

The Effect of Noise Exposure on Signal to Noise Ratio Changes of Distortion Product Otoacoustic Emissions (DPOAE) Examination in *Rattus Norvegicus*

Tengku Siti Hajar Haryuna^{1*} and Susi Mulyana¹

¹Department of Otorhinolaryngology-Head and Neck Surgery, Faculty of Medicine, Universitas Sumatera Utara, Jl. dr. Mansur Kampus USU Indonesia Medan 20155

Keyword: Noise-exposure, Outer hair cell, Otoacoustic emissions, Signal to noise ratio.

Abstract: Distortion Product Otoacoustic Emission (DPOAE) was used to assess outer hair cells. The aim of this study is to assess the damage to outer hair cells of the noise model *Rattus norvegicus* by assessing the Signal to noise ratio (SNR). Twenty-seven *Rattus norvegicus* were grouped into 3: controls; 2-hours 100 dB noise-exposure; and 2-hours 110 dB noise-exposure. DPOAE was assessed 3 times in the beginning before treatment, 2 hours after the second day of exposure (day 2), and after 3 days of no exposure (day 5). The results showed that the SNR of DPOAE after the noise exposure decreases at all frequencies compared to before the treatment. A significant decrease in SNR especially at 2000Hz - 4000Hz frequencies. These results suggest that DPOAE may be used as a sensitive procedure at various frequencies for early diagnosis and screening of NIHL.

1 INTRODUCTION

Noise is an important issue in occupational health; excessive exposure to noise has the social and psychological impacts caused by noise-induced hearing loss (NIHL) (Nassiri *et al.*, 2016). The bilateral sensorineural hearing loss may cause irreversible and progressive cochlear damage (Souza Alcaraz *et al.*, 2012; Jahani *et al.*, 2016) In NIHL, a notch at 3000, 4000, or 6000 Hz is a common finding in audiometric examination (Derekoy *et al.*, 2004; Lee Preel & Miller, 2016).

Physiological changes from noise exposure can be assessed on cochlear cells, where the outer hair cells are more susceptible to damage than inner hair cells. Degenerative outer hair cell is the most sensitive initial process against sound (Balatsouras, 2004). Otoacoustic emission measures the microscopic biochemical activity of healthy outer hair cell. OAE provides bicochlear mechanical stimulation that flow from the tympanum to the external ear through the external acoustic meatus (Nassiri *et al.*, 2016; Moussavi-Najarkola *et al.*, 2012).

The DPOAE measures the bicochlear intensified emissions recording of a different frequency (f1 and f2) (Moussavi-Najarkola *et al.*, 2012). The DPOAE examination uses two simultaneous pure sound stimuli, of different frequencies and intensities, called f1 and f2 (f1 < f2) (Janssen *et al.*, 2006; Prieve & Fitzgerald, 2015).

Studies on animal noise-model have used high-frequency DPOAE of 1-8 kHz and assess the Ldp (level distortion product). In this study, the group and intensity of noise exposure were different from prior studies which use a low-frequency DPOAE of 1-5 kHz, which is expected to be able to assess the damage to cochlear outer hair cell of noise model *Rattus norvegicus* by Signal to Noise Ratio (SNR) from DPOAE.

2 METHOD

Twenty-seven *Rattus norvegicus* aged 2-3 months with 200-300 gr of weight. All *Rattus norvegicus* were examined by the veterinarian and declared

healthy. The animals are placed in polycarbonate cage at 25°C with humidity ranging from 40%-60%. In its care, they get sufficient light source and free access to water and food until the end of the study. All procedures are carried out in accordance to Animal Research Ethicss Committees/AREC Faculty of Science University of North Sumatera (No.560/KEPH-FMIPA/2017).

The study group was divided into 3 groups ($n = 9$), Group I (control); Group 2 (2-hours 100 dB noise-exposure); and Group 3 (2-hours 110 dB noise-exposure). The noise was exposed to *Rattus norvegicus* in a noise-box of 64.5 x 45 x 40 cm made by the Faculty of Electro University of North Sumatra. The source of sound provided by Compact Disc (CD) containing sound recordings, CD player, and amplifier which generate the noise with a frequency of 1-10 kHz and an intensity of 100 dB and 110 dB as measured by a sound level meter.

The Distribution Product Otoacoustic Emission (DPOAE) examination (Elios Elito Otodia, Echodia Ltd., London, UK) were performed on all animal under anesthetic drugs with ketamine 50 mg/kg body weight and Xylazine 10 mg/kg body weight intraperitoneal in a soundproof room. The probe was modified from a plastic pipe adjusted to the size and placed in the ear canal. DPOAE was assessed 3 times: early before treatment; 2 hours after the second day of noise exposure and after 72 hours of the noise-free period. Acoustic emission results that can be evaluated at frequencies of 1.5, 2, 3, 4, 5 kHz. All study samples must have normal SNR (equal to or more than 6) before noise exposure.

To assess the SNR differences between groups and between frequency, the data were averaged between right and left SNR then analyzed with independent K Kruskal Wallis followed by Mann Whitney Test, significancy demonstrated as $P < 0.05$.

3 RESULTS AND DISCUSSIONS

Table 1 shows the mean and standard deviation of SNR on DPOAE at the frequency of 1500-5000 Hz in the control group. Table 2 and Table 3 show the mean and standard deviation of SNR DPOAE at 100 dB and 110 dB, respectively; there was a decrease of SNR value on the second and fifth day in both groups of 100 dB and 110 dB.

Table 1. The mean and standard deviation of SNR in control group.

Frequency (Hz)	Signal to noise ratio (dB)		
	Day 0	Day 2	Day 5
1500	18,3±9,71	16,7±7,95	9,2 ±8,53
2000	4,1 ± 1,28	8,1 ± 4,81	6,1 ±2,02
3000	9,7 ± 1,88	8,6 ± 3,2	9,7 ±4,11
4000	10,4±3,97	15,8 ±3,25	11,1±2,80
5000	12,1±5,72	11,7 ±7,91	10,7 ±5,12

Table 2. The mean and standard deviation of SNR in 100 dB group.

Frequency (Hz)	Signal to noise ratio (dB)		
	Day 0	Day 2	Day 5
1500	16,6±10,91	6,8 ±4,54	7,9 ±8,69
2000	10,5 ± 4,27	4,8 ±2,74	5,8 ±4,07
3000	11,00 ±4,32	3,0 ±4,34	6,4 ±2,87
4000	12,7 ± 4,92	2,0 ±2,68	5,4 ±2,17
5000	6,7 ± 9,29	5,3 ±1,81	6,2± 4,00

Table 3. The mean and standard deviation of SNR in 110 dB group.

Frequency (Hz)	Signal to noise ratio (dB)		
	Day 0	Day 2	Day 5
1500	18,2 ±9,42	6,8 ± 8,29	7,1 ±8,69
2000	9,1 ± 2,01	5,9 ± 6,23	6,3 ± 4,81
3000	8,9 ± 3,78	2,3 ± 2,42	4,3 ±5,66
4000	11,8±3,25	1,2 ± 3,39	5,2 ± 3,17
5000	7,1 ± 6,1	5,2 ± 6,47	6,0 ±4,73

Figure 1 and 2 show the SNR value after the noise exposure (day 2) in the 100 dB and 110 dB group was decreased at frequencies of 1500-4000 Hz compared to the SNR value before the exposure (day 0), this occurs particularly on 3000 Hz and 4000 Hz frequencies where the SNR value is 3.0 dB and 2.0 dB, respectively. At the 3000 Hz and 4000 Hz frequencies, the SNR value was 2, 3 dB and 1.2 dB in the 100 dB and 110 dB group, respectively. At the 5000 Hz frequency, there is a decrease of SNR but not significant either in 100 dB or 110 dB group.

Meanwhile, after 3 days of noise-free period (day 5), the SNR value was slightly improved compared to after exposure (day 2); the SNR value was decreased and did not return to baseline value before the noise exposure (day 0) on all frequencies in both group of 100 dB and 110 dB. This study showed that the noise-break was able to improve the SNR value after the noise exposure.

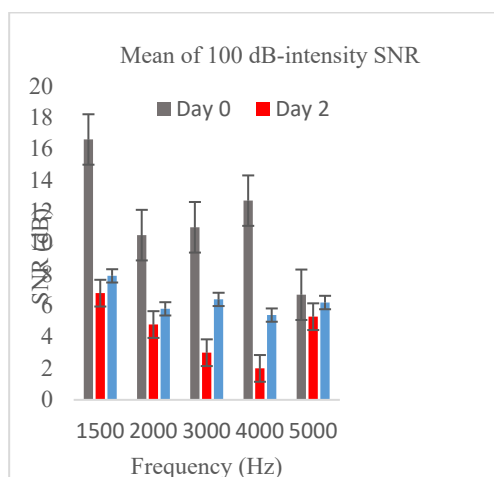


Figure 1. Average SNR changes in 100 dB group on day 0, day 2, day 5.

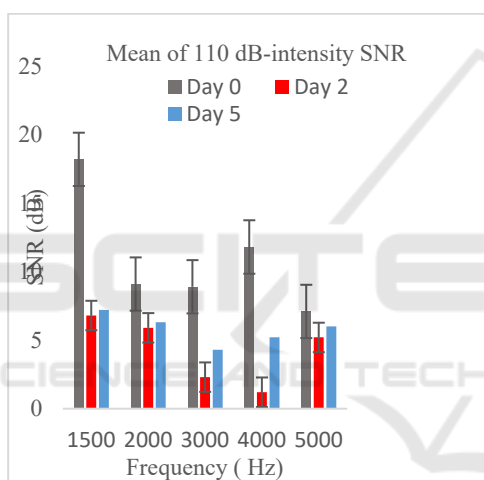


Figure 2. Average SNR changes in 110 dB group on day 0, day 2, day 5.

There is a difference of SNR value at the frequency of 1500 Hz after noise exposure (day 2) ($p < 0.005$) both in the 100 dB and 110 dB group. A similar finding was found at the frequency of 2000, 3000 and 4000 Hz. At the frequency of 5000 Hz, there were not any significant differences in SNR value ($p > 0.005$) both in the 100 dB and 110 dB group.

At all 1500-5000 Hz frequencies, no significant differences in SNR value were found in days 0, day 2 and day 5 compared between groups of 100 dB with 110 dB ($p > 0.005$). This shows the difference in intensity of 100 dB and 110 dB does not demonstrate the difference of SNR value.

In this study, DPOAE examination showed a decrease of the SNR after noise exposure at every frequency, especially at 2000-4000 Hz. NIHL occurs at high frequencies, especially at the frequency of

3000 Hz-6000 Hz; on audiometric examination, there was a notch at 4000 Hz frequency (Dobie, 2014). Several theories have been proposed to understand why the 4000 Hz frequency is the more susceptible region to noise exposure. The most popular theory is that the anatomical structure in the area is weaker (Dhingra, 2015). Damage to the cochlear structure due to the noise was observed, especially in the initial 8-10 mm diameter at the cochlear base which is a sensitive area caused by metabolic, vascular, and anatomical factors, with corresponding topography to the frequency of 4000 Hz (Yilmaz *et al.*, 2015).

Salehi *et al.* (2012) assessed the performance of outer hair cells in the noise-exposed rabbits, the result showed that the noise exposure reduces the DPOAE threshold at a frequency of 4-10 KHz (Salehi *et al.*, 2011). Nassiri *et al.* (2016) assessed the effect of noise exposure on mice by assessing SNR changes at the intensity of 65 dB, 95 dB and 105 dB concluded that higher noise intensity will decrease DPOAE level and sensitive DPOAE examination at various frequencies (Nassiri *et al.*, 2016).

Distortion product otoacoustic emission (DPOAE) is an initial and rapid test to detect the cochlear damage due to noise exposure. DPOAE sensitively measures the activity of cochlear outer hair cells recorded in the ear canal (Doosti *et al.*, 2014). Simultaneous stimulus response to the cochlea with 2 pure tone frequencies f_1 and f_2 , where frequency f_2 is slightly higher than f_1 ($2f_1-f_2$) (Moussavi-Najarkola *et al.*, 2012; Janssen *et al.*, 2006). The DPOAE examination measures the Signal to noise ratio (SNR) which defines the emission levels generated by noise levels as the background or ratio of signal strength to noise expressed in decibels (Nassiri *et al.*, 2016).

The noise impact depends on some sound characteristics i.e. intensity, spectrum/frequency, duration and pattern of exposure (Demirel *et al.*, 2009; Agrawal *et al.*, 2008). The damage to the outer hair cells depends on the intensity of noise. High-frequency noise exposure also causes damage to stereocilia and hair cells that eventually cause permanent damage. Primary exposure to noise will damage cochlear hair cells. Initially, the damage occurs to the outer hair cells, but if the exposure is continuing, it could evolve to destruct the hair cell (Nandi&Dhatrak, 2008). Noise-induced cochlear injury not only affects outer and inner hair cells, but also damage the fibroblasts of lateral cochlear wall (Haryuna *et al.*, 2015; Haryuna *et al.*, 2016; Haryuna *et al.*, 2016; Haryuna *et al.*, 2016). In this study, the SNR value after the exposure and the noise-free period was decreased compared to before exposure.

This confirms that there was a cochlear outer hair cell injury measured by DPAOE at both groups.

This experimental study used mice as an animal model. The mice also have similar inner ear structures to humans and have been used as animal models for genetic deafness studies (Vazquez et al., 2001; Salvi & Boettcher, 2008). The most common gender in the animal model is males. A study by Najarkola et al. using only male animal model showed that gender affects the measurement of level distortion product (Ldp). The Ldp is greater in females than in male. Several studies have reported these differences due to hormonal influences, and some reported due to the difference in electromotility of outer hair cells, and the mechanisms responsible for stereocilia motility, due to gender difference of lipid membrane that alter lipid-protein interactions (Moussavi-Najarkola et al., 2012).

4 CONCLUSIONS

In this study, the SNR of DPOAE examination decreased at each frequency after the noise exposure, the decrease was especially seen at the frequency of 3000 and 4000 Hz. There was an improvement in SNR values after the break period, and between the 100 dB and 110 dB group, there were not any SNR differences in each frequency.

ACKNOWLEDGMENTS

The authors express their greatest gratitude to the Research Institute of the Universitas Sumatera Utara for their full support in this research. The support is under the TALENTA of the Universitas Sumatera Utara Year 2017 Number: 5338/UN5.1.R/PPM/2017.

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