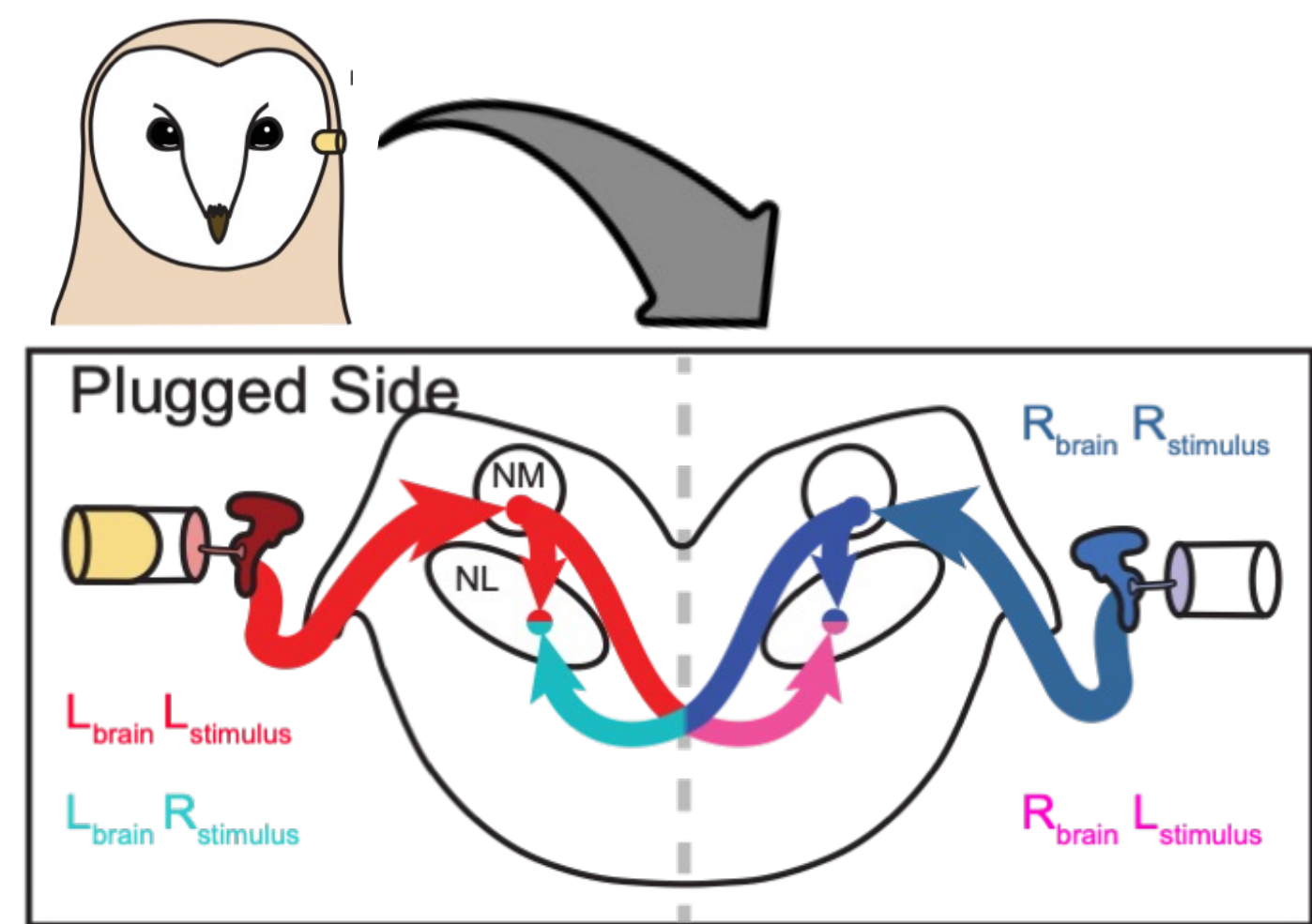


## Abstract

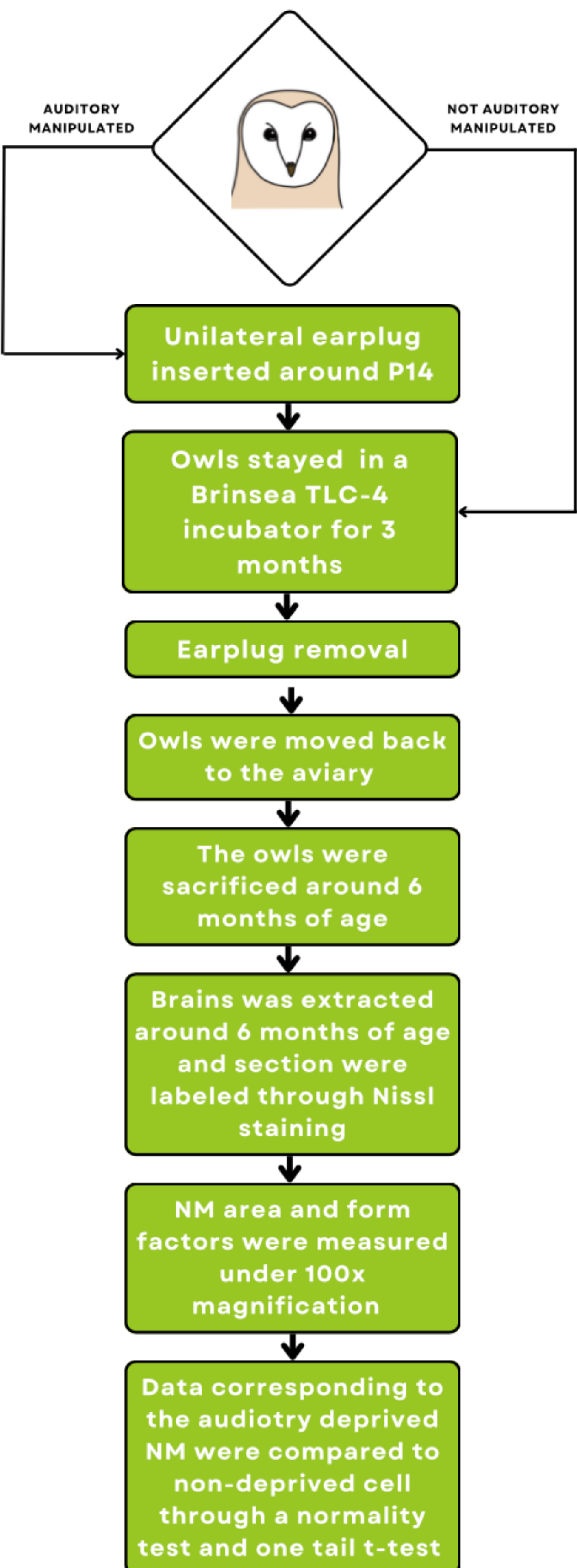
During development in barn owls (*Tyto furcata*), the interaural time differences (ITDs) circuit undergoes a progressive augmentation. At the core of the ITD detection mechanism lies the nucleus magnocellularis (NM) and nucleus laminaris (NL). NM and NL neurons together orchestrate precise auditory localization. The plasticity of the brain prompted an exploration into how the experience of barn owls influence the modulation of their ITD detection circuit. Auditory researchers have long suspected the dependence of NM neurons on auditory input. Since complete auditory isolation leads to significant changes in the morphology and physiology of NM cells, an investigation into mild acoustic deprivation could show the degree of auditory dependence NM cells require to develop [2,3,4]. A methodological approach involving the rearing of owls with unilateral ear inserts was designed to dampen the acoustic signal on one side. Upon reaching maturity, examination of the owls' brainstem revealed that NM cells situated ipsilateral to the earplug exhibited a decrease in size by approximately 7% compared to their contralateral counterparts. However, this only confined to regions corresponding to low frequency tonotopic domains. The rostral NM remained unchanged in size despite such alterations. Notably, the overall roundness or form factor of neurons within both caudal and rostral NM remained unaltered.

## Methodology

Leveraging Neuroanatomy to Create a Plan

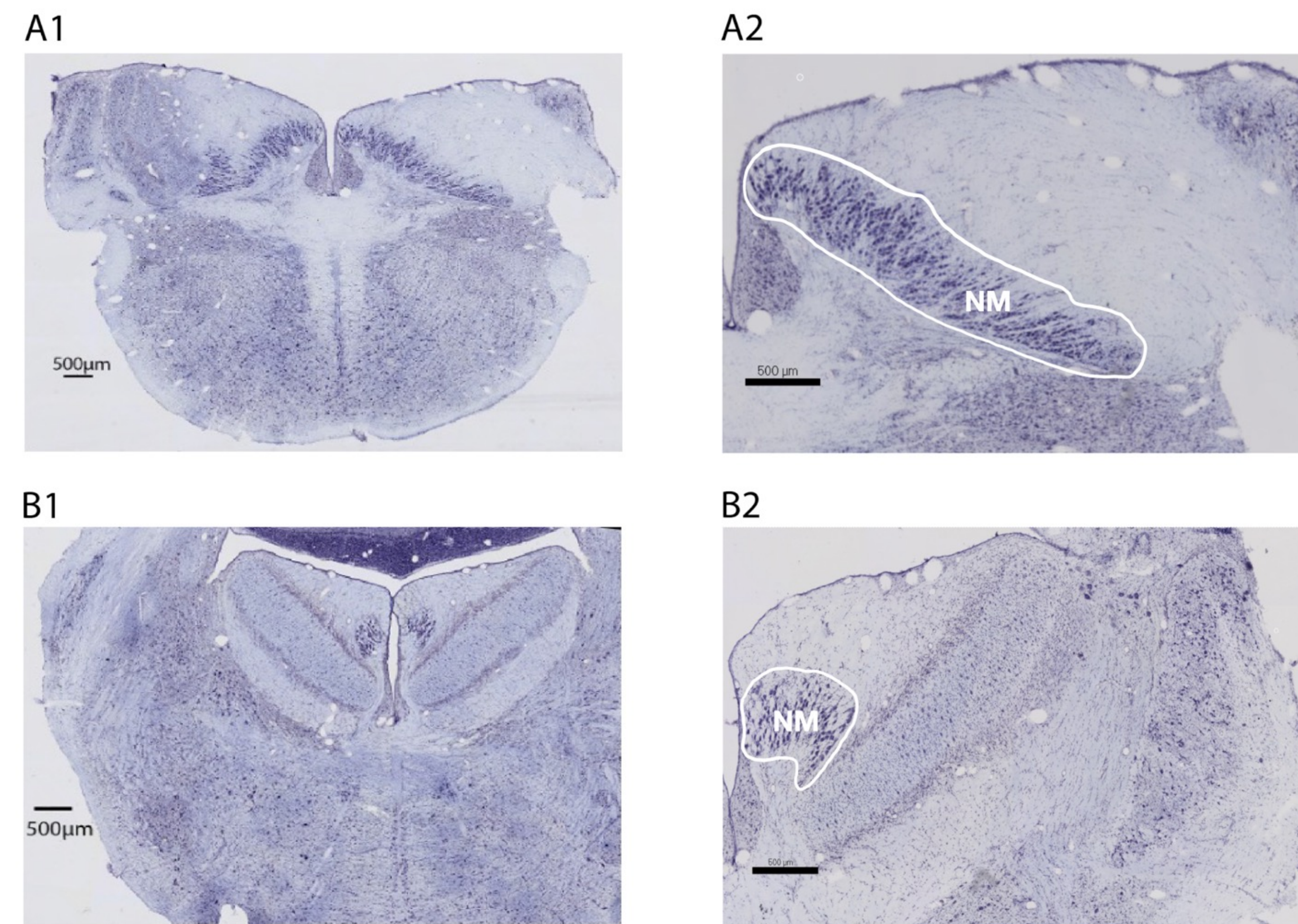


**Figure 1. ITD Circuit.** Earplug is placed in the left ear. Red lines represent left ear inputs to the left NM, which sends signals to both the ipsilateral (red) and contralateral (magenta) NL. Right ear inputs, shown in blue, reach the right NM which projects to both the ipsilateral NL (blue) and contralateral NL (cyan). From Carr et al., 2023.



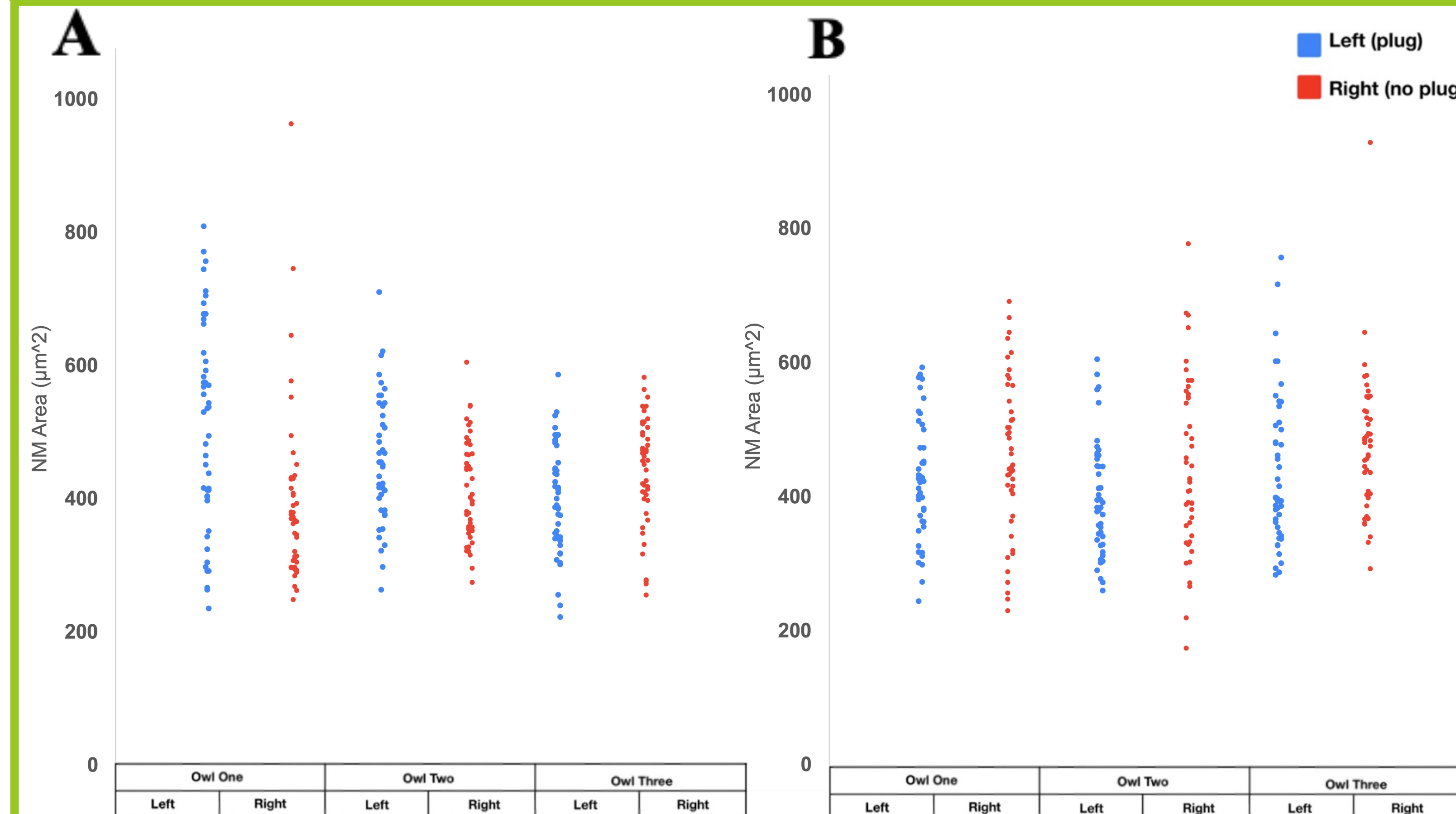
**Figure 2. Methodology Process.** Owl picture from Carr et al., 2023.

## Visualizing NM Neurons

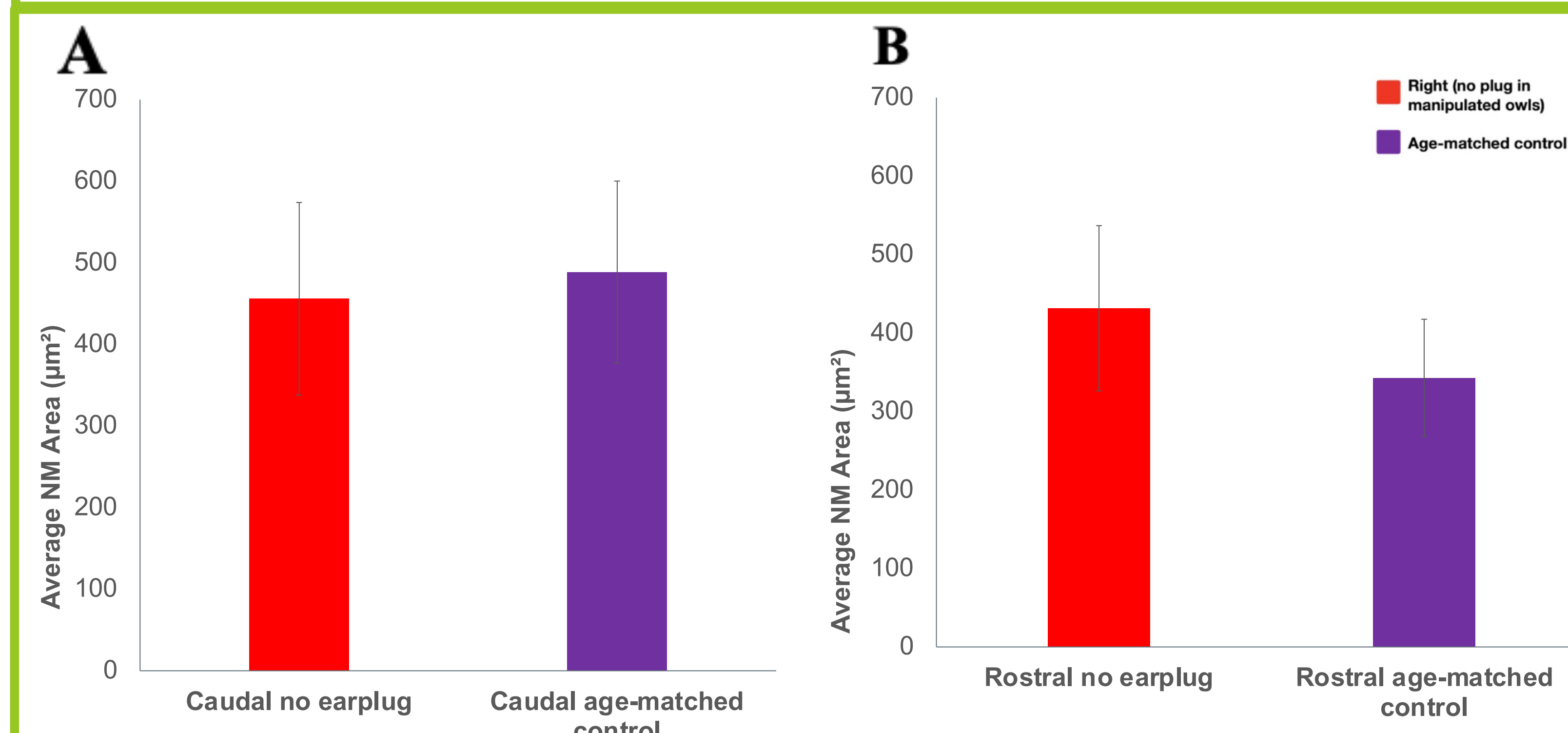


**Figure 3. NM Tonotopic Visualization.** Sections were stained with Nissl staining. Scale bars are found on each picture. A1 & A2) Caudal section at 1.25x and 10x respectively. B1 & B2) Rostral section at 1.25x and 10x respectively.

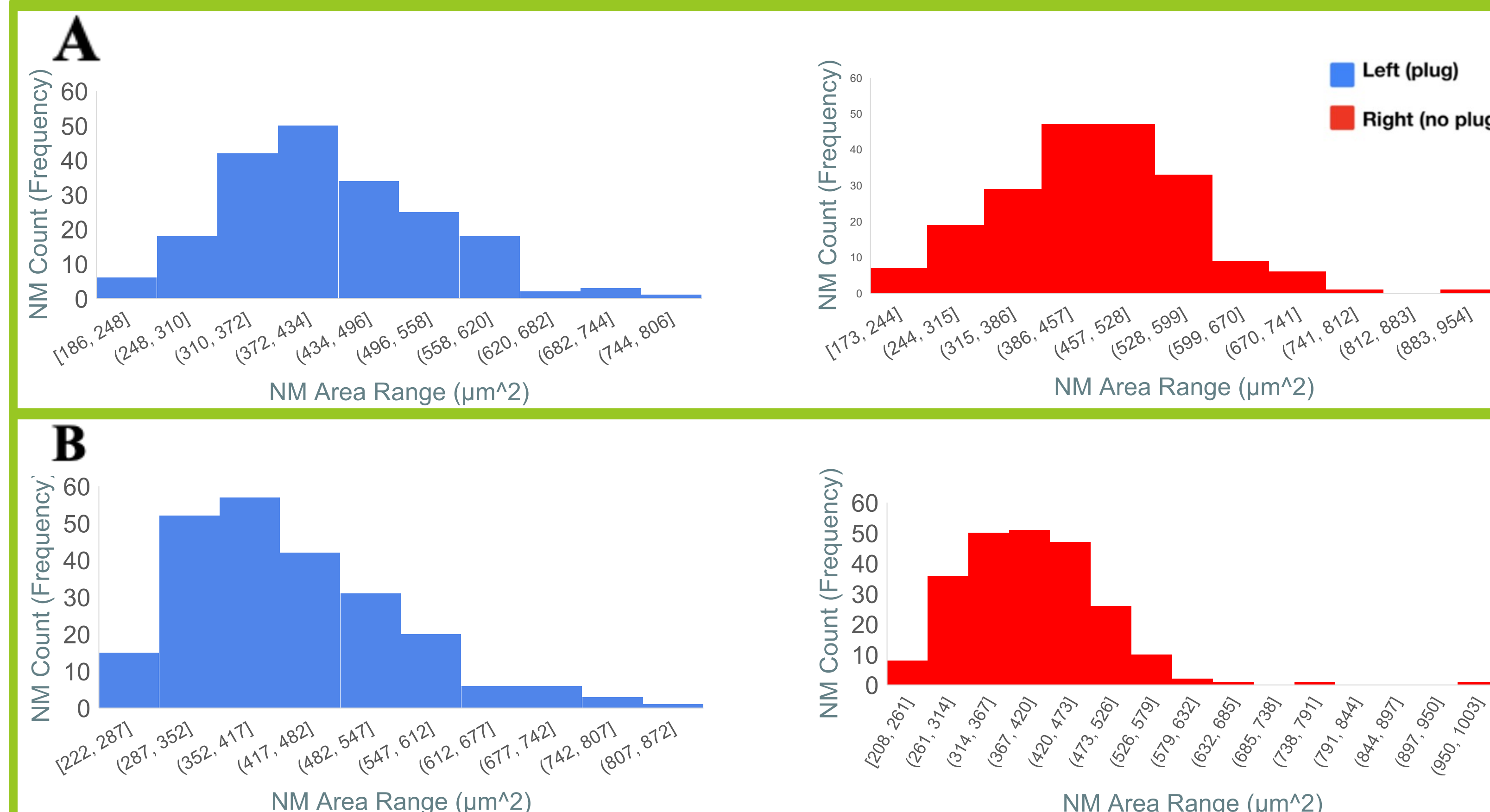
## Results



**Figure 4. NM cell area distribution.** n=3. Left=ear plug, right=no earplug. A) The area of caudal NM cells is plotted with respect to presence or absence of the earplug. B) The area of rostral NM cells is plotted with respect to presence or absence of the earplug.



**Figure 5. Comparing contralateral NM to age-matched control NM.** There is not a statistically significant difference between the area of the NM contralateral to the earplug and in the age-matched control NM. The same is true for the rostral NM.



**Figure 6. NM frequency of each area range.** All the plots are symmetrical, monomodal, and represent a normal distribution. n=3 for each distribution. A) The plot on the left corresponds to caudal NM. The mode is 372-434  $\mu\text{m}^2$ . The graph on the right portrays information about the right caudal NM with the mode being 386-528  $\mu\text{m}^2$ . B) Represents rostral NM. The right graph has a mode of 352-417  $\mu\text{m}^2$ . The right graph has a mode of 368-420  $\mu\text{m}^2$ .

	Area ( $\mu\text{m}^2$ )	Statistics	Form Factor	Statistics
Caudal plug side	426 $\pm$ 21	p=0.017	0.85 $\pm$ 0.01	p=0.423
Caudal no plug side	458 $\pm$ 25		0.85 $\pm$ 0.02	
Rostral plug side	442 $\pm$ 58	p=0.471	0.80 $\pm$ 0.02	p=0.808
Rostral no plug side	404 $\pm$ 28		0.80 $\pm$ 0.01	

**Table 1. NM morphology analysis.** n=3. The caudal NM ipsilateral to the earplug is smaller than the contralateral NM. The difference between the earplug and no earplug side is statistically significant. There is no statistical difference between the average area of rostral NM ipsilateral and contralateral to the earplug. The average form factor of caudal & rostral NM is the same on both sides. The p values were obtained using a paired 2-tailed t-test using the average area from each animal as a data point to account for dependence.

## Conclusion

Area

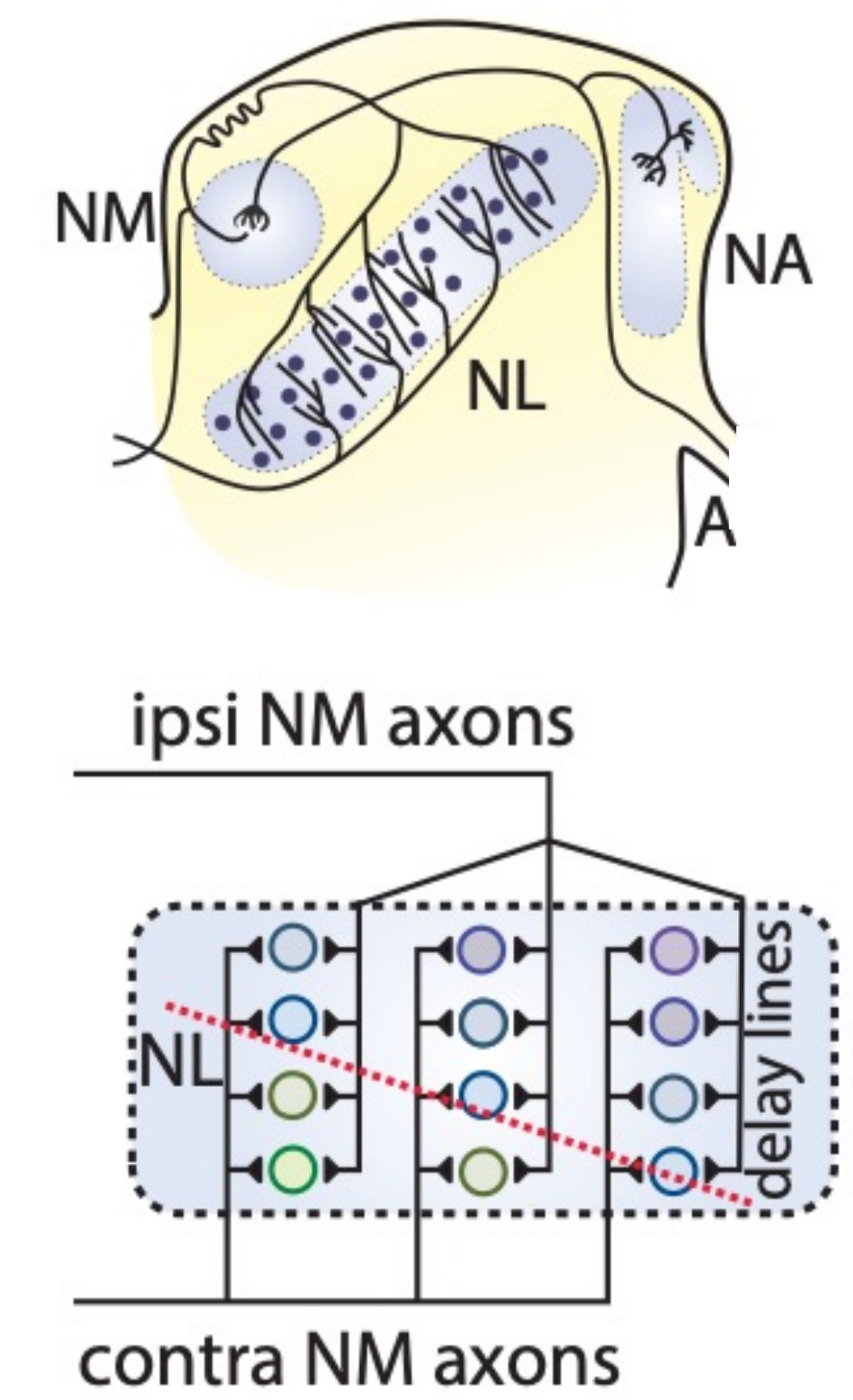
- Caudal NM cells ipsilateral to the earplug were 7% smaller than NM cells contralateral to the earplug.
- Rostral NM cells ipsilateral to the earplug were the same size as the NM cells contralateral to the earplug.

Form Factor

- The form factor of NM cells ipsilateral to the earplug was the same as NM cells contralateral to the earplug. This was true for both caudal and rostral NM cells.

Overall

- Change in the area of caudal NM cells could be explained by a compensatory shift to reestablish hemostasis [5].
- No change in the area of rostral NM cells could potentially be attributed to the small auditory reduction (15 dB) [5].
- Sample size needs to be increased (n=3).



**Figure 7. ITD Detection Circuit.** NM= delay line. NL= coincidence detectors. Adopted from Carr et al., 2023.

## Acknowledgement

This work was supported by The National Institutes of Health, Grant number RO1 DC-019341 to CEC.

## References

1. Carr, C. E., Wang, T., Kraemer, I., Kuokkanen, P. T., Capshaw, G., Ashida, G., Kemper, R., & Köppl, C. (2023). Experience-dependent plasticity in nucleus Laminaris of the barn owl. <https://doi.org/10.1101/2023.02.02.526884>
2. Edmonds, J. L., Jr, Hoover, L. A., & Durham, D. (1999). Breed differences in deafening-induced neuronal cell death and shrinkage in chick cochlear nucleus. *Hearing research*, 127(1-2), 62-76. [https://doi.org/10.1016/S0378-5955\(98\)00180-4](https://doi.org/10.1016/S0378-5955(98)00180-4)
3. Hamburger, V., & Levi-Montalcini, R. (1949). Proliferation, differentiation and degeneration in the spinal ganglia of the chick embryo under normal and experimental conditions. *Journal of Experimental Zoology*, 111(3), 457-501. <https://doi.org/10.1002/jez.1401110308>
4. Parks T. N. (1979). Afferent influences on the development of the brain stem auditory nuclei of the chicken: otocyst ablation. *The Journal of comparative neurology*, 183(3), 665-677. <https://doi.org/10.1002/cne.901830313>
5. Saunders, J. C., Coles, R. B., & Gates, G. R. (1973). The development of auditory evoked responses in the cochlea and cochlear nuclei of the chick. *Brain research*, 63, 59-74. [https://doi.org/10.1016/0006-8993\(73\)90076-0](https://doi.org/10.1016/0006-8993(73)90076-0)