Hearing Research 392 (2020) 107960

Contents lists available at ScienceDirect

Hearing Research

journal homepage: www.elsevier.com/locate/heares

Research Paper

The ongoing search for cochlear synaptopathy in humans: Masked thresholds for brief tones in Threshold Equalizing Noise

Frederic Marmel [a, b], Daniela Cortese [a, c], Karolina Kluk [a, *]

[a] Manchester Centre for Audiology and Deafness (ManCAD), School of Health Sciences, Faculty of Biology, Medicine and Health, University of Manchester, UK
[b] Institut Jean Le Rond ∂'Alembert (UMR 7190), Sorbonne Université, Paris, France
[c] Departamento de Fonoaudiología, Facultad de Medicina, Universidad de Chile, Chile

ARTICLE INFO

Article history:
Received 20 November 2019
Received in revised form
31 March 2020
Accepted 1 April 2020
Available online 17 April 2020

Keywords:
Cochlear synaptopathy
Hidden hearing loss
Tinnitus
Noise exposure
Audiometry
Temporal integration

ABSTRACT

This study aimed to advance towards a clinical diagnostic method for detection of cochlear synaptopathy with the hypothesis that synaptopathy should be manifested in elevated masked thresholds for brief tones. This hypothesis was tested in tinnitus sufferers, as they are thought to have some degree of synaptopathy. Near-normal-hearing tinnitus sufferers and their matched controls were asked to detect pure tones with durations of 5, 10, 100, and 200 ms presented in low- and high-level Threshold Equalizing Noise. In addition, lifetime noise exposure was estimated for all participants. Contrary to the hypothesis, there was no significant difference in masked thresholds for brief tones between tinnitus sufferers and their matched controls. Masked thresholds were also not related to lifetime noise exposure. There are two possible explanations of the results: 1) the participants in our study did not have cochlear synaptopathy, or 2) synaptopathy does not lead to elevated masked thresholds for brief tones. This study adds a new approach to the growing list of behavioral methods that attempted to detect potential signs of cochlear synaptopathy in humans.

1. Introduction

Cochlear synaptopathy, sometimes named "hidden hearing loss" (Schaette and McAlpine, 2011), is a subclinical hearing pathology that could potentially explain some listening difficulties observed despite (near) normal audiometric thresholds. These include difficulties understanding speech in challenging situations (e.g, noisy or reverberant environments), tinnitus, and hyperacusis (Plack et al., 2014; Kujawa and Liberman, 2015; Bramhall et al., 2019; Wu et al., 2019). Cochlear synaptopathy has been mostly studied in animals, as in humans it can only be observed post-mortem using histological techniques (Kujawa and Liberman, 2015; Bramhall et al., 2019). Several electrophysiological and behavioral measurements have been used in an effort to indirectly observe cochlear synaptopathy in humans (Plack et al., 2016; Bramhall et al., 2019). However, to this day there is no experimental method that is promising as a clinical diagnostic test for cochlear synaptopathy. The aim of this research was to investigate the potential of testing masked thresholds for brief tones to advance towards a clinical diagnostic method for the detection of cochlear synaptopathy. Following theoretical reasoning by Lopez-Poveda and Barrios (2013) and perceptual model simulations by Marmel et al. (2015), our main hypothesis was that masked thresholds for brief tones, but not for long tones, would be elevated in case of synaptopathy. As one of the predicted functional consequences of cochlear synaptopathy is tinnitus (Kujawa and Liberman, 2015; Bramhall et al., 2019), we measured and compared audiometric thresholds for brief and long tones between tinnitus sufferers and matched controls. As synaptopathy has been associated with noise exposure and ageing in animals, we also assessed how the results for the masked thresholds were related to age and to lifetime noise exposure, estimated using the Noise Exposure Structured Interview (NESI; Guest et al., 2018).

Our main hypothesis was based on work by Lopez-Poveda and collaborators (Lopez-Poveda and Barrios, 2013; Lopez-Poveda, 2014; Marmel et al., 2015). To study the effects of synaptopathy

Abbreviations: ABR, auditory brainstem responses; ANFs, auditory nerve fibres; DPOAEs, distortion product otoacoustic emissions; EFR, envelope following responses; HT, hearing thresholds; MEMR, middle-ear muscle reflex; NESI, noise exposure structured interview; OHC, outer hair cells; PTA, pure tone audiometry; TEN, threshold equalizing noise

* Corresponding author.
E-mail address: karolina.kluk@manchester.ac.uk (K. Kluk).

https://doi.org/10.1016/j.heares.2020.107960

on the neural representation of sound, Lopez-Poveda and Barrios (2013) used a signal processing analogy; according to them, the perceptual effects of synaptopathy could be compared to the effects of a stochastic undersampling of the auditory signal. As individual auditory nerve fibres (ANFs) fire stochastically, they provide an incomplete representation of the mechanical response waveform in the cochlea. In a healthy auditory nerve, a high-quality representation of the cochlear mechanical waveform is obtained from the combination of the spike trains from all ANFs. However, in a deafferented nerve due to synaptopathy, the reduced number of ANFs would be less able to compensate for the limited information encoded by individual fibres, thus imposing a limit to information encoding in the auditory nerve. Lopez-Poveda and Barrios (2013) argued that the neural representation of brief sound features would be especially degraded in the case of a deafferented nerve. They tested their stochastic undersampling analogy in perceptual experiments that used sound stimuli processed to be stochastically undersampled versions of themselves, in order to simulate a loss of stimulus information that could impact perception in a similar way as a loss of functional ANFs. Stochastically undersampled speech sounds and 100-ms pure tones led to impairments of speech recognition in noise but not in quiet, and left tone detection thresholds within the normal range, consistent with the expected perceptual consequences of cochlear synaptopathy (Kujawa and Liberman, 2015; Bramhall et al., 2019). In a later study (Marmel et al., 2015), stochastically undersampled broadband noises of various durations resulted in an elevation of absolute detection threshold for brief durations (5–10 ms) but not for longer durations. Finally, reducing the number of ANFs in a physiological model of the auditory periphery (the MAP model, Meddis et al., 2013) resulted in elevated simulated detection thresholds for brief broadband noises but not for longer ones (unpublished). Although the stochastic undersampling analogy is not a physiological analogy and does not aim to simulate the physiology of synaptopathy, the studies of Lopez-Poveda and collaborators suggest that measurements of audiometric thresholds for brief tones (<10 ms) might help detect cochlear synaptopathy in humans.

The detection of cochlear synaptopathy in humans might also be helped by measuring detection thresholds in noise. Elevated thresholds in Threshold Equalizing Noise (TEN; Moore et al., 2000) have been reported for tinnitus participants with normal audiometric thresholds (Weisz et al., 2006; Buzo and Carvallo, 2014). Similar results have been reported for patients with auditory neuropathy (Vinay and Moore, 2007). The elevation of thresholds in presence of TEN was explained as poor processing efficiency either caused by a reduction of neural synchrony or by synaptopathy. Therefore, the use of TEN might assist in the uncovering of synaptopathy. Another reason to measure thresholds in noise is that synaptopathy is thought to selectively affect auditory nerve fibers with high thresholds (Furman et al., 2013). The addition of TEN allows the measurement of detection thresholds at a high sound level, which should make the measurements more sensitive to synaptopathy.

The present study measured masked thresholds for brief and long tones, in low- and high-level TEN. We hypothesized that tinnitus sufferers would have elevated thresholds compared to the control group for the brief tones but not for the longer tones. The threshold elevation might be observed mostly or only in high-level TEN.

2. Methods

2.1. Participants

The participants were recruited via advertising in local newspapers, via email through the hearing research volunteer database of the Manchester Centre for Audiology and Deafness and via the daily announcements of the Faculty of Biology, Medicine and Health at the University of Manchester. Participants were required to have clinically near-normal hearing thresholds between 0.5 and 8 kHz, as well as normal otoscopic findings, no middle ear pathology, no history of ear surgery, no psychiatric disorders or claustrophobia. Normal or near-normal hearing was defined as hearing thresholds (HT) $\leq$ 25 dB HL up to 2 kHz, $\leq$ 30 dB HL at 3 kHz, $\leq$ 35 dB HL at 4 kHz and $\leq$40 dB HL at 6 kHz (Moore et al., 2012). Participants in the experimental group reported having experienced unilateral or bilateral non-pulsatile tinnitus (ringing, buzzing in their ears) lasting more than 5 min at least once a week for more than 6 months. Some participants in the control group had experience of tinnitus but it had never lasted more than a minute and it occurred less than twice a month.

Forty-three participants completed the study (20 in the control group and 22 in the experimental group). However, only eighteen tinnitus participants could be matched with controls, hence the final sample consisted of thirty-six participants (seven females per group). Groups were matched by sex, age (±5 years), and hearing thresholds (HT) (±10 dB up to 8 kHz). However, two pairs of participants could not be matched for age (10 and 11 years of difference) and eleven pairs of participants had at least one HT frequency that could not be matched (1.8 frequencies on average, with HT differences ranging from 15 to 25 dB HL). The mean age was 38 ± 3.0 years for the control group and 39 ± 3.5 years for the experimental group (mean ± standard error of the mean). The mean audiometric HTs (from 0.5 to 8 kHz) for the control and the experimental group were 9.2 ± 2.0 and 8.8 ± 1.7 dB HL, respectively. The study was conducted at the University of Manchester and was approved by the University Research Ethics Committee (Reference: 16289). All the participants provided written consent and were either reimbursed for their time and/or received their hearing test report.

2.2. Equipment and procedures

A generic health-check protocol was used to determine the health history of the participants. Lifetime noise exposure was estimated using the Noise Exposure Structured Interview (NESI) (Guest et al., 2018). Participants were asked to identify noisy activities (>80 dBA) in which they had been involved in their life, including both recreational and occupational noise. The total hours of noise exposure was calculated by asking the participant to recall a period of their life in which the activity was fairly stable and to estimate the number of days per week, and the number of hours per day, spent in the activity. The use of hearing protection for each activity was also considered. For a complete description of the procedures, see Guest et al. (2018).

HTs were obtained using a Kamplex Clinical Audiometer KC50 (U.K Edition – BS EN 60645) and TDH 39P supra-aural headphones, following the British Society of Audiology (2017) recommended procedure for the following frequencies: 0.5, 1, 2, 3, 4, 6, and 8 kHz.

Distortion product otoacoustic emissions (DPOAEs) were recorded from 2 to 10 kHz (8 frequencies/octave) using Titan (software v3.4.0, Interacoustics, DK). An f2/f1 ratio equal to 1.22 was used with stimulus levels L1/L2 of 65/55 dB SPL. The response was considered to be present when the DPOAE response reached 98% reliability with a distortion product (DP) level above −10 dB SPL, as set by the manufacturer.

Measurement of masked thresholds for tones in Threshold Equalizing Noise (TEN) was performed using custom MATLAB scripts (The Mathworks, USA), a Creative E-MU 0202 sound card and Sennheiser HD650 headphones. Sixteen test conditions were

included, combining four tone durations (5, 10, 100 and 200 ms), two frequencies (2 and 6 kHz) and two TEN levels (20 and 50 dB SPL/ERB_N, Glasberg and Moore, 1990). The tones were sine waves and included 2.5-ms raised-cosine rise/fall times (included in the tone-duration values specified above). The noise was TEN (Moore et al., 2000) calibrated in SPL, with a spectrum extending from 50 to 16 000 Hz. The TEN was turned on 50 ms before the start of the tone and was turned off 50 ms after the end of the tone. The TEN also had 2.5-ms raised-cosine rise/fall times. Dichotic stimulation was used, with the tone in the tested ear and TEN in both ears to prevent cross-hearing of the tone. Independent samples of TEN were presented to each ear. A two-interval two-alternative forced-choice task was used. The inter-stimulus interval was 500 ms. A two-down one-up adaptive procedure was used to track the 70.7% point of the psychometric function (Levitt, 1971). The level of the tone was varied in 6-dB steps for the first four reversals (i.e., changes of the adaptive track's direction) and then in 2-dB steps for the following twelve reversals. The final threshold estimate was the mean of the levels at the last twelve reversals. One run per condition was collected except if the standard deviation of the levels at the last twelve reversals exceeded 6 dB, in which case the run was repeated once and the final threshold estimate from the second run was used.

The ear with the better mean audiometric HTs (from 0.5 to 8 kHz) was selected as the tested ear. Twenty-four right ears and twelve left ears were assessed. All participants had a practice trial with two example conditions: 4 kHz, 100 ms, 20 dB SPL/ERB_N and 4 kHz, 5 ms, 50 dB SPL/ERB_N. The test conditions were pseudo-randomized for each participant to avoid starting with a very challenging test condition (i.e., a duration $\leq$ 100 ms). All the procedures were conducted in one session. Participants were encouraged to take regular breaks during the session.

2.3. Statistical methods

The 16 sets of masked threshold data corresponding to every combination of "Frequency", "Tone Duration" and "TEN Level" for both groups of participants combined were checked for normality by combining tests of skewness and kurtosis to produce an omnibus test of normality (D'Agostino, 1971; D'Agostino and Pearson, 1973). Deviations from normality were observed, so logarithmic transformations were applied to the data sets. A four-way mixed ANOVA was computed on the transformed thresholds, with "Frequency", "Tone Duration" and "Noise Level" as within-participants factors and "Group" (Tinnitus/Control) as a between-participants factor. Greenhouse-Geisser corrected *p*-values are reported whenever sphericity was violated.

The distributions of age, Pure Tone Audiometry (PTA) average from 0.5 to 8 kHz, and NESI scores for both groups of participants combined (3 sets of data) were also checked for normality. Statistical analyses involving these data sets used logarithmic transformations, and non-parametric statistical tests when required. Statistical analyses were performed using SPSS v.23 or Python v.2.7.

3. Results

3.1. Group matching

Statistical analyses were performed to assess whether age and HT mismatches could result in significant differences between groups. No significant differences were found between the mean age and the mean audiometric HT of tinnitus and control participants, using independent sample two-tailed t-tests ($t_{df=34} = -0.05$, $p = 0.95$ and $t_{df=34} = -0.06$, $p = 0.94$, respectively).

3.2. Noise exposure

Units of estimated lifetime noise exposure (NESI) obtained from all thirty-six participants varied from 0 to 202. The mean score and standard error of the mean (SEM) were 29 ± 8.0 for the control group, and 42 ± 12 for the tinnitus group. The NESI scores were not normally distributed, even after the logarithmic transformation. Therefore, a non-parametric test was performed. An independent-sample Mann-Witney *U* test (two-tailed significant value) showed no significant difference between the NESI scores for the control and the experimental groups ($U = 168$, $p = 0.86$).

3.3. DPOAEs

DPOAEs were categorized as present or absent for each participant. All participants had DPOAEs for at least two of the tested frequencies. For frequencies from 2 to 6.8 kHz, both groups had at least 13 participants with present DPOAEs. For higher frequencies, fewer participants had DPOAEs (for 7.5, 8.3, 9.1 and 10 kHz, respectively, 11, 8, 9 and 10 participants in the control group and 7, 8, 6 and 7 participants in the tinnitus group). Chi-squared tests revealed no significant association ($p > 0.05$) between group and the presence or absence of DPOAEs for each tested frequency, i.e., the two groups had equally present DPOAEs at all frequencies.

3.4. Masked thresholds for short tones in TEN

Mean masked thresholds for each group and condition are plotted in Fig. 1. Typical temporal integration curves were measured for both control and tinnitus groups for the two TEN levels. Similar masked thresholds were obtained for the two groups across all tested conditions. As described earlier, a four-way mixed ANOVA was performed. The masked thresholds were significantly affected by frequency [$F(1, 34) = 4.55$, $p < 0.05$], tone duration [$F(1.79, 61.0) = 836$, $p < 0.001$] and TEN level [$F(1, 34) = 876$, $p < 0.001$]. Masked thresholds increased as frequency increased, although this effect seemed to have been driven solely by the two longest durations at the low-TEN level. Masked thresholds increased as tone duration decreased, and were higher at the high-TEN level than at the low-TEN level. There were significant interactions between frequency and tone duration [$F(1.78, 60.4) = 18.4$, $p < 0.001$], between frequency and TEN level [$F(1, 34) = 9.38$, $p < 0.01$], between duration and TEN level [$F(2.05, 69.8) = 39.0$, $p < 0.001$], and between frequency, duration and TEN level [$F(2.33, 79.4) = 7.88$, $p < 0.001$]. There was no significant effect of group on the masked thresholds [$F(1, 34) = 0.24$, $p = 0.63$]. There was no significant interaction between group and any other factor.

To assess whether noise exposure history affected masked detection thresholds, the sample was divided into two groups according to their NESI score. Participants falling into the bottom 25% of the scores were classified as the low-exposed group (9 participants, 4 females) and participants from the top 25% were classified as the high-exposed group (9 participants, 2 females). The means ± SEM of the NESI scores for each group were 0.61 ± 0.21 and 95.6 ± 14.0, respectively. The mean age and PTA of the groups were not significantly different. Fig. 2 shows the mean masked thresholds for each group for all tested conditions. Typical temporal integration curves and similar masked thresholds were obtained for the two groups for all conditions. A four-way mixed ANOVA like the one reported above but with the participants grouped into low/high exposure (instead of control and tinnitus) showed no main effect of noise exposure [$F(1, 16) = 0.48$, $p = 0.50$], nor any interaction effect involving noise exposure.

Finally, the effect of age was assessed by creating a younger and an older group. The younger group included the participants falling

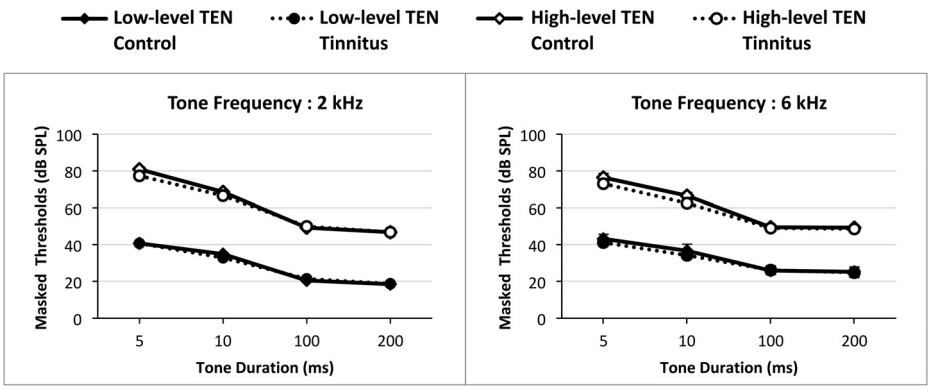

Fig. 1. Mean masked thresholds as a function of tone duration. The different symbols represent the control and the experimental group for each TEN level, as indicated on the legend of each plot. Error bars show ±1 SEM of the group mean for each condition.

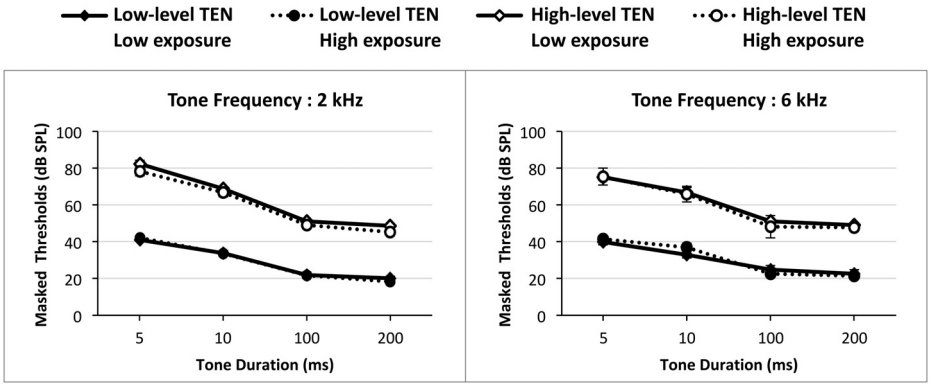

Fig. 2. Mean masked thresholds as a function of tone duration for low- and high-noise exposure groups. Otherwise as Fig. 1.

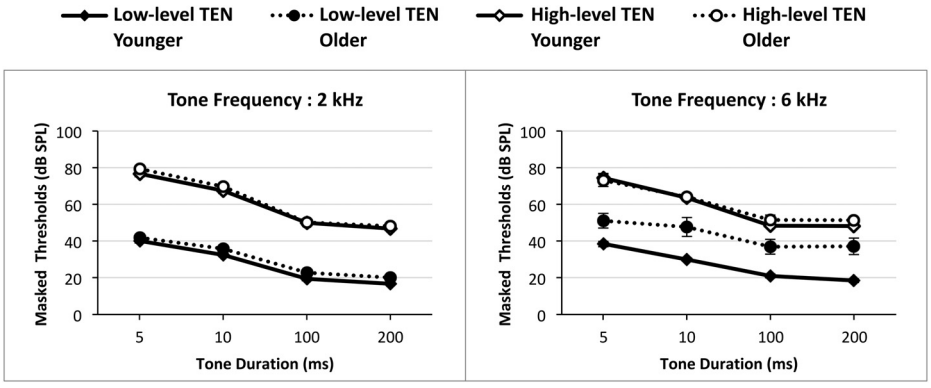

Fig. 3. Mean masked thresholds as a function of tone duration for the younger and the older groups. Otherwise as Fig. 1.

into the first quartile (25th percentile: 27 years and below) and the older group included the participants falling above the upper quartile (75th percentile: 50 years and above). With this division, the sample consisted of twenty participants, ten per group (4 females in the younger group and 6 in the older group). The PTA was significantly different between groups (independent sample two-tailed *t*-test: $t_{df=18} = -4.62$, $p < 0.001$). Fig. 3 illustrates the mean masked thresholds for each group for all tested conditions. Mean masked thresholds at 2 kHz for all durations and at both TEN levels were similar for the two groups. At 6 kHz, the two groups had similar masked thresholds at the high-TEN level but the younger group had lower masked thresholds than the older group at the low-TEN level. The older group's higher PTA may have limited their masked thresholds at 6 kHz, leading to the difference with the younger group at the low-TEN level. A four-way mixed ANOVA as described above but with PTA added as a covariate (to control for the difference in PTA between groups) showed no main effect of age [$F(1, 17) = 2.27$, $p = 0.15$] nor any interaction effect involving age.

4. Discussion

4.1. Looking for signs of cochlear synaptopathy in tinnitus sufferers

We hypothesized that tinnitus sufferers would have elevated masked thresholds for brief tones, especially in high-level TEN. However, as shown in Fig. 1 and supported by the outcomes of the statistical analyses, this was not the case. Contrary to the hypothesis, thresholds were not associated with tinnitus, noise exposure, or age. If the stochastic undersampling analogy proposed by Lopez-Poveda and Barrios (2013) represented the perceptual consequences of synaptopathy, and if tinnitus sufferers had some degree of synaptopathy, then elevated thresholds for the brief tones (10 and 5 ms) should have been observed for the experimental group (Marmel et al., 2015, p. 5). However, as pointed out by Marmel et al. (2015, pp. 9–10), the elevation of thresholds experimentally observed for their stochastically undersampled stimuli were obtained for degrees of undersampling that would simulate degrees of synaptopathy larger than the ones observed experimentally in rodents (Kujawa and Liberman, 2009; Furman et al., 2013; Sergeyenko et al., 2013) and than the ones observed post-mortem by analyzing human temporal bones (Makary et al., 2011). It is possible that brief-tone audiometry/masked thresholds would only be able to detect degrees of synaptopathy larger than those that were present in the tinnitus sufferers of the present study. Other human studies have tried several different approaches to indirectly detect synaptopathy in tinnitus sufferers, with mixed results (see Bramhall et al., 2019 for a review). These approaches were based on physiological changes observed in animals with synaptopathy, such as reduced amplitudes of the wave I of auditory brainstem responses (ABR), reduced amplitudes and phase-locking values of envelope following responses (EFR), and elevated middle-ear muscle reflex (MEMR) thresholds and maximal strength. For each approach, some studies reported relationships between tinnitus and the physiological change, and some others did not. Bramhall et al. (2019) pointed out that the lack of consistency across studies might be related to the variability of the underlying etiologies of tinnitus in the tinnitus populations studied, with noise exposure being only one of the possible etiologies. It is worth noting that, in the present study, there was no significant difference in the NESI scores between the tinnitus and control groups. It is thus possible that tinnitus in the participants of the present study was unrelated to noise exposure. This would suggest that either tinnitus in the present study's participants was related to synaptopathy caused by other factors than noise exposure, or that the tinnitus and control groups did not differ in their degree of synaptopathy (if any).

4.2. Looking for signs of cochlear synaptopathy associated with noise exposure and age

Experimental animal studies have described cochlear synaptopathy as a result of acoustic overexposure (in rodents: Kujawa and Liberman, 2009, Lin et al., 2011; Furman et al., 2013; Shaheen et al., 2015; Valero et al., 2016, 2018; Lobarinas et al., 2017; in rhesus monkeys: Valero et al., 2017) or as occurring with aging (Sergeyenko et al., 2013; Kujawa and Liberman, 2015). In the last few years, those experimental animal findings have motivated human studies to look for associations between lifetime noise exposure, age, and signs of noise-induced synaptopathy. These human studies have used the same approaches as those summarized in the previous section for studies on tinnitus sufferers. Results have been mixed and overall this line of research has not provided conclusive evidence of noise-induced synaptopathy in humans (see Bramhall et al., 2019 for a review and discussion). The present study assessed the potential effect of lifetime noise exposure and aging on masked thresholds of brief tones via post-hoc analyses, and did not find evidence that higher noise exposure or greater age affected brief-tone detection thresholds in noise. Hence, the present study contributes to the body of research that did not find any relationship between possible signs of synaptopathy (even indirect ones) and acoustic exposure or age. The present study's contribution is limited by it not being primarily designed to investigate noise exposure and age, which raises the question of whether participants were old enough, or had a lifetime noise exposure high enough, for synaptopathy to have perceptual consequences. Regarding whether participants were old enough, Valderrama et al. (2018, pp.43–44) discussed how their inclusion of participants who were older than in some previous studies (such as: Fulbright et al., 2017; Grinn et al., 2017; Grose et al., 2017; Prendergast et al., 2017) may have made the effects of noise exposure on the human ABR morphology more evident and allowed them to observe a negative correlation between lifetime noise exposure and the amplitude of the ABR wave I. Participants in Valderrama et al. (2018) were 20–55 years old with a mean of 43 years. The older group in the present study's post-hoc analysis was even older (50–69 years, mean of 57 years), which makes it unlikely that the absence of difference between older and younger listeners in the present study was a consequence of the older participants not being old enough. Regarding whether sufficient participants had a high enough lifetime noise exposure, the comparison between noise exposure values in the present study and that of Valderrama et al. (2018) is difficult because the noise exposure assessment methods were different. However, it is possible to compare the spread of lifetime noise exposure values across participants: the participant with the highest noise exposure in the present study had more than 8000 times the exposure of the participant with the lowest noise exposure, whereas this ratio was 3000 in Valderrama et al. (2018). Thus it seems unlikely that insufficient noise exposure could explain the lack of significant results in the present study. Bramhall et al. (2019) pointed that it might only be possible to reveal noise-induced cochlear synaptopathy non-invasively for audiometrically normal listeners in an auditory system alteration "sweet spot"; i.e. when the right combination of noise exposure and aging have produced sufficient synaptopathy whilst overall cochlear damage is low enough for outer hair cells (OHCs) to be intact and hearing thresholds to be normal. The present study may not have controlled strictly enough for OHC damage and for normal hearing thresholds. Hearing thresholds were not measured above 8 kHz and OHC function was assessed by scoring DPOAEs as pass/fail. The present study tested participants with broader ranges of age and lifetime noise exposure than some previous studies, and it is possible that Brahmall et al.'s "sweet spot" was "missed", i.e. some participants might have had some synaptopathy but also some cochlear damage. Bramhall et al. (2019) stressed the importance of using more stringent criteria than a pass/fail when using DPOAEs to measure the OHC function. It is likely that some participants in the present study had OHC damage. Unfortunately, insufficient control of OHC damage might be particularly crucial in the present study because the hypothesized pattern of an elevation of detection thresholds specific to brief tones (augmented temporal integration) is the opposite of the pattern usually observed in patients with cochlear hearing loss. Patients with cochlear hearing loss usually show elevated detection thresholds for all sound durations, with the elevation being larger for longer durations than for short durations, resulting in shallower threshold/duration functions (reduced temporal integration) than normal-hearing listeners (Florentine et al., 1988; Gerken et al., 1990; Plack and Skeels, 2007). This reduced temporal integration can be explained by OHC damage (Moore, 2007). Opposite effects of

OHC damage and of cochlear synaptopathy on temporal integration could potentially explain the lack of difference between threshold/duration functions for older *vs.* younger participants, and for participants with high-*vs.* low-lifetime noise exposure. The comparison of thresholds for older *vs.* younger participants at 6 kHz (Fig. 3) actually looks more consistent with OHC damage, as the threshold/duration function for the older participants looks a bit shallower than the younger participants' function, although this is not statistically significant.

4.3. Looking for signs of cochlear synaptopathy with brief-tone audiometry

One final explanation for why the present study did not observe elevated thresholds for brief tones is that this study's rationale might have been incorrect. The threshold elevation for brief tones in Marmel et al. (2015) was observed with stochastically undersampled stimuli, using young normal-hearing participants not suspected to have any cochlear synaptopathy. The stochastic undersampling aimed to impair their performance in a way that would simulate the perceptual effect of a loss of auditory nerve fibers, qualitatively and without distinguishing between fibers with low and high thresholds. The stochastic undersampling simulations did not aim to model cochlear synaptopathy (Lopez-Poveda, 2014). To our knowledge, no study has sought to extend the results of Marmel et al. (2015) with a physiological model of cochlear synaptopathy. This might be worth pursuing; the first author has preliminary (unpublished) simulations using the MAP model (Meddis et al., 2013) that are consistent with the results of Marmel et al. (2015). There is also a need for more experimental studies using brief-tone audiometry in cohorts suspected to have cochlear synaptopathy. Wong et al. (2019) measured temporal integration from 20 to 160 ms in budgerigars that had auditory nerve damage induced by kainic acid and found no evidence that the damage altered temporal integration. However, Marmel et al. (2015) only observed elevated thresholds for durations of 5 and 10 ms (there was no difference in their study for 20 ms). Thus the data of Wong et al. (2019) do not undermine the present study's rationale. Further studies properly controlling for OHC damage are needed to conclusively accept or reject the present study's rationale.

5. Conclusions

The present study was not successful in its search for cochlear synaptopathy in humans: masked thresholds for brief tones in TEN were similar for tinnitus sufferers and controls, and no relation was observed between masked thresholds and lifetime noise exposure or age. These findings do not support the study's rationale that brief-tone masked audiometry could help detect cochlear synaptopathy in humans.

CRediT authorship contribution statement

Frederic Marmel: Conceptualization, Methodology, Software, Writing - review & editing, Formal analysis, Supervision. **Daniela Cortese:** Investigation, Formal analysis, Writing - original draft. **Karolina Kluk:** Conceptualization, Writing - review & editing, Supervision, Project administration, Funding acquisition.

Acknowledgments

The authors would like to thank Dr Garreth Prendergast for advice on the statistical analyses, and Prof Brian C. J. Moore and one anonymous reviewer for very helpful comments on a previous version of this manuscript. Marmel and Kluk were supported by EPSRC Research Grant EP/M026728/1 and Kluk was additionally supported by MRC Programme Grant MR/K018094/1. The research was funded by an MRC Programme Grant MR/K018094/1 and supported by the NIHR Manchester Biomedical Research Centre.

References

Bramhall, N., Beach, E.F., Epp, B., Le Prell, C.G., Lopez-Poveda, E.A., Plack, C.J., et al., 2019. The search for noise-induced cochlear synaptopathy in humans: mission impossible? Hear. Res. 377, 88–103.

British Society of Audiology, 2017. Pure-tone Air-Conduction and Bone-Conduction Threshold Audiometry with and without Masking [Online] Available from: http://www.thebsa.org.uk/wp-content/uploads/2017/02/Recommended-Procedure-Pure-Tone-Audiometry-Jan-2017-V2.pdf. (Accessed 9 October 2017).

Buzo, B., Carvallo, R., 2014. Psychoacoustic analyses of cochlear mechanisms in tinnitus patients with normal auditory thresholds. Int. J. Audiol. 53 (1), 40–47.

D'Agostino, R., 1971. An omnibus test of normality for moderate and large sample size. Biometrika 58, 341–348.

D'Agostino, R., Pearson, E.S., 1973. Testing for departures from normality. Empirical Results for the Distributions of b2 and $\sqrt{b}$ 1. Biometrika 60, 613–622.

Florentine, M., Fastl, H., Buus, S.R., 1988. Temporal integration in normal hearing, cochlear impairment, and impairment simulated by masking. J. Acoust. Soc. Am. 84 (1), 195–203.

Fulbright, A.N., Le Prell, C.G., Griffiths, S.K., Lobarinas, E., 2017. In: Effects of Recreational Noise on Threshold and Suprathreshold Measures of Auditory Function. In Seminars in Hearing, vol. 38. Thieme Medical Publishers, pp. 298–318. No. 04.

Furman, A., Kujawa, S., Liberman, M., 2013. Noise-induced cochlear neuropathy is selective for fibers with low spontaneous rates. J. Neurophysiol. 110 (3), 577–586.

Gerken, G.M., Bhat, V.K.H., Hutchison-Clutter, M., 1990. Auditory temporal integration and the power function model. J. Acoust. Soc. Am. 88, 767–778.

Glasberg, B.R., Moore, B.C.J., 1990. Derivation of auditory filter shapes from notched-noise data. Hear. Res. 47 (1–2), 103–138.

Grinn, S.K., Wiseman, K.B., Baker, J.A., Le Prell, C.G., 2017. Hidden hearing loss? No effect of common recreational noise exposure on cochlear nerve response amplitude in humans. Front. Neurosci. 11, 465.

Grose, J.H., Buss, E., Hall III, J.W., 2017. Loud music exposure and cochlear synaptopathy in young adults: isolated auditory brainstem response effects but no perceptual consequences. Trends Hear. 21, 1–8.

Guest, H., Dewey, R., Plack, C., Couth, S., Prendergast, G., Bakay, W., Hall, D., 2018. The noise exposure structured Interview (NESI): an instrument for the comprehensive estimation of lifetime noise exposure. Trends Hear. 22, 1–10.

Kujawa, S., Liberman, M., 2009. Adding insult to injury: cochlear nerve degeneration after "temporary" noise-induced hearing loss. J. Neurosci. 29 (45), 14077–14085.

Kujawa, S.G., Liberman, M.C., 2015. Synaptopathy in the noise-exposed and aging cochlea: primary neural degeneration in acquired sensorineural hearing loss. Hear. Res. 330, 191–199.

Levitt, H., 1971. Transformed up-down methods in psychoacoustics. J. Acoust. Soc. Am. 49 (2B), 467–477.

Lin, H.W., Furman, A.C., Kujawa, S.G., Liberman, M.C., 2011. Primary neural degeneration in the Guinea pig cochlea after reversible noise-induced threshold shift. J. Assoc. Res. Otolaryngol. 12 (5), 605–616.

Lobarinas, E., Spankovich, C., Le Prell, C.G., 2017. Evidence of "hidden hearing loss" following noise exposures that produce robust TTS and ABR wave-I amplitude reductions. Hear. Res. 349, 155–163.

Lopez-Poveda, E., 2014. Why do I hear but not understand? Stochastic undersampling as a model of degraded neural encoding of speech. Front. Neurosci. 8 (348), 1–13.

Lopez-Poveda, E., Barrios, P., 2013. Perception of stochastically undersampled sound waveforms: a model of auditory deafferentation. Front. Neurosci. 7 (124), 1–13.

Makary, C., Shin, J., Kujawa, S., Liberman, C., Merchant, S., 2011. Age-related primary cochlear neuronal degeneration in human temporal bones. J. Assoc. Res. Otolaryngol. 12 (6), 711–717.

Marmel, F., Rodríguez-Mendoza, M., Lopez-Poveda, E., 2015. Stochastic undersampling steepens auditory threshold/duration functions: implications for understanding auditory deafferentation and aging. Front. Aging Neurosci. 7 (63).

Meddis, R., et al., 2013. A computer model of the auditory periphery and its application to the study of hearing. In: Moore, B.C.J., Patterson, R., Winter, I., Carlyon, R., Gockel, H. (Eds.), Basic Aspects of Hearing. Advances in Experimental Medicine and Biology, vol. 787. Springer, New York, NY.

Moore, B.C.J., Creeke, S., Glasberg, B.R., Stone, M.A., Sek, A., 2012. A version of the TEN test for use with ER-3A insert earphones. Ear Hear. 33 (4), 554–557.

Moore, B.C.J., Huss, M., Vickers, D.A., Glasberg, B.R., Alcantara, J.I., 2000. A test for the diagnosis of dead regions in the cochlea. Br. J. Audiol. 34, 205–224.

Moore, B.C.J., 2007. Cochlear Hearing Loss: Physiological, Psychological and Technical Issues, second ed. John Wiley & Sons, Chichester, UK, p. 137. Chapter vol. 3.

Plack, C., Barker, D., Prendergast, G., 2014. Perceptual consequences of "hidden" hearing loss. Trends Hear. 18, 1–11.

Plack, C., Leger, A., Prendergast, G., Kluk, K., Guest, H., Munro, K., 2016. Toward a diagnostic test for hidden hearing loss. Trends Hear. 20, 1–9.

Plack, C.J., Skeels, V., 2007. Temporal integration and compression near absolute threshold in normal and impaired ears. J. Acoust. Soc. Am. 122, 2236–2244.
Prendergast, G., Guest, H., Munro, K.J., Kluk, K., Léger, A., Hall, D.A., Heinz, M.G., Plack, C.J., 2017. Effects of noise exposure on young adults with normal audiograms I: Electrophysiology. Hear. Res. 344, 68–81.
Schaette, R., McAlpine, D., 2011. Tinnitus with a normal audiogram: physiological evidence for hidden hearing loss and computational model. J. Neurosci. 31 (38), 13452–13457.
Sergeyenko, Y., Lall, K., Liberman, M., Kujawa, S., 2013. Age-related cochlear synaptopathy: an early-onset contributor to auditory functional decline. J. Neurosci. 33 (34), 13686–13694.
Shaheen, L.A., Valero, M.D., Liberman, M.C., 2015. Towards a diagnosis of cochlear neuropathy with envelope following responses. J. Assoc. Res. Otolaryngol. 16 (6), 727–745.
Valderrama, J.T., Beach, E.F., Yeend, I., Sharma, M., Van Dun, B., Dillon, H., 2018. Effects of lifetime noise exposure on the middle-age human auditory brainstem response, tinnitus and speech-in-noise intelligibility. Hear. Res. 365, 36–48.
Valero, M.D., Hancock, K.E., Liberman, M.C., 2016. The middle ear muscle reflex in the diagnosis of cochlear neuropathy. Hear. Res. 332, 29–38.
Valero, M.D., Burton, J.A., Hauser, S.N., Hackett, T.A., Ramachandran, R., Liberman, M.C., 2017. Noise-induced cochlear synaptopathy in rhesus monkeys (Macaca mulatta). Hear. Res. 353, 213–223.
Valero, M.D., Hancock, K.E., Maison, S.F., Liberman, M.C., 2018. Effects of cochlear synaptopathy on middle-ear muscle reflexes in unanesthetized mice. Hear. Res. 363, 109–118.
Vinay, Moore, B.C.J., 2007. TEN(HL)-test results and psychophysical tuning curves for subjects with auditory neuropathy. Int. J. Audiol. 46 (1), 39–46.
Weisz, N., Hartmann, T., Dohrmann, K., Schlee, W., Norena, A., 2006. High-frequency tinnitus without hearing loss does not mean absence of deafferentation. Hear. Res. 222 (1–2), 108–114.
Wong, S.J., Abrams, K.S., Amburgey, K.N., Wang, Y., Henry, K.S., 2019. Effects of selective auditory-nerve damage on the behavioral audiogram and temporal integration in the budgerigar. Hear. Res. 374, 24–34.
Wu, P.Z., Liberman, L.D., Bennett, K., De Gruttola, V., O'Malley, J.T., Liberman, M.C., 2019. Primary neural degeneration in the human cochlea: evidence for hidden hearing loss in the aging ear. Neuroscience 407, 8–20.