

Impact of Intrascalar Blood on Hearing

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Objective/Hypothesis: The objective of this controlled animal study was to evaluate the effects of intrascalar blood on hearing. **Material and Methods:** Eight guinea pigs underwent intrascalar administration of their own blood in one ear and control solution in the contralateral ear. Solutions were applied through cochleostomy to the scala tympani. Compound action potential (CAP) thresholds were determined before administration and at different intervals for 2 months thereafter. **Results:** Immediate deterioration of thresholds was seen mainly in the high-frequency range, averaging 27 dB and 20 dB in the study and control groups, respectively. At day 3, threshold shifts recovered in the control group but remained in the low-frequency range in the study group. An extensive recovery was seen in both groups. However, permanent threshold shifts persisted. There was an enhanced shift of thresholds of up to 7 dB in the study group. **Conclusions:** Even small amounts of intrascalar blood seem to cause transient and permanent detrimental effects on cochlear function. In procedures involving opening of the otic capsule—like stapes surgery and cochlear implantation with hearing preservation—minimizing surgical blood admixture to intracochlear compartments seems therefore fundamental. **Key Words:** Cochlear implant, combined stimulation, EAS, CI, blood, guinea pig, hearing preservation, electric—acoustic stimulation, compound action potentials, CAP.

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INTRODUCTION

Cochlear implantation has evolved into a standard procedure in the treatment of deaf and profoundly hearing impaired individuals. Recently, the combined electric—acoustic stimulation (EAS) of the auditory system was established for a subgroup of patients with profound high-

frequency hearing loss and relatively good residual low-frequency hearing.^{1–4}

As a result of severely impaired hearing within the frequency range of spoken language, some of these patients do not show adequate benefit from conventional hearing aids. With the combined stimulation, missing high-frequency information is provided electrically, whereas conventional low-frequency amplification delivers information to the still functioning areas of the cochlea; thus, bimodal stimuli are delivered to the same ear. The addition of acoustic information showed significant performance benefits, especially in background noise. This was uniformly reported among various investigators.^{2,3}

During regular cochlear implantation, residual hearing is usually lost. Because the combined stimulation depends on low-frequency hearing, great efforts have been undertaken to preserve residual cochlear function. Although different surgical protocols of atraumatic procedures have been developed,^{2,4} some EAS patients still lost their residual hearing. However, the exact underlying mechanisms are not fully revealed at this time.

As shown in the central nervous system, some blood constituents like ferrous ions can act neurotoxic as a result of a strong oxidative potential.^{5,6} Additionally, there are several case reports of inner ear hemorrhage leading to severe hearing loss or even deafness.^{7,8}

Therefore, we evaluated the effect of blood admixture to the perilymphatic fluid of the scala tympani in an animal model over a period of 8 weeks.

MATERIALS AND METHODS

Animals

Eight adult guinea pigs (*Cavia porcellus*) were obtained from a local breeder (Charles River Wiga GmbH, Sulzfeld, Germany). The weight of the animals ranged from 230 to 480 g before the first procedure. The study protocol and care facilities were approved by the designated local authorities.

Anesthesia

All preparations and measurements were performed under general anesthesia. Each animal was anesthetized using a composition of ketamine and xylazine (initial dose: 62.5 ± 6.5 mg/kg body weight) applied intraperitoneally. Additional doses of 25% of the initial amount were given as necessary.

Implantation of Measurement Wires

Hearing thresholds were measured by means of eight nerves' compound action potentials (CAPs). To obtain measurements, a

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gold-wire electrode was implanted in both ears. Therefore, the skin was incised along the midline of the head and the caudal part of the incision was enlarged laterally to each side. Connective tissue and muscles overlying the middle ear cavity were pushed aside and a hole was made into the occipital bone using a scalpel and tweezers. A prepared gold-wire electrode was placed in close vicinity of the round window membrane and fixed to the skull using tissue adhesive. The procedure was repeated on the contralateral side. Then a plug was fixed to the skull and soldered to the electrodes. All compartments remained in situ during the entire time of the experiment.

Administration of Experimental and Control Solutions

A cochleostomy was drilled in both ears using a 1.0-mm diamond burr. Thereafter, blood was drawn from the pinna using a stab incision. In each animal, 3 μ L of blood was administered to one ear (study ear) and 3 μ L modified Hank's solution (Sigma-Aldrich Chemie GmbH, Munich, Germany) to the other (control ear) using Hamilton syringes. A small piece of muscle was placed onto the cochleostomy and the middle ear cavity was sealed with dental acrylic.

Measurements, Timeline, and Evaluation

CAP recordings were performed before opening of scala tympani, directly after blood/control administration, at days 3 and 7, and then weekly until 8 weeks after topical application of the experimental solutions. Tone bursts (bandwidth two-thirds octaves, length 3 ms) presented at 19 logarithmically equidistantly spaced middle frequencies in the range from 0.125 to 64 kHz were used for stimulation. Tone bursts at each frequency were presented at different sound pressure levels over a range of 100 dB in steps of 5 dB. The sound system consisted of a Beyerdynamic DT 48 sound source (Beyerdynamic, Heilbronn, Germany), which was connected to each ear with a speculum. Responses to tone bursts were amplified by 80 dB, band-pass filtered (0.1–3 kHz), and recorded through an A/D converter with 20 kHz sampling rate. Responses to 20 stimulus presentations at each level were averaged and stored for further data analysis. The hearing threshold at each frequency was defined as the minimum sound pressure level necessary to visually detect a response in the averaged signal.

Differences between study and control groups were analyzed using the Wilcoxon matched pairs test. A *P* value of $<.05$ was considered statistically significant.

RESULTS

The results refer to seven ears in each group. Two ears (one in each group) had to be excluded as a result of excessive permanent threshold shifts caused by the surgical procedure.

An overview about the time course of CAP thresholds is given in Figure 1. Postapplication thresholds in study and control groups were deteriorated mainly in the high-frequency range between 8 kHz and 64 kHz (Fig. 2). The average high-frequency threshold shifts were 27 dB and 20 dB for the study and control groups, respectively. Values were 12 dB and 9 dB for the lower frequencies. The difference between mean values calculated over the entire frequency range of both groups was statistically significant ($P < .05$).

Measurements at day 3 revealed a change in the pattern of the threshold shifts (Fig. 3); in control ears, recovery of threshold shifts was observed. Threshold shifts

nearly halved to 11 dB in the high frequencies and less than 5 dB in the lower frequencies. In contrast, average threshold shifts in the study group showed only slight recovery to 25 dB in the high frequencies and a worsening to 18 dB in the lower frequencies. Again, the differences between study and control ears were statistically highly significant ($P < .001$).

Stable permanent threshold shifts remaining until the end of the experiment were reached 5 weeks after surgery for both groups (Fig. 4). Within the control group, the threshold shift averaged 1 to 3 dB in the frequency range of 0.125 to 4 kHz. A more pronounced threshold loss of 8 to 12 dB was seen for the frequencies between 5.65 and 16 kHz. In even higher frequencies between 22.6 and 64 kHz, threshold loss was inconsistent and averaged +1 to –3 dB.

In the study group, the threshold shift averaged 3 to 5 dB in the frequency range of 0.125 kHz to 1.41 kHz. A more pronounced threshold shift of on average 9 to 12 dB was seen in the frequency range of 2 to 22.6 kHz. Again, an inconsistent threshold loss of +2 to –1 dB was observed in the frequency range of 32 to 64 kHz.

Differences in residual threshold shifts between both groups were clearly visible for the frequency range of 0.3 to 8 kHz. The threshold differences between both groups in this frequency range averaged from 3 to 7 dB during weeks 3 through 8. Differences of threshold shifts over the entire frequency range between study and control groups were statistically significant at weeks 6 and 7 ($P < .05$) and were marginal above significance level at weeks 3 to 5 and 8 ($P < .06$).

DISCUSSION

This study documents the effects of intrascalar blood on hearing in an animal model. According to the present report, threshold shifts can be subdivided into three different phases: 1) severe threshold shift mainly observed in the high frequencies in both groups immediately after intrascalar administration. Those shifts were more distinct in the study group. 2) At day 3, recovery was observed in the control group, whereas threshold loss was extended toward the low frequencies in the study group. 3) No change in threshold shifts was observed after 5 weeks after surgery. These threshold shifts remained stable until the end of the observation period and are thus considered permanent. Permanent inner ear damage was greater in the study group; however, differences were smaller than during the first week immediately after administration.

Thus, blood admixture to the scala tympani had a significant impact on hearing thresholds during the three different periods of the observation time. Postoperative measurements showed average hearing deterioration of 27 dB in the high-frequency range after cochleostomy and application of 3 μ L blood. This may in part be related to the surgical procedure, which is accompanied by a certain amount of fluid loss from the perilymphatic space. Additionally, temporary threshold shifts resulting from noise exposure during drilling and ossicular manipulation is expected. Kiefer et al.⁹ found an average threshold shift of 10 dB in the high-frequency range after cochleostomy alone in guinea pigs. The difference from the present data

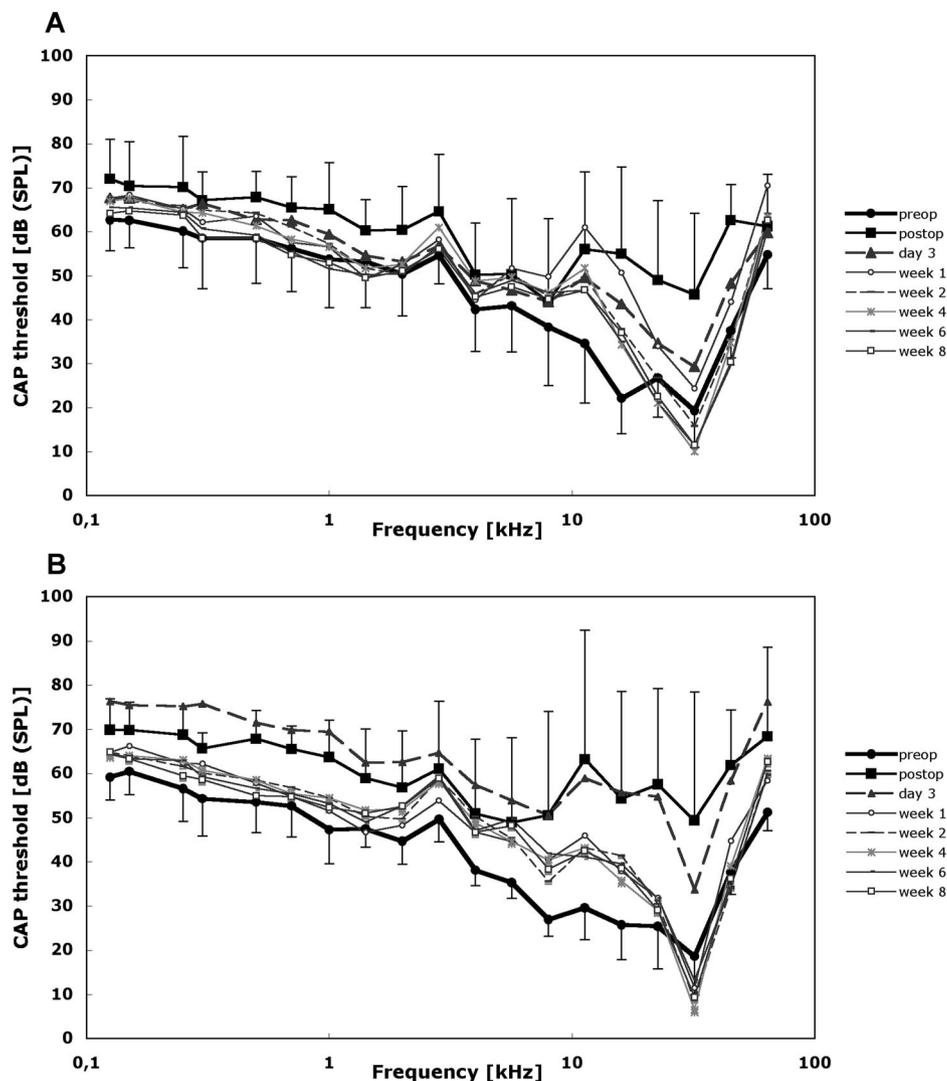


Fig. 1. Time course of hearing thresholds of (A) control and (B) blood group during the whole experiment. Bars indicate 1 standard deviation (SD).

is explainable by the additional manipulation caused by the application procedure.

At day 3, significant threshold recovery was observed in the control group. In the study group, however, only slight improvement of thresholds, which was limited to

high frequencies, was observed. Hearing thresholds for the lower frequencies of the blood group worsened noticeably. This is likely to be the result of an ongoing degradation process of erythrocytes and the subsequent release of potassium to the perilymphatic space. A distribution of

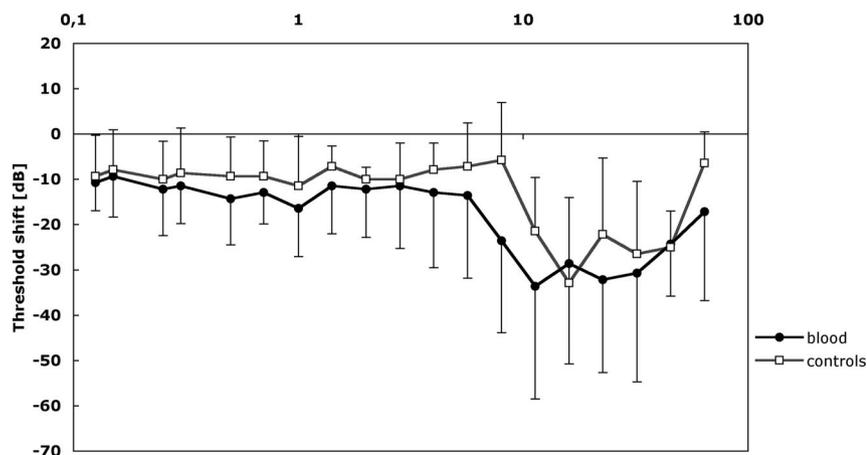


Fig. 2. Threshold shifts within the blood versus the control group at different frequencies immediately after application of experimental solutions. Differences are statistically significant ($P < .05$). Bars indicate standard deviation.

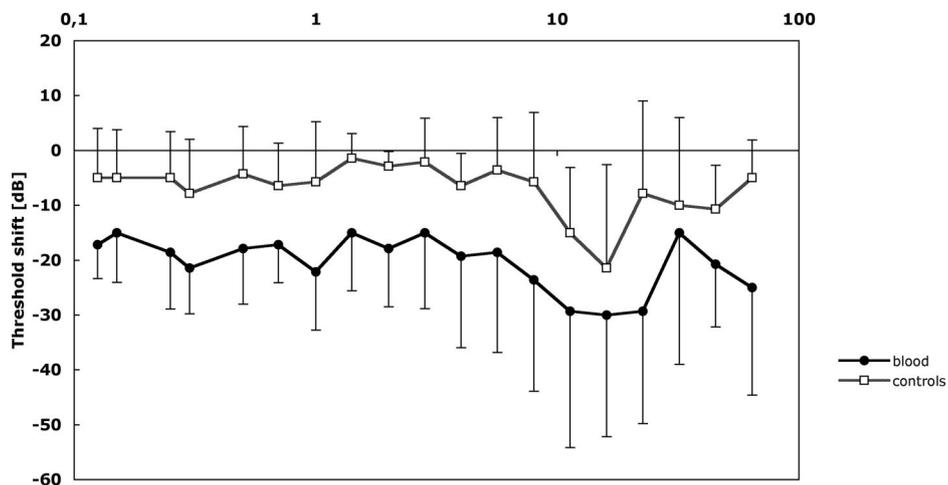


Fig. 3. Threshold shifts within the different groups at day 3 ($P > .001$). Bars indicate standard deviation.

blood cells and/or their contents to more apical regions of the cochlea is possible, leading to the delayed effects in the low-frequency range.

Carvalho and Lalwani¹⁰ found a frequency-dependent threshold shift of ≤ 10 dB in midfrequencies (4 and 8 kHz) and of ≥ 30 dB in the high-frequency range (16, 20, and 30 kHz) after cochleostomy and implantation of a miniosmotic pump delivering artificial perilymph. The threshold loss was observed on postoperative days 3 and 7, being the only two measurements in this acute experiment.

A permanent threshold shift was observed in the control group for frequencies of 5.65 to 16 kHz. We think that this loss was caused by mechanical damage resulting from the surgical procedure. Permanent threshold shifts were extended toward low frequencies in the study group. The admixture of blood caused an additional average threshold shift of 3 to 7 dB (weeks 3–8) in the frequencies of 0.3 to 8 kHz. The reason for this permanent threshold shift remains unknown. Possible explanations include oxidative effects of hemoglobin as demonstrated in central nervous system (CNS) hemorrhage. Sadrazadeh et al.⁵ have shown that hemoglobin reduces the activity of the Na/K-ATPase in the CNS. Additionally, it carries the potential to catalyze substantial per-oxidation reactions, e.g., of CNS lipids. These deleterious effects were shown to

be caused at least in part by ferrous ions that can oxidize oxygen to reactive oxygen species or directly lead to lipid per-oxidation. Ferric ions that arise through this reaction are subsequently recycled to ferrous ions by ascorbic acid.⁶ Thus, even small amounts of hemoglobin are likely to have harmful effects. Furthermore, there is some evidence that blood constituents are not easily eliminated from the perilymphatic space but are detectable for several months.⁷

Other conceivable reasons for the observed permanent threshold shift in the study group are inflammatory reactions resulting from the degradation process of blood constituents. In general, the detrimental effect of inflammation of the inner ear on hearing thresholds is well known. After surgery of the inner ear, Smouha¹¹ found greater degrees of hearing loss correlated with extended inflammatory responses in a guinea pig model. Additionally, it could be demonstrated that expression of tumor necrosis factor alpha (TNF α) was increased after CNS hemorrhage.¹² This is a strong mediator in the pathway of cochlear inflammation with its well-known consequences.^{13,14}

A limitation of this study might be the relatively small sample size. Although study and control groups have been treated similarly, factors unrelated to the application of blood may have contributed to the results.

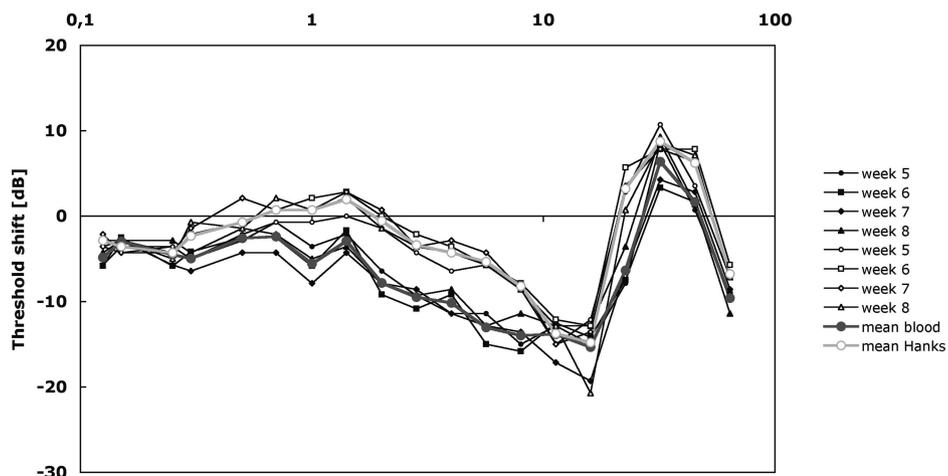


Fig. 4. Permanent threshold shifts measured at weekly intervals (week 5–8) within both groups. Bold lines show the mean values of each group ($P < .05$ for weeks 6 and 7; $P < .06$ for weeks 3–5 and 8).

CONCLUSIONS

In this study, the acute and long-term effect of intrascalar blood on hearing thresholds could be demonstrated. Transient threshold shifts were observed during the first postoperative days, whereas limited recovery occurred in the study ears. Hence, even small amounts of intrascalar blood caused significant permanent threshold shifts in this relatively small population.

Applications in this report were performed through a cochleostomy and thus imitate cochlear implantations (with hearing preservation). However, the results of the current report might be valid for other procedures, which involve unsealing of the otic capsule—like stapes surgery.

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