Transtympanic sodium thiosulfate for prevention of cisplatin-induced ototoxicity: a randomized clinical trial

Duinkerken, C.W.; Weger, V.A. de; Dreschler, W.A.; Molen, L. van der; Pluim, D.; Rosing, H.; ... ; Zuur, C.L.

Transtympanic Sodium Thiosulfate for Prevention of Cisplatin-Induced Ototoxicity: A Randomized Clinical Trial

*†Charlotte W. Duinkerken, ‡§||Vincent A. de Weger, ¶Wouter A. Dreschler, *Lisette van der Molen, §Dick Pluim, #Hilde Rosing, #Bastiaan Nuijen, **††Michael Hauptmann, ‡Jos H. Beijnen, *‡‡Alfons J.M. Balm, §§Jan Paul de Boer, ||||Jacobus A. Burgers, ‡§§Serena Marchetti, ¶¶Jan H.M. Schellens, and *‡‡##Charlotte L. Zuur

*Department of Head and Neck Surgery and Oncology, the Netherlands Cancer Institute, Amsterdam; †Department of Otolaryngology, Leiden University Medical Centre, Leiden; ‡Division of Clinical Pharmacology; §Division of Pharmacology, the Netherlands Cancer Institute, Amsterdam; ||Department of Internal Medicine, Noordwest Ziekenhuisgroep, Alkmaar; ¶Department of Audiology, Amsterdam University Medical Centre; #Department of Pharmacy and Pharmacology; **Department of Epidemiology and Biostatistics, the Netherlands Cancer Institute, Amsterdam, the Netherlands; ††Institute of Biostatistics and Registry Research, Brandenburg Medical School, Neuruppin, Germany; ‡‡Department of Maxillofacial Surgery, Amsterdam University Medical Centre; §§Department of Medical Oncology; ||||Department of Thoracic Oncology, the Netherlands Cancer Institute, Amsterdam; ¶¶Utrecht Institute of Pharmaceutical Sciences, Utrecht University, Utrecht; and ##Cell Biology and Immunology, Netherlands Cancer Institute, Amsterdam, the Netherlands

Objectives: To determine safety, feasibility, and preliminary activity of transtympanic injection of sodium thiosulfate (STS) against cisplatin-induced hearing loss (CIHL).

DESIGN Randomized controlled trial.

SETTING Tertiary cancer hospital.

PATIENTS Adults to be treated with high-dose cisplatin ($\geq$ 75 mg/m^2).

INTERVENTION Selected by randomization, 0.1 M STS gel on one side and placebo gel on the other side was transtympanically applied to the middle ear 3 hours before cisplatin administration. After amendment, the placebo ear was left untreated.

Main Outcome Measure: Primary outcome was safety and feasibility. Secondary outcomes included pharmacokinetic analysis of systemic cisplatin and preliminary activity of STS. Clinically relevant CIHL was defined as a $\geq$ 10 dB threshold shift at pure-tone average 8-10-12.5 kHz ($PTA_{8\text{-}12.5}$). Response to STS was defined as a threshold shift at $PTA_{8\text{-}12.5}$ in the STS-treated ear of $\geq$ 10 dB smaller than the untreated ear.

Results: Twelve patients were treated. Average CIHL at $PTA_{8\text{-}12.5}$ was 12.7 dB in untreated ears and 8.8 dB SPL in STS-treated ears ($p = 0.403$). Four patients did not develop CIHL. Four out of eight patients with CIHL responded to STS: CIHL at $PTA_{8\text{-}12.5}$ in STS-treated ears was 18.4 dB less compared to untreated ears ($p = 0.068$). Grade 1 adverse events were reported. Pharmacokinetic results were available for 11 patients.

Conclusion: Transtympanic application of STS was safe and feasible. Based on our pharmacokinetic analysis, we postulate that transtympanic STS does not interfere with the systemically available cisplatin. Our results provide a preliminary proof of concept for transtympanic application of STS in preventing CIHL and warrants further evaluation on a larger scale.

Key Words: Cisplatin-induced hearing loss—Ototoxicity—Sodium thiosulfate—STS—Transtympanic injection.

Otol Neurotol **42:**678–685, 2021.

Cisplatin-induced hearing loss (CIHL) occurs in 75 to 80% of the cisplatin-treated patients (1–4). CIHL is dose-dependent and characterized by symmetric, bilateral, and irreversible sensorineural hearing loss (SNHL), starting shortly after treatment (1,5). Cisplatin destructs the hair cells (HCs) within the organ of Corti (1,5,6). First, the outer HCs located at the basal cochlear turns are affected, leading to SNHL at ultrahigh frequencies. After subsequent doses also the apical windings are involved and CIHL progresses to lower frequencies (1,7,8). Furthermore, cisplatin may damage the inner HCs, spiral ganglion, and stria vascularis (1,5).

There is an increasing interest in the research field of (preventive) strategies against SNHL, including CIHL (9). The pathophysiology of CIHL consists of the formation of

Address correspondence and reprint requests to Charlotte L. Zuur, M.D., Department of Head and Neck Oncology and Surgery, the Netherlands Cancer Institute, Antoni van Leeuwenhoek, Plesmanlaan 121, 1066 CX Amsterdam, the Netherlands; E-mail: c.zuur@nki.nl

C.W.D. and V.A.d.W. contributed equally to this work.

Financial support: The Netherlands Cancer Institute, Antoni van Leeuwenhoek.

The authors disclose no conflicts of interest.

DOI: 10.1097/MAO.0000000000003069

toxic levels of reactive oxygen species (ROS) and the depletion of otoprotective antioxidants (1,4,5,7). Distinct antioxidants protect the cochlea from ototoxic stress, but are incapable of dealing with cisplatin-induced cochlear damage (10). The use of antioxidants that aim to reduce damage caused by ROS may therefore prevent CIHL. Various antioxidants, including sodium thiosulfate (STS) and N-acetylcysteine, have been shown to scavenge ROS and reverse endogenous antioxidant depletion (5,11–13). Furthermore, they inactivate cisplatin by binding to its active form (5,12,13).

Two recent phase III trials on the prevention of CIHL in children showed promising otoprotective effects of the antioxidant sodium thiosulfate (STS) when administered intravenously during cisplatin chemotherapy (14,15). Brock et al. (15) showed that the incidence of CIHL was 48% lower in children treated with cisplatin plus intravenous STS compared with cisplatin alone (relative risk, 52%; 95% confidence interval, 0.33–0.81, $p = 0.002$). Similarly, in the study of Freyer et al. (14) CIHL occurred in 56% of the children treated with cisplatin alone and in 29% when treated with simultaneous STS ($p = 0.00022$). Clinical application of intravenous STS may however be restricted by its side-effects and potential interference with cisplatin's antitumor activity (5,15). Accordingly, Freyer et al. (14) reported lower overall survival in disseminated disease when treated with additional STS (45%) compared with cisplatin alone (84%) ($p = 0.009$). Adverse events have been reported that were likely attributed to intravenous STS administration, including tumor progression, grade 3 infection, neutropenia, electrolyte disturbances, and anemia (14,15).

A topical approach of STS application may be advantageous in preventing CIHL while preserving cisplatin's antineoplastic effect. Several proof-of-principle studies showed that transtympanic application of antioxidants is safe and feasible (11,16,17). Interestingly, in guinea pigs, higher perilymph STS concentrations were achieved after transtympanic application when compared to intravenous infusion (12).

This phase I study evaluated the safety and feasibility and aimed to determine preliminary activity of transtympanic application of STS gel in adults treated with cisplatin dosed $\geq 75\ mg/m^2$ for advanced solid tumors.

METHODS

Study Design

This proof-of-concept phase I trial consists of two cohorts. Cohort A was a single-blind, placebo-controlled study. One ear was treated with STS gel and the other with placebo gel. Cohort B was a nonblinded, non-placebo-controlled study. Here, one ear was treated with STS gel and one was left untreated.

Setting

The study was performed at the Netherlands Cancer Institute in Amsterdam, the Netherlands. The protocol was approved by the institutional medical research ethics committee and registered in the European Clinical Trials Database (EudraCT: 2012-004653-80).

Patients

Patients of 18 years or older who were to be treated with cisplatin at a dose of $\geq 75\ mg/m^2$ for lung or head and neck (HNSCC) cancer were eligible. If patients were to receive concomitant radiotherapy, the maximum cochlear dose was 30 Gray as to avoid radiotherapy-induced hearing loss (2,18,19). Exclusion criteria were symptomatic brain or leptomeningeal metastases and relevant otological history (e.g., conductive hearing loss). All patients gave written informed consent. Patients were considered evaluable after the completion of one cycle of cisplatin including study medication.

Intervention

The ear to be treated with the STS gel was assigned by simple unstratified randomization in both cohorts at the institutional trial center using ALEA Clinical (Forms Vision BV). Patients were enrolled by their treating physician. In cohort A, two syringes with 2.0 ml 0.5% sodium hyaluronate (HYA) based gels were used: one without STS (placebo) and one with 0.1 M STS. In cohort B only the STS gel was prepared. Syringes with study medication were warmed up to 37°C for 30 minutes in an incubator (CULTURA M, Almedica AG) to prevent caloric symptoms during injections. The syringe was connected to the needle (Braun, Pencan 25G) via a 10 cm infusion line (BD Becton Dickenson Connecta). The needle was bended to approach the eardrum perpendicularly under sight.

Topical anesthesia was applied by 3×3 mm gauzes soaked in xylocaine 10% (lidocaine 100 mg/ml, AstraZeneca) applied on the eardrum before the placement of the grommet and injections. In cohort A, the gels were administered through a grommet, which was placed for venting air to prevent barotrauma while injecting. During this procedure there was backflow of gel along the infusion needle into the external ear canal. A different protocol was chosen for cohort B: the grommet was still placed for ventilation, but STS was injected directly through the posterior part of the eardrum (Fig. 1). During administration of the gel the patient was positioned with the upper body 30 degrees upward. After injection the patient's head was turned 45 degrees contra-laterally to allow the gel to reach the round window. Patients remained in this position for 30 minutes and were instructed to keep swallowing and talking to a minimum.

Cisplatin was given 3 hours after STS administration. Concomitant chemotherapy or radiotherapy was administered as per local protocol. Follow-up was performed within 7 days before start of each cisplatin cycle, and within 1 and 3 months after the last cycle. This consisted of audiometry, physical examination, registration of adverse events, and laboratory assessments (hematology, chemistry).

Outcome Measures

We aimed to determine safety and feasibility and the preliminary activity of transtympanic injection of sodium thiosulfate (STS) against CIHL.

Safety and feasibility were evaluated using adverse events registered according to the Common Terminology Criteria for Adverse Events version 4.03 (CTCAE) (20).

To evaluate whether the transtympanically administered STS does interfere with systemically available cisplatin, pharmacokinetic (PK) sampling of cisplatin was performed for comparison with previously published data. Samples were drawn

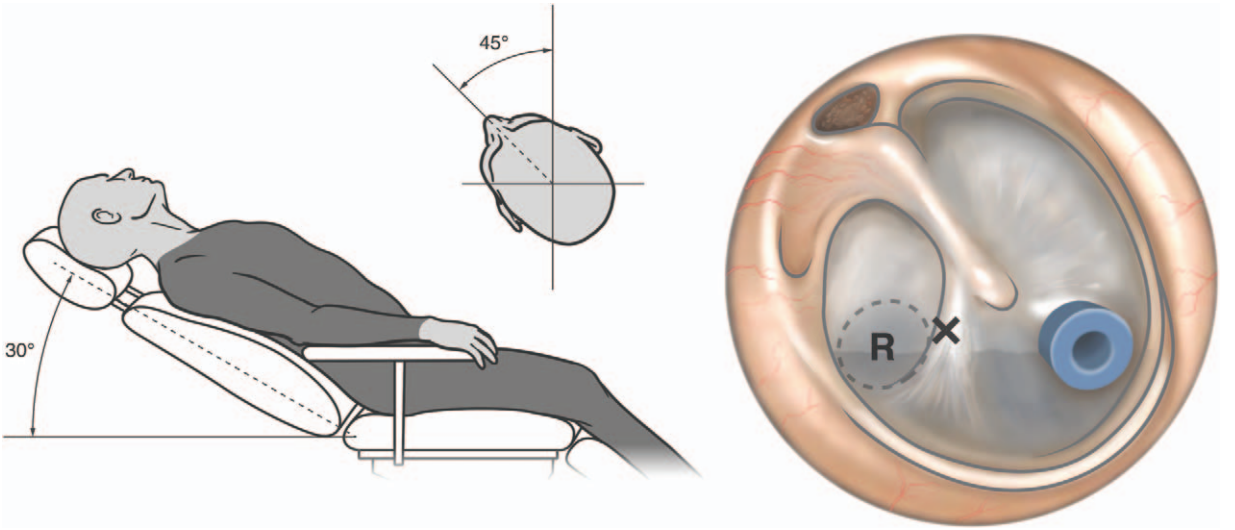

FIG. 1. The position of the patient during transtympanic injection (left) and the eardrum (right). *Right:* The patient is lying with the upper body positioned 30 degrees upward and the patient's head is turned 45 degrees contralaterally. *Left:* The grommet is placed for ventilation of the middle ear and the sodium thiosulfate containing gel is injected via the needle directly through the posterior part of the eardrum (X). The gel fills the middle ear, which enables exposure of the round window (R) to the drug.

predose, at the end of cisplatin infusion and 1, 2, 3, 4, and 18 hours thereafter. Blood was collected in a 10.0 ml heparin tube, which was centrifuged at 1,500 g for 10 minutes at 4°C. Of the plasma 2.0 ml was transferred to a 2.0 ml Eppendorf tube and stored at −20°C (total platinum). A plasma ultra-filtrate tube (Centrifree ultra-filtrate tubes, Merck Millipore Ltd.) was filled with plasma and centrifuged at 1,800 g for 10 minutes. The ultra-filtrate was transferred to a 2.0 ml Eppendorf and stored at −20°C (unbound platinum). Platinum levels were measured using a validated inductively coupled plasma mass spectrometer method (ICP-MS) (21). The lower level of quantification was 7.50 ng/L (21). The maximum observed plasma concentration (C_{max}) and area under the plasma concentration time curve from the start of cisplatin infusion (time = 0) to 22 hours ($AUC_{0\text{-}22h}$) were reported.

Efficacy was assessed using standard audiometry, including air conduction (AC) and bone conduction (BC) thresholds, performed in a sound-proof booth using the Decos Audiology Workstation. If thresholds at 8 kHz were not available for ultrahigh frequency audiometry, we converted them from pure-tone audiometry thresholds into dB SPL following ISO 389-1 (22). If the threshold level was beyond the audiometer's maximum output, we computed the threshold by adding 5 dB to this maximum. Audiometric testing was performed pretreatment (baseline), after each cisplatin cycle (posttreatment) and after 3 months (follow-up). We marked a conductive component if the average AC threshold at 0.5, 1, 2, and 4 kHz was ≥10 dB poorer than the average BC threshold. If there was a conductive component during or after therapy, we took BC thresholds for analysis.

Hearing thresholds of < 10 dB are accepted to indicate (sub)-normal hearing, according to the CTCAE and ASHA guidelines (20,23). Since cisplatin first affects the ultrahigh frequencies, clinically relevant CIHL was defined as ≥ 10 dB SNHL at pure-tone average 8-10-12.5 kHz ($PTA_{8\text{-}12.5}$). Clinically relevant response to STS was defined as a SNHL in the placebo (cohort A) or untreated (cohort B) ear exceeding SNHL in the STS-treated ear by ≥ 10 dB at $PTA_{8\text{-}12.5}$. Next, the patients were divided into three groups: 1) patients without CIHL, 2) patients with CIHL who responded to STS, and 3) patients with CIHL but no response to STS.

Statistics

The low standard errors for audiometric differences before and after cisplatin that were shown in a previous study allowed us to use a small number of patients (24). To study whether there was a significant difference in $\Delta PTA_{8\text{-}12.5}$ between the STS ears and the untreated ears, an exact Wilcoxon signed-rank test for paired samples was used. p values of ≤ 0.05 were considered statistically significant.

RESULTS

Sixteen patients were enrolled. Four patients withdrew consent, of whom three did not start therapy: one due to pain from bone metastases, one considered logistics to be troublesome, and one without a formal reason. Another patient with a narrow ear canal withdrew consent after a painful grommet insertion and was not available for evaluation. Patients were treated between June 2013 and October 2018. For baseline characteristics see Table 1. Eight patients were male (67%). The median age was 60 (range 46–67) years. Cisplatin was discontinued in two patients due to nephrotoxicity.

In total, 34 STS injections were given in 12 patients. In cohort A there was backflow of gel into the external meatus after application through the grommet. An average volume of 0.2 ml (range, 0.1–0.3 ml) was injected. The technique was improved in cohort B: The gel was injected by direct transtympanic puncture of the eardrum. A mean volume of 0.37 ml (range, 0.3–0.5 ml) could be administered.

After the insertion of the grommet and application of the gel(s) temporary, modest adverse events (AEs) were reported. After placement of the grommet and upon request, patients reported modification of sound perception in

TABLE 1. *Baseline characteristics*

	Cohort A (n = 6)	Cohort B (n = 6)	Total (n = 12)
Age median (range), years	60 (46–67)	59 (46–63)	59 (36–67)
Sex			
Male	5 (83%)	3 (50%)	8 (67%)
Female	1 (17%)	3 (50%)	4 (33%)
WHO Performance Score			
0	5 (83%)	6 (100%)	11 (92%)
1	1 (17%)	0	1 (8%)
Tumor type			
NSCLC	3 (50%)	1 (17%)	4 (33%)
Mesothelioma	3 (50%)	0	3 (24%)
Thymus carcinoma	0	1 (17%)	1 (8%)
HNSSC	0	4 (67%)	4 (33%)
Number of cycles cisplatin			
Median (range)	3 (2–4)	3 (1–4)	3 (1–4)
Cisplatin dose			
75 mg/m^2	6 (100%)	2 (33%)	8 (67%)
Number of cycles, median (range)	3 (1–4) cycles	4 (4)	3·5 (1–4)
100 mg/m^2	0	4 (67%)	4 (33%)
Number of cycles, median (range)	–	2 (1–3)	2 (1–3)

HNSSC indicates head and neck squamous cell carcinoma; NSCLC, non-small cell lung cancer; WHO, World Health Organization.

quality (not quantity), which could not be objectified by audiometry. One patient needed to receive subcutaneous local anesthesia with lidocaine 2% before grommet insertion and experienced grade 1 vertigo, which resolved within 4 hours. Only grade 1 AEs were reported for the transtympanic injections, which resolved within few hours. One patient with a narrow ear canal reported grade 1 pain, but continued therapy. Some patients reported fullness of the middle ear after the application of the gel (grade 1), which resolved within 1 hour. No persistent otitis media as a result of gel application occurred. Four patients reported grade 1 tinnitus after therapy (three bilateral and one in the placebo-treated ear). AEs to be attributed to cisplatin doublet treatment or malignancy and unrelated to STS injection, were renal failure, electrolyte disturbances, anorexia, dermatitis, and dysphagia. No grade $\geq$2 neither serious AEs (SAEs) related to STS injections were observed. The six reported SAEs occurred in patients treated with concomitant radiotherapy and 100 mg/m^2 cisplatin and included hospitalization due to renal failure, neutropenia, and dehydration.

For all 12 patients, the mean thresholds for the STS-treated ears and the untreated/placebo ears as measured at baseline, posttreatment and at follow-up are depicted in Figure 2A and Table 2. The mean threshold shift at PTA 8-10-12.5 kHz ($\Delta PTA_{8\text{-}12.5}$) was 12.7 dB in the untreated ears and 8.8 dB in the STS-treated ears ($p = 0.402$). Four patients did not develop CIHL. Their platinum PK curves were comparable to the rest of the group. Eight patients developed CIHL, of whom four (50%) responded to STS. The average difference in $\Delta PTA_{8\text{-}12.5}$ between STS-treated ($\Delta PTA_{8\text{-}12.5} = 14.1$) and untreated ears ($\Delta PTA_{8\text{-}12.5} = 20.2$ dB) in these eight patients was 6.1 dB in favor of the STS-treated ears ($p = 0.141$) (Fig. 2B). Regarding the four responders, the average difference in $\Delta PTA_{8\text{-}12.5}$ between the STS-treated ears ($\Delta PTA_{8\text{-}12.5}$ of 6.8 dB) and untreated ears ($\Delta PTA_{8\text{-}12.5}$ of 25.2 dB) was 18.4 dB ($p = 0.068$) (Fig. 2C).

Four patients did develop CIHL, but did not respond to STS. This group included one patient treated in cohort A who received an estimated volume of 0.1 ml STS gel during all three injections due to backflow of the gel through the grommet into the ear canal. Another nonresponder was unable to stay in the desired position for 30 minutes due to grade 1 vertigo after anesthetics with subcutaneous lidocaine 2% injection. His movements may have troubled absorption of the gel by the round window.

PK parameters C_{max} and $AUC_{0\text{-}22h}$ are shown in Table 2. PK data of one patient treated with 100 mg/m^2 are missing, as PK samples were not taken due to logistic reasons.

DISCUSSION

Application of transtympanic STS was safe and feasible. Four out of eight patients with clinically relevant CIHL showed a relevant interauricular difference in posttreatment hearing capacity. In these four patients, the STS ear benefited compared with the other ear with an average difference in hearing loss of 18.4 dB at $PTA_{8\text{-}12.5}$. Although not significantly different, the results of the above-mentioned four patients rise above random observations in four single patients and suggest the first in-human clinically relevant efficacy of transtympanic application of STS to prevent CIHL in adults (Table 3).

The pathophysiology of CIHL is multifactorial. Cross-linking between platinum and DNA and the accumulation

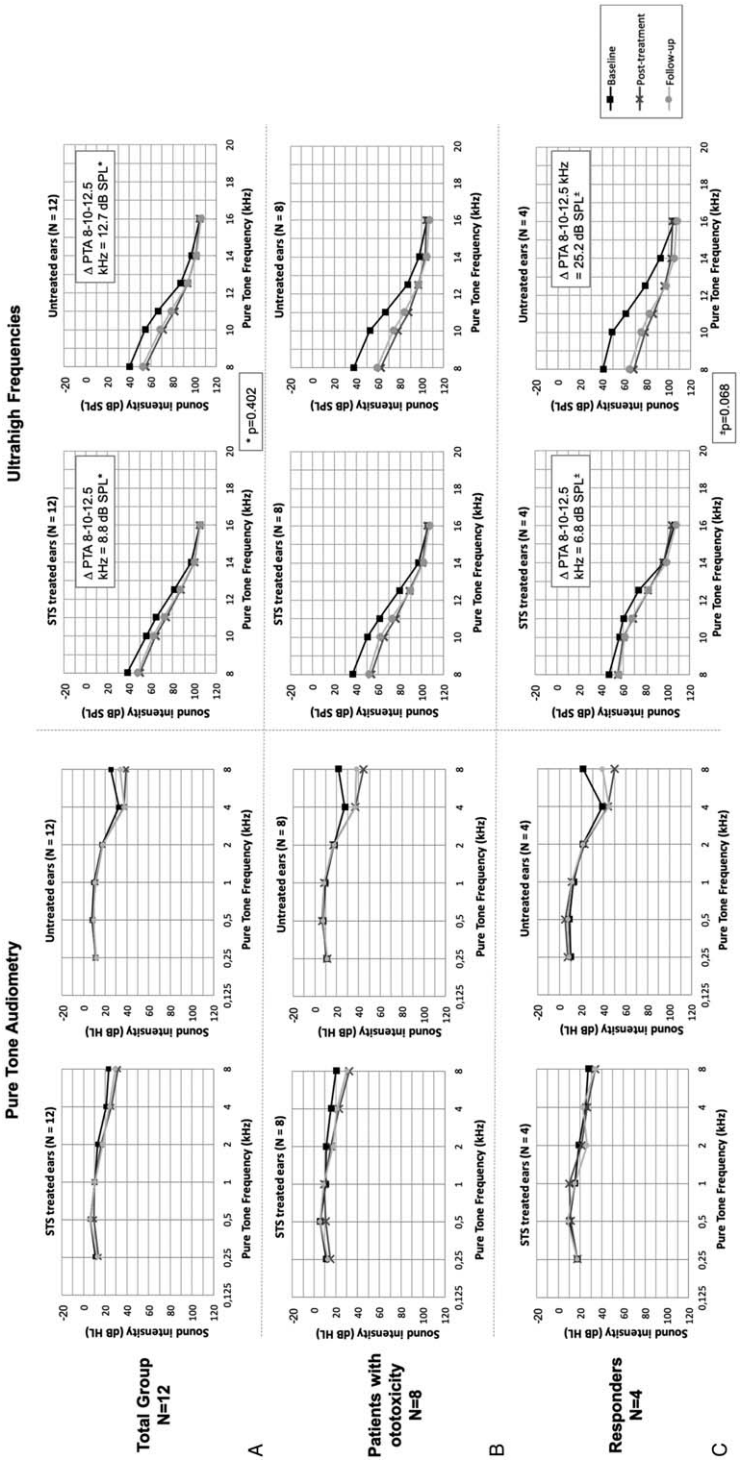

FIG. 2. Audiometric results. The curves show air conduction thresholds at baseline, directly after the last cycle of cisplatin (posttreatment) and at follow-up. *Left:* Pure-tone audiometry, mean of the thresholds in dB hearing level (HL). *Right:* Ultrahigh frequency audiometry, mean of the thresholds in dB sound pressure level (SPL). In some patients the thresholds at 8 kHz were not available for ultrahigh frequency audiometry. These values have been converted from the threshold measured with regular pure-tone audiometry into dB SPL following ISO 389-1. *A*, Audiometric results of the 12 patients in both cohorts A and B. The difference between the posttreatment pure-tone average (PTA) 8-10-12.5 kHz and baseline PTA 8-10-12.5 kHz is 3.9 dB SPL (12.7–8.8 dB, $p = 0.402$, Exact Wilcoxon test for matched pairs). *B*, Results of the eight patients who developed ototoxicity. Ototoxicity is defined as a shift of ≥ 10 dB at Δ PTA 8-10-12.5 kHz (Δ PTA 8-10-12.5 kHz = post-posttreatment PTA 8-10-12.5 kHz minus baseline PTA 8-10-12.5 kHz). *C*, Audiometric results of the four patients who developed ototoxicity and responded to transtympanic sodium thiosulfate (STS) injection. Response is defined as patients with ototoxicity with Δ PTA 8-10-12.5 kHz in the untreated ear exceeding Δ PTA 8-10-12.5 kHz in the STS-ear by ≥ 10 dB. The difference between posttreatment PTA 8-10-12.5 kHz and baseline PTA 8-10-12.5 kHz is 18.4 dB SPL (25.2–6.8 dB, $p = 0.068$, Exact Wilcoxon test for matched pairs).

TABLE 2. *Pharmacokinetic parameters*

		Cisplatin 75 mg/m^2 (n = 8)		Cisplatin 100 mg/m^2 (n = 3)	
		Total Platinum	Unbound Platinum	Total Platinum	Unbound Platinum
C_{max} ug/ml	Mean ± SD (CV)	2.09 ± 0.57 (27.1%)	0.77 ± 0.18 (22.8%)	1.91 ± 0.31 (16.1%)	1.10 ± 0.52 (4.7%)
$AUC_{0\text{-}22h}$ ug*h/ml	Mean ± SD (CV)	29.77 ± 5.90 (19.7%)	3.02 ± 0.59 (19.5%)	22.39 ± 6.64 (28.4%)	4.29 ± 1.85 (43.2%)

C_{max} indicates maximum observed concentration; $AUC_{0\text{-}22}$, area under the plasma concentration time curve from t = 0 to 22 hours; n, number of patients; SD, standard deviation; CV, coefficient of variatiation.

of cisplatin in cochlear structures induce the formation of toxic levels of reactive oxygen species (ROS) (1,4,7,10). Excessive ROS leads to depletion of otoprotective cochlear antioxidants (1,4,7,10). This is followed by apoptosis of HCs and the stria vascularis (1,6,7). Also, cisplatin is responsible for adenine dinucleotide phosphate oxidase 3 (NOX3)-mediated generation of ROS in the organ of Corti and spiral ganglion (1,10). Next, hydroxyl radicals are produced, causing HC damage by destructive calcium influx (1). Cell death may also occur after calcium influx into HCs due to activation of the transient receptor potential vanilloid 1 channel (TRPV1) (1).

Preventive strategies aiming to reduce the production or activity of ROS within the inner ear can be of value to prevent CIHL. Antioxidants with a thiol group, including STS and N-acetylcysteine, have been shown to scavenge ROS and reverse endogenous antioxidant depletion (5,11–13). Furthermore, they inactivate cisplatin by binding to its active form (5,12,13). Preclinical in vivo studies showed that both intravenous and transtympanic administered antioxidants are able to prevent CIHL (25,26).

The current study was a phase I study, designed to assess safety and feasibility. Therefore, the study was not powered aiming to prove efficacy of transtympanic STS against cisplatin-induced hearing loss. Furthermore, ideally a double blinding and placebo-controlled procedure would have been used to assess efficacy. No placebo was used in cohort B as this was found too troublesome and time-consuming. One patient withdrew consent as the insertion of the grommet was too painful and was therefore not included in the analysis. In future studies an intention to treat analyses should be included. Also, a larger phase II trial is needed to adequately prove the efficacy of transtympanic STS against CIHL.

Several other clinical trials assessed transtympanic drugs for the prevention of CIHL. Two studies reported significant hearing preservation by transtympanic application of N-acetylcysteine. Riga et al. (11) showed that the threshold change at 8 kHz was 7 dB greater in patients treated with transtympanic N-acetylcysteine compared to untreated patients ($p = 0.005$). Sarafraz et al. (16) showed significantly better hearing preservation at 4 and 8 kHz when transtympanic N-acetylcysteine was injected compared with transtympanic dexamethasone. As both studies did not perform PK analysis, it remains uncertain whether transtympanic N-acetylcysteine interferes with the systemic exposure to cisplatin.

Rolland et al. (17) also evaluated transtympanic STS injections in 13 patients treated with concomitant radiotherapy and cisplatin for HNSCC. Hearing loss was 1.3 dB less in STS-treated ears compared to untreated ears at frequencies from 3 to 10 kHz. They injected a higher concentration of STS (0.5 M versus 0.1 M) in a smaller volume (0.1 ml versus 0.3–0.5 ml). We think that a larger volume results in improved exposure of the round window to the gel. Also, it seems important to use a high-viscosity gel that does not rapidly flow through the Eustachian tube. The timing of injection differed to ours: Rolland et al. injected mean 20.5 hours before cisplatin infusion, whereas we injected 3 hours before cisplatin infusion. We chose this timing since Berglin et al. (26)

TABLE 3. *The threshold shifts of the pure-tone averages (PTAs) of all 12 patients (up) and the 4 patients that developed ototoxicity and responded to the STS gel (down)*

	STS-Treated Ears	Untreated Ears	p Value[a]
		Total group (n = 12)	
Δ PTA 0.5-1-2 kHz	2.1 dB HL	0.7 dB HL	
Δ PTA 1-2-4 kHz	2.5 dB HL	2.2 dB HL	
Δ PTA 8-10-12.5 kHz	8.8 dB SPL	12.7 dB SPL	0.402
		Responding patients (n = 4)	
Δ PTA 0.5-1-2 kHz	−0.4 dB HL	−1.3 dB HL	
Δ PTA 1-2-4 kHz	−0.4 dB HL	1.7 dB HL	
Δ PTA 8-10-12.5 kHz	6.8 dB SPL	25.2 dB SPL	0.068

Ototoxicity is defined as Δ PTA 8-10-12.5 ≥ 10 dB. Response is defined as patients with ototoxicity in which Δ PTA 8-10-12.5 kHz in the untreated ear exceeds Δ PTA 8-10-12.5 kHz in the STS-ear with ≥ 10 dB. Δ PTA is measured as the PTA directly after the last cycle of cisplatin minus the baseline PTA.

[a]Exact Wilcoxon test for matched pairs.

HL indicates hearing level; kHz, kilohertz; PTA, pure-tone average; SPL, sound pressure level; STS, sodium thiosulfate.

showed impressive HC protection in guinea pigs when injecting 3 hours before cisplatin. Preclinical PK results of transtympanic STS in guinea pigs are inconsistent: Berglin et al. showed stable perilymphatic STS concentrations between 1 and 3 hours after injection, while Schroeder II et al. (12) reported that perilymphatic STS has a short half-life of 44.4 minutes (dose, 250 mg/ml). Furthermore, Viglietta et al. recently published their results of a phase I study evaluating transtympanic application of STS in 42 healthy volunteers. Application of different doses of STS (0.15 M, 0.5 M, 1.0 M, 1.5 M) was safe and feasible (27).

Since cisplatin can be detected in the cochlea immediately after infusion and its elimination rate from the inner ear is slow (28), one may advocate to inject STS directly before cisplatin infusion. However, these preclinical results cannot be translated directly to humans, as the human's otic capsule is thicker and the round window permeability is lower compared to guinea pigs (5,26). We think that adequate timing of STS administration is essential and demands future studying.

Furthermore, both the exposure of the gel to the round window and the uptake of STS in the perilymph may depend on anatomic variations, the patient's position and otologic pathology (e.g., otosclerosis, otitis media). These factors might explain the inter-individual differences in response to transtympanic STS that we found.

One of the strengths of the study is that we performed PK analysis of systemic cisplatin. When considering an average body surface area of 1.8 m^2, patients received about 0.43 mmol cisplatin ($\geq$75 mg/m^2). The amount of STS administered ranged from 0.01 to 0.05 mmol. Since STS binds to platinum in a 1:1 ratio, <10% of the molar weight of cisplatin could be neutralized. However, the low oral bio-availability of STS restricts the amount of STS available in the systemic circulation (29). A comparison of our PK results with literature is difficult, as STS-bound platinum may be detected as unbound platinum by the ICP-MS method (30). A comparison of the unbound fraction is therefore not useful. The levels of unbound platinum were however in line with previously reported results. Interestingly, Viglietta et al. performed PK analysis of systemically available STS and state that the measured plasma STS levels are expected to be too low for interference with the antitumor effect of cisplatin (27). Based on literature and the poor oral bioavailability of STS, we postulate that transtympanic STS does not interfere with the systemically available cisplatin.

CONCLUSION

Transtympanic injection of STS was safe and feasible. In this small population of 12 patients, we were able to show hearing preservation by transtympanic STS in 4 of 8 patients enduring clinically relevant CIHL. Our PK data indicate that transtympanic STS does not interfere with the antineoplastic activity of cisplatin. Future research is needed to confirm the efficacy of transtympanic STS aiming to prevent CIHL. Variables including the optimal dose, viscosity, and timing require further investigation.

Acknowledgments: The authors thank the speech- and language pathologists for contribution in making audiograms and P. M. van Brussel for providing the illustrations shown in Figure 1.

REFERENCES

1. Paken J, Govender CD, Pillay M, Sewram V. Cisplatin-associated ototoxicity: A review for the health professional. *J Toxicol* 2016; 2016:1809394.
2. Schmitt NC, Page BR. Chemoradiation-induced hearing loss remains a major concern for head and neck cancer patients. *Int J Audiol* 2018;57 (sup4):S49–54.
3. Theunissen EA, Bosma SC, Zuur CL, et al. Sensorineural hearing loss in patients with head and neck cancer after chemoradiotherapy and radiotherapy: A systematic review of the literature. *Head Neck* 2015;37:281–92.
4. Trendowski MR, El Charif O, Dinh PC Jr, Travis LB, Dolan ME. Genetic and modifiable risk factors contributing to cisplatin-induced toxicities. *Clin Cancer Res* 2018;25:1147–55.
5. Callejo A, Sedo-Cabezon L, Juan ID, Llorens J. Cisplatin-induced ototoxicity: Effects, mechanisms and protection strategies. *Toxics* 2015;3:268–93.
6. Karasawa T, Steyger PS. An integrated view of cisplatin-induced nephrotoxicity and ototoxicity. *Toxicol Lett* 2015;237:219–27.
7. Lanvers-Kaminsky C, Zehnhoff-Dinnesen AA, Parfitt R, Ciarimboli G. Drug-induced ototoxicity: Mechanisms, pharmacogenetics, and protective strategies. *Clin Pharmacol Ther* 2017; 101: 491–500.
8. Zuur CL, Simis YJ, Lansdaal PE, et al. Ototoxicity in a randomized phase III trial of intra-arterial compared with intravenous cisplatin chemoradiation in patients with locally advanced head and neck cancer. *J Clin Oncol* 2007;25:3759–65.
9. Schilder AGM, Su MP, Blackshaw H, et al. Hearing protection, restoration, and regeneration: An overview of emerging therapeutics for inner ear and central hearing disorders. *Otol Neurotol* 2019;40:559–70.
10. Sheth S, Mukherjea D, Rybak LP, Ramkumar V. Mechanisms of cisplatin-induced ototoxicity and otoprotection. *Front Cell Neurosci* 2017;11:338.
11. Riga MG, Chelis L, Kakolyris S, et al. Transtympanic injections of N-acetylcysteine for the prevention of cisplatin-induced ototoxicity: A feasible method with promising efficacy. *Am J Clin Oncol* 2013;36:1–6.
12. Schroeder RJ 2nd, Audlin J, Luo J, Nicholas BD. Pharmacokinetics of sodium thiosulfate in Guinea pig perilymph following middle ear application. *J Otol* 2018;13:54–8.
13. Rybak LP, Whitworth CA, Mukherjea D, Ramkumar V. Mechanisms of cisplatin-induced ototoxicity and prevention. *Hear Res* 2007;226:157–67.
14. Freyer DR, Chen L, Krailo MD, et al. Effects of sodium thiosulfate versus observation on development of cisplatin-induced hearing loss in children with cancer (ACCL0431): A multicentre, randomised, controlled, open-label, phase 3 trial. *Lancet Oncol* 2017;18:63–74.
15. Brock PR, Maibach R, Childs M, et al. Sodium thiosulfate for protection from cisplatin-induced hearing loss. *N Engl J Med* 2018;378:2376–85.
16. Sarafraz Z, Ahmadi A, Daneshi A. Transtympanic injections of N-acetylcysteine and dexamethasone for prevention of cisplatin-induced ototoxicity: Double blind randomized clinical trial. *Int Tinnitus J* 2018;22:40–5.
17. Rolland V, Meyer F, Guitton MJ, et al. A randomized controlled trial to test the efficacy of trans-tympanic injections of a sodium thiosulfate gel to prevent cisplatin-induced ototoxicity in patients with head and neck cancer. *J Otolaryngol Head Neck Surg* 2019;48:4.
18. Jereczek-Fossa BA, Zarowski A, Milani F, Orecchia R. Radiotherapy-induced ear toxicity. *Cancer Treat Rev* 2003;29:417–30.
19. Landier W. Ototoxicity and cancer therapy. *Cancer* 2016;122: 1647–58.

20. U.S. Department of Health and Human Services, Common terminology criteria for adverse events (CTCAE). Version 4.03. Available at: https://www.eortc.be/services/doc/ctc/CTCAE_4.03_2010-06-14_QuickReference_5x7.pdf; 2010. Accessed May 28, 2009.
21. Brouwers EEM, Tibben MM, Rosing H, Hillebrand MJX, Joerger M, Schellens JHM. Sensitive inductively coupled plasma mass spectrometry assay for the determination of platinum originating from cisplatin, carboplatin, and oxaliplatin in human plasma ultrafiltrate. *J Mass Spectrom* 2006;41:1186–94.
22. ISO 389-1, Acoustics - Reference zero for the calibration of audiometric equipment. 1998.
23. American Speech-Language-Hearing Association. Audiologic management of individuals receiving cochleotoxic drug therapy. *ASHA* 1994;36:11–9.
24. Rademaker-Lakhai JM, Crul M, Zuur CL, Baas P, Beijnen JH, Simis YJW. Relationship between cisplatin administration and the development of ototoxicity. *Clin Oncol* 2006;24:918–24.
25. Van den Berg JH, Beijnen JH, Balm AJM, Schellens JHM. Future opportunities in preventing cisplatin induced ototoxicity. *Cancer Treat Rev* 2006;32:390–7.
26. Berglin CE, Pierre PV, Bramer T, et al. Prevention of cisplatin-induced hearing loss by administration of a thiosulfate-containing gel to the middle ear in a guinea pig model. *Cancer Chemother Pharmacol* 2011;68:1547–56.
27. Viglietta V, Shi F, Hu QY, et al. Phase 1 study to evaluate safety, tolerability and pharmacokinetics of a novel intra-tympanic administered thiosulfate to prevent cisplatin-induced hearing loss in cancer patients. *Invest New Drugs* 2020;38:1463–71.
28. Hellberg V, Wallin I, Ehrsson H, Laurell G. Cochlear pharmacokinetics of cisplatin: An in vivo study in the guinea pig. *Laryngoscope* 2013;123:3172–7.
29. Farese S, Stauffer E, Kalicki R, et al. Sodium thiosulfate pharmacokinetics in hemodialysis patients and healthy volunteers. *Clin J Am Soc Nephrol* 2011;6:1447–55.
30. Brouwers EEM, Huitema ADR, Schellens JHM, Beijnen JH. The effects of sulfur-containing compounds and gemcitabine on the binding of cisplatin to plasma proteins and DNA determined by inductively coupled plasma mass spectrometry and high performance liquid chromatography-inductively coupled plasma mass spectro. *Anticancer Drugs* 2008;19:621–30.