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Serum Levels of Stereocilin as a Hearing Biomarker

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Abstract

Noise-induced hearing loss (NIHL) often presents with an insidious onset, resulting from the cumulative effect of chronic, high-level noise exposure regardless of etiology. Stereocilin (STRC) is a protein that supports stereocilia attachment and cochlear hair cell function, 2 common targets of noise trauma. In this study, we explored the relationship between STRC and daily noise exposure in young, healthy adults. We found that higher noise exposure levels were associated with lower serum levels of STRC, as was the case for another inner-ear protein, prestin. There was a statistically significant positive correlation between serum STRC and prestin levels. These results support a biomarker approach for the diagnosis and monitoring of NIHL. The ability to detect and measure STRC in the blood also has implications for targeted gene therapy. STRC mutations are known to be associated with autosomal recessive deafness, a condition that is now amenable to targeted gene therapy.

Keywords

biomarker, noise-induced hearing loss, prestin, stereocilin

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One in 4 adults in the United States aged 20 to 69 years old have audiological features of noise-induced hearing loss (NIHL).[1] High-level noise exposure (>85 dB) can result in damage to the outer hair cells (OHCs), resulting in ineffective sound amplification and permanent hearing loss.[2] Blood-based biomarkers are noninvasive, inexpensive, and may be able to identify changes in the inner ear caused by noise exposure before clinically noticeable symptoms.[3] Inner ear proteins have been demonstrated to cross the labyrinth-blood barrier and enter the systemic circulation.[3] An example is prestin, an 81 kDa OHC membrane protein responsible for cochlear amplification. Among young adults, average serum prestin levels demonstrated a negative correlation to the average daily level of noise exposure.[3]

Stereocilin (STRC) is a 193 kDa inner ear-specific protein encoded by the STRC gene that is present in all 6 sensory areas of the inner ear.[4] On a structural level, STRC plays a role in the connection between the stereocilia bundle of OHCs as well the crowns that connect the tallest stereocilia to the tectorial membrane.[5] Noise exposure results in splaying, fusion, and loss of functional stereocilia.[6] We hypothesize that STRC will be detectable in the blood of normal-hearing human subjects and that its levels will be influenced by daily noise exposure. The findings of this study have the potential to add to the diagnostic potential of serum inner ear markers in the assessment of cochlear health.

Methods

The details of the Methods of this research were previously published.[3,7] Participants included 33 undergraduate students ages 18 to 24 years old at the University of Connecticut consented under Institutional Review Board approval (#H14-214). Exclusion criteria included students with clinically diagnosed hearing loss, chronic ear infections, ear surgery, head trauma, seizures, or neuropathy. Average daily noise exposure was collected for 3, nonconsecutive, weeks using a body-worn noise dosimeter. The dosimeters were configured to an 85-dBA criterion level and 3-dB exchange rate, in conformance with the National Institute for Occupational Safety and Health (NIOSH) recommended criteria[8] and a 70-dBA threshold. From the daily dosimeter data, we derived 2 related measurements: an 8-h-normalized A-weighted equivalent continuous sound level (LAeq,8h; dB), and the daily noise dose (expressed as a percentage) derived from this normalized sound level. Exposures at or exceeding the NIOSH recommended exposure limit of 85 LAeq,8h (dB), had noise doses >100%. The risk of hearing damage is assumed to be higher in those with higher doses. One of 5 nonfasting blood samples collected from each participant throughout the investigation was selected for STRC quantification because we previously demonstrated the stability of otologic biomarker levels across multiple

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measurements over 6 months in these subjects.[7] STRC levels in the serum were measured using the MBS9714058 ELISA kit, which has a detection range of 0.5 to 7 μg/mL (MyBioSource). Prestin levels in the serum were measured using the MBS167508 ELISA kit, which has a detection range of 10 to 3000 pg/mL (MyBioSource) and averaged across the 5 sessions.[3] Two subjects with outlier values above the STRC upper detection range were excluded. Two-tailed significance levels were applied for statistical analyses.

Results

Serum STRC levels ranged from 3.16 to 5.72 μg/mL, with a mean ± standard error of the mean (SEM) of 4.11 ± 0.11 μg/mL and a median of 4.08 μg/mL. Serum prestin levels ranged from 35.14 to 118.23 pg/mL, with a mean ± SEM of 196 ± 44.77 pg/mL and a median of 98.82 pg/mL.

Shapiro-Wilk tests of normality demonstrated statistically significant differences from normal for both distributions ($P < .001$), necessitating nonparametric statistical analyses.

Correlation analysis demonstrated a strong positive correlation between serum STRC and prestin levels (Spearman's $\rho = 0.59$, $P < .001$) (**Figure 1**). For both, STRC and prestin, higher noise exposure levels were associated with lower levels of both STRC and prestin (**Figure 2**). There was a statistically significant negative correlation between serum STRC and noise exposure levels as expressed in percent daily dose with a

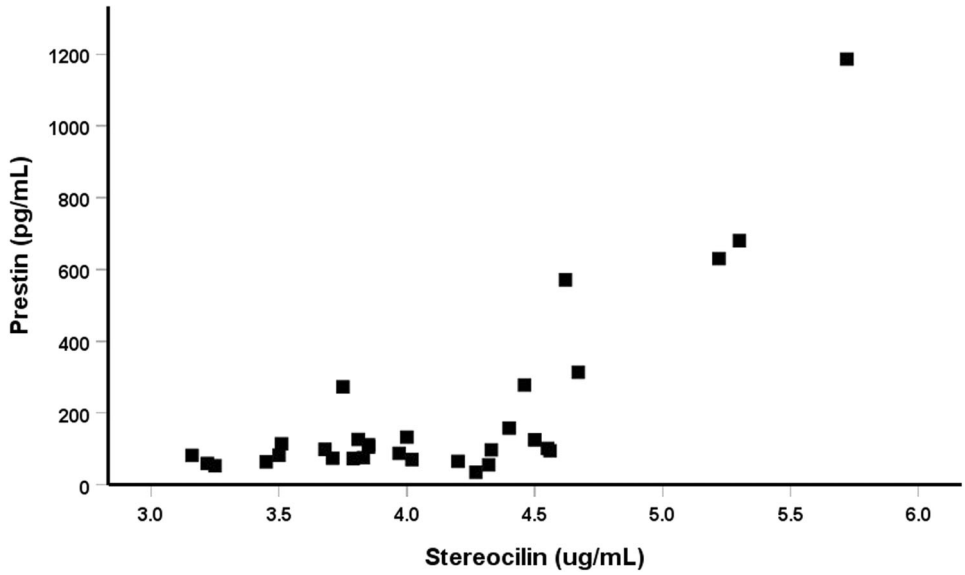

Figure 1. The correlation between stereocilin and prestin serum levels.

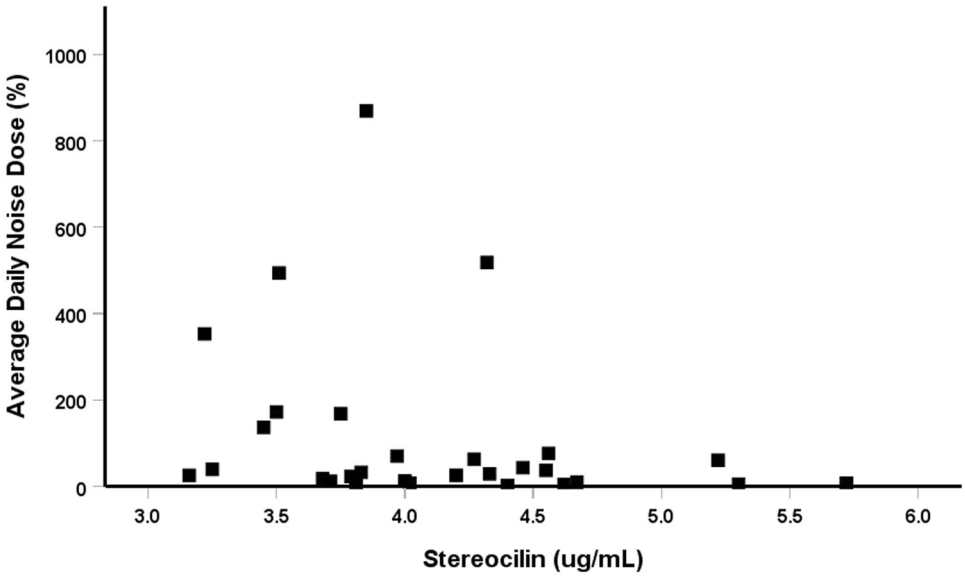

Figure 2. The correlation between serum stereocilin levels and average daily noise dose.

Spearman's ρ of −0.37 ($P < .048$). When comparing the correlation between average serum prestin levels and noise exposure, Spearman's ρ was −0.38 ($P = .039$).

Discussion

We previously reported that noise exposure is inversely related to serum prestin levels.[3] Our main findings here are that blood levels of key inner ear proteins, prestin and STRC, appear to be correlated and that these levels are influenced by noise exposure levels in young, healthy individuals. The latter implies that the expression of these key proteins in the inner ear may be modulated by auditory experience across the board. Our results add further support to the proposed role of inner ear proteins found in the blood as biomarkers for inner ear disorders.

The significance of developing an inner ear biomarker approach extends beyond NIHL. For example, gene therapy has proven successful in improving hearing thresholds and protein production in mouse models of DFNB1-related hearing impairment.[9] A recent clinical trial demonstrated the safety and efficacy of gene therapy with an adeno-associated virus serotype 1 carrying a human OTOF transgene as a treatment for children with autosomal recessive deafness 9 improving hearing thresholds by 40 to 50 dB.[10] Serial measurement of inner ear gene products and proteins in the blood may serve to track the response to gene therapy in both research and clinical settings in advance of audiometric biomarkers, that is, noninvasively assess the efficacy of gene therapy in a target-specific manner. While our approach could be adapted to DFNB1, it is worth noting that mutations in the STRC gene, DFNB16, are considered second to gap junction protein beta 2 gene mutations in contributing to autosomal recessive nonsyndromic hearing loss.[11]

Limitations of our study include the characteristics of the study population and sample size, which affect generalizability. Furthermore, many inner ear proteins are expressed elsewhere in the body. STRC is expressed to a lesser extent in the central nervous system and reproductive organs, limiting specificity.[12] The correlation with noise exposure, however, strongly implies cochlear origin.

We excluded 2 outliers with STRC values above the ELISA kits detection range. Prestin values for these 2 outliers were also near or above the upper limit, across all 5 time points. If both subjects were included Spearman's ρ would have been stronger at 0.66. Thus, their exclusion represents a more conservative treatment of our results and conclusions. A key question is why some subjects have very high blood levels of these markers. While we have demonstrated that noise can influence otologic marker levels,[3] it is tempting to speculate that very high blood levels may reflect vulnerability to other cochlear phenomena. One example is tinnitus which has been suggested to be associated with the upregulation of cochlear prestin expression in salicylate toxicity.[13] To examine this hypothesis, we are prospectively investigating blood prestin levels in tinnitus patients.

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Author Contributions

Carly Malesky, data analysis, wrote the first draft of the manuscript; **Diana Daniel**, data collection and analysis; **Erika Skoe**, study design, data collection, and analysis, edited the draft; **Kourosh Parham**, conceived the study, study design, data collection, and analysis, revised the manuscript for publication.

Disclosures

Competing interests: The authors declare that there is no conflict of interest.

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References

1. National Institute on Deafness and Other Communication Disorders. Noise-induced hearing loss. NIDCD. December 14, 2018. Accessed March 17, 2024. http://www.nidcd.nih.gov/health/noise-induced-hearing-loss
2. Jain R, Pingle S, Tumane R, et al. Cochlear proteins associated with noise-induced hearing loss: an update. *Indian J Occup Environ Med*. 2018;22(2):60-73. doi:10.4103/ijoem.IJOEM_43_18
3. Parker A, Parham K, Skoe E. Noise exposure levels predict blood levels of the inner ear protein prestin. *Sci Rep*. 2022;12(1):1154. doi:10.1038/s41598-022-05131-z
4. Verpy E, Leibovici M, Michalski N, et al. Stereocilin connects outer hair cell stereocilia to one another and to the tectorial membrane. *J Comp Neurol*. 2011;519(2):194-210. doi:10.1002/cne.22509
5. Avan P, Le Gal S, Michel V, et al. Otogelin, otogelin-like, and stereocilin form links connecting outer hair cell stereocilia to each other and the tectorial membrane. *Proc Natl Acad Sci*. 2019;116(51):25948-25957. doi:10.1073/pnas.1902781116
6. Wang H, Yin S, Yu Z, Huang Y, Wang J. Dynamic changes in hair cell stereocilia and cochlear transduction after noise exposure. *Biochem Biophys Res Commun*. 2011;409(4):616-621. doi:10.1016/j.bbrc.2011.05.049
7. Parker A, Parham K, Skoe E. Reliability of serological prestin levels in humans and its relation to otoacoustic emissions, a functional measure of outer hair cells. *Ear Hear*. 2021;42:1151-1162.
8. NIOSH. *Criteria for a Recommended Standard: Occupational Noise Exposure (Revised Criteria 1998)*. No. 98e126. NIOSH; 1998.

9. Lahlou G, Calvet C, Giorgi M, Lecomte MJ, Safieddine S. Towards the clinical application of gene therapy for genetic inner ear diseases. *J Clin Med*. 2023;12(3):1046. doi:10.3390/jcm12031046
10. Lv J, Wang H, Cheng X, et al. AAV1-hOTOF gene therapy for autosomal recessive deafness 9: a single-arm trial. *Lancet*. 2024;403(10441):2317-2325. doi:10.1016/S0140-6736(23)02874-X
11. Han S, Zhang D, Guo Y, Fu Z, Guan G. Prevalence and characteristics of STRC gene mutations (DFNB16): a systematic review and meta-analysis. *Front Genet*. 2021;12:707845. doi:10.3389/fgene.2021.707845
12. The Human Protein Atlas. Tissue expression of STRC—summary. 2023. Accessed June 4, 2024. https://www.proteinatlas.org/ENSG00000242866-STRC/tissue
13. Yu N, Zhu ML, Johnson B, Liu YP, Jones RO, Zhao HB. Prestin up-regulation in chronic salicylate (aspirin) administration: an implication of functional dependence of prestin expression. *Cell Mol Life Sci*. 2008;65(15):2407-2418.