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RESEARCH****Research Report****Follow-up of latency and threshold shifts of auditory brainstem responses after single and interrupted acoustic trauma in guinea pig****Boris Gourévitch, Thibaut Doisy, Marie Avillac, Jean-Marc Edeline\****Laboratoire de Neurobiologie de l'Apprentissage, de la Mémoire et de la Communication, UMR CNRS 8620, Université Paris-Sud, 91405 Orsay cedex, France*

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## ABSTRACT

Thresholds of auditory brainstem responses (ABRs) are widely used to estimate the level of noise-induced hearing loss or the level of acquired resistance to acoustic trauma after repeated exposures, i.e., the “toughening” effect. Less is known about ABR latencies and their relation to threshold changes. Guinea pigs were exposed to a traumatic pure tone at 5 kHz, 120 dB SPL, as either single (2 h, 4 h) or repeated (1 h every 48 h, four times) sessions. Thresholds and latencies of ABRs were monitored up to 45 days following the acoustic trauma. We show that latencies are prolonged in the case of large temporary threshold shifts observed in the days following trauma. The latency shift decreases after several repeated exposures, then stabilizes, similar to thresholds, suggesting that the “toughening” effect also applies to latencies. Permanent latency shift is usually very small compared to the permanent threshold shift. This effect could produce a recovery in the ability to process auditory information through the precise timing of neuronal events. Our study indicates that when estimated at suprathreshold stimulation level (70 dB SPL), latency provides complementary information to the sole threshold.

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**1. Introduction**

In young adults, acoustic trauma is the most common cause of permanent hearing loss (World Health Organization, 1980; U.S. Environmental Protection Agency, 1981; Franks, 1988; International Organization for Standardization, 1990) and an impressive amount of studies has used animal models to determine the physiological, morphological, and biochemical/molecular consequences of acoustic trauma (for review see Dancer et al., 1992). However, many studies examined the consequences of acoustic trauma after a single exposure to

traumatizing sounds, whereas repeated exposure to traumatic noise (>85 dB SPL) is the most common situation in humans, especially for teenagers (concerts, night clubs), musicians, and factory workers (Schmuziger et al., 2007; Ologe et al., 2008).

On the other hand, studies using repeated exposures in animal models of hearing loss have rapidly discovered that the equal energy principle (Eldred et al., 1957), which assumes that hearing loss is a function of the total energy received, does not apply in these situations. That is, hearing loss from a given exposure is determined, in part, by the subject's previous history to noise exposure (e.g., Clark et al., 1987;

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Abbreviations: ABR, auditory brainstem response; PTS, permanent threshold shift; SPL, sound pressure level; TTS, temporary threshold shift

Canlon et al., 1988; Subramaniam et al., 1991a,b; see for review Attanasio et al., 1998). Results from two different protocols support this view. First, “conditioning protocols,” i.e., pre-exposing subjects to the acoustic stimuli at a lower non-traumatic level, were shown to protect from the damaging effects of high-level stimulation (Rajan and Johnstone, 1983; Canlon et al., 1988; Campo et al., 1991; Ryan et al., 1994; Zuo et al., 2008). Second, results from “interrupted repetitive protocols” revealed that the threshold shift induced by a traumatic sound during the initial days of exposure gradually recovers during the following days of exposure, a phenomenon described as the “toughening” effect (Miller et al., 1963; Clark et al., 1987; Subramaniam et al., 1991a).

Previous studies mostly investigated trauma and recovery effects using the threshold shift of auditory brainstem responses (ABRs). ABRs represent a complex summation of neuroelectric activity in the brainstem auditory pathways. They can be used as a functional assay of cochlear and retrocochlear pathologies. Significant correlations were found between ABR thresholds and (i) behavioral thresholds in chinchilla (Henderson et al., 1983), (ii) auditory nerve fiber thresholds at the same frequency in cats (Ngan and May, 2001), and (iii) the amplitudes of the compound action potentials of the eighth nerve in the guinea pig (Fredelius et al., 1988). Also, permanent threshold shift (PTS) measured with ABRs long after acoustic trauma is associated with damage or loss of inner hair cells and, less often, outer hair cells (Canlon et al., 1987; Harding et al., 2002; Chen et al., 2003). In addition, it has been shown that during toughening, results obtained from ABR thresholds parallel those obtained from behavioral thresholds (Clark et al., 1987; Sinex et al., 1987).

The present study had two main purposes. First, most studies using conditioning or interrupted noise protocols have used massive exposure conditions, such as 6 h/day for 10 days (e.g., see in Subramaniam et al., 1991a,b; McFadden et al., 1997; Ahroon and Hamernik, 1999, 2000; Hamernik and Ahroon, 1999). Yet, several daily situations (such as use of hammer drills, attendance to night clubs, or festivals of music) expose the auditory system to only a few acoustic traumas that still may have temporary, or even permanent, effects. Thus, we wanted to evaluate whether a “toughening” effect can be observed with only a few exposures to a traumatic sound and to what extent this toughening has long-term effects when tested more than a month later.

Second, the consequences of toughening on ABRs latencies are still unclear. Permanent sensorineural hearing loss was found to induce delays in ABRs of humans and guinea pigs, even in utero (see for example in Picton et al., 1976; Sohmer et al., 1981; Cook et al., 1982; Attias and Pratt, 1984). However, the existence of a correlation between the amount of hearing loss and the latency shift remains controversial (Donaldson and Ruth, 1996; Francois, 1999). To clarify this correlation, this paper evaluates whether latencies of ABRs provide an insight into toughening effects complementary to thresholds values.

To this aim, groups of guinea pigs were exposed to a traumatic pure tone (5 kHz, 120 dB SPL) either during a single session (2 h or 4 h in duration) or during four 1 h sessions separated by 48 h, inducing various hearing loss levels and toughening effects. For each group, the evolution of threshold

shift and latencies (computed at 70–85 dB SPL) was followed from 24 h to 45 days post-exposure.

## 2. Results

### 2.1. Effects observed on individual examples

Fig. 1 shows typical ABRs to clicks and to tone pips at 16 kHz from two animals before and after acoustic trauma. Typically, ABRs to clicks were of higher amplitude and displayed more distinct waves than ABRs to pure tones (control conditions, compare Fig. 1A to Figs. 1B–D and Fig. 1H to Fig. 1I). In control conditions, stability of ABRs obtained at 16 kHz over 15 days is shown in Figs. 1B–D. For the animal M003 (Figs. 1A–G), after a 4 h trauma, ABRs disappeared for frequencies above 5 kHz (16 kHz here) even at high sound intensities (up to 85 dB here, Fig. 1E). Responses started to recover a few days after the trauma but the threshold (65 dB, Fig. 1G) remained above the pre-trauma values (10 dB, Figs. 1B–D). Note that first deflection and N3 wave taken at 70 dB SPL (or higher) were still delayed relative to control values (compare Fig. 1G to Figs. 1B–D).

Figs. 1H–N displays control (Figs. 1H, I) and post-trauma (Figs. 1J–N) ABRs of another animal (M009) exposed to four 1 h trauma. The acoustic threshold was largely increased from 10 dB to 80 dB after the first two 1 h exposures (compare Fig. 1K to Fig. 1I), but the threshold tended to recover after the fourth exposure (60 dB, Fig. 1L). On the following days (Figs. 1M and N), the ABR displayed waves that had disappeared immediately after the first exposure. However, 10 days after trauma threshold was still 50 dB SPL (Fig. 1N), i.e., additional recovery of thresholds after the fourth exposure was limited to 10 dB. In the following paragraphs, data obtained on the whole database are presented.

### 2.2. Reliability of auditory brainstem responses

For each animal, two or three control experiments over 15 days were conducted before trauma. On average, the ABR thresholds observed over the tested frequencies were in the range of, or 5–10 dB better than, previously published results on guinea pigs (Sliwinska-Kowalska et al., 1992; Yamasoba et al., 1999) and there was no difference between genders (Fig. 2A; Mann–Whitney bilateral test,  $p > 0.05$ ). On average, for a given animal, variability (mean absolute deviation) of thresholds across the two or three control experiments reached about 5 dB around the mean (Fig. 2C) and was slightly less for stimuli inducing the best thresholds (click, 8–16 kHz). The mean latency of the first deflection, and that of the N3 wave measured at 70 dB SPL, did not differ between 2 and 16 kHz and did not differ between genders (Fig. 2B). Variability of latencies across experiments reached about 0.05 ms around the mean (Fig. 2D). In the control condition, decreasing the sound intensity from 70 dB to threshold (down to 0 dB) delayed the N3 waves (Fig. 2E). Examination of Fig. 2E reveals that the better the threshold, the smaller the latency shift with intensity decrease. For instance, the latency shift observed at 2 kHz is greater than that at 8 kHz and the difference roughly corresponds to a 15 dB shift between the two curves in Fig. 2E, which also is the averaged threshold difference between 2 and 8 kHz in Fig. 2A.

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