THE REGISTRY

Newsletter of the NIDCD National Temporal Bone, Hearing and Balance Pathology Resource Registry

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CONTENTS

MISSION STATEMENT

The NIDCD National Temporal Bone, Hearing and Balance Pathology Resource Registry was established in 1992 by the National Institute on Deafness and Other Communication Disorders (NIDCD) of the National Institutes of Health (NIH) to continue and expand upon the former National Temporal Bone Banks (NTBB) Program. The Registry promotes research on hearing and balance disorders and serves as a resource for the public and the scientific community about research on the pathology of the human auditory and vestibular systems.

Correlation of CT and Histopathology in Resorption of the Distal Long Process of the Incus

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In our recently published paper in *Clinical Neuroradiology*[1], we discuss the computed tomography (CT) and histopathologic correlation of resorption of the distal long process of the incus. While complete loss of the distal long process and lenticular process of the incus is a known radiologic and clinical cause of a conductive hearing loss with an increased air bone gap, we report a case of incomplete resorption of the distal incus that also resulted in hearing loss.

The case studied was of a patient who had experienced a mixed hearing loss in her left ear with a conductive component and an increased air bone gap. Following donation of the patient's temporal bones to the National Temporal Bone Registry at Massachusetts Eye and Ear, temporal bone removal and processing were performed. After removal and during fixation, a high-resolution CT of the temporal bones was performed and the temporal bones were then processed, serially sectioned, and stained with hematoxylin and eosin (H&E).

Upon initial observation of the CT of the donated temporal bone specimen, the incudostapedial articulation was preserved bilaterally. However, in comparison with the right ear, there was marked thinning and diminished density measured in Hounsfield units of the left incus, suggesting resorption (Figures 1–3). The average Hounsfield unit (HU) measurement of the left incus was 544 HU as opposed to 1023 HU in the right incus (Figure 3). Correlation with the histopathologic findings in the same patient demonstrated marked areas of bony resorption and presence of Howship's lacunae in the long process of the left incus, indicating areas of osteoclastic resorption (Figure 4a). On the right, the long process of the incus was preserved without evidence of osteoclastic resorption (Figure 4b).

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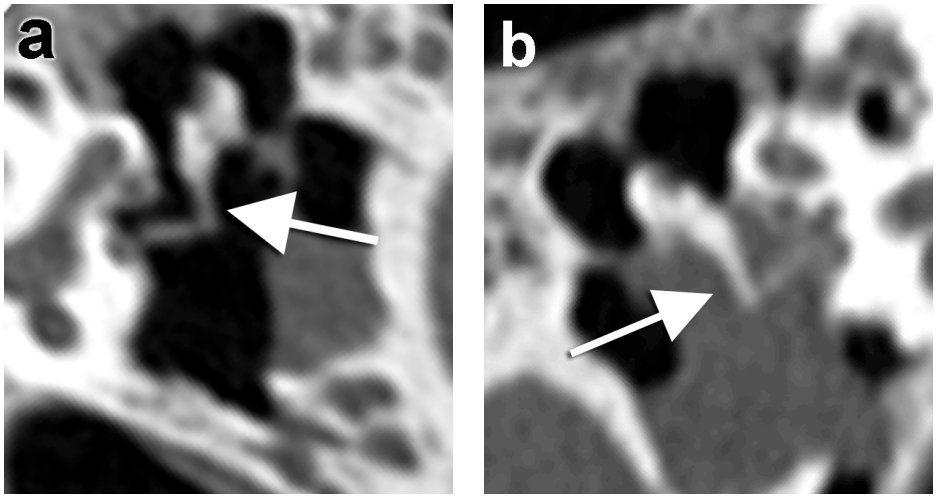

Figure 1. Pöschl plane CT reformat of the pathologic specimen demonstrates marked tapering of the distal left incus (a, arrow) when compared with the right (b, arrow). The incudostapedial relationships are maintained.

Reinshagen KL, Nadol JB, Juliano AF, Curtin HD. Correlation of CT and histopathology in resorption of the distal long process of the incus. Clin Neuroradiol. 2017 Sep 15. doi: 10.1007/s00062-017-0627-3.

The incus is the most vulnerable of the middle ear ossicles. In particular, the distal long process is commonly eroded by middle ear pathologies. However, in the absence of a known pathology, idiopathic resorption of the distal incus can still occur.[2] The cause of resorption of the distal incus has been debated. Previous theories about the lack of blood supply resulting in resorption have since been refuted.[3] In fact, increased vascular supply and canal openings within the distal incus may be a potential cause of resorption.[3,4] Alternatively, reduced expression of osteoprotegerin may cause increased resorption in the middle ear ossicles.[5] Furthermore, the incus, unlike the stapes, undergoes remodeling throughout life and progressive resorption with advancing age.[3,6,7] Impaired remodeling of the distal long process may be responsible for impaired age-related bone resorption.[2,6] Partial but incomplete ossicular discontinuity has been shown to cause an air bone gap with high-frequency conductive hearing loss.[8]

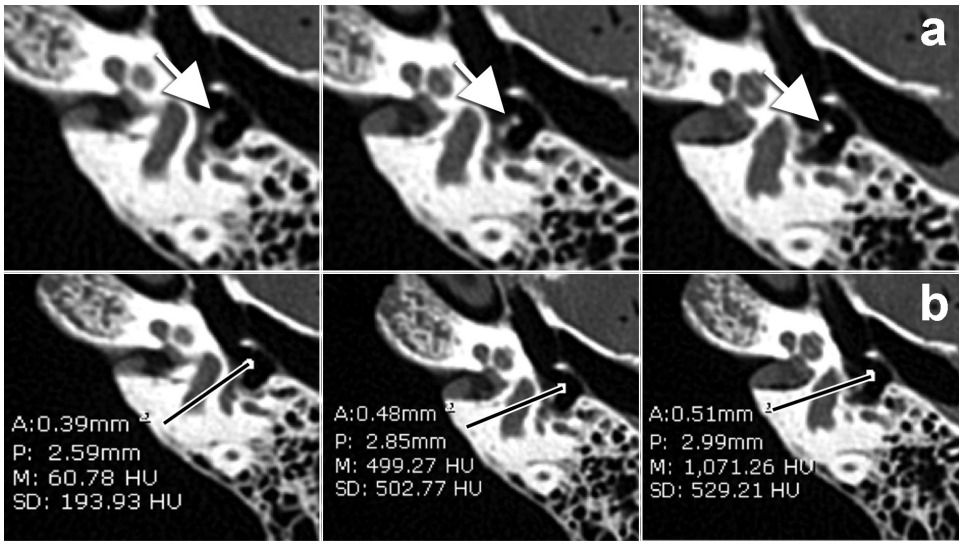

Figure 2. Axial CT images (a) with density measurements (b) of three contiguous images through the distal long process of the left incus (arrow).

Reinshagen KL, Nadol JB, Juliano AF, Curtin HD. Correlation of CT and histopathology in resorption of the distal long process of the incus. Clin Neuroradiol. 2017 Sep 15. doi: 10.1007/s00062-017-0627-3.

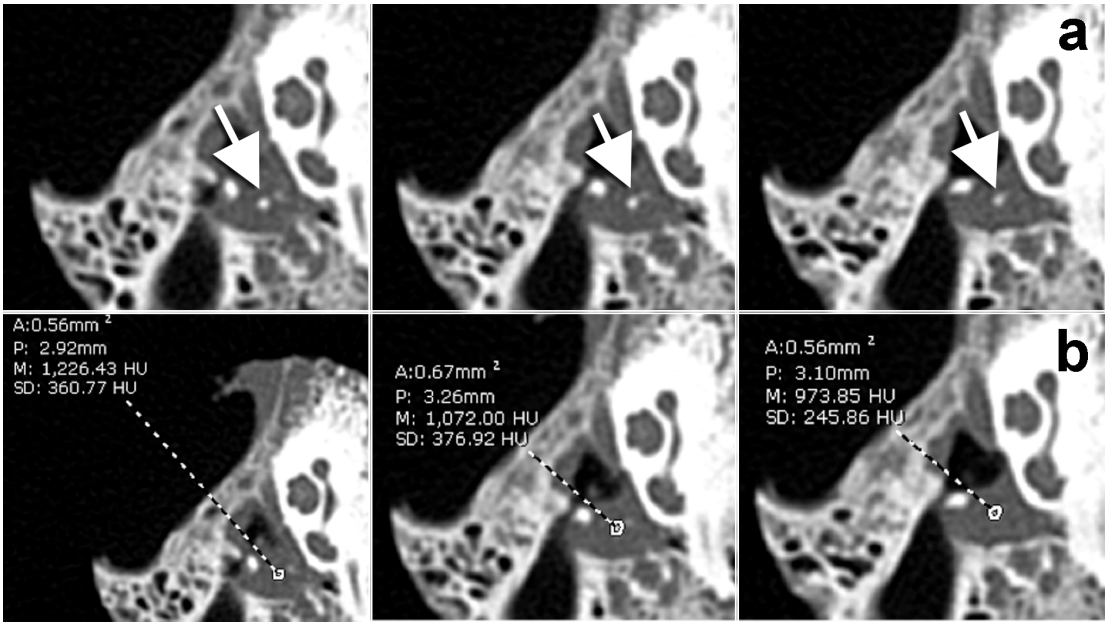

Figure 3. Axial CT images (a) with density measurements (b) of three contiguous images through the distal long process of the right incus (arrow).

Reinshagen KL, Nadol JB, Juliano AF, Curtin HD. Correlation of CT and histopathology in resorption of the distal long process of the incus. Clin Neuroradiol. 2017 Sep 15. doi: 10.1007/s00062-017-0627-3.

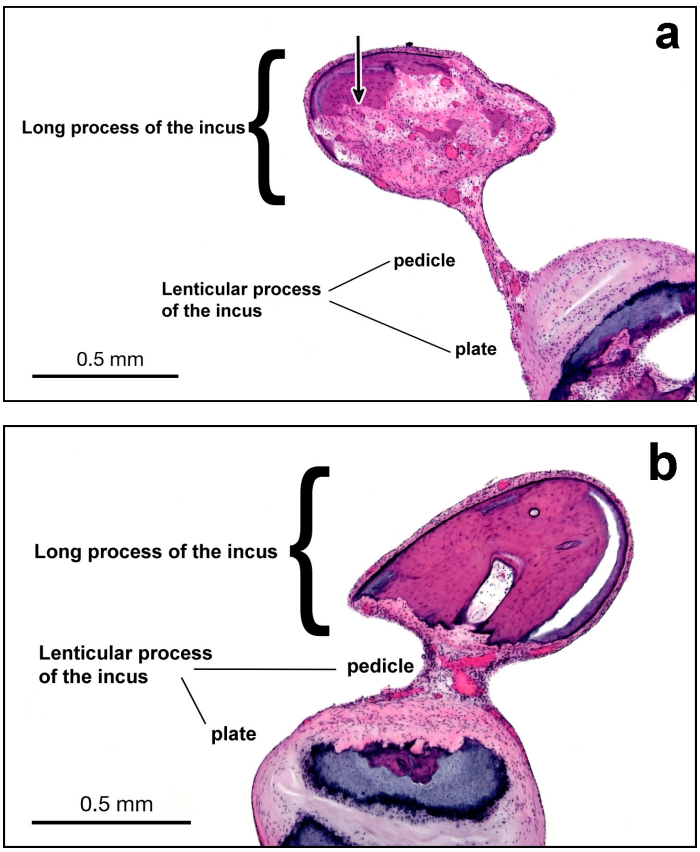

Figure 4. Histology of the distal long process of the incus on the left (a) and right (b) at 4x power. On the left, there are marked areas of bony resorption and presence of Howship's lacunae (arrow) at the site indicating margins of osteoclastic resorption. On the right, there are preserved osteocytes and absence of osteoclastic resorption.

Reinshagen KL, Nadol JB, Juliano AF, Curtin HD. Correlation of CT and histopathology in resorption of the distal long process of the incus. Clin Neuroradiol. 2017 Sep 15. doi: 10.1007/s00062-017-0627-3.

This finding can be an important cause of conductive hearing loss that may be underrecognized by the radiologist as a result of the preserved incudostapedial articulation. Marked tapering of the distal incus as well as decreased attenuation may be key radiographic features that can help diagnose this subtle cause of conductive hearing loss. Further studies will help to confirm the reproducibility of CT to detect this abnormality *in vivo*.

ACKNOWLEDGEMENTS

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REFERENCES

1. Reinshagen KL, Nadol JB, Juliano AF, Curtin HD. Correlation of CT and histopathology in resorption of the distal long process of the incus. Clin Neuroradiol. 2017 Sep 15. doi: 10.1007/s00062-017-0627-3.

2. Imauchi Y, Karino S, Yamasoba T. Acquired atrophy of the long process of the incus. Otolaryngol Head Neck Surg. 2005;132(1):156–8.

3. Chien W, Northrop C, Levine S, Pilch BZ, Peake WT, Rosowski JJ, Merchant SN. Anatomy of the distal incus in humans. J Assoc Res Otolaryngol. 2009;10(4):485–96.

4. Chen H, Okumura T, Emura S, Shoumura S. Scanning electron microscopic study of the human auditory ossicles. Ann Anat. 2008;190(1):53–8.

5. Kanzaki S, Ito M, Takada Y, Ogawa K, Matsuo K. Resorption of auditory ossicles and hearing loss in mice lacking osteoprotegerin. Bone. 2006;39(2):414–9.

6. Lannigan FJ, O'Higgins P, Oxnard CE, McPhie P. Age-related bone resorption in the normal incus: A case of maladaptive remodeling? J Anat. 1995;186(Pt3):651–5.

7. Anson BJ, Bast TH. Development of the incus of the human ear: Illustrated in atlas series. Q Bull Northwest Univ Med Sch. 1959;33(2):110–9.

8. Farahmand RB, Merchant GR, Lookabaugh SA, Röösli C, Ulku CH, McKenna MJ, de Venecia RK, Halpin CF, Rosowski JJ, Nakajima HH. The audiometric and mechanical effects of partial ossicular discontinuity. Ear Hear. 2016 Mar–Apr;37(2):206–15.

Human Cochlear Histopathology Reflects Clinical Signatures of Primary Neural Degeneration

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Damage to the auditory nerve, such as that caused by noise trauma, genetic conditions, or the effects of normal aging, can lead to problems in the functional transmission of sound to the brain. People who respond normally to tests evaluating the sensory cells of the ear but abnormally to tests singling out the auditory nerve can be diagnosed with auditory neuropathy, a significant and understudied form of human hearing loss.[1] It is difficult to estimate the population-level prevalence of auditory neuropathy because the most widely used diagnostic test in clinical audiology, the pure tone audiogram, does not yield results specific enough to determine which cells are damaged in a given patient.[2] Because the inner ear cannot be biopsied in life, the histopathological study of human temporal bones forms the basis of our scientific understanding of many auditory disorders. The purpose of this study was to identify patients with severe primary neural degeneration and model their neuronal loss in relation to their audiometric thresholds and word recognition scores.

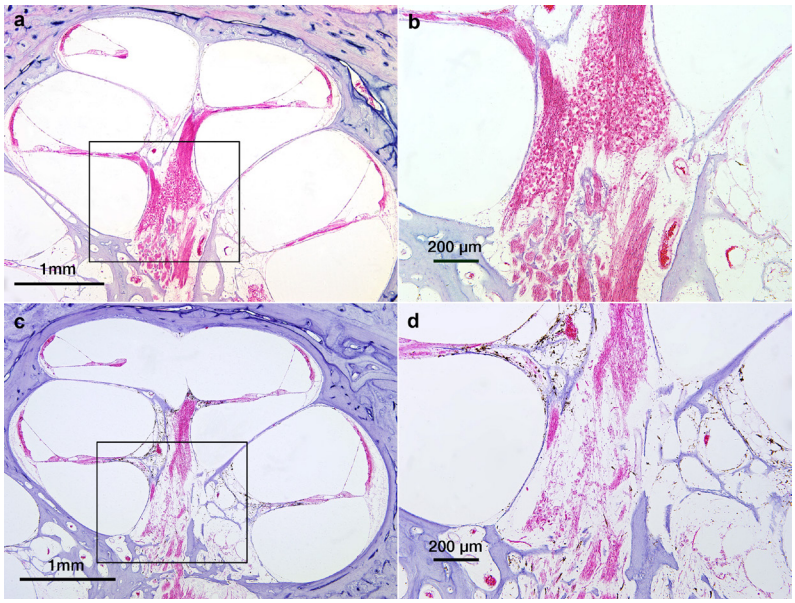

Figure 1. Representative cochlear histopathology in severe primary neural degeneration compared to age-matched control. In both examples, sensory structures are normal. **A.** Mid-modiolar section (4X) from the cochlea of a patient with appropriate SGN numbers for age; scale bar 1 mm. The boxed area is shown magnified in B. **B.** 10X magnification of the modiolus in A; scale bar 200 µm. **C.** Mid-modiolar section (4X) from an age-matched patient with severe primary neural degeneration showing 85 percent fewer total SGN cell bodies than expected for age; scale bar 1 mm. The boxed area is shown magnified in D. **D.** 10X magnification of the modiolus in C; scale bar, 200 µm.

Sagers JE, Landegger LD, Worthington S, Nadol JB, Stankovic KM. Human cochlear histopathology reflects clinical signatures of primary neural degeneration. Sci Rep. 2017 Jul 7;7(1):4884.

In normally aging individuals, sectioning the cochlea along its central axis reveals a large, healthy population of spiral ganglion neuron (SGN) cell bodies inside Rosenthal's canal, a spiraling channel within the temporal bone. In individuals with severe neural degeneration, few neuronal cell bodies are observed in this canal, though sensory cells of the organ of Corti appear in normal numbers (Figure 1). We identified 30 ears from 23 patients with spiral ganglion neuron populations that fell at least one standard deviation below the mean number of neuronal cell bodies expected for age[3], but demonstrated age-appropriate populations of cochlear sensory cells. These patients ranged in age (11–99 years, median 67 years), sex, and auditory diagnosis—from neural presbycusis to near-complete auditory neuropathy due to Mohr-Tranebjaerg syndrome. We then generated hypothesis-driven statistical models to evaluate the relationship of primary neuronal degeneration with pure tone audiometric thresholds and word recognition scores.

Specifically, we used a linear mixed effects regression model to correlate audiometric thresholds at each of six audiometric test frequencies with total neuronal loss, represented for each patient

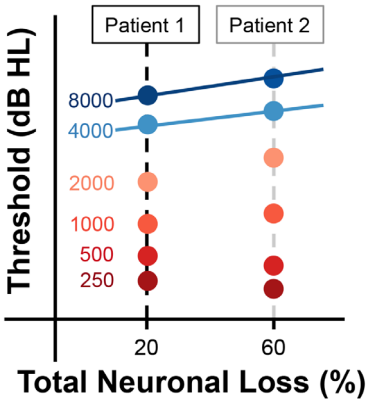

Figure 2. Schematic demonstrating the configuration of the linear mixed model depicted in Figure 3. Four to six hearing threshold observations per patient (depending on which frequencies were clinically examined), color-coded by each individual audiometric test frequency, were graphed vertically on the y-axis at the point along the x-axis representing total neuronal loss for that patient (as percent of age-matched control). In this way, relationships among patients at individual audiometric test frequencies can be examined overall as a function of total neuronal loss.

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as a percentage of the mean number of spiral ganglion neuron cell bodies expected for age.[3] For each ear, hearing thresholds were color-coded to represent the frequency at which each threshold was recorded and then graphed in a vertical line along the y-axis at the single point along the x-axis representing the amount of total neuronal loss observed in that ear (Figure 2). This allowed us to visualize audiometric threshold relationships among patients as a function of total neuronal loss.

We found that neuronal loss in patients with severe primary neural degeneration correlated significantly with elevated hearing thresholds and poor word recognition scores (Figure 3). Averaging thresholds across all six audiometric test frequencies, our model yielded a mean threshold increase of 6.0 dB HL per 10 percent neuronal loss, though the rate of threshold increase was steeper among low audiometric test frequencies (0.5-2 kHz) than high audiometric test frequencies (4-8 kHz) (Figure 3a–b). Word recognition scores were available for 15 ears, in which word recognition decreased by 6.8 percent per 10 percent neuronal loss (Figure 3c). Importantly, as observed in Figure 3a, a significant elevation in the y-intercept for hearing thresholds measured at high test frequencies (4–8 kHz) highlighted the contribution of threshold-elevating factors that cannot be attributed solely to neuronal loss.

Though the positive relationship in this population between neuronal loss, word recognition scores, and hearing thresholds is clear, a simple linear increase is not well explained by animal models of auditory neuropathy.[4–6] Importantly, though the

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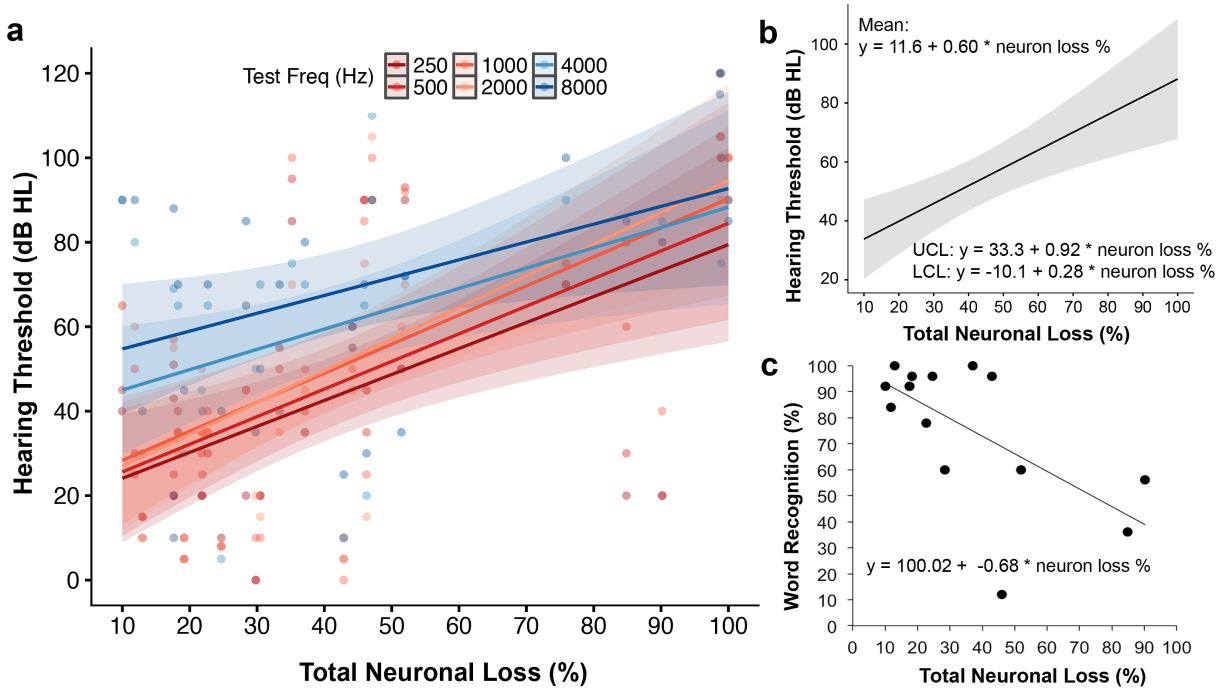

Figure 3. Primary neuronal loss correlates significantly with elevated audiometric thresholds and poor word recognition scores. Confidence intervals are represented as shaded areas around regression lines and are measures of 95 percent precision for predicted slopes and intercepts. **A.** Linear mixed regression model correlating total neuronal loss (percent of age-based mean) with audiometric thresholds (dB HL) within patients at each individual test frequency (n=30 ears). Solid lines, linear regressions per audiometric test frequency (Freq); data points, individual threshold observations color-coded by audiometric test frequency. Conditional f test with Kenward-Rogers correction for degrees of freedom reveals that slopes of these lines are significantly different than zero ($p<0.001$), but not significantly different from one another ($p=0.10$). **B.** Averaging across all six audiometric test frequencies yields a mean threshold increase of 6.0 dB HL per 10 percent total neuronal loss; shaded area, 95 percent confidence interval. **C.** Total neuronal loss (percent of age-based mean) correlates with poor word recognition (n=15 ears). Mean word recognition score decreases by 6.8 percent per 10 percent total neuronal loss ($r = -0.644$).

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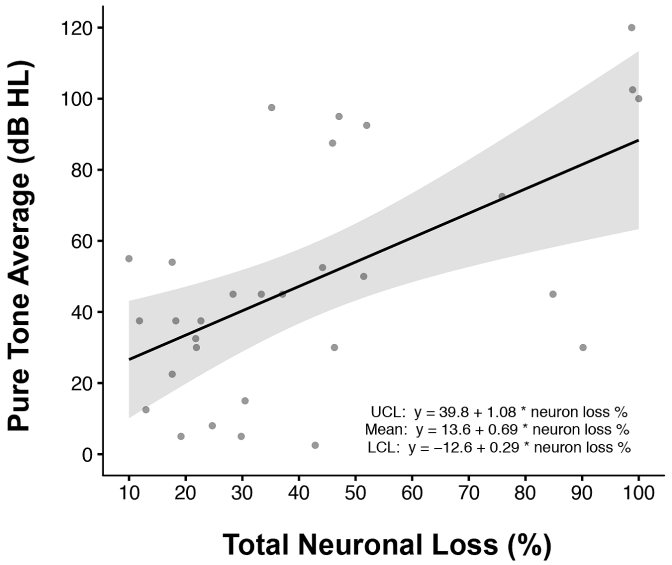

Figure 4. Relationship between pure tone average (dB HL) and total neuronal loss (percent of age-based mean) in severe neural degeneration (n=30 ears). In patients with severe neural degeneration, mean hearing thresholds increase by 6.9 dB HL per 10 percent total neuronal loss. Shaded area, 95 percent confidence interval; UCL, upper confidence limit; LCL, lower confidence limit.

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sensory cells of patients presented in this study were determined to be morphologically intact and present in age-appropriate numbers, we cannot determine to what degree these cells were functional in life. As our patients ranged widely in age and diagnosis, it is also helpful to remember that diseases thought to be primarily neurodegenerative can also negatively affect the function of and communication between neighboring or interconnected cells. For example, our model does not account for damage to the stria vascularis, which has been documented in other studies to contribute to hearing loss in humans.[7]

Our models have important clinical implications for patients considering cochlear implantation. If severe neural degeneration is suspected in a patient, his or her total neuronal loss could potentially be approximated using pure tone threshold average (Figure 4). Currently, patients who score below 60 percent on a word recognition test are considered implant candidates, and auditory rehabilitation following cochlear implantation is correlated with the number of surviving cochlear neurons.[8] Our data suggest that for people with severe primary neuronal degeneration, the 40 percent drop in word recognition that precedes cochlear implant candidacy corresponds with a 58.8 percent loss of spiral ganglion neurons.

Because there is currently no way for a clinician to confirm a diagnosis of primary neural degeneration or verify predicted loss in a living patient, our findings also highlight the need to develop clinically relevant imaging tools that enable cellular-level resolution of structures within the inner ear.[9] Additionally, the fact that neuronal cell bodies can remain alive in the modiolus for years after losing connections with sensory cells[10] motivates a fascinating direction for therapeutic innovation. Viral expression of neurotrophic factors may catalyze the successful regeneration of connections between remaining neurons and sensory cells, as recently shown.[11]

REFERENCES

1. Starr A, Picton TW, Sininger Y, Hood LJ, Berlin CI. Auditory neuropathy. Brain. 1996;119,741–53.

2. Landegger LD, Psaltis D, Stankovic KM. Human audiometric thresholds do not predict specific cellular damage in the inner ear. Hear Res. 2016;335,83–93.

3. Makary CA, Shin J, Kujawa SG, Liberman, MC, Merchant SN. Age-related primary cochlear neuronal degeneration in human temporal bones. JARO. 2011;12,711–717.

4. Kujawa SG, Liberman MC. Adding insult to injury: Cochlear nerve degeneration after "temporary" noise-induced hearing loss. J Neurosci. 2009;29,14077–85.

5. Woellner RC, Schuknecht HF. Hearing loss from lesions of the cochlear nerve: An experimental and clinical study. Trans Am Acad Ophthalmol Otolaryngol. 1955;59,147–9.

6. Lobarinas E, Salvi R, Ding D. Insensitivity of the audiogram to carboplatin induced inner hair cell loss in chinchillas. Hear Res. 2013;0,113–120.

7. Pauler M, Schuknecht HF, White JA. Atrophy of the stria vascularis as a cause of sensorineural hearing loss. Laryngoscope. 1988;98,754–759.

8. Seyyedi M, Viana LM, Nadol JB Jr. Within-subject comparison of word recognition and spiral ganglion cell count in bilateral cochlear implant recipients. Otol Neurotol. 2014;35,1446–50.

9. Iyer JS, et al. Micro-optical coherence tomography of the mammalian cochlea. Sci Rep. 2016;6,3328.

10. Felix H, Pollak A, Gleeson M, Johnsson LG. Degeneration pattern of human first-order cochlear neurons. Adv Otorhinolaryngol. 2002;59,116–23.

11. Suzuki J, Corfas G, Liberman MC. Round-window delivery of neurotrophin 3 regenerates cochlear synapses after acoustic overexposure. Sci Rep. 2016;25,6;24907.

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To learn more or view the database, visit
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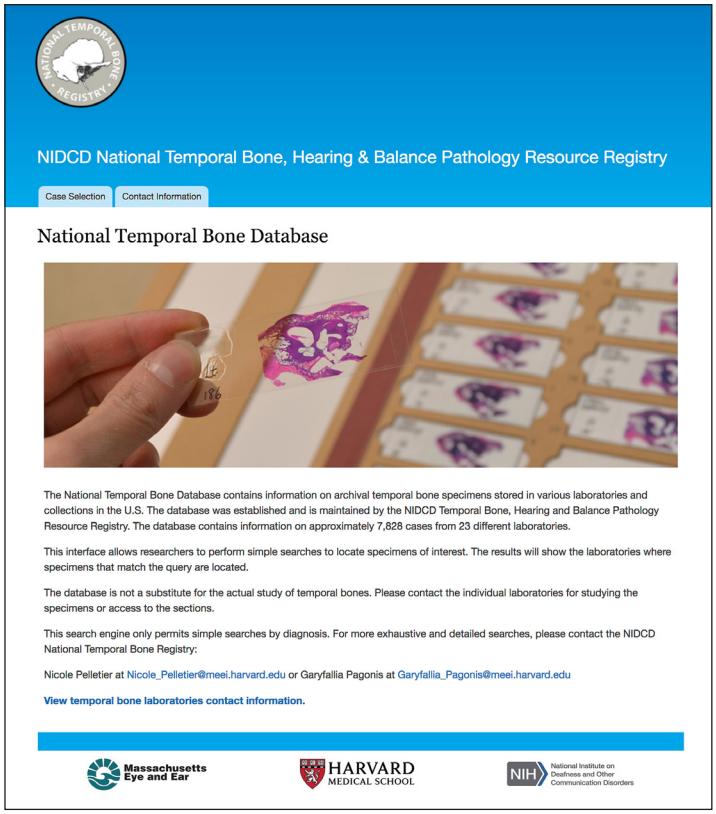

Otopathology Mini-Travel Fellowship Program

The NIDCD National Temporal Bone Registry's mini-travel fellowships provide funds for research technicians and young investigators to visit a temporal bone laboratory for a brief educational visit, lasting approximately one week. The emphasis is on the training of research assistants, technicians, and junior faculty.

These fellowships are available to:

- U.S. hospital departments who aspire to start a new temporal bone laboratory
- Inactive U.S. temporal bone laboratories who wish to reactivate their collections
- Active U.S. temporal bone laboratories who wish to learn new research techniques

Up to two fellowship awards will be made each year ($1,000 per fellowship). The funds may be used to defray travel and lodging expenses. Applications will be decided on merit.

Interested applicants should submit the following:

- An outline of the educational or training aspect of the proposed fellowship (1–2 pages)
- Applicant's curriculum vitae
- Letter of support from temporal bone laboratory director or department chairman
- Letter from the host temporal bone laboratory, indicating willingness to receive the traveling fellow

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