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Relations between cochlear histopathology and hearing loss in experimental cochlear implantation

S.J. O'Leary*, P. Monksfield [1], G. Kel, T. Connolly [2], M.A. Souter [3], A. Chang, P. Marovic [4], J.S. O'Leary, R. Richardson [5], H. Eastwood

Department of Otolaryngology, University of Melbourne, 2nd Floor, Peter Howson Wing, Royal Victorian Eye and Ear Hospital, 32 Gisborne St, East Melbourne, Victoria 3002, Australia

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ABSTRACT

This study reviews the cochlear histology from four hearing preservation cochlear implantation experiments conducted on 73 guinea pigs from our institution, and relates histopathological findings to residual hearing. All guinea pigs had normal hearing prior to surgery and underwent cochlear implantation via a cochleostomy with a silastic-platinum dummy electrode. Pure tone auditory brainstem response (ABR) thresholds from 2 to 32 kHz were recorded prior to surgery, and at one and four weeks post-operatively. The cochleae were then fixed in paraformaldehyde, decalcified, paraffin embedded, and mid-modiolar sections were prepared. The treatment groups were as follows: 1) Systemic dexamethasone, 0.2 mg/kg administered 1 h before implantation, 2) Local dexamethasone, 2% applied topically to the round window for 30 min prior to cochlear implantation, 3) Local n-acetyl cysteine, 200 μg applied topically to the round window for 30 min prior to implantation, 4) inoculation to keyhole-limpet hemocyanin (KLH) prior to implantation, and 5) untreated controls. There was a significant correlation between the extent of the tissue reaction in the cochlea and the presence of foreign body giant cells (FBGCs), new bone formation and injury to the osseous spiral lamina (OSL). The extent of the tissue response, as a percentage of the area of the scala tympani, limited the best hearing that was observed four weeks after cochlear implantation. Poorer hearing at four weeks correlated with a more extensive tissue response, lower outer hair cell (OHC) counts and OSL injury in the basal turn. Progressive hearing loss was also correlated with the extent of tissue response. Hearing at 2 kHz, which corresponds to the region of the second cochlear turn, did not correspond with loco-regional inner hair cell (IHC), OHC or SGC counts. We conclude that cochlear injury is associated with poorer hearing early after implantation. The tissue response is related to indices of cochlear inflammation and injury. An extensive tissue response limits hearing at four weeks, and correlates with progressive hearing loss. These latter effects may be due to inflammation, but would also be consistent with interference of cochlear mechanics.

1. Introduction

Delayed hearing loss following cochlear implantation remains a problem for hearing preservation surgery. Although hearing may be preserved immediately after surgery in a significant proportion of patients undergoing implantation, it has been reported that approximately a third of recipients lose their residual hearing slowly over the next few months (Barbara et al., 2003; Gstoettner

Abbreviations: ABR, auditory brainstem response; CI, cochlear implant; FBGC, foreign body giant cell; IHC, inner hair cell; KLH, keyhole-limpet hemocyanin; NAC, n-acetyl cysteine; OHC, outer hair cell; OSL, osseous spiral lamina; SGC, spiral ganglion cell; ST, scala tympani.

* Corresponding author. Tel.: +61 3 9929 8366; fax: +61 3 9663 1958.
E-mail addresses: sjoleary@unimelb.edu.au (S.J. O'Leary), monksfield@talk21.com (P. Monksfield), gekel@unimelb.edu.au (G. Kel), timothyent@gmail.com (T. Connolly), msouter@me.com (M.A. Souter), achang_tw@yahoo.com (A. Chang), paul.marovic@gmail.com (P. Marovic), j.oleary3@unimelb.edu.au (J.S. O'Leary), rricharson@bionicsinstitute.org (R. Richardson), haydente@unimelb.edu.au (H. Eastwood).
[1] Present address: Queen Elizabeth Medical Centre, Birmingham, West Midlands B15 2TH, United Kingdom.
[2] Permanent address: Geelong Hospital, 72-322 Ryrie Street, Geelong, Victoria, Australia.
[3] Permanent address: Christchurch Hospital, Riccarton Avenue, Private Bag 4710, Christchurch, NZ.
[4] Present address: Southern Health, Monash Medical Centre, Victoria, Australia.
[5] Permanent address: Bionics Institute, 384-388 Albert St, East Melbourne, Victoria, Australia.

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et al., 2006; Woodson et al., 2010). The reasons for this are not well understood, but it has been proposed that the tissue response to implantation may be implicated. Studies of human temporal bones have demonstrated that the electrode is surrounded by a fibrous sheath (Nadol et al., 2001). It has been proposed that this tissue response[6] may adversely affect residual hearing in several ways. The fibrous tissue could dampen cochlear mechanics if it were to involve the basilar membrane at the site of implantation in the basal turn, where it has been predicted that high frequency hearing loss will result (Choi and Oghalai, 2005; Kiefer et al., 2006). Whereas fibrosis occupying a significant proportion of the scala tympani (ST) is predicted to cause a low frequency hearing loss, according to the modelling of Choi and Oghalai (2005). Alternatively, it has been proposed that biological activity within the tissue response may be toxic to the organ of Corti (Eshraghi et al., 2005). The data presented here provide some insights into how residual hearing may change over time after cochlear implantation.

Of the small numbers of human temporal bones available, most involve patients that have had little, if any, residual hearing and have been implanted years prior to their death (Fayad et al., 2009; Nadol and Eddington, 2006; Nadol et al., 2001). Therefore it is unlikely that this material will provide insights into postoperative delayed hearing impairment in the foreseeable future. In contrast, experimental animals have a histopathological response to an implant which resembles that seen in humans, with fibrosis, new bone growth (osteoneogenesis) and the presence of foreign body giant cells (FBGCs) being prominent features (James et al., 2008; Xu et al., 1997). Therefore an experimental approach is more likely to provide insights into the aetiology of delayed hearing loss and its relationship to the intracochlear tissue response from implantation.

In our laboratories, we now have a sizeable cohort of guinea pigs that have undergone hearing preservation cochlear implantation, where the nature and extent of the tissue response present within the basal turn of the cochlea has been assessed. These animals received a cochlear implant (CI) into the basal turn with or without a short application of a hearing preservation drug, such as dexamethasone or n-acetyl cysteine. A robust analysis of cochlear histopathology and its impact upon hearing loss has been precluded due to marked variability in the extent of the tissue response in separate experiments. Now, sufficient numbers of animals have been studied across a range of experiments to minimise the influence of inter-animal variability, so an analysis has been conducted incorporating subjects across study groups.

Here we correlate the histopathological appearance of the cochlea, with changes in auditory brainstem response (ABR) thresholds over time in an attempt to determine whether the cochlear histopathology has influenced these hearing outcomes. In all of these experiments there was an initial elevation in ABR threshold, which was still apparent one week after surgery. Over the next three weeks, ABR thresholds either partially recovered, remained unchanged or worsened slightly. These divergent hearing outcomes were correlated with the histopathological analyses performed on cochlear tissue collected four weeks after implantation. In addition, we examined whether there was a treatment effect upon the tissue response from the drugs applied to these animals.

[6] tissue response – this is the term used to describe the tissue response to cochlear implantation. It is thought to arise from both the response to surgery and also the response to the presence of the implant (i.e. the foreign body reaction). The tissue response was quantified as the percentage of the area of scala tympani (in the lower basal turn) that it occupied.

2. Methods

2.1. Animals

All procedures in this study were approved by the Animal Ethics Committee of the Royal Victorian Eye and Ear Hospital (Projects 05/113A, 06/132A, 07/140A and 07/146A). The guinea pigs were bred in the Biological Resource Centre at the Royal Victorian Eye and Ear Hospital and weighed ≥300 g. The anaesthesia used for all ABR testing was 60 mg/kg ketamine and 4 mg/kg xylazine, intramuscularly. For surgical procedures, after induction with ketamine and xylazine, gaseous anaesthesia was titrated against respiration with 0.5–1% isoflurane delivered in oxygen set at a rate of 500 ml/min.

2.2. Experimental groups

The experimental groups have been described in detail previously (Table 1) (Connolly et al., 2010; Eastwood et al., 2010; James et al., 2008; Maini et al., 2009; Souter et al., 2012). In the topical dexamethasone study (James et al., 2008), a carboxylmethylcellulose hyaluronic acid polymeric pledget (Seprapack™, Genzyme Corporation) was loaded with 5 μl of either 2% dexamethasone phosphate or saline and placed on the round window membrane for 30 min prior to cochlear implantation. In the n-acetyl cysteine (NAC) study (Eastwood et al., 2010) Seprapak pledgets were loaded with 5 μl of either 40 mg/ml n-acteyl cysteine or saline and again applied to the round window membrane for 30 min prior to cochlear implantation. In the systemic dexamethasone study (Connolly et al., 2010), a low dose of dexamethasone (0.2 mg/kg) or saline was injected into the internal jugular vein 60 min prior to cochlear implantation. The experimental groups described above were typified by there having been hearing protection observed at 32 kHz only. In the keyhole limpet hemocyanin (KLH) study (Souter et al., 2012), animals were injected subcutaneously with KLH at a dose of 1 mg a week prior to implantation. KLH-primed animals were then randomised to receive a 5 μl volume of either 20% dexamethasone or normal saline delivered to the round window on a Seprapak pledget for 30 min prior to cochlear implantation. The

Table 1
Summary of treatments applied to the animals included in this analysis. All animals underwent cochlear application. Some were pre-treated with either topical application of drug to the cochlea, via its application to the round window on a carboxymethylcellulose-hyaluronic acid polymer. Others were treated systemically, via intravenous injection. "KLH" animals were inoculated to induce an immune response against keyhole limpert hemocyanin.

Study	Treatment (all received implant)	Numbers	Descriptor
1. Topical dexamethasone (James et al., 2008)	Local application to round window 2% dexamethasone	14	2% dex
2. Topical NAC (Eastwood et al., 2010)	Local application to round window 200 μg n-acetyl cysteine	14	NAC
3. Systemic dexamethasone (Connolly et al., 2010)	Intravenous administration 0.2 mg/kg dexamethasone	6	Low dose
4. KLH/Topical dexamethasone (Souter et al., 2012)	Local application to round window 20% dexamethasone KLH primed	10	KLH steroid
5. KLH control (Souter et al., 2012)	Local application to round window normal saline KLH primed	10	KLH control
6. Controls from non-KLH studies		19	Control
Total		73	