

# Longitudinal changes in dynamic characteristics of neonatal external and middle ears

メタデータ	言語: English 出版者: 公開日: 2020-06-05 キーワード (Ja): キーワード (En): 作成者: Kanka, Nattikan, Murakoshi, Michio, Hamanishi, Shinji, RisakoKakuta, Matsutani, Sachiko, Kobayashi, Toshimitsu, Wada, Hiroshi, 村越, 道生 メールアドレス: 所属:
URL	<a href="https://doi.org/10.24517/00058227">https://doi.org/10.24517/00058227</a>

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**Title:** Longitudinal changes in dynamic characteristics of neonatal external and middle ears

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**Keywords:** longitudinal change, dynamic characteristics, neonatal ear

**Abstract:**

**Objectives:** Neonates have smaller and less mature ears than adults. Developmental changes in structure and function continually occur after birth and may affect the diagnostic results obtained by audiometric assessment instrumentation, such as tympanometry and otoacoustic emission. In the present study, we investigated longitudinal changes in external and middle ear dynamic characteristics by performing sweep frequency impedance (SFI) tests.

**Methods:** SFI tests were longitudinally performed on healthy Japanese neonates (1 female and 1 male) from birth to 3 and 5 months, respectively. A sound of sweeping sinusoidal frequency, ranging from 0.1 kHz to 2 kHz, was presented to the ear canal at 50-daPa intervals of static pressure from +200 to -200 daPa. Test results were expressed as an SPL curve showing the sound pressure level (SPL) relative to probe tone frequency.

**Results:** The first variation in resonance frequency ( $RF_1$ ) and change in SPL ( $\Delta SPL_1$ ) related to the external ear showed significant developmental changes as chronological

age increased:  $RF_1$  increased, while  $\Delta SPL_1$  decreased and thereafter became unmeasurable at approximately 5 months of age. In contrast, the second variation in RF and SPL ( $RF_2$  and  $\Delta SPL_2$ ) related to the middle ear showed negligible changes over the measurement period.

**Conclusions:** The present results suggest that the dynamic characteristics of the external ear canal wall changed with increases in chronological age; the resonance of the wall at approximately 0.3 kHz at birth tended to completely disappear by approximately 5 months of age, while the characteristics of the neonatal middle ear were considered to be adult-like. The results showing how neonatal ear conditions change with chronological age may be an important key in further hearing research and the development of hearing devices and diagnostic tools that are suitable for neonates.

## **Introduction**

The external and middle ear systems are immature at birth and undergo continued development postnatally [1-4]. The external ear canal is approximately 50% shorter in length and approximately 50% narrower in diameter than that of adults (approximately 35 and 9 mm, respectively) [1, 3, 5]. After birth, the diameter and length of the ear canal increase through to the oldest test age of 24 months [3]. The neonatal external ear canal is composed of elastic cartilage at its inferior wall and was found to be easily deformed by the application of negative pressure [2, 6-10]. The tympanic membrane lies superficially in a nearly horizontal plane relative to the external ear canal axis at an angle of approximately 20 degrees, while this angle is approximately 50 to 60 degrees in adults [11]. It becomes more adult-like by 3 years of age [4]. The tympanic membrane is thicker in neonates than in adults, ranging between 0.1 and 1.5 mm in the central region of the pars tensa [12], while it varies between 0.04 and 0.12 mm in adults [13]. The thickness of other regions of the pars tensa ranges between 0.4 and 0.7 mm in the posterior–superior region, 0.7 and 1.5 mm in the umbo region, and 0.1 and 0.25 mm in the posterior–inferior, anterior–superior and anterior–inferior regions [12]. The middle ear of neonates is not completely aerated and may be filled with amniotic fluid, mesenchyme, meconium, exudates and other

contents [14]. The middle ear cavity includes the tympanic cavity, aditus ad antrum, mastoid antrum and mastoid air cells. Its volume is approximately 330 mm<sup>3</sup> in a 22-day-old infant [7], 452 mm<sup>3</sup> in 3-month-old infants and 640 mm<sup>3</sup> in adults [15]. The Eustachian tube in neonates is approximately half the size of that in adults and is closer to being horizontal with the skull base at an approximately 10-degree angle [16]. The ossicular chain is histologically adult-like at birth; i.e. its formation is complete by approximately the sixth month of fetal life [17].

These developmental changes may affect the dynamic characteristics of the neonatal ear. In our previous studies on dynamic characteristics of neonatal ears, we identified two variations of frequency, revealing the presence of vibrating elements at approximately 0.3 and 1.1 kHz [18, 19]. These variations were considered to be related to the resonances of the external ear canal wall and middle ear, respectively. In adults, only one variation related to the resonance of the middle ear was identified at approximately 1 kHz [5]. We recently attempted to investigate the effects of chronological age on these dynamic characteristics and found that the first variation tended to disappear by approximately 4–6 months of age [20]. However, there were two limitations in that study; since we adopted a cross-sectional design (i.e., we examined neonates at birth with follow-up appointments scheduled at 1, 2, 4 and 6

months of age, and evaluated the averaged value for each age group), the investigation on the maturation of the external and middle ears, particularly during the fast developmental period from birth to 4 months, was restricted to some extent. Furthermore, 5-month-old neonates were not examined. Therefore, it currently remains unclear whether 5-month-old infants show SFI findings that are consistent with the changes in resonance frequency and mobility observed in that study.

We herein performed a longitudinal study on 2 healthy neonates to track developmental changes in external and middle ear dynamic characteristics using the SFI technique.

## **Materials and methods**

### **1. SFI meter**

The sweep frequency impedance (SFI) meter was initially developed to objectively and quickly evaluate the mobility of the middle ear in adults and children [5]. It was then successfully developed to measure dynamic characteristics in the external and middle ear systems of neonates [18, 19, 21-23]. Data obtained using the SFI technique contributes to the detection of middle ear dysfunctions.

A schematic diagram of the SFI meter is shown in Fig. 1. The SFI device consists of a probe containing a microphone and earphone (ER-10C, Etymotic Research, Elk Grove Village, IL, USA), amplifier, syringe pump, stepping motor (KR-3ML, Techno Drive, Kawasaki, Japan), pressure sensor (PA-100-500D-W, Nidec Copal Electronics, Tokyo, Japan), pressure relief valve (790 Dual Action Breather Valve, Halkey-Roberts, St. Petersburg, FL, USA), AD/DA converter (NI 6024E, National Instruments, Austin, TX, USA) and personal computer (CF-W7, Panasonic, Osaka, Japan).

The features of SFI data [5, 18, 19, 24] are briefly reviewed as follows. The SFI meter measures SPL by presenting a sound of sweeping sinusoidal frequencies, ranging from 0.1 kHz to 2 kHz, to the ear canal with a constant volume displacement. During this procedure, static pressure from +200 daPa to -200 daPa is applied at intervals of 50 daPa to the ear canal. The reflected sound is recorded by the microphone and then sent to the personal computer for data analysis. The measurement results show a sound pressure level (SPL) relative to the probe tone frequency, called the SPL curve, as shown in Fig. 2. Variations in SPL are calculated using the following equation:

$$\text{SPL} = 20 \log \frac{P}{P_{\text{REF}}} \quad (1)$$

where  $P$  is the sound pressure measured by the microphone and  $P_{\text{REF}}$  is the minimum audible sound pressure, which is  $2.0 \times 10^{-5}$  Pa. Dynamic characteristics, which are the resonance frequency (RF) and changes in SPL ( $\Delta\text{SPL}$  in dB), are measured using the following equations:

$$\text{RF}_n = \frac{F_{a,n} + F_{b,n}}{2} \quad (n = 1, 2) \quad (2)$$

$$\Delta\text{SPL}_n = P_{a,n} - P_{b,n} \quad (3)$$

where  $P_{a,n}$  and  $P_{b,n}$  are the maximum and minimum sound pressures,  $F_{a,n}$  and  $F_{b,n}$  are the frequencies corresponding to these sound pressures, and  $n$  is the number of variations.

## 2. Subjects

Recruitment of participants was performed by nurses of the Department of Obstetrics and Gynecology, Japanese Red Cross Sendai Hospital, Sendai, Japan, in accordance with the ethical guidelines approved by the Ethics Committees on Clinical Investigation of Tohoku University School of Medicine and the Japanese Red Cross Sendai Hospital. Parents of healthy neonates at the maternity unit of the Japanese Red Cross Sendai Hospital were informed of the study before the measurements. Participants were included in the study upon written consent from their parents.

In the present study, SFI tests were performed on 2 healthy Japanese neonates (1 female and 1 male). Both neonates were full-term and had normal perinatal histories with no high risk factors for hearing loss. Their birth weights were 3,056 and 3,312 g, respectively. They had passed newborn hearing screening tests; otoscopy tests by otorhinolaryngology doctors to confirm the normal development of the external ear and tympanic membrane, and automated auditory brainstem response (AABR) tests.

### **3. Measurements**

In the present study, SFI tests were performed longitudinally for different lengths of time during the first half year after birth. The female neonate underwent SFI tests from birth to 3 months (6, 14, 23, 38 and 92 days after birth), while the male neonate underwent SFI tests from birth to 5 months (9, 22, 58, 74, 96, 118, 124, 135, 143 and 152 days after birth). Tests were performed when neonates were in a good health condition and conducted in a quiet room while they were naturally sleeping.

## **Results**

### **1. Longitudinal changes in the female neonate (0–3 months)**

The SPL curves obtained from the female neonate are shown in Fig. 3(A). The SPL curve obtained from 6 days after birth showed the minimum SPL, 54.2 dB, at a frequency of 0.14 kHz (cross mark, Fig. 3(A)), which subsequently increased to approximately 1.5 kHz. The SPL curve obtained when the neonate was 92 days old showed some differences in shape and value. Regarding shape, at the frequency area below 0.28 kHz, SPL obtained from 92 days after birth was approximately 2–3 dB higher. At the higher frequency area, the SPL curve had a similar curve to the former, but with lower SPL by approximately 2–3 dB.

The first variation in RF ( $RF_1$ ) and change in SPL ( $\Delta SPL_1$ ) are shown in Fig. 3(B) and (C).  $RF_1$  was 0.27 kHz and  $\Delta SPL_1$  was 9.62 dB 6 days after birth. As chronological age increased to 92 days,  $RF_1$  increased approximately 1.59-fold to 0.43 kHz, while  $\Delta SPL_1$  rapidly decreased to 3.75 dB. Six days after birth, the second variation in RF ( $RF_2$ ) was 1.29 kHz (Fig. 3(B)) and the change in SPL ( $\Delta SPL_2$ ) was 2.15 dB (Fig. 3(C)). Ninety-two days after birth,  $RF_2$  increased 1.02-fold to 1.31 kHz (Fig. 3(B)), while  $\Delta SPL_2$  increased approximately 1.07-fold to 2.31 dB (Fig. 3(C)). Regarding the curves of RF and  $\Delta SPL$ ,  $RF_1$  appeared to increase proportionally and was slightly elevated even after 92 days.  $\Delta SPL_1$  monotonically decreased with an increase in age and then markedly decreased. The curves for  $RF_2$  and  $\Delta SPL_2$  showed negligible

changes over the time period tested.

## **2. Longitudinal changes in the male neonate (0–5 months)**

The SPL curves obtained from the male neonate are shown in Fig. 4(A). As chronological age increased to 135 days, SPL at a frequency below 1 kHz became 2–3 dB lower, while that at higher frequency up to 2 kHz became 3–10 dB lower than that obtained 9 days after birth. Additionally, at a frequency above 0.26 kHz, SPL obtained from 143 and 152 days after birth became 3–6 dB lower than that obtained 9 days after birth.  $RF_1$ , as shown in Fig. 4(B), was 0.27 kHz 9 days after birth. It subsequently increased approximately 2.15-fold to 0.58 kHz at 96 days, approximately 2.63-fold to 0.74 kHz at 135 days, and thereafter became unmeasurable.  $RF_2$  was 0.98 kHz 9 days after birth. It increased by approximately 1.06-fold to 1.04 kHz at 96 days and 1.08-fold to 1.06 kHz at 152 days.  $RF_2$  was not significantly different from that obtained 9 days after birth.

The change in SPL is shown in Fig. 4(C).  $\Delta SPL_1$  was 7.27 dB and  $\Delta SPL_2$  was 4.23 dB 9 days after birth.  $\Delta SPL_1$  rapidly decreased in the first 2 months, and then gradually decreased: 1.19 and 0.74 dB at 96 and 135 days, respectively. Regarding the grey line in Fig. 4(C),  $\Delta SPL_2$  changed in an uncertain pattern across the 5-month period,

but finally became 3.45 dB, which was approximately 0.82-fold lower than that obtained at 9 days.

## Discussion

In the present study, to track developmental changes in external and middle ear dynamic characteristics in neonates using the SFI technique, a longitudinal design, rather than the cross-sectional design in our previous study [20], was adopted in a limited number of healthy neonates from birth to 3 and 5 months. The present results showed that  $RF_1$  (black lines, Figs. 3(B) and 4(B)) and  $\Delta SPL_1$  (black lines, Figs. 3(C) and 4(C)), which were considered to be related to the external ear system, increased and decreased, respectively, as chronological age increased. In contrast,  $RF_2$  (gray lines, Figs. 3(B) and 4(B)) and  $\Delta SPL_2$  (gray lines, Figs. 3(C) and 4(C)), which were considered to be related to the middle ear system, showed negligible developmental changes. These results demonstrate that the resonance of the external ear canal wall at approximately 0.3 kHz at birth tends to completely disappear by 5 months of age.

In a study on changes in RF in the ear with age, Holte *et al.* [25] performed multifrequency tympanometry on healthy neonates at various time intervals in the first 4

months of life and found a resonance of approximately 0.45 kHz, which subsequently disappeared by 4 months after birth. Meyer *et al.* [26] performed conventional and high probe tone tympanometry on a healthy female neonate until 6.5 months of age. Resonance appeared below 0.55 kHz during the first 3 months, and then began to disappear at approximately 3.5 months of age. We recently reported [20] that  $RF_1$  was below 0.55 kHz until 4 months of age and then tended to disappear at approximately 4–6 months. The means and standard deviations of  $RF$  and  $\Delta SPL$  derived from Japanese (this study) and Australian neonates [20] are shown in Fig. 5. Their  $RF_1$ , shown in Fig. 5(A), and  $\Delta SPL_1$ , shown in Fig. 5(B), were similar to the values obtained at birth; 0.27 kHz in Japanese and  $0.28 \pm 0.05$  kHz in Australians. They also showed similar disappearance. The  $RF_1$  and  $\Delta SPL_1$  obtained from Australian neonates tended to disappear by approximately 4–5 months of age, while those derived from Japanese neonates disappeared after 4 months and completely disappeared by 5 months of age. The developmental changes that occur in the external and middle ears of neonates may be explained by regarding the ear as a simple harmonic oscillator. The natural frequency of this oscillator  $f$  is proportional to the characteristic of the oscillator, as expressed by the following equation:

$$f \propto \sqrt{\frac{k}{m}} \quad (4)$$

where  $k$  and  $m$  are the characteristic and the mass of the external or middle ear, respectively. Equation (4) shows that the frequency increases when the characteristic representing the stiffness of the ear increases; therefore, the results in Fig. 5(A) may be interpreted as the external ear canal wall becoming stiffer with chronological age as  $RF_1$  continuously increased. Additionally, from each bar height in Fig. 5(A), the stiffness of the external ear canal wall was larger in Australian neonates than in Japanese neonates during the first 2 months of age.  $\Delta SPL_1$ , which represents the mobility of the external ear in terms of changes in SPL, appeared to decrease, indicating that the mobility of the external ear canal wall became restricted.

$RF_2$ , as shown in Fig. 5(C), from Japanese and Australian neonates had similar values and tendencies.  $RF_2$  obtained from Japanese and Australians were  $1.14 \pm 0.15$  and  $1.22 \pm 0.24$  kHz at birth, respectively. Even with increases in chronological age,  $RF_2$  remained similar; at the age of 4 months, the  $RF_2$  of Japanese neonates was 1.12 kHz and that of Australian neonates was  $1.40 \pm 0.18$  kHz. These comparisons confirmed that RF in the middle ear did not significantly change, as each bar shows an almost constant level.  $\Delta SPL_2$  at birth, shown in Fig. 5(D), were  $3.2 \pm 1.0$  and  $5.0 \pm 2.0$  dB in Japanese and Australian neonates, respectively, and did not significantly change

thereafter. In comparison to the RF and  $\Delta$ SPL of Japanese adults [27], which were  $1.17 \pm 0.27$  kHz and  $2.18+3.84/-1.39$  dB, respectively, the measurement results from the present study confirmed that the middle ears of neonates did not only have an adult-like structure from after birth but had adult-like characteristics; e.g., stiffness and mobility [16-18, 26]. Additionally, from each bar height in Figs. 5(C)–(D), Australian neonates showed slightly larger  $RF_2$  and  $\Delta$ SPL<sub>2</sub> than Japanese neonates. Although the number of Japanese neonates examined was limited, Australian neonates may possibly to have a stiffer middle ear than Japanese neonates.

The present results are consistent with previously reported developmental changes [20]; the chronological age at which the oscillatory behavior of the external ear canal disappeared was approximately 5 months of age. This may be vital information for upcoming neonatal hearing research.

There were some limitations in the present study. Difficulties were associated with performing neonatal SFI tests at specific appointment times because these tests were only conducted in the present study when subjects were in a good health condition in order to reduce risk factors that may alter their ear condition; e.g., restless, cold, or fever, and when they were naturally sleeping in order to avoid jaw and suckling movements. Furthermore, the sample size was small. Since the longitudinal study

design for approximately one quarter or half a year challenging without the cooperation and efforts of both babies and parents, the difficulties associated with completing the entire battery of tests were markedly greater than those of a cross-sectional study design. A larger sample size is needed for more appropriate statistical analyses.

## **Conclusions**

In the present study, SFI tests were longitudinally performed on 2 Japanese neonates from birth to 3 and 5 months, respectively, in an attempt to investigate changes in external and middle ear dynamic characteristics accompanying chronological age. The results obtained suggest that the stiffness of the external ear canal wall increases with chronological age, and the oscillatory behavior of the external ear canal wall tends to completely disappear by approximately 5 months of age. In contrast, changes in the dynamic characteristics of the middle ear were not significant. Consistent with previous findings, the neonatal middle ear was considered to be adult-like after birth. These results on how neonatal ear conditions change with chronological age may be useful for further research on the mechanics of neonatal hearing and development of hearing devices.

## **Acknowledgements**

The authors are grateful to the nurses and other personnel of the Japanese Red Cross Sendai Hospital, Sendai, Japan, for their cooperation. This work was supported by JSPS KAKENHI Grant Number JP16K11194 to H.W, and by a grant from the TERUMO FOUNDATION for LIFE SCIENCES and ARTS and a grant from the MIKIYA Science And Technology Foundation to M.M.

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## Figure captions

**Fig. 1.** Sweep frequency impedance (SFI) meter. The meter consists of a probe containing a microphone and earphone, amplifier, syringe pump, stepping motor, pressure sensor, pressure relief valve, AD/DA converter and personal computer.

**Fig. 2.** Representative SPL curve in a healthy neonate. RF is the resonance frequency,  $\Delta$ SPL is the change in SPL,  $P_a$  and  $P_b$  are the maximum and minimum sound pressures, and  $F_a$  and  $F_b$  are the frequencies corresponding to these sound pressures.

Under ambient pressure, two variations in RF were observed at approximately 0.3 and 1.1 kHz. (From Murakoshi *et al.* (2013). *International Journal of Pediatric*

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**Fig. 3.** SFI results obtained from the female neonate. (A) SPL curve 6 and 92 days after birth. The cross mark, at approximately 0.14 kHz, shows the minimum SPL 6 days after birth. The SPL curve at 92 days showed higher SPL than that at 6 days at a frequency below 0.28 kHz, while its SPL thereafter became approximately 2–3 dB lower. (B) Longitudinal changes in RF<sub>1</sub> and RF<sub>2</sub>. RF<sub>1</sub> showed proportional changes

in SPL, while RF<sub>2</sub> did not significantly change. (C) Longitudinal changes in  $\Delta\text{SPL}_1$  and  $\Delta\text{SPL}_2$ .  $\Delta\text{SPL}_1$  steadily changed, whereas no significant change was observed in  $\Delta\text{SPL}_2$ .

**Fig. 4.** SFI results obtained from the male neonate. (A) SPL curve 9, 96, 135, 143 and 152 days after birth. SPL became lower as chronological age increased. (B) Longitudinal changes in RF<sub>1</sub> and RF<sub>2</sub>. RF<sub>1</sub> changed proportionally and became unmeasurable after 135 days, while RF<sub>2</sub> did not significantly change. (C) Longitudinal changes in  $\Delta\text{SPL}_1$  and  $\Delta\text{SPL}_2$ .  $\Delta\text{SPL}_1$  rapidly decreased in the first 2 months, while  $\Delta\text{SPL}_2$  showed no significant change.

**Fig. 5.** Means and standard deviations of RF and  $\Delta\text{SPL}$ ; (A) RF<sub>1</sub>, (B)  $\Delta\text{SPL}_1$ , (C) RF<sub>2</sub> and (D)  $\Delta\text{SPL}_2$ , derived from Japanese neonates (2 neonates (1 female and 1 male)) and Australian neonates (117 ears from 83 infants (47 males and 36 females) [20]). As chronological age increased, RF<sub>1</sub> increased and  $\Delta\text{SPL}_1$  decreased, while RF<sub>2</sub> and  $\Delta\text{SPL}_2$  did not significantly change.

Fig.1

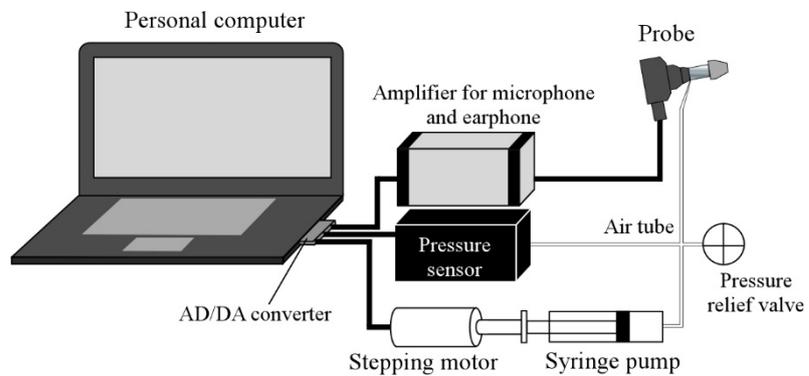


Fig. 2

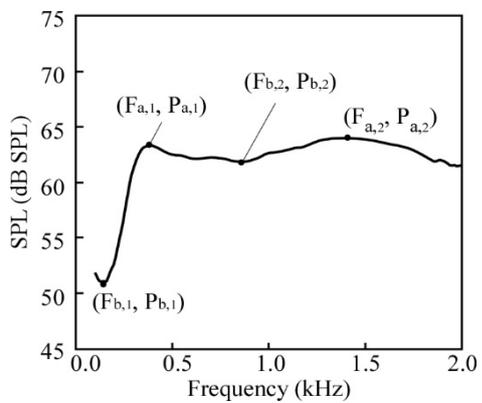


Fig. 3

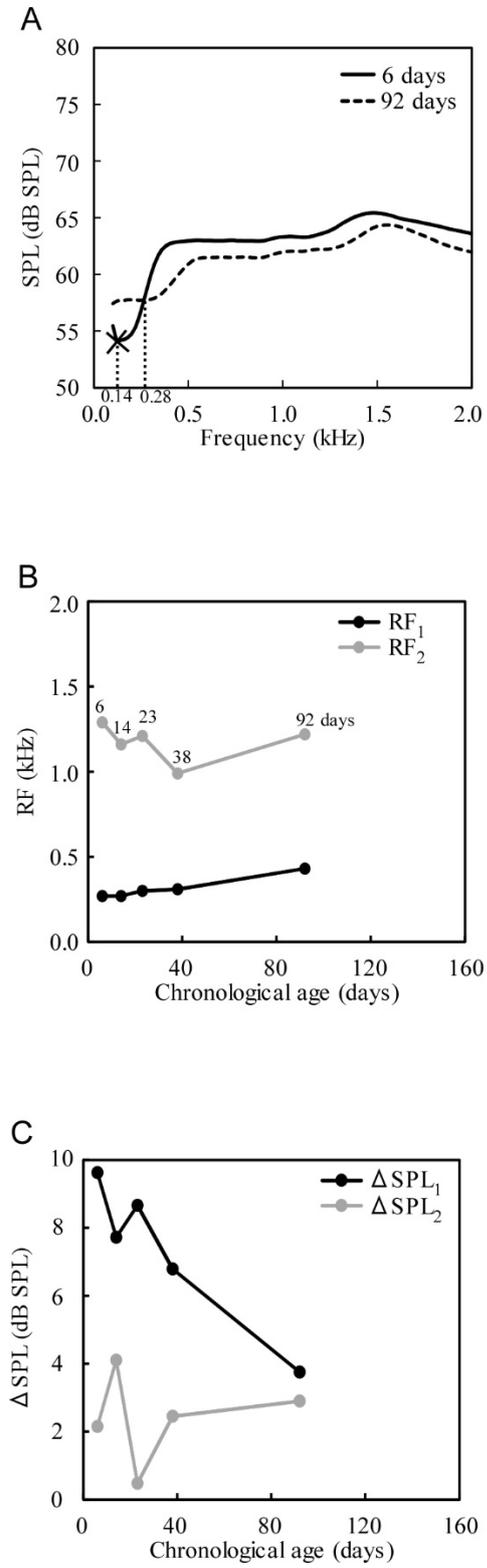


Fig. 4

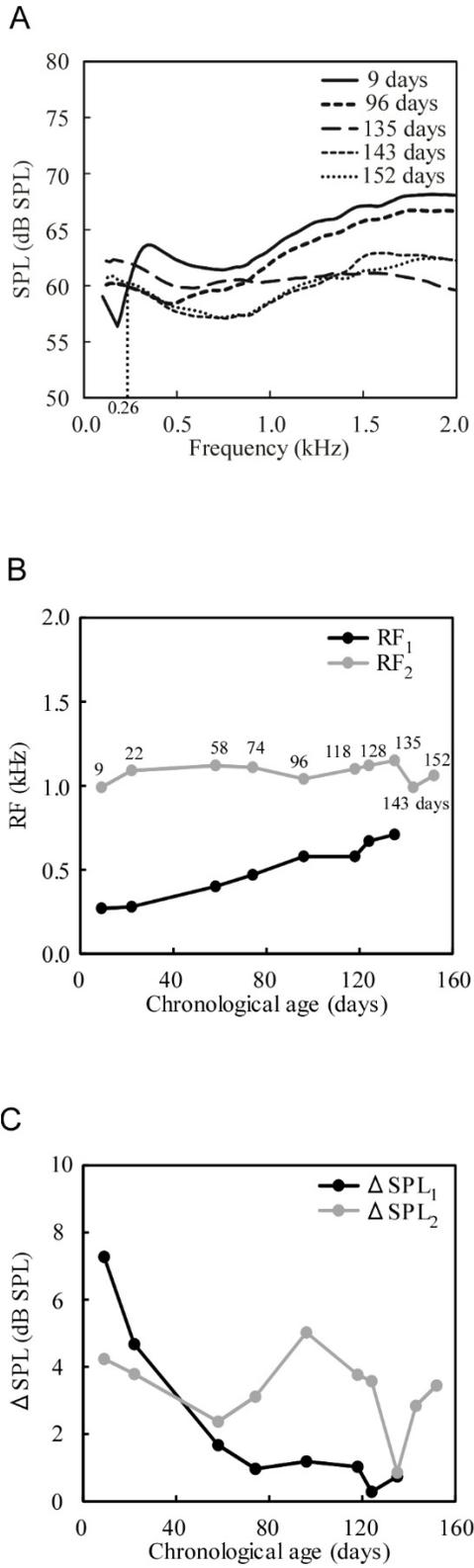


Fig. 5

