation was done on certain regions of the imported CAD which had unwanted noise shells, planar holes, overlapping triangles – all of which were fixed to ensure a high quality CAD file is produced for 3D-Printing.

The final baffle design included mediums of varying densities from solid walls and honeycomb latticing to 3D-Print in Nylon. The baffle was printed using Powder Bed Fusion technology using Formlabs Fuse machine, where Nylon material is sintered layer-by-layer to create a 3D-object. The design intent of the honeycomb lattice was to allow for nylon material to be enclosed inside the closed cells during the printing process. The inventors added a clearance design between the baffle and the resonant scanning components to allow for the fitment of polyurethane foam. This created a baffle where high-frequency sound waves had to penetrate through foam, solid nylon wall, enclosed nylon powder inside the honeycomb followed by a second solid wall.

The adhesive back foam used for the baffle was Polyurethane foam 1/8 inches thick and medium soft with 3/16 inch extra soft foam. The foam was cut to shape using a laser cutter to adhere to the 3D-Printed components. High-frequency noise data were collected with a microphone and an oscilloscope. The microphone was fixtured in the general area of the location where the mice would perform auditory tasks.

The baffle is a first-of-its-kind attachment to the Thorlabs Mesoscope (US Pats. 10901194 and 10295811, incorporated herein by reference) and is not commercially available. Since 3D printing allows for mass customization, this baffle can be adapted for any resonant scanning microscopes on the market. This will reduce unwanted noise levels from the experiments yielding to better experimental results for auditory research and also allow for a safer environment for users since the high-frequency noise can be detected by researchers and users of such devices.

The target goal was to lower the noise level to below 16 dB when performing auditory tasks on mice. The results indicate that the 12.5 kHz noise went from 60 dB without the baffle down to an average of 10 dB with the baffle installed.