A Comparison of Pure Tone Thresholds and Distortion Product Otoacoustic Emission Measures in Patients with Tuberculosis Receiving Aminoglycosides

A Report on a Research Project Presented to

The Division of Communication Sciences and Disorders

School of Health and Rehabilitation Sciences

Faculty of Health Sciences

University of Cape Town

University of Cape Town

In Fulfillment of the Requirements for the Degree of Masters in Science (Audiology)

By

Lucretia Petersen

August 2005

The copyright of this thesis vests in the author. No quotation from it or information derived from it is to be published without full acknowledgement of the source. The thesis is to be used for private study or non-commercial research purposes only.

Published by the University of Cape Town (UCT) in terms of the non-exclusive license granted to UCT by the author.

University of Cape Town

Abstract

Patients with multidrug resistant tuberculosis (MDR-TB) in South Africa receive ototoxic medication as part of their treatment (Department of Health, 1999). Therefore, it is essential to monitor these patients' hearing with a sensitive test. The study aimed at comparing the relative sensitivity of conventional pure tone audiometry, distortion product-gram (DPgram) absolute amplitudes and distortion product input-output (DP I/O) function thresholds to determine which test detected abnormal cochlear function first. Other aims were to examine the nature of DP I/O function slopes and the stimulus parameters (frequency and intensity) of the DP I/O function test. Twelve participants were included in a repeated measures study design, where each of the abovementioned tests was conducted every two weeks for an eight-week period. Results indicated that the DPgram detected abnormal cochlear function first (z = 2.1; p = .03), as determined statistically by the Wilcoxon signed ranks test. It was found that the DP I/O function slopes of individuals whose hearing remained normal, as well as those who developed hearing loss over time, were predominantly compressive. With DP I/O function thresholds, the high frequency range (3284, 4102 and 4919 Hz) seemed to detect the most ears with abnormal cochlear function. Research and clinical implications, as well as limitations, of the current study are discussed.

University of Cape Town

Acknowledgements

Firstly, I want to thank the Almighty for giving me the opportunity to learn so much about myself through this research process.

I also want to thank the following people who have all played a crucial role in the completion of this project:

- All of the participants without whom this study would have been impossible
- My supervisor, Prof. Shajila Singh, whose guidance, patience, encouragement, and willingness to share her knowledge will always be an inspiration to me
- The statisticians Dr. S. Isaacs and G. Distiller for their input regarding the data analysis
- My colleagues for their ongoing support and interest
- My husband, Gillian, for all the drafts that he had to proof-read and his ongoing love and support throughout this whole process
- My family for their motivation
- My father, for the example that he set in being a true scholar

University of Cape Town

Table of contents

University of Cape Town

University of Cape Town

University of Cape Town

University of Cape Town

List of tables

University of Cape Town

List of figures

University of Cape Town

List of appendices

University of Cape Town

Glossary of terms

Active cochlear process	Caused by the micromechanical feedback loop between the motile outer hair cells and basilar membrane vibration (Dorn et al., 2001). This process is responsible for compressive nonlinearities (Dorn et al., 2001; Durrant & Lovrinic, 1995).
Cochleotoxic	Having a poisonous action on the hair cells of the Organ of Corti (Stach, 2003).
Cochlear amplifier	Collection of processes that cause physical amplification of travelling waves on the basilar membrane (Mills & Rubel, 1996). Active processes in the cochlea responsible for frequency resolution, sensitivity and dynamic range of hearing (Stach, 2003).
Early detection	Prior to the appearance of audiometric hearing loss (Hall, 2000).
Hearing sensitivity	The capacity of the auditory system to detect a stimulus, most often described by audiometric pure tone thresholds (Stach, 2003).
Nephrotoxic	Poisonous to the kidney (Online Merriam-Webster dictionary, http://ww2.merriam-webster.com/cgi-in/mwmednlm?book&va=nephrotoxic)

University of Cape Town

Ototoxic	Having a poisonous action on the ear, particularly the hair cells of the cochlea and vestibular end organs (Stach, 2003). The tendency of certain therapeutic agents and other chemical substances to cause functional impairment and cellular degeneration of the inner ear (Arslan, Orzan & Santarelli, 1999)
Passive cochlear process	Basilar membrane activities that are potentially occurring due to macromechanical properties of the cochlear energy transmission system (Dorn et al., 2001). This process is not well understood (Dorn et al., 2001).
Vestibulotoxic	Having a poisonous action on the hair cells of the cristae and maculae of the vestibular labyrinth (Stach, 2003)

University of Cape Town

Introduction

Tuberculosis (TB) is a major public health problem in South Africa (Zwarenstein, Schoeman, Vundule, Lombard & Tatley, 1998). Until the rise of the human immune-deficiency virus (HIV), TB was the most commonly reported disease in South Africa (Zwarenstein et al.).

A serious complication of the TB problem in South Africa has been the emergence of multi-drug resistant TB (MDR-TB) (Fourie, n.d.). Patients with MDR-TB in South Africa receive ototoxic medication affecting the hair cells in the cochlea, which may eventually lead to permanent hearing loss, and will impact on an individual's ability to communicate. Therefore, it is important to detect hair cell damage early, before noticed by the individual. The main aim of early detection of abnormal cochlear function is to prevent permanent hearing loss and, therefore, minimise / eliminate the impact on quality of life.

University of Cape Town

Various methods of audiological testing are available, which to date, include conventional and high frequency audiometry, and otoacoustic emissions (OAEs). This study set out to investigate the relative sensitivity of conventional pure tone audiometry and distortion product OAE (DPOAE) measures (distortion product-gram [DPgram] and distortion product input/output [DP I/O] function thresholds) to detect abnormal cochlear function early in TB patients who receive ototoxic medication, as well as to investigate the nature of DP input/output (I/O) function slopes during aminoglycoside treatment. Another aim was to identify the DP I/O test parameters that best identify ototoxicity. A

longitudinal study with a repeated measures design was conducted in 12 participants with TB who received ototoxic medication.

University of Cape Town

Literature Review

Worldwide, tuberculosis (TB) is an increasingly serious disorder (Zwarenstein, Schoeman, Vundule, Lombard & Tatley, 1998), especially in less developed countries (Jaramillo, 1999). In 2000, the World Health Organization (WHO) identified South Africa as the country with the ninth highest prevalence of estimated tuberculosis cases in the world (WHO, 2001, in Herselman, 2002). According to the WHO report (2001) the national incidence of TB in South Africa was estimated at 492 per 100 000, with an incidence in the Western Cape of 678 per 100 000, in 2003 (PAWC, 2003).

TB is an infectious disease that mainly affects the lungs (Jaramillo, 1999; Elender, Bentham & Langford, 1998). The three important stages of the disease are infection, breakdown of cells and death (Jaramillo, 1999). The latter occurs in instances where there is a lack of appropriate and/or timely intervention. TB infection is an asymptomatic life-long condition, but around 10% of infected people will develop the disease during their lifetime, at a time when their immune system is not optimal (Jaramillo, 1999).

"TB in South Africa mainly affects the economically active age group, where 86.6% of the TB patients reported in 1999" were in the age group between 20 to 59 years (Kironde, 2000, p.336). This phenomenon has economic implications (Kironde), because if a person is ill, he or she cannot work, and is therefore, during the time of illness, not contributing to the economy of his/her family and country.

University of Cape Town

The epidemic of TB is boosted by the concurrent presence of the human immune-deficiency virus (HIV) infection (Fätkenheuer, Taelman, Lepage, Schwenk & Wenzel, 1999; WHO, 2003). Development of active TB in latently infected persons is amplified by the deterioration of the immune system in the course of HIV infection. According to estimations of the Medical Research Council (MRC), 32.8% of all TB sufferers in 1997 were co-infected with HIV (http://www.sahealthinfo.org/tb/tbburden.htm, 2004/07/16). The presence of HIV increases the susceptibility for TB infection and facilitates the progression of latent TB infection to active disease (Fätkenheuer et al.). With increased likelihood of reinfection, the chances of developing multi-drug resistant TB (MDR-TB) is greater (Herselman, 2002).

MDR-TB is defined as TB that is resistant to isoniazid and rifampicin, with or without resistance to other drugs (Fätkenheuer et al. 1999; Khan et al. 2001). Isoniazid and rifampicin form part of the standard anti-tuberculosis regimen (Zwarenstein et al., 1998), and the removal of these two drugs from TB treatment (because of resistance to them) has serious implications for prognosis, as well as for cost of TB management (Department of Health, 1999). MDR-TB is a man-made problem (Department of Health) and there are a variety of ways in which drug-resistant strains of the bacteria Mycobacterium tuberculosis have evolved (Mukherjee et al. 2004, Weyer, 1997).

University of Cape Town

MDR-TB mainly develops due to over-usage and misuse of antibiotics (Herselman, 2002; Khan, et al., 2001). Non-compliance with treatment also plays a role in the development of acquired resistance to anti-tuberculous drugs (Van Rie et al., 2000). Drug resistant strains of TB in patients without a history of previous treatment also exist, but this prevalence is consistently less than that of acquired resistance (Dye, Espinal, Watt, Mbiaga & William, 2002; Van Rie et al.). The incidence of new MDR-TB cases was estimated to be as high as 2.4% of all cases in 2000 (Dye et al.). According to K. Sheen (personal communication, 27 July 2004) the prevalence of MDR-TB was 301 per 100 000 in the Western Cape in 2003.

Due to increasing bacterial resistance to the standard anti-tuberculosis drugs, medical treatment has to rely on an increasing diversity of drugs, which include aminoglycosides (Frieden, et al., 1993; Sha & Schacht, 1997). The aminoglycoside group of antibiotics, including among others, kanamycin and streptomycin, are known to have accompanying adverse reactions, such as nephrotoxicity and ototoxicity (Frieden et al., 1993; Kucers & Bennett, 1987; Sha & Schacht, 1997). Advantages of the aminoglycosides are their effectiveness against aerobic gram-negative bacilli that cause TB and their low cost (Barclay & Begg, 1994; Bennett, 1996). These advantages, relative to other antibiotics with less toxicity, have lead to the persistent use of aminoglycoside antibiotics in the treatment of MDR-TB, despite their negative side effects (Barclay & Begg, Song, Sha & Schacht, 1998). An increase in the use of aminoglycosides can be expected, due to the fact that HIV has now resulted in a worldwide resurgence of TB (Sha & Schacht).

University of Cape Town

In South Africa, streptomycin and kanamycin form part of the drug regimen administered to MDR-TB patients (Department of Health, 1999). Streptomycin, an aminoglycoside generally restricted to the treatment of tuberculosis (Lerner & Matz, 1979), has been found to be primarily vestibulotoxic, and has a fairly minor cochleotoxic effect (Kucers & Bennett, 1987; Bennett, 1996; Voogt & Schoeman, 1996). Even for patients with normal renal function receiving the usual dosage of about 1 gram per day, there is the possibility of vestibular toxicity and, less frequently, of cochlear or renal toxicity (Barclay & Begg, 1994; Lerner & Matz, 1979; Voogt & Schoeman, 1996). Reported incidence of cochleototoxicity due to streptomycin varies and is generally below 20% (Sha & Schacht, 1997).

Kanamycin, on the other hand, is predominantly cochleotoxic, and destroys outer hair cells in the cochlea (ASHA, 1994; Barclay & Begg, 1994; Voogt & Schoeman, 1996). From this point onwards the term ototoxicity will be used to refer to cochleotoxicity only, for ease of reference. Hotz, Harris and Probst (1994) found an ototoxicity rate of 90% in five humans (9 out of 10 ears) on amikacin treatment (which has been likened to kanamycin by Bennett, 1996) for more than 12 days by measuring transient evoked otoacoustic emissions (TEOAEs) using 4 kHz tone bursts. Moore, Smith and Lietman (1984) reported that 30 out of 135 participants, i.e., 22.3%, who received amikacin, developed ototoxicity, defined as a decrease of 15 dB in hearing threshold over time.

University of Cape Town

Fausti, et al. (1984) reported that 62.5% of their participants demonstrated a deterioration of pure tone thresholds as measured with high frequency audiometry. However, they did not report on the actual aminoglycoside used in their study.

Wide variability exists in the reported incidence of aminoglycoside ototoxicity. According to ASHA (1994) the actual frequency of cochleotoxicity associated with aminoglycosides is unclear "because of inconsistencies in reported data" (p. 83). Rates of ototoxicity caused by aminoglycosides range from 0% (Powell, Thompson, & Luthe, 1983, in ASHA, 1994) to 63% (Tablan, Reyes, Rintelmann & Lerner, 1984, in ASHA, 1994). The variability was ascribed to a lack of standardised guidelines for the monitoring of ototoxicity (Bennett, 1996). Different aminoglycosides differ in terms of their level of ototoxicity, with e.g., kanamycin being more ototoxic than streptomycin (Bennett). Another possible contributor to the variability in rates of ototoxicity due to aminoglycosides could be the use of different tests to detect ototoxicity (ASHA, 1994). Other reasons could be the differences in dosage, courses of administration and individual susceptibility to aminoglycoside ototoxicity (Arslan, Orzan & Santarelli, 1999).

University of Cape Town

In order to fully understand the effect of aminoglycosides on the cochlea, as well as the role of different tests for ototoxicity monitoring, it is essential to briefly discuss the relevant anatomy and physiology of the ear, in particular the cochlea. The inner ear consists of three semicircular canals, the vestibule and the cochlea (Bess & Humes, 1995). The cochlea consists of a bony,

coiled tube that is subdivided into three fluid-filled compartments, namely the scala tympani, scala vestibuli and scala media (Hall, 2000). The first two compartments are filled with perilymph and the latter with endolymph. The inward-outward motion of the stapes conveys the mechanical energy that travels through the middle ear to the scala vestibuli. These pressure vibrations are then transmitted from the base to the apex of the cochlea via the perilymph of the scala vestibuli, through the endolymph of the scala media until it reaches the perilymph of the scala tympani (Durrant & Lovrinic, 1995). "The stimulus-related vibrations in the perilymph deform the flexible basilar membrane with a wavelike motion ..." (Hall, p. 37) called the travelling wave (Durrant & Lovrinic). The Organ of Corti, which rests on the basilar membrane, contains inner (IHCs) and outer hair cells (OHCs), which are responsible for the enhancement of hearing sensitivity and frequency selectivity (Hall). The OHCs have active properties that increase the mechanical energy within the cochlea, thereby increasing the stimulus-specific vibrations of the basilar membrane. The mechanical energy is transmitted to the IHCs and hearing sensitivity and tuning (frequency selectivity) are enhanced (Hall).

Each region of the basilar membrane responds selectively to a narrow range of frequencies (Abdala & Sininger, 1996). The tonotopic organisation entails mapping of the frequency of the sound wave to the place of maximum activity within the basilar membrane (Bess & Humes, 1995). The primary determinant of the tonotopic organisation is the stiffness of the basilar membrane (Durrant & Lovrinic, 1995). The basilar membrane is stiffest and narrowest at the basal

University of Cape Town

area of the cochlea, which vibrates easiest at high frequencies and least stiff in the wider, apical region, which can thus, vibrate more easily at low frequencies (Durrant & Lovrinic). These properties explain why high frequency stimulus frequencies cause maximum displacement at the base of the cochlea, and low frequencies at the apex.

With respect to the inner ear, aminoglycosides may injure either the cochlea or the vestibular system (Barclay & Begg, 1994; Bennett, 1996). The aminoglycosides enter the inner ear fluids via the bloodstream and result in intracellular biochemical and morphological changes of the cochlear OHCs (Barclay & Begg, 1994). Aminoglycosides bind with iron, forming an oxidative compound that contributes to the formation of free radicals, which are involved in tissue damage in the body due to oxidative activities with proteins and other targets (Halliwell & Gutteridge, 1986, in Hall, 2000).

Initial ototoxic effects of aminoglycosides occur in the OHCs and supporting cells at the basal end of the cochlea, where high frequencies are transduced (Barclay & Begg, 1994; Hashino, Tinitan & Salvi, 1995; Henley, Weatherly, Martin & Lonsbury-Martin, 1996). Damage progresses toward the apical portion of the cochlea responsible for the processing of low frequencies with prolonged use of aminoglycosides (Barclay & Begg, 1994; Hall, 2000).

This progression of damage was found animals (Hashino, et al., 1995; Kakigi, Hirakawa, Harel, Mount & Harrison, 1998), as well as in humans (Hotz, Harris & Probst, 1994; Stavroulaki, et al., 2002) receiving aminoglycosides.

University of Cape Town

Therefore, the hair cells responsible for processing high frequencies are affected first. The reasons for this pattern were explored in animal studies.

Wu et al. (2001) found that when all the OHCs of animal cochleas (adult mice and rats) were equally exposed to aminoglycosides (kanamycin), the OHCs in the basal area were affected before the apical ones. The authors speculated that the basal OHCs might have an intrinsic susceptibility to ototoxic damage (Wu et al.). A study by Sha, Taylor, Forge and Schacht (2001) reported similar results with the cochleas of guinea pigs. They found that the survival of basal OHCs was significantly improved by the addition of radical scavengers. This finding led them to speculate that the protection afforded by radical scavengers implies that the accelerated death of basal OHCs is due to free-radical damage. They, therefore, concluded that the basal OHCs seem to be more susceptible to damage by free radicals, but the possible reasons for this phenomenon are still elusive (Sha et al.).

As stated earlier, the OHCs responsible for analysing high frequencies are damaged first when exposed to aminoglycosides (Henley et al., 1996). Damage to hair cells diminishes the cochlea's ability to perform a frequency analysis on incoming sounds and reduces the sensory response for sounds of low and moderate intensity (Bess & Humes, 1995). These ototoxic effects in the cochlea might remain undetected, and might not immediately result in a noticeable hearing loss (Fausti et al, 1984).

University of Cape Town

Changes in cochlear function due to OHC destruction are most likely irreversible since it has not been firmly established that the cochlear hair cells are capable of regeneration (Konrad-Martin et al., 2005; Probst, Harris & Hauser, 1993). Research studies have provided evidence of some extent of OHC recovery after exposure to ototoxic agents has been terminated in animals (Nicol, Hackney, Evans & Pratt, 1992) and in humans (Dreschler et al., 1985; Fausti et al., 1984; Voogt, 1987), but not to the same levels of functioning as before treatment started. Some researchers have found that ototoxicity is reversible in more than half of all cases, when the ototoxicity is detected early enough (before permanent, i.e., irreversible damage to the cochlea has occurred) and appropriate steps are taken to reduce or terminate medication (Barclay & Begg, 1994; Fee, 1980, in Voogt & Schoeman, 1996).

Hashino et al. (1995) and Marean, Cunningham, Burt, Beecher and Rubel (1995) stated that, in avian ears, damaged hair cells are replaced by new hair cells, but in mammals this hair cell regeneration fails to occur in vivo. It is speculated that, at best, the human cochlear system has only a limited capacity for recovering from severe hair cell damage (Probst et al.). If cochlear damage is not detected in a timely fashion, cumulative and combined effects of harmful influences causing severe damage to the cochlear hair cells (outer and inner) can eventually lead to a permanent loss of hearing (Probst et al.). Detection of communication deficits by individuals probably signifies a marked hearing loss for frequencies between 250 to 8000 Hz, the frequencies generally considered to be essential for perception of speech (Katz, 2002).

University of Cape Town

Risk factors for developing ototoxic hearing loss include genetic susceptibility, poor nutrition, excessive noise exposure, advanced age, pre-existing hearing loss and vestibular dysfunction (Barclay & Begg, 1994; Sha & Schacht, 1997). Previous aminoglycoside treatment is also an established risk factor for ototoxicity (Barclay & Begg, 1994; Lerner & Matz, 1979). Ototoxic damage also depends on several other factors such as dosage in relation to weight, and prolonged exposure to ototoxic medication (Lopez-Gonzalez, Delgado & Lucas, 1999). Concomitant treatment with ethacrynic acid or other nephrotoxic drugs, as well as renal impairment, also increase the likelihood of ototoxicity, as impaired renal function may lead to an accumulation of the aminoglycoside in the body (Barclay & Begg, 1994; Lerner & Matz, 1979). Aminoglycoside-induced nephrotoxicity is correlated with the duration of therapy and serum concentrations in the blood (Barclay & Begg, 1994).

Due to inter-participant variability, even when all the risk factors for a given patient are known, the degree of ototoxicity cannot be fully predicted (Campbell & Durrant, 1993), as is the case with aminoglycoside-induced nephrotoxicity. Aminoglycosides clear slowly from inner ear fluids, and according to Roland (2004) and Wang et al. (1999) aminoglycosides can remain detectable in cochlear hair cells from three months to one year after administration of the drug. Therefore, damage to the auditory system can continue even though drug administration has ceased (ASHA, 1994; Hall, 2000). This delay in onset of ototoxic damage was demonstrated in an animal study where Song et al. (1998) reported that progressive ABR threshold shifts occurred in guinea pigs, which began after the cessation of kanamycin

University of Cape Town

treatment. A study by Dulon (1993, in Hashino et al., 1995) similarly showed that the clearance of aminoglycosides from the mammalian inner ear tissue was slow with an estimated half-life of five to six months after gentamicin treatment was stopped.

Therefore, careful monitoring of hearing and cochlear status is important for purposes of hearing conservation during and after aminoglycoside treatment (Lerner & Matz, 1979; ASHA, 1994). The only way to ensure that ototoxic damage is not missed is to provide hearing testing for all individuals who are treated with aminoglycosides. ASHA (1994) recommends follow-up testing for at least six months after the cessation of aminoglycoside treatment in order to assess possible long-term residual effects.

The number of audiological methods available for ototoxicity monitoring is increasing, but the relative sensitivity and specificity of each test for this specific purpose is not well established (Campbell & Durrant, 1993). An auditory monitoring procedure that permits early identification of OHC damage due to ototoxicity, prior to participative detection by the patient, could potentially prevent permanent hearing loss (Probst, et al., 1993). However, hearing conservation is not always possible, e.g., when the cessation of aminoglycoside treatment could be life threatening or when finances are not available to change treatment to a more expensive, but less ototoxic drug. In these instances a sensitive test of OHC damage will allow more time for counselling and management in preparation of an eventual hearing loss than a test that identifies the damage after the individual has started noticing

University of Cape Town

changes in hearing (Campbell & Durrant). In the case of MDR-TB patients, preservation of life presumably supersedes the preservation of hearing, and counselling prior to the development of a hearing loss might aid in the acceptance process of the individual and his/her significant others, and prepare the person with strategies to maximise communication.

In order to explore tests that detect hearing loss due aminoglycoside exposure early, it is necessary to discuss sensitivity and specificity. Sensitivity refers to the true positive rate, i.e., the percentage of positive results of all true positive results, which is identified by a measure (Katzenellenbogen, Joubert & Karim, 1997), e.g., tests of cochlear function. In other words, sensitivity refers to the ability of a test to detect the disorder that it was designed to detect, expressed as the percentage of positive results in patients with the disorder (Stach, 2003). Specificity refers to the true negative rate, i.e., the percentage of negative test results of all true negatives (Katzenellenbogen et al.). Therefore, specificity reflects a test's ability to differentiate between a normal condition and "the disorder that the test was designed to detect" (Stach, p. 246). Test performance for ototoxicity monitoring can be determined by examining the sensitivity and specificity obtained using a particular criterion threshold shift or deterioration in cochlear function to identify ototoxic hearing loss (Konrad-Martin et al., 2005). The percentage of instances when individuals that exhibit change in cochlear function are identified as showing change, using a specified criterion shift, is a measure of that test's sensitivity. Specificity or the correct rejection rate refers to the percentage of times individuals with stable or unchanging cochlear function are correctly labelled using the criterion shift.

University of Cape Town

Sensitivity and specificity have related diagnostic errors. Failure to correctly identify change in cochlear function results in a misdiagnosis; diagnosing the presence of cochlear abnormality when cochlear function is unaltered results in a false positive. The likelihood of making diagnostic errors in ototoxicity monitoring depends on how a criterion threshold shift relates to normal test-retest variability intrinsic to serial testing (Konrad-Martin et al., 2005). A statistical method for examining test performance involves the construction of receiver-operator characteristic (ROC) curves, in which hit rates for a range of criterion threshold shifts or shifts in cochlear function can be plotted as a function of the corresponding false alarm rates (Konrad-Martin et al.).

In order to monitor ototoxicity it is important that other possible causes of decreased auditory function, e.g., middle ear problems, be ruled out (Konrad-Martin et al., 2005). Due to the fact that ototoxicity affects the cochlea, the clinician must ensure that the hearing loss is sensorineural in nature. As sensitivity and specificity of the various tests are not well established, a test battery approach is essential (ASHA, 1994).

In compiling a test battery for ototoxicity monitoring, apart from the relative sensitivity and specificity of the tests, costs of procedures should be considered, as well as the cumulative cost of multiple procedures and multiple test sessions. Individuals receiving ototoxic medication are frequently seriously ill, as is often the case with individuals with MDR-TB, thus, extensive testing might be too stressful (Campbell & Durrant, 1993). Therefore, one

University of Cape Town

needs to consider the most humane method(s). It is important that the selected test(s) yield the maximum useful data with the least cost and stress to the patient. The cost of the equipment, ease of use and applicability in the clinical environment are also factors that must be taken into account when compiling a test battery for ototoxicity monitoring. In addition, South Africa's developing nation context must also be considered, where resources for hearing testing are considered a luxury relative to other more immediate needs or healthcare costs. Factors like the length of the test, the level of participation required from the individual need to be considered. It is also important that the test(s) detect OHC damage early (Probst, et al., 1993), before the individual detects the damage, i.e., by experiencing hearing loss due to speech perception being affected.

University of Cape Town

Hearing loss can affect an individual's quality of life. In the United States of America (USA), a survey was conducted to determine the effect of communication disorders on the US economy, as well as the cost of lost or degraded employment for people with such disorders (Ruben, 2000). It was found that 36% of individuals with hearing difficulty were unemployed in comparison to an unemployment rate of 25% for individuals with no disability (Ruben). According to Ruben, communication disorders will become a major public health concern in the 21st century, because untreated communication disorders will adversely affect the economic well-being of a society that functions primarily on communication. Over time, a shift from manual labour to communication-based employment was reported in the USA, and will continue

during the 21st century (Ruben) and one can argue that a similar trend may develop in South Africa.

Miyakita, Ueda, Zusho and Kudoh (2002) found that a relationship exists between hearing loss and determinants of quality of life. Their study included 210 participants who had to complete a scale on hearing disabilities and handicap, and answer questions regarding quality of life. The results indicated that, regardless of the aetiology and type of hearing loss, hearing-impaired individuals are likely to suffer from anxiety, lack of self-confidence, depression and/or social isolation (Miyakita et al.). Similarly, Cohen, Labadie, Dietrich and Haynes (2004) found that hearing impairment impacted negatively on social function, emotional state and communication. Their results also indicated that individuals with more significant hearing loss experience greater impairment in their quality of life (Cohen et al.). According to Mulrow, et al. (1990) the ability to communicate is of vital importance to quality of life, especially for social interaction.

University of Cape Town

Herbst (1983, in Cohen, Labadie, Dietrich & Labadie, 2004) found that the hearing impaired adults with untreated hearing loss were more likely to suffer from sadness, depression, anxiety, social isolation and insecurity than those whose hearing loss has been treated. This author concluded that management of hearing loss was directly associated with quality of life. Therefore, even in the case of individuals where permanent hearing loss due to aminoglycoside exposure cannot be avoided, early detection of cochlear damage is important to enhance quality of life with timely intervention.

Ototoxic hearing loss may go undetected by the individual until a communication problem develops, signifying a hearing loss within the frequency range important for speech perception (Fausti et al., 1984; Katz, 2002; Konrad-Martin et al., 2005). Due to the fact that symptoms of ototoxicity are poorly correlated with variables like drug dosage and peak serum levels, early detection of ototoxicity is only possible by monitoring auditory function directly (Konrad-Martin et al.). Early identification of ototoxic damage can improve treatment outcome by preventing or minimising hearing loss (Konrad-Martin et al.).

Various protocols for monitoring ototoxicity can be found in the literature (American Speech-Language-Hearing Association (ASHA), 1994; Fausti et al., 1984; Lerner & Matz, 1979). ASHA, Fausti et al. and Lerner and Matz recommend conventional audiometric testing between 250 to 8000 Hz to monitor ototoxicity. ASHA recommends that frequencies above 8000 Hz should also be measured, to allow for detection of hearing loss in this frequency region. High frequency pure tone audiometry (frequencies >8000 Hz) presents challenges in the South African context, though, and these challenges will be discussed later. According to ASHA, and Lerner and Matz, a baseline should be determined before, or within the first three days of the treatment course. Fausti et al. (1984) recommended that a baseline must be obtained before, or within the first 48 hours after initiation of treatment. No clear rationale for why monitoring has to be initiated within the stated number of hours after the start of ototoxic drug administration is provided in any of the

University of Cape Town

abovementioned literature. Commencement of ototoxicity monitoring should be related to when a drug begins to have an impact on cochlear function.

Research in animals indicated that the occurrence of ototoxicity due to kanamycin treatment in an animal study (rats) in male rats was seen as early as Day 10 of Treatment, and at Day 22 in female rats, as measured by distortion product otoacoustic emission thresholds and absolute amplitudes (Mills, Loos & Henley, 1999). Ototoxic damage in all ears was noted first in the higher frequencies. Hashino, Tinitan and Salvi (1995) used scanning electron microscopy to detect OHC loss in chicks after six days of kanamycin treatment. Similar studies on humans are needed with different aminoglycosides to accurately determine when cochlear damage occurs.

University of Cape Town

Suggested hearing test intervals during aminoglycoside treatment vary from two to three days (ASHA, 1994; Fausti et al., 1984) to once a week (Lerner and Matz, 1979). Fausti et al.'s protocol suggests that the testing interval should be increased to once weekly, then twice monthly, thereafter once monthly after the initial administration period. They do not specify when one must change from one test interval to the next, though. If any hearing changes are detected the schedule should be reverted to the initial test interval of 2 to 3 days (ASHA; Fausti et al).

Post-treatment audiological evaluations are recommended on completion of treatment and 4 to 6 weeks after treatment Fausti et al. (1984). ASHA (1994) recommends that conventional pure tone audiometry should be done as soon

as possible after treatment has been discontinued and at approximately 3- and 6-months intervals after the cessation of aminoglycoside treatment to assess possible long-term residual effects. As is noted in the above discussion, no one agreed protocol for ototoxic monitoring currently exists.

Apart from the fact that a universal protocol for ototoxicity monitoring does not exist (Vasquez & Mattucci, 2003), a standard definition of what constitutes change in hearing threshold due to ototoxicity is currently lacking. The minimum criterion for detecting hearing sensitivity change due to ototoxicity recommended by Fausti et al. (1984) is a decrease in any audiometric threshold of 10 dB HL. ASHA (1994), however, proposed three criteria in ototoxicity monitoring to indicate what constitutes a hearing loss as determined with pure tone audiometry: "a) 20 dB HL decrease at any one test frequency, or b) 10 dB HL decrease at any two adjacent test frequencies, or c) loss of response at three consecutive test frequencies where thresholds were previously obtained (specifically the highest frequencies tested)" (p. II-54). It is important that the detected change is confirmed by repeat testing (ASHA, 1994). The criteria for a clinically significant hearing change suggested by ASHA (1994) are based on results of large clinical research studies, reported normal test-retest variability in healthy subjects not receiving ototoxic drugs, and to a limited extent on ROC curves constructed for pure tone threshold shift data obtained in drug- or noise-exposed individuals (Konrad-Martin et al., 2005). However, the data obtained from drug-exposed individuals are limited (Konrad et al.). Similar evidence is not available for the criteria suggested by Fausti et al.

University of Cape Town

Although the ASHA guidelines have been implemented in many clinical settings, these criteria are not standard (Konrad-Martin et al., 2005). In order to standardise criteria for hearing loss due to exposure to ototoxic drugs that are universally accepted, statistical methods will need to be employed to compare pure tone thresholds of patients receiving ototoxic drugs with that of control patients who receive non-ototoxic medication (Konrad-Martin et al.). In order to achieve this, a large-scale longitudinal study is necessary that could be costly, time-consuming and logistically difficult. This could be a reason why standardised criteria for ototoxic hearing loss do not currently exist.

The following discussion will concentrate on tools that can be used to measure ototoxicity. Until recently, ototoxicity could only be monitored by conventional pure tone audiometry, between the frequencies of 250 and 8000 Hz. Conventional pure tone testing's popularity was based on the relative ease of the test, the relatively low cost of equipment and the fact that the equipment and norms are readily available (ASHA, 1994).

In determining pure tone thresholds, the audiologist relies on the behavioural responses of the patient, which may not always be accurate (Gelfand, 1997). Individuals receiving ototoxic medication are often too sick to concentrate for the required period of time (approximately ten minutes), and could, therefore, yield unreliable behavioural responses (Konrad-Martin et al., 2005). Pure tone testing with sick individuals can be a time-consuming procedure (Vaughan et al., 2002) because of the unreliable behavioural responses (Vasquez &

University of Cape Town

Mattucci, 2003). This test does not include all the frequencies that the cochlea can process, but only those frequencies important for speech perception (Katz, 2002). For efficient ototoxicity monitoring, it is important to detect OHC damage before the range of speech frequencies is affected, therefore, before the individual's speech perception is affected (Fausti, et al., 1984; Konrad-Martin et al.).

Generally, detection of a change in cochlear function with the use of conventional audiometry only occurs when irreparable damage to the OHCs responsible for processing the high frequencies has occurred (Vasquez & Mattucci, 2003; Voogt & Schoeman, 1996), because it does not include the ultra-high frequencies above 8000Hz (Voogt, 1987). As mentioned, ototoxic substances first cause a decline in the high frequency hearing range above 10kHz (Barclay & Begg, 1994; Dreisbach & Siegel, 2001). By the time that cochlear damage is visible on a conventional pure tone audiogram, valuable time for preventative measures has been lost (Stavroulaki, et al., 1999). Permanent cochlear damage may be prevented with regular monitoring of high frequency hearing sensitivity (i.e., hearing >8000Hz) (Voogt & Schoeman).

In animal studies, varying the frequency stimulus parameter seems to be a contributing factor to the sensitivity of detecting the presence of ototoxicity. Osako et al. (1979, in Aran, Erre, Da Costa, Debbarh, & Dulon, 1999) found that 15% of rats exhibited ototoxicity due to treatment with kanamycin, as measured with auditory brainstem response recordings (ABR) using an 8 kHz

University of Cape Town

tone burst. The presence of ototoxic damage was noted if ABR thresholds were significantly elevated after exposure to kanamycin in comparison to the baseline measurement. By using a 16 kHz tone burst, 55% of rats displayed ototoxicity (Osaka et al., 1979).

High frequency audiometry (HFA) constitutes the means of evaluating cochlear functioning between 8 and 20 kHz by obtaining behavioural auditory thresholds of pure tone frequencies (ASHA, 1994).

Dreschler et al. (1985) reported that more participants were identified with hearing loss due to ototoxic damage with HFA (68%) than with conventional pure tone audiometry (44%) in their study. Fausti, Frey, Henry, Olson & Schaffer (1993) found similar results in the 131 ears they studied, on which they reported 62.5% of ears having ototoxic damage as detected by high frequency audiometry. Voogt and Schoeman (1996) found no abnormal cochlear function (thus, hearing of all 92 participants were normal) when conventional pure tone audiometry was used to monitor the hearing of participants with TB who were exposed to kanamycin or streptomycin. HFA, however, revealed a significant difference between their participants' baseline and final audiograms at the highest frequencies where thresholds could be obtained. However, the actual difference in dB was not provided.

It is evident from the preceding discussion that HFA has been established as a more sensitive method for early detection of cochlear damage due to ototoxicity than conventional pure tone audiometry (Dreschler et al., 1985;

University of Cape Town

Tonndorf & Kurman, 1984, Voogt & Schoeman, 1996), because it detects a higher numbers of ears with abnormal cochlear function. HFA also detects cochlear damage earlier than pure tone audiometry (Fausti et al., 1984; Voogt & Schoeman). The ASHA guidelines (1994) for ototoxicity monitoring emphasize the increased test sensitivity achieved by using high-frequency audiometry monitoring to detect ototoxicity. Test-retest differences for high-frequency thresholds using modern equipment are generally reported to be within 10 dB for frequencies between 8 kHz and 14 kHz (Konrad-Martin et al., 2005). Therefore, HFA could potentially assist in preventing or minimising irreversible cochlear damage in patients receiving aminoglycosides. If ototoxicity could be detected before it progresses to involve the frequencies essential for communication, it might be possible for the medical practitioner to lower the dose or change to another antibiotic to prevent a permanent hearing loss that might impact on an individual's communication ability (Vasquez & Mattucci, 2003; Voogt & Schoeman).

HFA is not without disadvantages. A problem with HFA is that special audiometers and transducers are needed that are capable of producing high frequency stimuli with the necessary amplitude and fidelity, and still maintaining a reasonably flat frequency response curve (Voogt, 1987). The equipment is more expensive than standard audiometers and transducers, which is challenging in a country like South Africa where healthcare resources are limited. Another disadvantage is that high frequency audiometry cannot be used to the exclusion of conventional pure tone audiometry, because normal or unchanged high frequencies do not guarantee unchanged thresholds in the

University of Cape Town

conventional frequency range (< 8000 Hz) (Fausti et al., 1984). In some patients, the ototoxic hearing loss starts in the mid-frequency range (< 8000 Hz), or the hearing loss could show asymmetry in the conventional frequency range, which would pose a problem if only high frequency audiometry was employed in ototoxicity monitoring. This procedure with additional frequencies is therefore, more time-consuming than conventional pure tone audiometry alone. The long test-time poses a problem in very ill populations, like patients with MDR-TB, where attention span is not optimal (Vasquez & Mattucci, 2003). Because high frequency testing is a behavioural measure, one has to rely on the active participation of the testee, which could be compromised in individuals who are ill. HFA might be limited in managing the elderly, because a portion of the geriatric population might not have measurable hearing thresholds beyond 8 kHz to allow for high frequency monitoring, due to presbyacusis (Campbell & Durrant, 1993). Test-retest variability of high-frequency audiometry is generally poorer in young children when compared to healthy older children and adults (Konrad-Martin et al., 2005).

University of Cape Town

Apart from the abovementioned limitations, there is no set HFA test protocol, or universally accepted normative data, which also pose difficulties in the application of HFA (Voogt, 1987). Due to differences in calibration techniques, equipment and test procedures, comparisons between different studies cannot be easily drawn (Voogt & Schoeman, 1996). Comparisons between participants are complicated by individual patterns of standing waves in the ear canal (Dreschler et al., 1985). Thus, the need for objective, reliable

measures that require minimal participation of the individual being tested is evident in a population where active participation cannot always be ensured.

Otoacoustic emissions (OAEs) constitute the only non-invasive means of objective cochlear investigation to date (Stavroulaki et al., 1999). OAEs are sounds that originate due to activity of the outer hair cells (OHCs) in the cochlea (Brownell, 1990). These sounds travel from their point of generation in the cochlea, through the middle ear and into the ear canal, where they are measured (Abdala & Sininger, 1996; Berlin, 1998; Hall, 2000; Kemp, 1978; Kon, Inagaki & Kaga, 2000). OAEs can emerge spontaneously, but more commonly they are evoked by acoustic stimulation. Evoked OAEs for example, transient evoked OAEs (TEOAEs) and distortion product OAEs (DPOAEs), are low level sounds emitted from the cochlea as a result of acoustic stimulation (Berlin, 1998; Hall, 2000; Robinette & Glattke, 2002). TEOAEs and DPOAEs are the evoked OAEs that are most commonly used in the clinical setting (Hall, 2000; Satoh, Kanzaki, O-Uchi & Yoshihara, 1998).

The presence of evoked OAEs is associated with normal, or near normal, mechanically active functioning of the OHCs (Kemp, 1978; Robinette & Glattke, 2002), and can only be measured reliably with normal middle ear functioning (Hall, 2000). An animal study (Trautwein, Hofstetter, Wang, Salvi & Nostrant, 1996) has provided proof that IHC loss doesn't alter DPOAEs. In this study, eight adult chinchillas were injected with carboplatin, an ototoxic drug, which resulted in IHC loss while leaving the OHCs intact. Pre- and post-carboplatin DPOAE measurements were both within normal limits, indicating

University of Cape Town

that the generation of these OAEs only depend on OHC function, regardless of whether the IHCs are intact or not (Trautwein et al.).

According to Stavroulaki, et al. (1999) histopathological studies have shown that the OHCs are the cochlear components most susceptible to injury from ototoxic drugs like aminoglycosides. Therefore, the evoked OAE tests seem to be appropriate tools in the monitoring of ototoxic effects on the cochlea. TEOAEs and DPOAEs are evoked in different ways, but essentially yield the same information regarding cochlear function (Hall, 2000).

TEOAEs are elicited by brief broadband stimuli, most frequently clicks, which simultaneously stimulate the cochlea across a wide frequency region (Hall, 2000; Robinette & Glattke, 2002). According to Hall, normal-amplitude TEOAEs up to approximately 5000 Hz should be recordable in the normal ear. Due to the nature of the eliciting stimuli, TEOAEs represent an averaged overview of cochlear activity across the speech frequency range between 1 to 3 kHz (Tlumak & Kilney, 2001).

DPOAEs, on the other hand, are defined as acoustic energy in the ear canal arising from the non-linear interaction of two simultaneously applied pure tones, of frequency f_1[a] and f_2[b], within the cochlea (Zhao & Stephens, 1999). These frequencies, f_1 and f_2, are known as primaries, with intensities, L_1[c] and

[a] f_1 refers to the lower frequency of the pair of eliciting stimuli
[b] f_2 refers to the higher frequency primary
[c] L_1: intensity of f_1

University of Cape Town

L_2[d], respectively. Several combination tones may be predicted mathematically with such stimulation, but the tone at the frequency $2f_1$-f_2[e] is the most robust and the easiest to detect in human ears (Beattie & Bleech, 2000; Harris, 1990). The generation of $2f_1$-f_2 depends upon active non-linear processes within the cochlea at the frequency place on the basilar membrane where f_1 and f_2 interact (Hall, 2000). When this cochlear region is altered, then DPOAEs are either reduced in amplitude or eliminated (Hall, 2000; Lonsbury-Martin, Martin, Probst & Coats, 1987).

The changes in DPOAE-amplitude are specific to the frequencies affected by the damage, and the amplitude of DPOAEs generated in other frequency regions remains unchanged (Harris, 1990). Due to this frequency specificity and sensitivity to changes within the cochlea, Harris proposed that the measurement of DPOAEs could be used as a means of evaluating cochlear function objectively in humans.

Research suggests that DPOAEs are more frequency-specific than TEOAEs for determining minor cochlear dysfunction (Probst, Harris & Hauser, 1993; Stavroulaki, et al., 2002; Suckfüll, Schneeweiss, Dreher & Schorn, 1996) because TEOAEs represent the activation of broad areas of hair cells responsible for processing a wide frequency region, whereas DPOAEs represent the stimulation of more frequency-specific, restricted regions of hair cells (Tlumak & Kilney, 2001). Franklin, McCoy, Martin and Lonsbury-Martin

[d] L_2: intensity of f_2
[e] An equation that reflects the place on the basilar membrane where the DPOAE is expressed.

University of Cape Town

(1992) evaluated test-retest reliability of evoked OAEs in 12 participants. They found that DPOAEs could be recorded more reliably at high frequencies above 3000 Hz than TEOAEs (Franklin et al., 1992).

Currently two methods of DPOAE-measurement are used in clinical applications (Ozturan & Lam, 1998; Robinette & Glattke, 2002). One method is where the intensity levels of the primary tones are kept constant, and DPOAE data are recorded for different frequency regions. The resultant representation of amplitude recorded across the frequency range is called a DPgram (Hall, 2000). With a second method, the DP input/output (I/O) function or growth function, the frequencies of the primary stimuli are kept constant and the intensity is increased/decreased systematically in regular increments (Hall). Due to the fact that the cochlear amplifier functions in an intensity-level dependent fashion, DPOAEs recorded as a function of the stimulus level (i.e., DPOAE I/O function) may provide important information about the range and operational characteristics of the cochlear amplifier (Abdala, 2000). However, the specifics of the nature of the information that could be derived have not been provided.

With appropriate stimulus parameters, DPOAEs can be measured in nearly all normally hearing ears (Hall, 2000). To date, reasons for absent DPOAEs from approximately 1% of normal-hearing individuals with no obvious otological dysfunction have not been explored (Hall), which is an implication for validity of this measure. When normal cochlear processes are disrupted, the generation of DPOAEs is affected (Harris, 1990). Careful probe placement

University of Cape Town

and adjustment of stimulus (i.e., intensity and frequency) levels are necessary prerequisites for monitoring cochlear function with OAEs.

High stimulus levels (> 70 dB SPL) tend to stimulate passive cochlear processes (Dorn et al. 2001). Although the stability of the response may be improved by using higher stimulus levels, the sensitivity of detecting functional changes in the cochlea may be compromised (Probst et al., 1993). Research has shown that DPOAE measurements separate ears with normal hearing from those with hearing loss most accurately when moderate intensity level primaries are used, i.e., between 40 to 70 dB SPL, and that DPOAEs decrease for either lower or higher level primaries (Stover, Gorga, Neely & Montoya, 1996). Moderate stimulus levels of 65 dB sound pressure level (SPL) across all frequencies are recommended for clinical DPOAE-measurement (Lonsbury-Martin et al., 1990; Probst et al., 1993). Stover et al. found that the primary levels, L_1 and L_2, of 65 and 55 dB SPL separated normal from impaired ears better than either lower or higher level stimuli.

University of Cape Town

Apart from the intensity of the primaries, i.e., L_1 and L_2, the relationship between L_1 and L_2 also plays a role in the amplitude and sensitivity of DPOAEs. Three relationships between L_1 and L_2 are possible, i.e., $L_1 = L_2$, $L_1 < L_2$ and $L_1 > L_2$. In comparison to $L_1 = L_2$, $L_1 > L_2$ usually yield DPOAE amplitudes that are 3 dB larger than when L_1 and L_2 are equal in intensity (Gaskill & Brown, 1990; Hauser & Probst, 1991), and there is heightened sensitivity to cochlear dysfunction (Robinette & Glattke, 2002). A difference of 10 dB between L_1 and L_2 yields the highest DPOAE amplitudes. Thus, the

stimulus levels of L_1=65 and L_2=55 are generally used in clinical applications (Stover et al., 1996). DPOAE amplitude is more dependent on the intensity of L_1 than the intensity of L_2 (Hall, 2000).

The frequency separation of the primaries also plays a critical role in DPOAE measurement (Dreisbach & Siegel, 2001). Hall (2000) states that, "a DP(OAE) will not be recorded if the two tones are spaced too far apart or if they are too close together" (p. 111), in terms of frequency. The relationship between the primaries, expressed as the f_2/ f_1 ratio, influences DPOAE amplitude. Some research has shown that an f_2/ f_1 ratio of 1.22 yields maximum DPOAE amplitudes between 1 to 4 kHz (Harris, Lonsbury-Martin, Stagner, Coats & Martin, 1989). The cumulative results of previous studies confirm that a clinically effective f_2/ f_1 ratio is between 1.20 and 1.23 (Hall, 2000; Harris et al.).

Theoretically, OAEs should be helpful in the detection of ototoxicity, because most ototoxins are thought to affect the OHCs of the inner ear (Campbell & Durrant, 1993). This ability to detect OHC damage due to ototoxicity has been demonstrated in animal studies (Aran et al., 1999; Henley et al., 1996), as well as in humans (Berninger, Karlsson, Hellgren & Eskilsson, 1995; Hotz, Harris & Probst, 1994; Stavroulaki, et al., 1999; Stavroulaki et al., 2002). One advantage of using DPOAEs to investigate cochlear functioning is that it is objective and, thus, does not rely on the testee's co-operation or judgement. Another advantage is the fact that measurement of cochlear status is obtained in a non-invasive manner. It is also a relatively quick (i.e., a few seconds per

University of Cape Town

ear) and reliable test of cochlear function, especially with individuals who are ill, when compared to pure tone audiometry (Hall, 2000). DPOAE-measurement is straightforward when using modern commercially available test equipment (Probst et al., 1993). The cost of DPOAE equipment needed for ototoxicity monitoring is comparable to that required for conventional pure tone audiometry. Research evaluating repeatability of DPOAEs found no significant changes in DPOAE amplitude across recording sessions (Zhao & Stephens, 1999). Thus, because of high test-retest reliability, as well as the high sensitivity and specificity of DPOAEs to the status of the cochlea, it is an ideal tool to use in the monitoring of dynamic changes in cochlear responsiveness (Probst et al., 1993).

Since OAEs are reduced or absent in the presence of a middle ear problem, DPOAE measurements have to be preceded by tympanometry in order to rule out any conductive elements (Robinette & Glattke, 2002). When a middle ear problem is present, any DPOAE results will be unreliable, and not indicative of cochlear function (Hall, 2000).

DPOAEs can also not be used effectively in ototoxicity monitoring when the baseline hearing loss is greater than 40 dB HL (Hall, 2000; Konrad-Martin et al., 2005; Robinette & Glattke, 2002). Due to the fact that DPOAEs are absent with a moderate hearing loss, ototoxicity monitoring with DPOAEs can only be conducted with individuals who have, at the worst, a minimal hearing loss at baseline. Currently, there are no accepted protocols or criteria for ototoxic change using objective measures like OAEs. Most reports in individuals

University of Cape Town

receiving ototoxic drugs have focused on ABR or OAE test sensitivity, in which sensitivity was defined as a clinically significant change in the value of the objective measure (Mulheran & Degg, 1997; Stavroulaki et al., 1999; Stavroulaki et al., 2002). Test-retest variability in subjects not receiving ototoxic drugs has been used to provide criteria for a clinically significant change and to estimate false positive rates (Konrad-Martin et al., 2005). Such studies have been useful for developing potential objective protocols for ototoxicity, which need to be validated. Further research is needed to compare test sensitivity and specificity for objective tests to a behavioural standard (Konrad-Martin et al.).

A study with guinea pigs demonstrated reduced DPOAE-amplitude after exposure to aminoglycosides (Campbell & Durrant, 1993). In a study done by Mulheran and Degg (1997), pure tone thresholds in the conventional frequency range and DPOAE I/O function thresholds of participants receiving gentamicin treatment were compared with those of control participants with normal hearing. Significant elevation of stimulus levels for DPOAE-growth functions at f_2 = 4 kHz were found in the gentamicin group when compared to the control group after gentamicin treatment. No significant differences for pure tone thresholds were found between the two groups. The authors argued that the significant increases in stimulus levels necessary to elicit DPOAEs in the gentamicin group could reflect the early onset of an ototoxic effect of gentamicin in these participants. In a study with participants receiving hemodialysis, usually associated with sensorineural hearing loss, pre- and post-treatment measures of pure tone thresholds and DPOAE-amplitude were

University of Cape Town

compared (Ozturan & Lam, 1998). While pure tone thresholds revealed no significant changes, the DP-gram showed significantly lower amplitude in frequencies above 2000 Hz, and a specifically remarkable decline in the DPOAE-amplitude at 6000 Hz.

DP I/O function slopes are another source of information on cochlear function. According to Dorn et al. (2001) and Kummer, Janssen and Arnold (1998), DP I/O function slopes in individuals with normal cochlear function are linear with low input stimuli ($L_2 \leq 30$ dB SPL), compressive with moderate input stimuli (L_2 between 30 to 60/70 dB SPL), and linear again with high input stimuli ($L_2 \geq$ 60/70 dB SPL). In both these studies linear DP I/O function slopes were found at all stimulus levels with hearing-impaired participants. These researchers suggested that the compressive growth observed in normal ears reminded them "of the nonlinear gain attributed to the cochlear amplifier" (Kummer et al., p. 3441) and therefore, suggest normal cochlear function. Dreisbach and Siegel (2001), on the other hand, found almost linear slopes at all stimulus levels with their study of six normal-hearing individuals. A reason for the differences could be the difference in number of normal-hearing participants included in the studies: Dorn et al. had 27 participants and Kummer et al. 20 participants, in contrast with Dreisbach and Siegel's six.

Stover et al. (1996) reported varied overall shapes of DPOAE I/O function slopes with a fixed 10 dB stimulus level difference in 103 normal-hearing participants. The shapes included: non-monotonic (compressive), saturated (no increase in DP amplitude with increase in stimulus intensity) and continual

University of Cape Town

growth for even the highest stimulus levels. Therefore, it can be deduced that variability does exist in DP I/O function slopes in normally functioning cochleas when a stimulus level difference of 10 dB is used.

Different equipment and stimulus parameters were used in these studies on DP I/O function slopes, which could have contributed to the differences in results. The methods of determining the DP I/O function slopes also differed among the studies, and this could also give rise to the contradiction in the results. Therefore, it is necessary to standardise equipment and stimulus parameters used in the recording of DP I/O function slopes in order to establish the utility of these measures in the clinical setting.

Clinical observations in humans of reduced OAEs in normal-hearing patients, who typically have been exposed to noise, or potentially ototoxic drugs, and who complain of hearing difficulties, which are detected by conventional pure tone audiometry, provide support for the notion that OAEs act as early detectors of imminent cochlear dysfunction (Arnold et al., 1999). Due to the ability of OAEs to detect abnormal cochlear function before conventional pure tone audiometry, the assumption is made that the DPOAE amplitude indicates the summed activity of a substantial number of OHCs, rather than only a few OHCs as in the case with conventional pure tone audiometry (Arnold et al., 1999).

When comparing DPOAEs with conventional and high frequency pure tone audiometry in terms of suitability as a test to monitor ototoxicity, test reliability,

University of Cape Town

the relative test time, cost of equipment and the independence of the test need to be considered. DPOAE recording would have higher reliability than the other two tests in individuals who, due to illness, have compromised attention spans (Vasquez & Mattucci, 2003). The reason is that the DPOAE test is an objective test and only requires passive participation of the individual being tested, whereas conventional and high frequency pure tone audiometry are subjective tests requiring active participation. For this reason, test time with DPOAEs will be shorter than the two subjective tests, on average one minute for DPOAEs in comparison to roughly ten minutes for conventional pure tone audiometry in cooperative individuals. The relative cost of DPOAE equipment is comparable to that required for conventional pure tone audiometry, and could be less expensive than equipment for high frequency audiometry. When monitoring ototoxicity, it is important to establish that the hearing loss is sensorineural in nature. With pure tone audiometry, it is, therefore, necessary to conduct air conduction and either bone conduction or tympanometry. When high frequency audiometry is conducted, conventional pure tone audiometry and tympanometry also need to be measured. DPOAE testing has to be preceded by tympanometry, in order to rule out the presence of a middle ear problem (Hall, 2000).

To date, conventional pure tone audiometry has been used to monitor the ototoxic effects of streptomycin and kanamycin once monthly in TB-patients at a TB-hospital in the Western Cape. Due to limited resources, more regular monitoring of ototoxicity in these patients has not been possible to date. With sick patients who cannot participate actively in testing, monitoring has to be

University of Cape Town

delayed until they are able to yield reliable responses with conventional pure tone audiometry. As a result, cochlear damage may often be detected at a late stage, when the cochlear damage is probably irreversible (Vasquez & Mattucci, 2003). At this late stage, the frequencies important for speech perception are affected, which in turn leads to possible permanent hearing loss experienced by the individual (Fausti et al., 1984). This hearing loss could potentially affect communication, and, potentially, employment negatively.

With the use of OAEs, it would be possible to monitor the patients more often, because of the shorter test time (seconds) in comparison with pure tone audiometry (minutes). Monitoring the very ill patients will also be possible because OAEs require only passive participation. Cochlear damage will be detected sooner with OAEs, because of its increased sensitivity to cochlear changes (Hall, 2000).

In addition, the time difference in detecting cochlear damage with DPOAEs vs. conventional pure tone audiometry with aminoglycoside administration in humans has not yet been explored extensively. OAE application as a monitoring tool for ototoxicity has not been evaluated sufficiently to enable formulation of specific guidelines (ASHA, 1994). In addition, the DP I/O function test, to date, has enjoyed limited clinical utility, due to the length of the test and lack of sufficient research on its role in monitoring cochlear activity (Hall, 2000). Thus, the aim of this study was to compare the relative sensitivity, i.e., difference in time that it takes for DPOAEs to detect cochlear damage to that of conventional pure tone audiometry, and the relative number

University of Cape Town

of ears with abnormal cochlear function detected. The sensitivity of the DPgram vs. the DP I/O function was also compared. The nature of DP I/O function slopes was examined and, lastly, the stimulus parameters of the DP I/O function were investigated to determine whether it would be possible to shorten the length of this test. The overall purpose of the study was to evaluate different measures of cochlear function, in an attempt to improve the monitoring of ototoxic damage in individuals with TB exposed to aminoglycosides and, thereby, minimising the impact of hearing loss on the quality of their lives.

University of Cape Town

Methodology

Aims

In order to facilitate early intervention to prevent or minimise hearing loss, and maximise quality of life of individuals receiving ototoxic drugs, this study aimed to determine, in participants with TB who were receiving aminoglycosides:

1) the presence and nature of auditory dysfunction (hearing loss and abnormal cochlear function) during streptomycin or kanamycin treatment
2) the relative performance of three tests of auditory function (namely: pure tone thresholds, DPgram absolute amplitudes and DP I/O function thresholds) in first detecting ototoxicity and comparing the number of ears with abnormal cochlear function detected
3) the nature of DP I/O function slopes during streptomycin or kanamycin treatment
4) which DP I/O test parameters best identify ototoxicity (intensity levels and/or frequencies)

University of Cape Town

Study design

This study used a descriptive one-tail repeated measures design (Katzenellenbogen, Joubert & Karim, 1997), and a quantitative approach. It was a one-tailed study because only deterioration of cochlear function was expected. With a repeated measures design, one was able to monitor the cochlear function of the individuals over time. The participants therefore, acted as their own controls. The problems associated with variability between

participants were eliminated with the use of this design, as participants were compared to themselves, and not to other participants (Howell, 1999). The research conducted was basic.

A known disadvantage of using a repeated measures design is the possibility of a learning effect (Howell, 1999), which could have affected the conventional pure tone threshold results. Providing the same instructions at each test interval, in part, controlled for learning effects, i.e., participants were encouraged to respond to the softest sound that they were able to detect. Learning effects do not influence OAEs, because it is an objective measure that does not require active participation of the person being tested.

University of Cape Town

Participants

Population and sampling

Selection Criteria:

All the participants had to:

a) have a diagnosis of tuberculosis and be scheduled for tuberculosis treatment with streptomycin or kanamycin as decided by a medical doctor. Streptomycin and kanamycin have ototoxicity as a known side effect. He/she had to receive the treatment for a minimum of 2 weeks, in order to monitor possible ototoxic effects.

b) have normal non-diagnostic otoscopic results, to rule out possible outer or middle ear problems that could obscure ototoxicity monitoring (ASHA, 1994)

c) have normal hearing at baseline, which is defined as thresholds of ≤ 15 dB hearing level (HL) (Clark, 1981, in Katz, 2002) for the frequency range 250Hz to 8000Hz, in order to monitor deterioration of hearing sensitivity

d) yield reliable responses, that were replicable with conventional pure tone audiometry, in order to monitor hearing performance during aminoglycoside exposure over time (ASHA, 1994)

e) have normal middle ear functioning, which is defined as a Type A tympanogram, with compliance between 0.2 and 1.4 cm^3, middle ear pressure ranging from –150 daPa to 100 daPa and ear canal size between 1.0 and 2.0 cm^3 (GSI 38 manual, 2003) because OAEs can not be measured in the presence of a middle ear abnormality (Hall, 2000)

f) exhibit present DPOAEs for at least 8 of the 9 frequencies tested, in order to monitor change in DPOAEs over time, i.e., DPOAE amplitudes have to be at least 3 dB above the noise floor to be considered valid (Hall, 2000)

g) be 18 years or older in order for the individual to consent to participate in non-invasive research (N. Abrahams, personal communication, August, 2001; Medical Research Council, 2002).

University of Cape Town

Sample size

An a priori power calculation performed with GPOWER Version 2.0 software indicated an ideal sample size of 42 ears. Power analysis was conducted with an alpha-level of .05, power of .80 and effect size of .90. Since each ear of the participant could be viewed separately, a sample size of 21 participants was ideal, rendering results for 42 ears. Each ear of the participant was considered individually because it was found in earlier research that individual ears could

react differently to streptomycin administration (Fausti et al., 1984), and also because of ear effects on DPOAEs (Hall, 2000).

Recruitment

The 21 participants were recruited from a TB-hospital in the Western Cape following ethics approval from the Faculty of Health Sciences' Research Ethics Committee, University of Cape Town and written permission was received from the superintendent of the institution in question. The resident audiologist provided the researcher with a list of newly admitted patients with normal hearing who were to be treated with either streptomycin or kanamycin each week. Since the resident audiologist only referred those individuals who fitted the selection criteria, all individuals that were referred were considered for inclusion in the study.

University of Cape Town

Sampling

The researcher met potential participants individually to describe the purpose and nature of the current study. Each person who met the selection criteria and was willing to participate in the study was accepted. All 21 individuals who were referred agreed to participate in the study. Thus, convenience sampling was employed (Katzenellenbogen et al., 1997). Convenience sampling is a form of non-random sampling where all available individuals are selected to participate in a study. Due to the limited time and resources for data collection, and the relatively limited number of individuals with TB receiving aminoglycosides that have normal hearing at baseline, convenience sampling was chosen. According to Katzenellenbogen et al., convenience sampling

may be used when it is difficult to locate sufficient cases that can be included in a study by sampling randomly, especially in instances when time and resources are limited.

The referred sample consisted of 21 participants who had already been screened and met the selection criteria, but subsequently four had to be excluded. Among those excluded was one participant who developed binaural middle ear problems by Week 2, and had to be referred for medical treatment. Two participants chose to go home after the baseline test, and, thus, were not available for testing after they left the hospital. One participant withdrew from the study after the baseline test, because she believed that she would acquire a hearing loss if she underwent pure tone testing. This misconception was subsequently cleared up, but the participant still withdrew from the study. Due to a limited time frame for data collection additional participants were not recruited for the study.

University of Cape Town

Participant description

The study sample consisted of 17 participants with TB, six males and 11 females, ranging in age from 18 to 58 years (mean age of males = 36 years; females = 25 years). Nine participants had MDR-TB and received the aminoglycoside kanamycin during the course of the study. One participant's medication was changed from kanamycin to clarithromycin after she developed a hearing loss. This participant was still included in the study, because she had received kanamycin for a period of three weeks during the study. The remaining eight participants received streptomycin. Fifteen

participants had received previous aminoglycoside treatment for TB. Renal status was not known for any of the participants, because renal function was not routinely tested at the study site. For a summary of biographical details, refer to Table 1.

Table 1. Biographical information of participants

Participant nr.	Age (years)	Gender	Aminoglycoside	Retreatment	Other
1	29	F	Kanamycin	yes	MDR
2	33	M	Streptomycin	yes	
3	25	M	Streptomycin	yes	
4	49	M	Streptomycin	no	
5	26	M	Kanamycin	yes	MDR
6	31	F	Streptomycin	no	
7	32	F	Kanamycin	yes	MDR
8	18	F	Kanamycin; Clarithromycin	yes	MDR
9	58	M	Streptomycin	yes	
10	18	F	Kanamycin	yes	MDR
11	19	F	Kanamycin	yes	MDR
12	36	F	Kanamycin	yes	MDR
13	27	M	Kanamycin	yes	MDR
14	45	F	Kanamycin	yes	MDR
15	28	F	Streptomycin	yes	
16	28	F	Streptomycin	yes	
17	33	F	Streptomycin	yes	
Mean age	31				
Age range	18-58				

Note: F = Female; M = Male

MDR = Multi-drug resistant tuberculosis

University of Cape Town

Data collection

Equipment

All audiometric tests were conducted in a single sound-treated booth in the Audiology department at the TB-hospital.

A Heine Minilux otoscope was used for non-diagnostic otoscopic examinations. This otoscope provided sufficient illumination of the outer ear and tympanic membrane in order to determine whether any obstructions or abnormalities were present in these ear structures.

The Madsen Screen-Tymp portable tympanometer was used for tympanometry. As the institution where the study was conducted did not have a tympanometer, a portable tympanometer was used. Only screening tympanometry was required, as the purpose of tympanometry for the study was to detect whether middle ear problems were present or not. This detection was necessary to determine whether DPOAEs could be performed reliably.

An Amplaid 308 audiometer and TDH-39 headphones were used for audiometry. These were available at the institution where the study was conducted.

University of Cape Town

All DP-measurements were done with an Otodynamics ILO92. The ILO92 allowed the researcher to conduct diagnostic DPOAEs and manipulate the stimulus parameters for the current study. ILO 292 probe tips were used. All of the equipment was calibrated according to the manufacturers' specifications, within the three months prior to testing.

No normative data for the DPOAE stimulus parameters employed in this study were available for the ILO92 equipment at the time of data collection (ILO office, personal communication, 2 June 2004). Therefore, subsequent to data collection, data for the DPgram amplitudes and DP I/O function amplitudes and thresholds were collected from ten individuals with normal hearing (thresholds ≤ 15 dB HL) and middle ear function (Type A-tympanogram), and no history of aminoglycoside treatment or noise exposure. This data was used to constitute the normative data for the DPOAE measures of this study. See Tables 2 and 3 for normative data on DPgram absolute amplitudes and DP I/O function thresholds respectively. The normative data were used to determine if DPOAEs were normal or abnormal and thus, to determine the presence of abnormal cochlear function.

University of Cape Town

Table 2. Normative data for DPgram absolute amplitudes

	DPgram absolute amplitudes (dB SPL)		
Frequency (Hz)	Mean (*SD*)	Minimum	Maximum
818	9.3(6)	1.1	18.7
1038	11.9(7)	-1.2	22.8
1306	12.0(6)	1.5	22.8
1636	9.8(5)	0	16.3
2063	9.1(6)	0.4	21.1
2600	9.0(7)	1.3	21
3284	9.4(6)	1.1	19.8
4126	15.5(5)	7.2	22.2
5200	11.5(7)	0	20.6

Note: n = 20 ears

Table 3. Normative data for DP I/O function thresholds

	DP I/O function thresholds (dB SPL)		
Frequency (Hz)	Mean (*SD*)	Minimum	Maximum
818	50(6)	≤ 45	65
1636	≤ 45(7)	≤ 45	55
2454	≤ 45(6)	≤ 45	60
3284	≤ 45(5)	≤ 45	60
4102	≤ 45(6)	≤ 45	55
4919	≤ 45(3)	≤ 45	50

Note: n = 20 ears

Procedure

Prior to commencing the study, ethics approval was obtained from the Faculty of Health Sciences' Research Ethics Committee, University of Cape Town. Thereafter written permission to conduct research was obtained from the

University of Cape Town

Medical Superintendent of the hospital. Written informed consent was obtained from each participant. The researcher verbally conveyed information regarding the purpose, protocol and the requirements of the study in the participant's language (English or Afrikaans), which was also shared with each participant in written form. In the case of Xhosa-speaking participants, a native Xhosa-speaking individual explained the relevant information regarding the study, and the written format of the participant information sheet was in Xhosa. The researcher trained the Xhosa "interpreter" by firstly explaining the purpose and procedures of the study, as well as the instructions employed with all the tests. In the training session, the interpreter had to convey all the information to a second native Xhosa speaker, and this person had to repeat all the conveyed information in Afrikaans. This allowed the researcher to evaluate whether the correct information was conveyed. All discrepancies and uncertainties were addressed. See Appendix A1, A2 and A3 for the participant information sheets and A4, A5 and A6 for the consent forms.

University of Cape Town

Numbers from 1 to 17 were assigned to each participant at the point where he/she was included in the study. Only this number appeared on all test results in order to maintain participant anonymity and confidentiality.

Before commencing the test protocol, the nature of all test procedures were explained to the participant in the native language in order to familiarise the participant with the session. The participant was then given an opportunity to seek clarification, and was also reminded that questions could be asked at

any stage during data collection. The participant was reminded that he/she could withdraw at any point in the study.

Clear instructions were given before each procedure, and repeated during the test if necessary. The same instructions were given to all clients to improve reliability. The instructions used for pure tone testing were: "You will hear soft sounds, some with a high pitch and some with a low pitch. Each time you hear a sound, please raise your hand immediately. Even if the sound is very soft and you can barely hear it, you must still raise your hand. We will start with your right ear." If it was deemed necessary, the instructions were repeated during pure tone testing.

Before DPOAE measurement, the participant was instructed as follows: "I am going to place a probe in your ear. It will not be painful. Your ear will just feel blocked up. Please remain quiet and still while the probe is in your ear. You do not have to do anything, even if you hear a sound. You can sit back and relax."

The participant was seated in a comfortable chair for the duration of the tests. A glass of water was provided for the participant in the case of coughing. Testing was stopped in the case of coughing (frequent in individuals with TB) and continued after coughing ceased.

The following tests were conducted at each of the baseline and evaluation sessions at Weeks 0, 2, 4, 6 and 8: otoscopy, tympanometry, conventional

University of Cape Town

pure tone audiometry, DPgram and DP I/O function. Speech audiometry to enhance reliability of the pure tone results, could not be conducted, due to equipment limitations.

Otoscopy

Non-diagnostic otoscopy involved visual inspection of the external auditory meatus and tympanic membrane. If any abnormality of these structures were observed, appropriate referral was made after the participant's consent to do so was obtained. No abnormality of the external auditory meatus or tympanic membrane was found in any of the participants.

Tympanometry

With tympanometry, a probe was inserted into the external auditory meatus, whereby pressure (-400 to +200 daPa) was automatically applied onto the tympanic membrane. Measurements of the compliance of the middle ear system, the ear canal volume and the middle ear pressure were subsequently made by the automatic tympanometer. If abnormalities were detected, referral to the relevant medical practitioner was made and the individual was excluded from the study at this point. Two participants had to be excluded from the study on the basis of abnormal middle ear function.

Pure tone audiometry

The resident audiologist at the TB-hospital administered the pure tone air conduction testing. This test involved presenting pure tones from 250 to 8000 Hz, at octave intervals, using the ANSI procedure (1978, in Gelfand, 1997).

University of Cape Town

The audiologist started testing at 30 dB HL at 1000 Hz, using an ascending approach. The participant had to raise his/her hand to indicate when the tone was detected. This response method was chosen because peripheral neuropathy, which is often present in individuals with TB, could interfere with their ability to press a button. The results were recorded in dB HL on an audiogram.

DPgram

The researcher conducted all DP-measurements on the same day as pure tone testing. A probe was placed in the external auditory meatus, and two pure tones were presented to one ear at a time. The frequency range for the input stimuli were from 1000 to 6000 Hz, with an f_2/f_1 ratio of 1.22 (Harris et al., 1989). Three points per octave were recorded (ILO92 Manual, 1992). L_1 was equal to 65 dB and L_2 was 55dB (Lonsbury-Martin et al., 1990). For the duration of this test the intensity of the presented signals were kept constant and the frequency was varied systematically, progressing from lowest to the highest frequency, resulting in a DPgram. The frequency pairs that were used are presented below in Table 4. The absolute amplitudes and the noise floor were recorded for each ear. The size of relative amplitude is dependent on the level of the noise floor, but the size of absolute amplitudes is not. Therefore, the preferred measurement for ototoxicity monitoring in this instance where noise cannot be controlled extensively would be absolute amplitudes. Each participant received 18 stimulus presentations, i.e., nine per ear at each test interval. Due to time constraints, only one sweep could be presented per ear.

University of Cape Town

Table 4. Stimulus frequency pairs for DPOAE measures

DPgram		DP I/O function	
f_1 (Hz)	f_2 (Hz)	f_1 (Hz)	f_2 (Hz)
818	1001	818	1001
1038	1257	1636	2002
1306	1587	2454	3003
1636	2002	3284	4004
2063	2515	4102	5005
2600	3174	4919	6006
3284	4004		
4126	5042		
5200	6348		

University of Cape Town

DP I/O function

The DP I/O function was measured with the same frequency range (i.e., 1000 to 6000 Hz) and f_2/f_1 ratio as the DPgram. The frequency pairs used are presented in Table 4. With the probe still in the ear from the previous test, two pure tones were presented simultaneously, while keeping the frequency pair constant and varying the intensity. The stimulus intensity levels of the DP I/O function ranged from L_2 = 45 to 75 dB SPL (L_1 = 35 to 65 dB SPL), with both L_1 and L_2 decreased in 5 dB increments. These stimulus levels were used because Kummer et al. (1998) found that the compressive range indicating normal cochlear function was between L_2 = 40 to 60 dB SPL. Another reason for the chosen stimulus levels, and for not including lower intensities for the

current study, was due to equipment limitations. Each participant received 84 stimulus presentations, i.e., 42 per ear at each test interval.

Noise floor

The noise floor was recorded at every session for all participants during both DPgram and DP I/O function measurements. In OAE measurement, it is crucial to monitor the noise floor level in order to determine whether the OAEs obtained are valid (Hall, 2000; Robinette & Glattke, 2002). DPOAE measurement was stopped when the average noise floor was –10 dB SPL for all participants, as this was an obtainable low noise level. DPOAEs were only regarded as valid (and therefore, present) if their amplitudes were 3 dB above the noise floor (Hall, 2000).

As soon as a decrease in hearing thresholds or a reduction in DPOAE amplitude to below the norms was observed, the participant, as well as the doctor, was notified. Criteria used to indicate hearing loss that developed during aminoglycoside treatment, was as defined by ASHA (1994): (a) a 20 dB decrease at any one test frequency, or (b) a 10 dB decrease at any two adjacent test frequencies, or (c) loss of response at three consecutive test frequencies (i.e., the three highest frequencies tested) at the limits of the audiometer where responses were previously obtained.

The doctor, together with the participant, then had to decide on the future path of treatment. The participants were free to withdraw from the study at any point in time, but were not excluded from the study by the researcher if

University of Cape Town

streptomycin or kanamycin treatment was terminated after two weeks of administration, because aminoglycoside exposure of five days can result in a hearing loss (Arnold et al., 2000).

All of the above tests were conducted once a fortnight over a two-month period during streptomycin or kanamycin administration. The whole test battery used in the study took approximately 40 minutes per participant.

Data recording

Participants' pure tone thresholds were recorded on-line and manually on an audiogram. All DP-measures were saved on-line on disk and analysed off-line.

University of Cape Town

Reliability and validity

Reliability refers to the degree of similarity of information obtained when the measurement is repeated on the same participant or same group (Katzenellenbogen et al., 1997). Reliability with regards to DPOAE measures was partially addressed by ensuring that the probe placement was stable, and done according to the manufacturers' specifications. The noise floor had to be ≤ -10 dB SPL to improve reliability and validity of the DPOAE results.

Replicable results are good indicators of reliability. Unfortunately, it was not possible to repeat measurements in order to ensure that all the results obtained were reliable. Due to the inclusion of DP I/O function measurement,

the testing session was fairly long, and would have been double in length if measurements were repeated.

In order to enhance test-retest reliability, the stimulus level was calibrated with the probe tip and probe in the participant's ear before DPOAE measures were conducted. The stability of the probe had to be acceptable before data collection started (ILO92 manual, 1992). DPOAE measures were stopped when the average noise floor was less than -10 dB SPL with each participant. This ensured that higher noise levels would not obscure visibility of present DPOAEs. The researcher, as well as the resident audiologist gave all the participants the same instructions to ensure that different instructions did not influence the test results.

The researcher followed the same procedures in the same order with each participant to enhance intra-tester reliability. For example, DPOAEs were always recorded first in the right ear. In all participants, DPgram absolute amplitudes and DP I/O functions were recorded in one ear, and then in the other.

In order to enhance inter-tester reliability, the results of all the pure tone threshold testing conducted by the resident audiologist were withheld from the researcher until after the completion of data collection to minimise bias. The researcher was the only person who conducted the objective measures, i.e., tympanometry and DPOAE measures, therefore, inter-tester reliability was irrelevant with these measures.

University of Cape Town

An independent person blind to the nature of the study reanalysed 10% of statistical data to determine reliability of data analysis. The researcher then compared the analysed results and found no difference between the initial statistical analysis and that conducted by the blind independent person.

Validity is the "extent to which a measure actually measures what it is meant to measure" (Katzenellenbogen et al., 1997, p. 90). Face validity requires that the measures utilised make sense (Katzenellenbogen et al.). Previous research has proven that DPOAEs measure OHC function (Trautwein et al., 1996), and is therefore, an appropriate tool to use in ototoxicity monitoring during aminoglycoside treatment, that is known to affect OHC function (Barclay & Begg, 1994; Hashino et al., 1995). Also, DPOAEs were only regarded as valid (and therefore, present) if their amplitudes were 3 dB above the noise floor (Hall, 2000).

University of Cape Town

Content validity refers to the inclusion of all the elements being investigated by the measure employed in a study (Katzenellenbogen et al., 1997). Cochlear function was evaluated in the current study, and it was therefore, appropriate to use DPOAEs as a measurement tool (Hall, 2000).

Criterion-related validity requires that the results of the index study be evaluated against the most valid measure available, i.e., the "gold standard" (Katzenellenbogen et al., 1997). Auditory brainstem response measurement (ABR) can be regarded as the standard against which the validity of OAEs

can be compared (Le Calvez, Avan, Gilain & Romand, 1998). Kakigi et al. (1998) did not find a correlation between DPOAE, TEOAE measurement and ABR threshold in chinchillas that received amikacin. The authors speculated that the reason for the low correlation between the three tests could be because the OAEs and ABR represent different mechanisms in the auditory pathway (Kakigi et al.). However, Le Calvez et al. found a correlation between the DPgram and ABR thresholds in CD1 mice if f_2 was used as the reference point. Thus, the DPgram can be regarded as a valid test for cochlear function.

"Predicted validity requires that the measure confirms a known or theoretically hypothesised association" (Katzenellenbogen et al., 1997, p. 92). Research has proven that reduced DPOAE amplitude is related to cochlear abnormality due to ototoxic damage (Campbell & Durrant, 1993; Mulheran & Degg, 1997). Therefore, DPOAEs would be a valid tool to use in the monitoring of cochlear function during aminoglycoside exposure.

Bias refers to any process that produces results or conclusions that differ from the truth in a one-sided way (Katzenellenbogen et al., 1997). DPOAEs are objective, therefore, the researcher could not influence results with feelings or expectations. Also, the fact that the researcher was blind to the participants' pure tone thresholds until data collection was completed also reduced bias. In data analysis, the significance level ($p < .05$) was decided upon before starting data analysis.

University of Cape Town

Measurement bias refers to methods employed that can influence the results in a systematic way (Katzenellenbogen et al., 1997), and relates to the previous explanation. Measurement error could potentially be problematic in individuals with TB, because they cough, and therefore, generate noise. In the case of a high noise floor, OAEs can seem to be absent, but are actually obscured by the noise floor. Therefore, measurement of OAEs was stopped during coughing, and resumed only when coughing subsided. A set criterion of an average noise floor of < -10 dB SPL was used when measuring DPOAEs in all participants before stopping the recording, in order to minimise the possibility of measurement bias.

Selection bias exists if the participants included in the study are not representative of the population from which they were selected (Katzenellenbogen et al., 1997). The current study included hospitalised individuals with TB who had normal hearing and received aminoglycosides. Therefore, the sample is not representative of all individuals with TB receiving aminoglycosides, because individuals with existing hearing loss at the start of the study were not selected. This fact has to be borne in mind in the interpretation of the results and conclusions drawn from this study.

Ethical considerations

Ethics approval was obtained from the Research Ethics Committee, Faculty of Health Sciences, University of Cape Town before initiating the study. Permission from the superintendent of the hospital and written informed consent was obtained from participants prior to data collection.

University of Cape Town

Autonomy refers to the competent individual's right to self-determination (Katzenellenbogen et al., 1997). Providing the participants with adequate information to give informed consent to participate in the study adhered to this principle of autonomy, and safeguarded their freedom of choice. In order to facilitate understanding of the study, information was presented in the first language of the participant. Participants were given the opportunity to ask for clarification at any time during the study. It was also explained to the participant that participation in the study was voluntary and that he/she could withdraw at any stage of data collection. The participants were informed that refusal to participate in or withdrawal from the study would not affect their management at the institution. Each participant was required to sign a consent form if he/she agreed to take part in the study.

The ethical principle of beneficence requires that the actions of the researcher are aimed at improving the well-being of the participant (Katzenellenbogen et al., 1997) and minimising harm (MRC, 2002). The participants were informed that if they developed cochlear damage due to aminoglycoside treatment, the damage could possibly be detected earlier while participating in the study because: 1) the monitoring interval of the study (once every two weeks) was shorter than the routine interval at the hospital (once a month) and (2) the DPOAE measurements used in the study could potentially be more sensitive to cochlear damage than the conventional pure tone audiometry used at the hospital. Thus, as soon as abnormal cochlear function was detected with

University of Cape Town

DPOAEs, the participant was referred to the relevant doctor to consider alternative management.

Non-maleficence is the principle that refers to the researcher's obligation to not do harm (Katzenellenbogen et al., 1997). No procedures were used that could harm the participants, and all tests were non-invasive. All necessary equipment e.g., probe tips and earphones were cleaned with alcohol swabs after use with each participant (according to hospital protocol), to ensure that bacteria were not spread from one participant to the next. The researcher washed her hands with an antiseptic agent for the same reason.

The ethics principle of justice can be interpreted to mean a fair distribution of benefits of the research (Katzenellenbogen et al., 1997). This principle "also leads to the interpretation that all who stand to benefit from the research should contribute to its risks and discomforts" (Katzenellenbogen et al.). This study included all individuals at the age of 18 or older with normal hearing that received aminoglycoside treatment during the period of data collection that consented to participating. Younger participants were not included in this study, due to difficulty obtaining proxy consent from parents or guardians. The institution in the study was chosen because most MDR-TB patients receiving long-term aminoglycoside treatment are hospitalised and are at risk of developing hearing loss.

Confidentiality was respected at all times and all details and results of participants were kept confidential and anonymous by assigning a number to

University of Cape Town

each participant. This number was used on all documentation pertaining to the study. Identifying information was only known to the researcher, and was kept separate from collected data in a locked filing cabinet in the researcher's office. The researcher also assured the participants that no identifying information would appear in any publication or presentation forthcoming from this study.

Researcher experience implies that the researcher is competent to conduct the research (MRC, 2002). The researcher was professionally qualified as an audiologist, and had sufficient theoretical and clinical expertise to conduct the research. The researcher acted in an accountable and responsible manner, and upheld professional standards.

University of Cape Town

Data analysis

Quantitative analysis

Pure tone thresholds were manually entered into MS Excel spreadsheets for ease of analysis. DPOAE results were converted from MS-DOS spreadsheets into Excel spreadsheets in order to facilitate ease of data analysis. The software SPSS v.9.0 was used for quantitative analysis.

Descriptive statistics (mean, mode and standard deviation) for the pure tone thresholds, DPgram absolute amplitudes, DP I/O functions and thresholds for all test intervals were calculated separately. Thereafter all the data was analysed to determine normal distribution, using skewness and kurtosis measures.

The DP I/O function slopes were calculated at each frequency by dividing the change in DPOAE amplitude (from one stimulus intensity level to the next one) by the increase in stimulus intensity, i.e., 5 dB SPL. The formula used was therefore: Δ DPOAE amplitude ÷ 5. The calculated slopes were plotted as line graphs.

The results of left and right ears for all the audiological measures were compared to determine whether results were different for the two ears. The Wilcoxon signed ranks test determines the relative magnitude of differences within pairs (Siegel & Castellan, 1988), and was therefore, suited to compare results of the two ears.

The pure tone thresholds, DPgrams and DP I/O functions were compared to determine whether any of these measures of auditory function deteriorated significantly over time. Statistical analysis included the Friedman ANOVA test, which tests the null hypothesis that the *k* repeated measures or matched groups come from the same population (Siegel & Castellan, 1988). For each case, the variables were then ranked.

The ears that developed hearing loss over time were compared with those that did not, to determine whether there was a difference between the changes in pure tone thresholds of these two groups. The Mann-Whitney *U* test is able to determine whether two groups differ from one another (Siegel & Castellan, 1988), and was therefore, used.

University of Cape Town

The three audiological measures were compared post hoc to determine which one contributed to the statistically significant difference obtained in the number of ears in which cochlear abnormalities were detected. The Wilcoxon signed ranks test was used because it allows one to determine which member of a pair is "greater than" the other, in other words it is able to indicate the sign of difference between any pair (Siegel & Castellan, 1988, p. 87).

A significance level of .05 was used for all of the abovementioned statistical tests. The Bonferroni method was used to correct the significance levels when multiple comparisons were made with one data set, in order not to violate assumptions regarding the alpha level due to multiple comparisons. Corrected significance levels were obtained by multiplying the original significance level with the number of comparisons that were made (Dr. S. Isaacs, personal communication, July 2004).

University of Cape Town

Nature of trends in cochlear function

The data were subjected to descriptive analysis to determine whether any trends were visible relating to the changes over time in pure tone thresholds, amplitudes of DPgrams and DP I/O function, as well as in DP I/O thresholds. The DP I/O results were also examined with regard to the frequency and stimulus parameters to see whether patterns indicating cochlear abnormality could be detected in order to reduce the testing time. DP I/O function slopes were visually examined to determine whether trends emerged that

corresponded with or were contrary to previous findings (Dreisbach & Siegel, 2001; Kummer et al., 1998; Popelka et al., 1993; Stover et al., 1996)

University of Cape Town

Results

The results will be described in accordance with the aims of the study. Firstly, the nature and presence of auditory function (hearing loss and abnormal cochlear function) during streptomycin or kanamycin treatment as shown with pure tone audiometry, DPgram absolute amplitudes and DP I/O function thresholds will be examined. Secondly, differences in sensitivity among the conventional pure tone audiometry, the DPgram absolute amplitudes and DP I/O function thresholds with regard to time of detection of ototoxicity, as well as number of ears with abnormal cochlear function will be presented. The DP I/O function slopes will then be examined for trends, i.e., whether the slopes are compressive or linear. Next, the DP I/O function will be examined to determine whether certain test parameters (stimulus frequency and intensity) contribute to creating a DP I/O function profile of ototoxicity.

University of Cape Town

Treatment of data

All the data were tested for normality. The data were positively skewed (Sprinthall, 1987), and was thus, not normally distributed (see Appendix B1 & B2). According to Tabachnick and Fidell (2001), transformation often hinders interpretation if the variable is measured in a scale that is not meaningful or widely used, which was the case with the measurements in this study. Hence, transformations were not done, because clinicians find it difficult to read transformed data (Dr. S. Isaacs, personal communication, March 2004). Therefore, non-parametric statistics were used to analyse the data.

In order to exclude pre-existing cochlear abnormalities, results of the total number of 34 ears were examined. Seventeen ears yielded amplitudes at Week 0 that were below the norm for either the DPgram absolute amplitudes or DP I/O function absolute amplitudes, and were excluded from the analysis. In some cases, only one ear of a participant yielded abnormal results, and only the abnormal ear was excluded. As a result, 17 ears (12 subjects) remained for analysis. This is a limitation of the study that should have been considered as part of the subject selection criteria in order to have a larger sample size.

Comparison of the left and right ears in all tests

The pure tone thresholds, DPgram absolute amplitudes and DP I/O function absolute amplitudes were statistically compared with the Wilcoxon signed ranks test to determine whether there was a difference between the left and right ears for each of the three tests. This comparison was done in order to determine whether the results of individual ears could be presented without regard for laterality. The Bonferroni method was used to correct the significance levels, as multiple comparisons were made with one data set, i.e., the left and right ears of the pure tone thresholds, DPgram absolute amplitude and DP I/O function thresholds were compared for each test individually (Wallenstein, Zucker & Fleiss, 1980). The Wilcoxon signed ranks test showed that the performance of the right and left ears did not differ significantly for the pure tone thresholds ($p > .016$) (see Table 5 for details of performance on pure tone thresholds), DPgram absolute amplitudes with $p > .016$ (Table 6) and DP I/O absolute amplitudes ($p > .016$) (Table 7) respectively, except for

University of Cape Town

the pure tone thresholds at 4000 Hz at Week 2 (Z = -2.2; p = .00), and four instances out of 210 in the DP I/O absolute amplitudes (818 Hz at Week 0, with stimulus intensity of 75 dB SPL; 1636 Hz at Week 4, with stimulus intensities of 45 and 50 dB SPL; 4919 Hz at Week 2, with stimulus intensity of 45 dB SPL). The statistically significant differences in the pure tone threshold and DP I/O absolute amplitudes appeared to occur randomly, i.e., without any clear pattern. Thus, the results of the pure tone thresholds, DPgram absolute amplitudes and DP I/O function thresholds will be presented without reference to the laterality of the ear.

Table 5. Differences in pure tone thresholds between the left and right ears over the 8-week period

	Wilcoxon's signed ranks test									
Week	0		2		4		6		8	
Frequency (Hz)	*Z*	*p*	*Z*	*p*	*Z*	*p*	*Z*	*p*	*Z*	*p*
250	-0.6	1.6	-0.4	2.1	-1.1	0.9	-1.1	0.8	-1.3	0.6
500	-1.0	1.0	-1.0	1.0	-0.8	1.2	-1.0	1.0	-1.3	0.5
1000	0.00	3.0	0.00	3.0	0.00	3.0	0.00	3.0	-0.6	1.7
2000	-0.4	2.0	0.00	3.0	-1.0	1.0	-0.4	2.0	0.00	3.0
4000	-0.7	1.4	-2.2	**.00**	-1.5	0.4	-0.1	2.7	-1.9	0.02
8000	-1.9	0.2	-0.4	2.2	-1.0	1.0	-0.6	1.6	-0.2	2.6

Bonferroni method correction: α = .016

Note: Bold $p \leq .016$, i.e., statistically significant

University of Cape Town

Table 6. Differences in DPgram absolute amplitudes between the left and right ears over the 8-week period

	Wilcoxon signed ranks test									
Week	0		2		4		6		8	
Frequency (Hz)	*Z*	*p*	*Z*	*p*	*Z*	*p*	*Z*	*p*	*Z*	*p*
818	-0.28	2.33	-1.40	0.49	-0.51	1.83	-0.28	2.33	-0.63	1.59
1038	-0.92	1.07	-0.24	2.44	-0.41	2.04	-0.66	1.53	-1.01	0.93
1306	-0.69	1.48	-0.45	1.96	-0.10	2.75	-0.41	2.05	-0.21	2.50
1636	-0.45	1.96	-0.62	1.61	-1.24	0.64	-0.16	2.63	-1.50	0.40
2063	-0.31	2.27	-0.28	2.33	-1.06	0.87	-0.47	1.91	-1.01	0.93
2600	-0.43	2.01	-1.37	0.51	-0.65	1.54	-0.09	2.77	-1.82	0.21
3284	-0.78	1.30	-0.64	1.57	-0.13	2.69	-0.60	1.65	-1.43	0.46
4126	-1.02	0.93	-0.50	1.86	-1.09	0.83	-0.22	2.48	-1.22	0.66
5200	-0.02	2.94	-2.20	0.08	-0.31	2.27	-1.19	0.70	-1.22	0.66

Bonferroni method correction: $\alpha = .016$

Note: Bold $p \leq .016$, i.e., statistically significant

University of Cape Town

Table 7. Differences in DP I/O function absolute amplitudes between the left and right ears over the 8-week period

		Wilson's signed ranks test									
	Week	0		2		4		6		8	
Frequency	Intensity										
(Hz)	(dB SPL)	z	p	z	p	z	p	z	p	z	p
818	75	-2.84	**0.00**	-0.47	0.64	-0.45	0.65	-1.08	0.28	-0.55	0.58
	70	-6.59	0.08	-0.85	2.33	-1.24	2.04	-2.31	1.33	-1.78	1.66
	65	-3.62	0.68	-2.20	1.39	-1.09	2.15	-3.48	0.74	-2.20	1.39
	60	-4.34	0.44	-0.64	2.49	-1.47	1.87	-2.54	1.19	-1.99	1.52
	55	-3.48	0.74	-2.06	1.48	-1.24	2.04	-0.85	2.33	-4.51	0.40
	50	-0.78	2.38	-3.27	0.83	-0.93	2.27	-3.01	0.94	-3.67	0.66
	45	-0.78	2.38	-0.21	2.83	-1.86	1.60	-0.28	2.77	-5.56	0.19
1636	75	-2.12	0.03	-1.78	0.07	-2.04	0.04	-0.03	0.97	-1.22	0.22
	70	-5.43	0.21	-3.34	0.80	-5.20	0.25	-1.98	1.53	-3.25	0.84
	65	-5.47	0.21	-0.50	2.61	-3.26	0.83	-1.04	2.19	-3.04	0.93
	60	-3.26	0.83	-1.35	1.96	-2.87	1.02	-1.65	1.75	-3.57	0.70
	55	-1.78	1.66	-0.31	2.75	0.00	3.00	-2.92	0.99	-6.18	0.12
	50	-1.99	1.52	-2.79	1.06	-9.23	**0.01**	-0.73	2.42	-6.18	0.12
	45	-4.47	0.41	-1.78	1.66	-8.53	**0.01**	-1.99	1.52	-3.67	0.66

University of Cape Town

Table 7. (cont.)

		Wilson's signed ranks test									
	Week	0		2		4		6		8	
Frequency (Hz)	Intensity (dB SPL)	z	p	z	p	z	p	z	p	z	p
2454	75	-1.14	0.26	-0.64	0.52	-0.62	0.53	-0.47	0.64	-0.45	0.65
	70	-3.10	0.90	-0.92	2.27	-4.19	0.49	-0.31	2.75	-2.41	1.26
	65	-2.77	1.07	-0.16	2.88	-4.11	0.51	-6.12	0.12	-3.36	0.79
	60	-1.21	2.06	-1.07	2.17	-3.65	0.67	-7.06	0.06	-0.42	2.67
	55	-0.50	2.61	-4.50	0.40	-1.40	1.92	-3.86	0.59	-3.88	0.59
	50	-2.20	1.39	-4.12	0.51	-0.93	2.27	-2.35	1.30	-2.83	1.04
	45	-5.18	0.25	-2.63	1.14	-5.43	0.21	-0.75	2.41	-3.67	0.66
3284	75	-1.01	0.31	-0.21	0.83	-0.36	0.72	-1.41	0.16	-0.03	0.97
	70	-2.64	1.14	-0.07	2.94	-2.33	1.31	-4.43	0.42	-1.15	2.10
	65	-0.50	2.61	-0.78	2.38	-1.55	1.82	-2.92	0.99	-0.42	2.67
	60	-0.07	2.94	-1.63	1.76	-0.16	2.88	-0.09	2.92	-2.20	1.39
	55	-0.14	2.89	-3.34	0.80	-2.87	1.02	-4.80	0.33	-1.89	1.59
	50	-1.07	2.17	-0.43	2.66	-0.62	2.51	-6.50	0.09	-0.31	2.75
	45	-1.42	1.91	-1.92	1.57	-2.56	1.18	-6.87	0.07	-4.09	0.52
4102	75	-1.88	0.06	-0.36	0.72	-0.03	0.98	-1.13	0.26	-0.17	0.86
	70	-3.72	0.64	-1.78	1.66	-0.39	2.69	-5.74	0.17	-4.51	0.40
	65	-2.77	1.07	-0.92	2.27	-4.11	0.51	-4.43	0.42	-4.09	0.52
	60	-1.42	1.91	-0.99	2.22	-2.09	1.46	-3.86	0.59	-5.77	0.16
	55	-6.28	0.11	-2.84	1.03	-2.48	1.22	-4.30	0.46	-5.77	0.16
	50	-2.41	1.26	-0.07	2.94	-4.58	0.38	-1.13	2.12	-1.57	1.80
	45	-1.63	1.76	-2.49	1.22	-3.72	0.64	-2.92	0.99	-0.63	2.50
4919	75	-0.10	0.92	-1.40	0.16	-1.14	0.26	-1.88	0.06	-1.47	0.14
	70	-1.78	1.66	-5.04	0.28	-2.56	1.18	-4.52	0.40	-3.67	0.66
	65	-0.50	2.61	-4.05	0.53	-1.55	1.82	-0.66	2.48	-5.03	0.28
	60	-1.07	2.17	-0.36	2.72	-3.10	0.90	-0.66	2.48	-5.14	0.26
	55	-0.28	2.77	-4.62	0.37	-5.59	0.19	-5.37	0.22	-5.06	0.27
	50	-1.78	1.66	-2.06	1.48	-3.10	0.90	-5.18	0.25	-0.52	2.58
	45	-1.92	1.57	-8.52	**0.01**	-6.05	0.13	-0.94	2.26	-5.77	0.16

Note: Bold $p \leq .016$, i.e., statistically significant; Bonferroni method correction: $\alpha = .016$

University of Cape Town

Presence and nature of auditory dysfunction

Pure tone thresholds

Frequency of hearing loss as depicted by pure tone thresholds

At the end of the eight-week period, 6 (35%) of 17 ears (i.e., 3 of the 12 participants) demonstrated binaural sensorineural hearing losses. The degree of hearing loss ranged from slight for one participant (20 dB HL) to moderate to severe for two participants (one with 60 dB HL at 8000 Hz, and one with a flat hearing loss with a pure tone average of 60 dB HL). See Table 8 for details. The hearing sensitivity, as measured by pure tones, for the other nine participants remained within normal limits over the eight-week test period.

Table 8. Pure tone threshold changes over eight weeks in participants with hearing loss

	Pure tone decrease (dB)											
	Right ear						Left ear					
Frequency (Hz)	250	500	1000	2000	4000	8000	250	500	1000	2000	4000	8000
Participant												
A	0	15	0	0	0	**50[a]**	**15[b]**	**10[b]**	0	0	0	**50[a]**
B	**10[b]**	**10[b]**	0	0	5	10	**10[b]**	**10[b]**	0	5	10	5
C	**50[a]**	**45[a]**	**45[a]**	**45[a]**	**60[a]**	**65[a]**	**60[a]**	**55[a]**	**50[a]**	**45[a]**	**65[a]**	**70[a]**

Note: Bold indicates clinically significant pure tone threshold changes as per ASHA criteria (1994) for determining clinically significant hearing loss in ototoxicity monitoring:

(a) a 20 dB decrease at any one test frequency, or

(b) a 10 dB decrease at any two adjacent test frequencies, or

(c) loss of response at three consecutive test frequencies where responses were previously obtained.

University of Cape Town

The nature of pure tone threshold changes over the eight-week period

All participants

Hearing sensitivity as a function of pure tone thresholds revealed the following:

The mean pure tone thresholds appeared to deteriorate over the eight-week test period (see Table 9), with the mean hearing thresholds at Week 8 being worse at all frequencies relative to those obtained at baseline. The decrease in the mean pure tone thresholds was not clinically significant, according to ASHA's guidelines for monitoring ototoxicity (1994). However, it is worth noting that all the mean thresholds had been within normal limits at baseline (see Table 9). By Week 6, however, the pure tone thresholds at 4000 and 8000 Hz were elevated to 17 dB HL and 21 dB HL, respectively, which exceeded the normal limit of 15 dB HL (Clark, 1981, in Katz, 2002). This deterioration in pure tone thresholds with exposure to aminoglycosides was not statistically significant (see Table 9).

University of Cape Town

Table 9. Mean pure tone thresholds over the 8-week period for all participants

	Mean dB SPL (*SD*)					Friedman's ANOVA		
Week	0	2	4	6	8	*F*	*df*	*p*
Frequency (Hz)								
250	10(*1*)	11(*2*)	12(*5*)	15(*12*)	18(*19*)	3.9	4	.42
500	10(*0*)	10(*0*)	11(*4*)	14(*11*)	18(*18*)	6.7	4	.15
1000	10(*0*)	10(*0*)	10(*0*)	12(*4*)	16(*17*)	4.0	4	.41
2000	10(*1*)	10(*1*)	10(*0*)	13(*7*)	16(*16*)	5.5	4	.23
4000	10(*1*)	11(*2*)	11(*4*)	17(*19*)	20(*23*)	9.0	4	.06
8000	12(*1*)	12(*2*)	19(*4*)	21(*19*)	21(*23*)	4.0	4	.41

Note: Bonferroni correction: $\alpha = .016$, N = 17

Underline indicates clinically abnormal pure tone threshold (Clark, 1981, in Katz, 2002)

Participants with hearing loss

In order not to mask the results of the three participants who developed a hearing loss as measured by pure tone thresholds, their results were extracted from those whose hearing sensitivity remained within normal limits. Thus, what follows is a description of the hearing sensitivity changes in those who developed a hearing loss with exposure to aminoglycosides.

Presence of cochlear function abnormalities as depicted by DPgram absolute amplitudes

By Week 2 examination of the results revealed that 2 of the 17 ears (12%) displayed ototoxic hearing loss, as per ASHA (1994) guidelines (see Table 10). By Week 8, the pure tone thresholds of 6 ears (35%) showed ototoxic hearing loss, indicating deteriorating cochlear functioning over time with exposure to aminoglycosides.

University of Cape Town

Table 10. Cochlear abnormalities across frequencies as detected by pure tone thresholds

	Participant's ear where cochlear abnormalities were detected over time				
Week	2	4	6	8	Total ears per frequency
Frequency (Hz)					
250			7l	12r 12l 8r 8l	5
500				12r 12l 8r 8l	4
1000				8r 8l	2
2000				8r 8l	2
4000				8r 8l	2
8000	7r 7l	8r 8l			4
Number of new ears	2	2	0	2	
Cumulative number of ears	2	4	4	6	

Note: # = Participant number

r = right ear; l = left ear

Inspection of the participants' pure tone thresholds (refer to Table 8) revealed that the deterioration of thresholds in the six ears was clinically significant over

University of Cape Town

time (ASHA, 1994). All the mean thresholds had been within normal limits at baseline (see Table 11). However, by Week 2, the mean pure tone threshold at 8000 Hz was elevated to 19 dB HL, which exceeded the normal limit of 15 dB HL (Clark, 1981, in Katz, 2002). By Week 8 the pure tone thresholds at all frequencies exceeded the normal limit. The difference between their thresholds at baseline and at Week 8 was statistically significant at all frequencies ($p < .05$), except for 1000 Hz (see Table 11 for details on Friedman's ANOVA).

Table 11. Mean pure tone thresholds over the 8-week period of ears with hearing loss

	Mean dB SPL (*SD*)					Friedman's ANOVA		
Week	0	2	4	6	8	*F*	*df*	*p*
Frequency (Hz)								
250	10 (1)	12.5 (3)	18 (8)	24 (18)	35 (26)	11.5	4	**.02**
500	10 (0)	10 (0)	18 (7)	24 (15)	34 (22)	18.9	4	**.00**
1000	10 (0)	10 (0)	10 (0)	14 (7)	26 (27)	8	4	.09
2000	10 (0)	10 (0)	10 (0)	17 (12)	26 (25)	10.7	4	**.03**
4000	10 (1)	10 (0)	13 (6)	28 (32)	35 (37)	13.4	4	**.01**
8000	10 (0)	19 (14)	48 (34)	52 (36)	52 (31)	18.4	4	**.00**

Note: Underline indicates clinically abnormal pure tone threshold (Clark, 1981, in Katz, 2002)

Bold $p \leq .05$, N = 6

University of Cape Town

Comparison of the pure tone thresholds in ears with and without hearing loss

At Week 8 mean pure tone thresholds in the nine ears that did not acquire hearing loss ranged from 16 to 21 dB HL (see Table 9 for details of mean pure tone thresholds), and in the six ears that developed hearing loss it ranged from 26 to 52 dB HL (see Table 11 for details of pure tone thresholds in ears that developed hearing loss). The difference in mean thresholds between those ears that acquired hearing loss by Week 8 and those that did not, was statistically significant, Mann-Whitney *U* test ($p \leq .05$) at all frequencies (see Table 12).

University of Cape Town

Table 12. Difference in mean pure tone thresholds at Week 8 between ears with and without hearing loss

	Mann-Whitney test									
Week	0		2		4		6		8	
Frequency (Hz)	*z*	*p*	*z*	*p*	*z*	*p*	*z*	*p*	*z*	*p*
250	0.5	.6	2.6	**.0**	2.5	**.0**	0.1	.2	3.4	**.0**
500	0.0	1.0	0.0	1.0	3	**.0**	0.0	.2	3.4	**.0**
1000	0.0	1.0	0.0	1.0	0	1.0	0.0	.2	2.2	**.0**
2000	0.7	.5	0.7	.5	0	1.0	0.1	.5	2.4	**.0**
4000	1.4	.2	1.0	.3	2	**.1**	0.1	.2	2.9	**.0**
8000	1.8	.1	1.1	.3	2.6	**.0**	0.0	.1	2.5	**.0**

Note: Bold $p \leq .05$

The *z*-scores were derived from the Mann-Whitney *U*-statistic, as per SPSS v.9.0.

DPOAE measures

Noise floor

The DPgram and DP I/O function noise floor levels for all tests (Weeks 0 to 8) were not statistically significant at all frequencies, Friedman's ANOVA (p > .05). See Appendix C for the details of the statistical comparison between the noise floor levels as recorded during DPgram measurement and Appendix D for details on the noise floor levels for the DP I/O function, for all test intervals. Thus, all DPOAE measures were not affected by differences in the noise floor level.

DPgram absolute amplitudes

Presence of cochlear function abnormalities as depicted by DPgram absolute amplitudes

By Week 2 examination of the results revealed that 4 of the 17 ears (25%) displayed abnormal (i.e., amplitudes lower than the norm minimum for at least one frequency) DPgram absolute amplitudes (see Table 13). By Week 8, the DPgram absolute amplitudes were below the norm for at least one frequency in 15 ears (88%), indicating deteriorating cochlear functioning over time with exposure to aminoglycosides. As can be seen in Table 13, 11 ears were affected at more than one frequency. The frequencies that detected the most cochlear abnormalities were 3284, 4126 and 5200 Hz (see Table 13 for exact numbers of ears). The mean DPgram absolute amplitudes at the three highest frequencies tested, i.e., 3284, 4126 and 5200 Hz were abnormal by Week 8 (see Table 14 for mean DPgram absolute amplitudes over the 8-week period).

University of Cape Town

Table 13. Cochlear abnormalities across frequencies as detected by DPgram absolute amplitudes

	Participant's ear where cochlear abnormalities were detected over time				
Week	2	4	6	8	Total ears per frequency
Frequency (Hz)					
818					0
1038					0
1306					0
1636					0
2063		6r		9r	2
2600	3r 3l		6r	4r 9r	5
3284	3r 3l		6r 6l 9l	1r 2r 2l 4r 7l	10
4126	3r 3l 6r		2r 6l 9r	1r 2l	8
5200	3r 6r 7r	7l 9r 9l 10r	2r	1r 2l 5l 12r	12
Number of new ears with abnormality	4	4	2	5	
Cumulative number of ears	4	8	10	15	

Note: # = Participant number

R = right ear; l = left ear

University of Cape Town

The nature of DPgram absolute amplitude changes over the eight-week period

The results of the DPgram absolute amplitudes over the eight-week period can be seen in Table 14. The mean absolute amplitudes deteriorated over this period at all frequencies, suggesting deteriorating cochlear function with exposure to aminoglycosides over time. At Week 0, the mean DPgram absolute amplitudes for all the frequencies were within normal limits. However, by Week 6, the mean DPgram absolute amplitude for one frequency, i.e., 4126 Hz was below the normal range, and by Week 8 three frequencies (3284, 4126 and 5200 Hz) were below the normal range (see Table 14). The difference between the DPgram absolute amplitudes at Week 0 and Week 8 was statistically significant at the low frequencies (818, 1038, 1306 and 1636 Hz) and at 5200 Hz, Friedman's ANOVA ($p < .016$). See Table 14 for details of mean DPgram absolute amplitude change over time.

University of Cape Town

Table 14. Mean DPgram absolute amplitudes for all participants over eight weeks

		Mean dB SPL (*SD*)					Friedman's ANOVA		
	Week	0	2	4	6	8	*F*	*df*	*p*
Frequency (Hz)	Norm range								
818	1.1 – 18.7	12.9(4)	12.6(4)	12.5(4)	12.4(5)	7.0(3)	16.7	4	**0.002**
1038	-1.2 – 22.8	13.3(4)	13.3(4)	13.6(4)	12.7(5)	7.9(4)	17.3	4	**0.002**
1306	1.5 – 22.8	13.7(3)	12.3(5)	12.7(3)	11.9(5)	7.1(3)	13.9	4	**0.008**
1636	0.0 – 16.3	13.0(5)	11.7(7)	12.0(5)	11.2(6)	6.7(4)	13.1	4	**0.011**
2063	0.4 – 21.1	10.5(6)	9.5(5)	8.9(6)	8.4(7)	3.6(9)	8.7	4	0.069
2600	1.3 – 21.0	9.1(6)	8.3(4)	7.1(3)	6.0(5)	2.3(8)	11.7	4	0.020
3284	1.1 – 19.8	7.2(6)	5.2(6)	5.8(4)	3.0(8)	-0.6(8)	11.5	4	0.022
4126	7.2 – 22.2	10.3(4)	9.6(5)	8.6(6)	6.1(9)	2.3(9)	11.1	4	0.025
5200	0.0 – 20.6	5.7(8)	5.1(8)	2.9(9)	0.8(10)	-2.0(9)	25.2	4	**0.000**

Note: Bonferroni correction: $\alpha = .016$

Bold $p \leq .016$

Underlined values indicate DPgram absolute amplitude levels outside the norm

University of Cape Town

DP I/O function thresholds

Presence of cochlear function abnormalities as depicted by DP I/O function thresholds

Examination of the participants' DP I/O function thresholds revealed that 1 of the 17 ears (5%) displayed a threshold below the norm range at Week 2 (see Table 15 for details on when cochlear abnormality was detected in the different participants). By Week 8, nine ears (53%) had abnormal DP I/O thresholds for at least one frequency, indicating that a larger number of participants demonstrated deteriorating cochlear function with exposure to aminoglycosides over time (Table 15).

Table 15. Week when cochlear abnormality at different frequencies was detected in participants

		Week when DP I/O function threshold detected cochlear abnormality					
Frequency (Hz)		818	1636	2454	3284	4102	4919
Participant	Ear						
1	Right	-	-	-	-	-	6
2	Right	-	-	-	6	-	-
3	Right	-	8	-	4	4	-
4	Right	-	-	-	-	-	8
5	Left	-	-	-	-	-	6
6	Right	-	-	-	6	-	2
	Left	6	8	-	6	4	4
7	Right	-	-	-	-	4	-
8	Left	-	4	-	6	-	-

University of Cape Town

The nature of DP I/O function changes over the eight-week period

Relative to the baseline, the mean DP I/O function thresholds deteriorated at all frequencies over the eight-week period (see Table 16). At Week 0, all DP I/O function thresholds were within normal limits, but from Week 2 onwards, the thresholds at 4919 Hz were below the norm range. There was a statistically significant deterioration in the mean DP I/O thresholds at 1636, 2454 and 3284 Hz over the eight weeks, Friedman's ANOVA ($p < .008$). See Table16 for details of the mean DP I/O function changes over time.

University of Cape Town

Table 16. Mean DP I/O thresholds for all participants over eight weeks

		Mean dB SPL (*SD*)					Friedman's ANOVA		
	Week	0	2	4	6	8	F	*df*	*p*
Frequency (Hz)	Norm range (dB SPL)								
818	<45 — 65	49 (5)	51 (7)	50 (5)	54 (9)	52 (7)	7	4	0.138
1636	<45 — 55	48 (4)	48 (4)	48 (4)	49 (5)	51 (6)	14.8	4	**0.005**
2454	<45 — 60	47 (3)	48 (4)	48 (4)	49 (5)	51 (5)	17.7	4	**0.001**
3284	<45 — 60	51 (8)	52 (6)	52 (6)	56 (9)	55 (8)	15.1	4	**0.005**
4102	<45 — 55	49 (4)	50 (5)	54 (6)	51 (8)	51 (5)	12.3	4	0.015
4919	<45 — 50	48 (5)	51 (8)	52 (9)	52 (9)	52 (8)	6.9	4	0.143

Note: Bonferroni correction: α = .008

Bold $p \leq .008$

Underlined indicates DP I/O thresholds outside the norm

University of Cape Town

The relative sensitivity of pure tone thresholds and DP measures in detecting ototoxicity

The relative sensitivity in all participants

The DPgram absolute amplitudes appeared to suggest the presence of cochlear damage earlier than the pure tone thresholds and DP I/O function thresholds (see Table 17). This suggestion is based on the greater number of ears presenting with DP abnormalities at each test interval. However, the differences in the number of abnormalities detected between the three tests was only statistically significant at Week 8, Friedman's ANOVA with $F(2, 17) = 9.5$, $p = .009$, indicating that DPgram absolute amplitudes detected significantly more ears with cochlear abnormalities than the other two tests at this test interval.

University of Cape Town

Table 17. Comparison of the performance of the three tests to detect cochlear abnormalities in all subjects over the 8-week period

	Total number of ears with cochlear abnormalities detected			Friedman's ANOVA		
Procedure / Week	PT	DP	IO	*F*	*df*	*p*
2	2	4	1	2.3	2	.311
4	4	8	6	4.8	2	.091
6	4	10	8	1.6	2	.459
8	6	15	9	9.5	2	**.009**

Note: Bonferroni correction: $\alpha = .013$;

Bold indicates a statistically significant difference

PT = pure tone thresholds; DP = DPgram absolute amplitudes; IO = DP input/output function thresholds

Post hoc testing was conducted in order to determine which of the three measures (pure tone testing, DPgram and DP I/O function) contributed to the statistically significant difference obtained in the ability to detect cochlear abnormalities. The number of ears with cochlear abnormalities detected by pure tone thresholds, DPgram absolute amplitudes and DP I/O function thresholds were compared. The Wilcoxon signed ranks test was conducted. The results revealed that the number of ears with cochlear abnormalities detected by pure tone thresholds were significantly lower than those detected by DPgram absolute amplitudes (z = 2.1; p = .03). In addition, the DPgram absolute amplitudes also detected a significantly higher number of ears with cochlear abnormality than DP I/O function thresholds (z = 2.8; p = .005), with a significance level of $p \leq .05$. There was no significant difference between the numbers of cochlear abnormalities detected by pure tone and DP I/O thresholds. Therefore, in this study the DPgram absolute amplitude measures were able to detect significantly more individuals with cochlear abnormalities than either pure tone or DP I/O function thresholds did. In 5 out of the 6 ears with hearing loss, the DPgram absolute amplitude detected the cochlear abnormality first, i.e., before pure tone thresholds were affected.

The relative sensitivity in ears that demonstrated hearing loss

The pure tone thresholds identified more ears with cochlear abnormalities than the DPgram absolute amplitudes and the DP I/O function thresholds (see Table 18) in terms of those ears that demonstrated hearing loss (as measured by pure tone thresholds). The DPgram absolute amplitudes also appeared to

University of Cape Town

identify more ears with cochlear abnormalities than the DP I/O function thresholds. However, none of these differences were statistically significant (see Table 18 for details on Friedman's ANOVA results).

Table 18. Number of cochlear abnormalities in ears with hearing loss (as determined by pure tone threshold testing)

Procedure	Total number of ears with cochlear abnormalities detected			Friedman's ANOVA		
	PT	DP	IO	*F*	*df*	*p*
Week						
2	2	3	1	.666	2	.716
4	4	4	2	3	2	.223
6	5	4	3	2	2	.367
8	6	5	3	4.666	2	.096

Note: Bonferroni correction: $\alpha = .013$

PT = pure tone thresholds; DP = DPgram absolute amplitudes; IO = DP input/output function thresholds

University of Cape Town

Nature of DP I/O function slopes

Studies conducted by Dorn et al. (2001) and Kummer et al. (1998) found that DP I/O function slopes are linear with low input stimuli ($L_2 \leq 30$ dB SPL) and non-linear with moderate input stimuli (L_2 between 30 to 60/70 dB SPL), representing the compressive ability of a normally functioning cochlea. In both studies, linear DP I/O function slopes were found with participants who had sensorineural hearing losses (Dorn et al.; Kummer et al.), indicating that the compressive function of the cochlea was affected.

Participants with normal hearing as measured by pure tone thresholds

Frequency trends in DP I/O function slopes

Inspection of the DP I/O function slopes, representing the compressive ability of the cochlea, revealed different patterns for the low frequencies of 818, 1636 2454 and 3284 Hz, relative to the higher frequencies of 4102 and 4919 Hz (see Table 19 for DP I/O slope values in participants with normal hearing). At the low frequencies calculations of the slopes (see Methodology section for the formula to calculate the DP I/O function slopes) revealed that the slopes initially increased to greater than 1dB/dB when the stimulus intensity was increased from 40 to 45 dB SPL, therefore, the DP I/O function slopes were linear at the lowest stimulus intensities. With the stimulus intensities higher than 55 dB SPL, the DP I/O function slopes were non-linear at the low frequencies, therefore, indicating that cochlear function became compressive at the higher intensities. For the high frequencies from 4102 to 4919 Hz, the DP I/O function slopes were predominantly linear (i.e., > 0.5 dB/dB, Ruggero et al., 1997) at all of the stimulus intensities, therefore, displaying a lack of compression in cochlear function.

University of Cape Town

In summary, the results of the low frequencies of 818 to 3284 Hz were equivocal in terms of the frequency of occurrence of non-linearity, ranging from 33 to 50%. However, at the highest frequencies of 4102 and 4919 Hz, the participants with normal hearing (as per pure tone thresholds) demonstrated linearity (100%), therefore, a lack of compressive cochlear

function. See Table 19 for details on the presence of non-linear DP I/O function slopes at the individual frequencies.

Trends in DP I/O function slopes over 8-week period

No distinctive trends could be detected for the DP I/O function slopes over the 8-week period for any of the frequencies (818 to 4919 Hz). See Figure 1 details on the DP I/O function slopes over time. Thus, all the DP I/O function slopes remained generally similar over the eight-week test period.

University of Cape Town

Table 19. DP I/O function slopes in participants who did not develop a hearing loss (as per pure tone thresholds)

	DP I/O function slopes (dB/dB)						
	Current study						Previous research (Dorn et al., 2001; Kummer et al., 1998)
L_2 stimulus intensity level	Frequency (Hz)						Frequency (Hz)
dB SPL	818	1636	2454	3284	4102	4919	1000 to 6000
40	Nlin (0.2)	Nlin (-1.1)	Nlin (-0.1)	Lin (0.7)	Lin (0.8)	Lin (0.6)	Nlin
45	Lin (1.2)	Lin (1.8)	Lin (1.6)	Lin (0.6)	Lin (0.6)	Lin (1.0)	Nlin
50	Lin (0.7)	Lin (0.9)	Lin (1.3)	Lin (1.2)	Lin (0.9)	Lin (0.8)	Nlin
55	Lin (0.7)	Lin (0.9)	Lin (0.8)	Nlin (0.5)	Lin (0.7)	Lin (0.7)	Nlin
60	Nlin (0.4)	Lin (0.9)	Nlin (0.3)	Lin (1.0)	Lin (0.8)	Lin (0.9)	Lin
65	Nlin (0.2)	Nlin (0.3)	Nlin (0.2)	Nlin (0.4)	Lin (0.8)	Lin (0.9)	Lin
% Nlin at each frequency	50	33	50	33	0	0	
Overall % Nlin	28						

Note: Lin = Linear slope with range > 0.5 dB/dB (Ruggero et al., 1997)

Nlin = Non-linear slope with range ≤ 0.5 dB/dB (Ruggero et al., 1997)

University of Cape Town

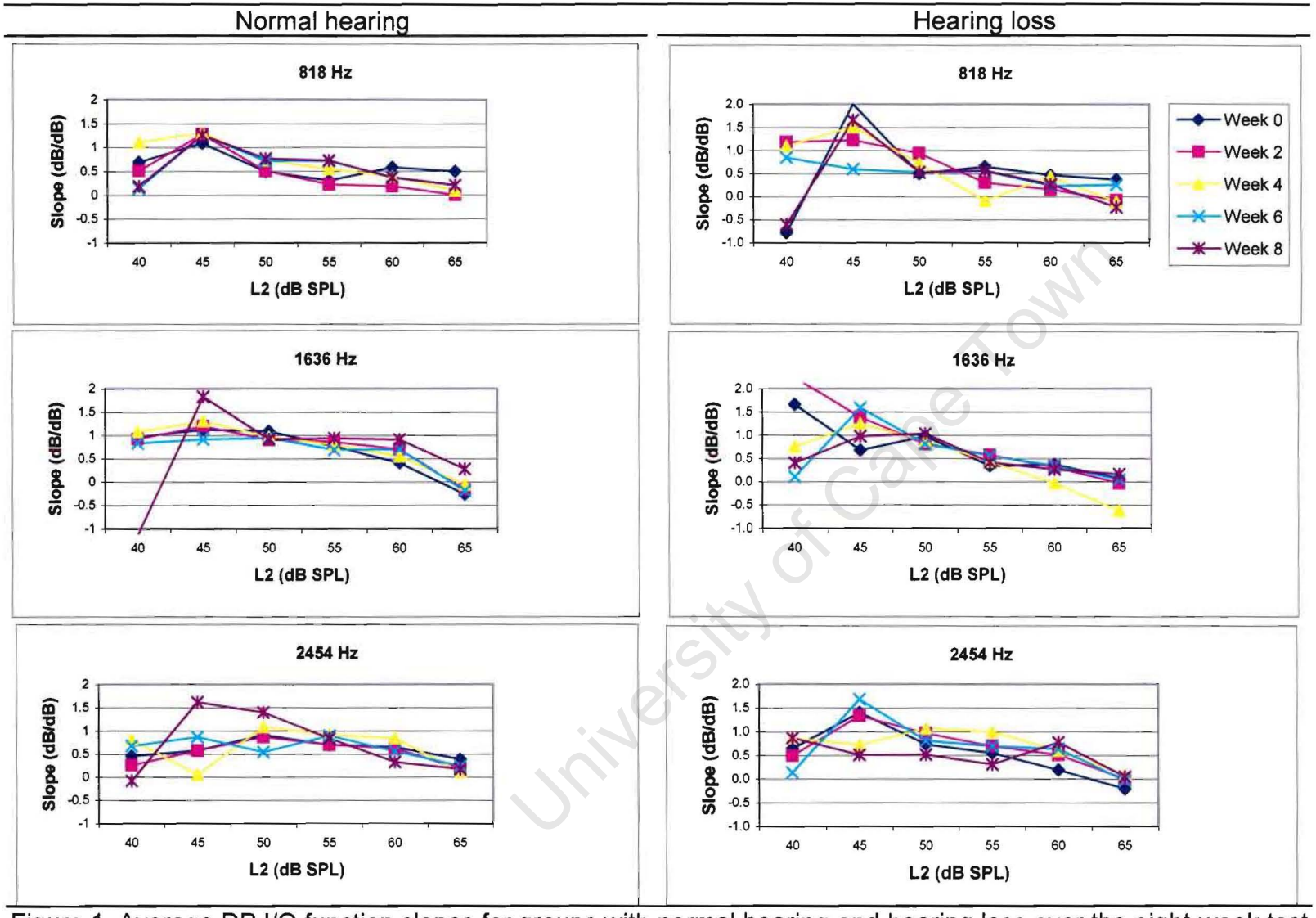

Figure 1. Average DP I/O function slopes for groups with normal hearing and hearing loss over the eight-week test

University of Cape Town

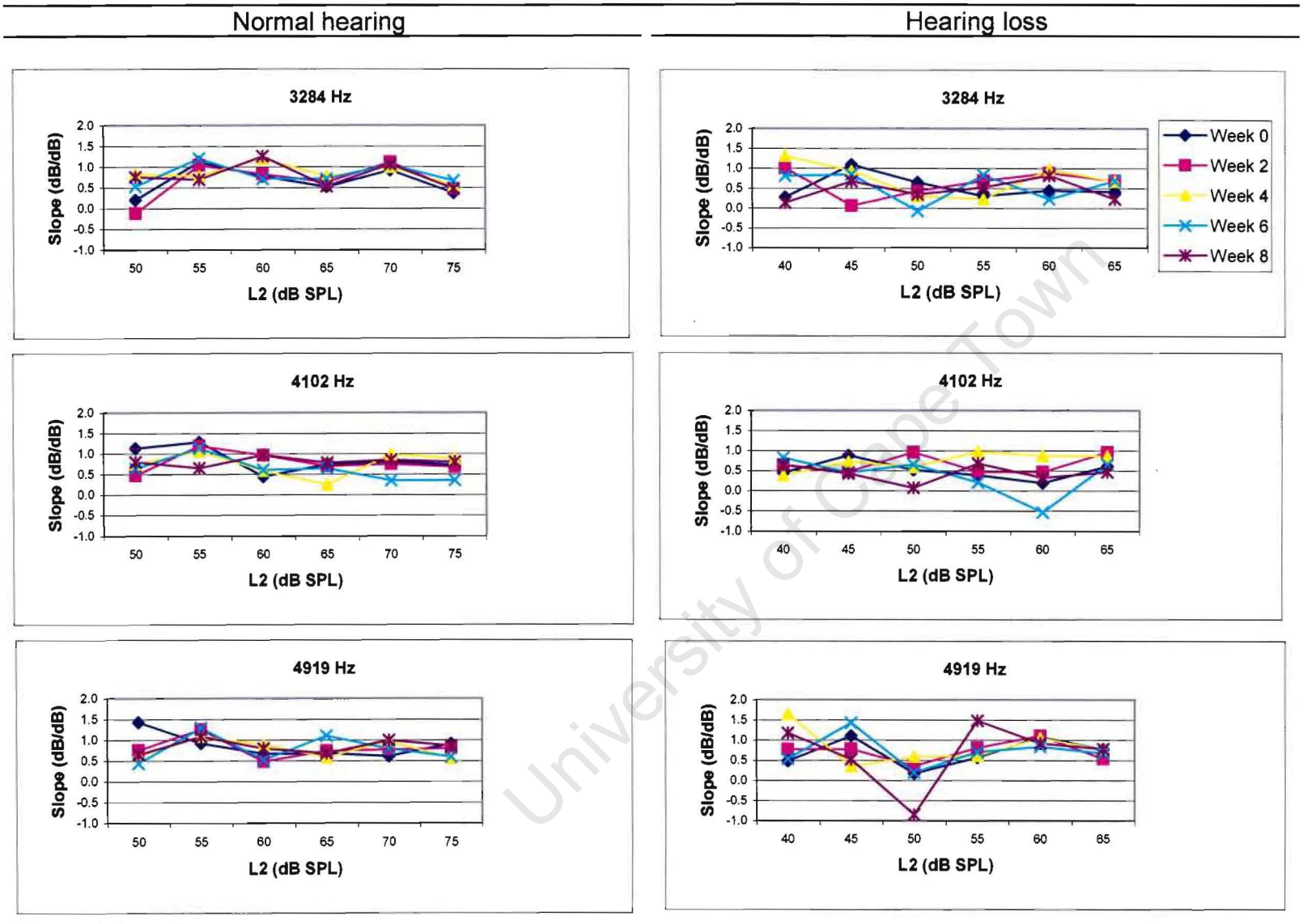

Figure 1 (cont.)
Note: Key applies to all the graphs

University of Cape Town

Participants who developed hearing loss as measured by pure tone thresholds

Frequency trends in DP I/O function slopes

From 818 to 4102 Hz the majority of the averaged DP I/O function slopes were non-linear (64%) between L_2 = 40 to 65 dB SPL, indicating predominantly compressive cochlear function (see Table 20 for values of DP I/O function slopes at Week 8). The remaining high frequency of 4919 Hz exhibited 67% linear DP I/O function slopes at stimulus intensities of L_2 = 40 and 55 to 65 dB SPL, displaying reduced cochlear compressive function. In these participants with hearing loss, fair degree of non-linearity was present in the low frequencies, which progressively decreased at higher frequencies. At 4919 Hz, the highest frequency, the presence of non-linear DP I/O function slopes was the least (33%). See Table 20 for the presence of non-linearity at all frequencies.

Visual inspection revealed more variability in the shapes of the DP I/O slopes over time in participants with hearing loss relative to the slopes of participants with normal hearing (see Figure 1 for DP I/O function slopes of all participants over the eight-week period).

Trends in DP I/O function slopes over 8-week period

No trends could be observed relating to slope changes over the eight-week period (see Figure 1), as the DP I/O function slopes present at Week 0 resembled those at Week 8. Therefore, the DP I/O function slopes did not change with aminoglycoside exposure over time. The only exception was at

University of Cape Town

2454 Hz, where the slopes at Week 8 for stimulus intensities from 45 to 55 dB SPL were less linear than those for the other weeks in participants with hearing loss (i.e., slopes of 0.5 on average at Week 8 in comparison to an average slope of 0.8 at the other weeks). This change suggests that, at 2454 Hz, the DP I/O function became non-linear with longer exposure to aminoglycosides, thus, demonstrating more compressive behaviour in the cochlea. This result was unexpected, and (seeing that this change only occurred at one frequency), the result at 2454 Hz may be regarded as a possible anomaly.

When analysing the DP I/O function slopes of the three participants with hearing loss individually, there did not appear to be any common trends among the participants (see Appendixes E1, E2 and E3 for DP I/O function slope values of the individual participants).

University of Cape Town

Table 20. DP I/O function slopes at Week 8 in participants who developed hearing loss (as per pure tone thresholds)

	DP I/O function slopes (dB/dB)						
	Current study						Previous research (Dorn et al., 2001; Kummer et al., 1998)
L_2 stimulus intensity level	Frequency (Hz)						Frequency (Hz)
dB SPL	818	1636	2454	3284	4102	4919	1000 to 6000
40	Nlin (-0.6)	Nlin (0.4)	Lin (0.9)	Nlin (0.1)	Lin (0.6)	Lin (1.1)	Lin
45	Lin (1.1)	Lin (0.98)	Nlin (0.5)	Lin (0.7)	Nlin (0.4)	Nlin (0.5)	Lin
50	Nlin (0.5)	Lin (1.0)	Nlin (0.5)	Nlin (0.3)	Nlin (0.1)	Nlin (-0.9)	Lin
55	Nlin (0.5)	Nlin (0.4)	Nlin (0.3)	Nlin (0.5)	Lin (0.7)	Lin (1.4)	Lin
60	Nlin (0.2)	Nlin (0.3)	Lin (0.8)	Lin (0.8)	Nlin (0.3)	Lin (0.9)	Lin
65	Nlin (-0.2)	Nlin (0.2)	Nlin (0.1)	Nlin (0.2)	Nlin (0.5)	Lin (0.8)	Lin
% Nlin at each frequency	83	67	67	67	67	33	
Overall % Nlin	64						

Note: Lin = Linear slope with range > 0.5 dB/dB (Ruggero et al., 1997)

Nlin = Non-linear slope with range ≤ 0.5 dB/dB (Ruggero et al., 1997)

University of Cape Town

DP I/O function test parameters

Stimulus frequency

The ears with cochlear abnormality, as detected by the DP I/O function thresholds, were examined at each frequency to determine whether there was a trend for specific frequencies to reflect cochlear abnormalities (i.e., abnormal DP I/O function thresholds present) earlier than other frequencies (see Table 21). The DP I/O function thresholds appeared to reflect most ears with cochlear abnormalities at 3284 and 4919 Hz, and the least at 2454 Hz (see Table 21). The frequencies could be grouped into a low frequency range (818, 1636 and 2454 Hz) and a high frequency range (3284, 4102 and 4919 Hz). The high frequency group reflected a total of 13 ears with cochlear abnormalities, while the low frequency group reflected four ears. Therefore, the higher frequencies reflected more cochlear abnormalities than the low frequencies. This finding was not analysed statistically due to the small sample size.

University of Cape Town

Table 21. Number of abnormal DP I/O function thresholds at different frequencies over eight weeks

	Number of ears with cochlear abnormality					
Frequency (Hz)	818	1636	2454	3284	4102	4919
Week						
2	-	-	-	-	-	1
4	-	1	-	1	3	1
6	1	-	-	4	-	2
8	-	2	-	-	-	1
Total number of ears	1	3	0	5	3	5

Stimulus intensity

The range of DP I/O function thresholds were between L_2 = 45 and 70 dB SPL for the frequencies from 818 to 2454 Hz (see Table 22), over the eight-week monitoring period. DP I/O function thresholds at 3284, 4102 and 4919 ranged between L_2 = 45 and 75 dB SPL (Table 22). As can be seen in Table 22 some of the DP I/O function thresholds were outside the normal range (higher than the norm) over the eight-week period.

Table 22. Range of DP I/O function thresholds over the eight-week period

Frequency (Hz)	Norm range (dB SPL)	Range in current study (dB SPL)
818	<45 – 65	≤ 45 - 70
1636	<45 – 55	≤ 45 – 60
2454	<45 – 60	≤ 45 - 60
3284	<45 – 60	≤ 45 – 75
4102	<45 – 55	≤ 45 - 75
4919	<45 – 50	≤ 45 - 75

University of Cape Town

Discussion

The purpose of the study was to improve the monitoring of ototoxicity in order to minimise the impact of hearing loss on the quality of life of individuals with TB receiving aminoglycosides as part of their treatment. Therefore, by comparing different measures used to monitor ototoxicity, the researcher attempted to evaluate their ability to detect abnormal cochlear function due to aminoglycoside exposure early enough to prevent or minimise permanent sensorineural hearing loss. This study attempted to determine: the presence and nature of auditory dysfunction as depicted by pure tone testing, the DPgram and DP I/O function; whether conventional pure tone testing or DPOAE measurements were the most effective in early detection of cochlear damage, the nature of the DP I/O function slopes and lastly, which DP I/O function test parameters best identify ototoxicity.

University of Cape Town

Presence and nature of auditory dysfunction

Presence of auditory dysfunction

While animal studies in rats (Henley et al. 1996; Mills, Loos & Henley, 1999) suggest that all those exposed to aminoglycosides may acquire a hearing loss, as determined by reduction in ABR thresholds, in this study with human participants, only 35% demonstrated hearing loss by the end of the eight-week test period, as manifested by elevated pure tone thresholds. However, the DP I/O function thresholds and DPgram absolute amplitudes at Week 8 indicated higher numbers of participants (53% and 88%, respectively) who demonstrated deterioration in cochlear function. The presence of ototoxicity in

the current study was higher than that reported by Sha and Schacht (1997). Using pure tone tests, they reported that less than 20% of their participants who had been exposed to aminoglycosides developed a hearing loss. The discrepancy possibly arises from the inclusion of participants exposed to streptomycin only (Sha & Schacht), whereas the current study included participants receiving either streptomycin or kanamycin.

Fausti et al. (1984) reported a high number (62.5%) of participants who developed hearing loss due to aminoglycoside exposure. However, they used high frequency audiometry to monitor ototoxicity. The type of aminoglycoside treatment was not specified. Fausti et al., and Voogt and Schoeman (1996) concluded that high frequency audiometry was more sensitive to cochlear damage than conventional pure tone audiometry. Such an explanation may account for the higher percentage obtained by Fausti et al. relative to the current study's pure tone results.

The current DPgram absolute amplitude results suggest a higher percentage (88%) of ears with cochlear damage relative to the percentages obtained with high frequency audiometry (62.5%) by Fausti et al. (1984). A possible reason for the discrepancy between the two studies could relate to difference in sample size (12 vs. 222 participants in the Fausti study). It could also be postulated that the DPgram absolute amplitudes may be more sensitive to cochlear damage than high frequency audiometry. However, the validity of such a suggestion needs to be established by further research with a large sample, which this study did not have.

University of Cape Town

The present DPgram results indicated the presence of auditory dysfunction in 88% of cochleas exposed to ototoxicity, could not be compared with other studies due to a lack of research in this area of study. However, one animal study reported that approximately 50% of chinchillas demonstrated auditory dysfunction after exposure to aminoglycosides as measured by DPOAE absolute amplitudes (Kakigi et al., 1998). This frequency of auditory dysfunction in animals detected with DPOAEs is lower than the results of the current study with human participants.

The DP I/O function threshold data from this study could not be compared with other human or animal studies, as at the time of this study such data were not available.

Hence, the current results suggest that pure tone, DPgram and DP I/O function measures all signal the presence of ototoxicity in those individuals receiving aminoglycoside treatment. There seems to be a differential ability to detect ototoxic effects and this aspect will be explored later in this discussion.

From the preceding discussion it can be seen that the frequency of hearing loss due to ototoxicity as reported in the current study and in the literature is usually less than 100%. This frequency implies that not everyone exposed to aminoglycosides develops a hearing loss, which could be linked to variable susceptibility to ototoxic drugs. A possible explanation could be ascribed to a genetic susceptibility to aminoglycoside ototoxicity (Barclay & Begg, 1994).

University of Cape Town

Bykhovskaya, et al. (2004) reported on the mutation (A15558G) of a gene (12S rRNA) associated with hearing loss, influenced by aminoglycosides. Therefore, it has been speculated (Prof. J. Greinwald, personal communications, 16 August, 2004) that those individuals who do not exhibit mutation of the specific gene, may not develop a hearing loss when exposed to aminoglycosides. Such speculation has not been confirmed and is pending further research.

Thus, future genetic research could investigate individuals' susceptibility to ototoxicity from various aminoglycosides. Genetic testing of at-risk individuals could enhance appropriate drug intervention and reduce the possibility of developing a hearing loss due to ototoxicity, and, thus, maximize human quality of life. With prevention of ototoxic hearing loss, health care costs could be reduced.

University of Cape Town

According to the findings of the current study and other studies (Fausti et al., 1984; Sha & Schacht, 1997) some individuals receiving ototoxic medication will develop a hearing loss. Accurate predictions on who will develop hearing loss cannot currently be made without monitoring cochlear function. Thus, it is important that the auditory function of all individuals who receive ototoxic medication be clinically monitored (ASHA, 1994; Konrad-Martin et al., 2005).

Nature of auditory dysfunction

Patterns of change demonstrated in terms of the frequencies affected in the pure tone tests and the DP measures were similar. The ototoxic damage (as

demonstrated by either hearing loss or auditory dysfunction) was first noticed at the highest frequency measured in all the tests of auditory function. With prolonged exposure to aminoglycosides cochlear abnormality then progressed to the adjacent lower frequencies. These findings are in agreement with the literature on the nature of cochlear changes due to ototoxicity, i.e., that the basal region is affected prior to the apical region (Kakigi, et al., 1998, Stavroulaki, et al., 2002).

The pattern of cochlear change is linked to the basal hair cells being more susceptible to ototoxic damage than the apical ones (Barclay & Begg, 1994), and has been confirmed in animal studies (Henley et al., 1996; Kakigi et al. 1998; Mills et al., 1999) and in research with humans (Berninger et al., 1995; Mulheran & Degg, 1997; Stavroulaki et al., 2002; Wang et al., 1999). In animals, the differential vulnerability of basal and apical hair cells to ototoxic damage is based on the intrinsic susceptibility to free radicals (Sha et al., 2001). Similar research in human cadavers has yet to be undertaken. Perhaps animal studies could also explore the physiology of the hair cells to determine why the basal and apical hair cells differ in terms of susceptibility to free radicals.

The findings of the current study provide further support for the inclusion of high frequencies in the clinical monitoring of ototoxicity. The high frequency range (3284 to 5200 Hz) is crucial for speech perception. If a decrease in functioning in this range can be detected early, i.e., before it impacts negatively on an individual's speech perception, it would be possible to

University of Cape Town

provide early intervention to minimise cochlear damage and to optimise the individual's ability to communicate.

In addition to the above findings, the mid-range of frequencies (1636 to 3284 Hz) of DP I/O function thresholds deteriorated significantly over the eight-week period. This frequency range is similar to that of the minimal audibility curve (MAC) in humans, which indicates that the highest sensitivity to sound is in the 2 to 5 kHz region (Durrant & Lovrinic, 1995). Most speech sounds are concentrated in this region. As DP I/O function thresholds in this frequency range are elevated due to ototoxicity, it is very likely that speech perception could also be affected. Therefore, clinicians should take this into account by including these frequencies in their protocol for ototoxicity when managing individuals who receive ototoxic medication.

University of Cape Town

The relative sensitivity of pure tone thresholds and DP measures in detecting ototoxicity

DPgram absolute amplitude measures were able to detect deterioration in cochlear function earliest in this study, i.e., at least two weeks prior to pure tone thresholds and the DP I/O function thresholds. While previous research has compared the ability of pure tone thresholds and DPOAE measures in terms of the number of ears or individuals with cochlear abnormalities (Mulheran & Degg, 1997; Stavroulaki et al. 2002), most studies that used DPgram absolute amplitudes and DP I/O function thresholds to monitor ototoxicity did not report on the nature of exact time difference in detection of the two DP measures (i.e., DPgram absolute amplitudes vs. DP I/O function

thresholds) and pure tone thresholds (Berninger et al., 1995; Stavroulaki et al. 2002). Thus, no direct comparison can be made with the current study.

Based on the results of the current study, it can be suggested that DPgram absolute amplitudes, in conjunction with tympanometry (to rule out middle ear problems), have the potential to be used as the primary tools to monitor ototoxicity in individuals with normal hearing at baseline testing. The DPgram absolute amplitudes in this study detected cochlear abnormality first, as well as in the highest number of ears when compared to pure tone thresholds and DP I/O function thresholds. While these measures were not significant, it could possibly be that the small sample size constrained the findings. Nonetheless, DPgram measures could identify abnormal cochlear function earlier than the other two tests, before a sensorineural hearing loss due to aminoglycoside exposure develops.

University of Cape Town

However, currently, it is not known what reduction in DPgram absolute amplitude constitutes significant ototoxic damage, i.e., when will it be necessary to notify a doctor to consider altering medication. It is also not clear how much reduction in DPgram absolute amplitudes indicates possible permanent hearing loss, which will affect an individual's speech perception ability. Thus, further research is required to establish criteria for DPOAEs that would indicate clinically significant ototoxic damage, similar to the criteria for pure tone testing (ASHA, 1994; Konrad-Martin et al., 2005). This information will enable clinicians to use DPOAEs as a routine monitoring tool for ototoxic damage.

Apart from the fact that there is currently no protocol with criteria for ototoxic damage, the issue of DPOAEs not being a stand-alone measure to monitor ototoxicity also has to be considered. DPOAEs cannot be measured accurately in the presence of middle ear problems, thus, precluding the use of DPOAEs in this population (Hall, 2000; Robinette & Glattke, 2002). In order to rule out middle ear problems, DPOAEs have to be performed in conjunction with tympanometry (Hall; Robinette & Glattke). DPOAEs are also absent in individuals with pre-existing hearing loss of 40 dB HL or more (Konrad-Martin et al., 2005), which precludes the use of DPOAEs in ototoxicity monitoring in these individuals. In cases of pre-existing hearing loss, the clinician has to use pure tone audiometry to monitor cochlear damage due to aminoglycoside exposure. Therefore, the use of pure tone audiometry is essential in monitoring ototoxicity in individuals with middle ear problems and pre-existing hearing loss ≥ 40 dB HL.

University of Cape Town

In theory, the DP I/O function should be more sensitive to detecting abnormal cochlear function than the DPgram test (Hall, 2000). With DPgram measurement a moderate stimulus level is normally used (e.g., 65 dB SPL), which only provides information about cochlear function in response to a single intensity pair, while the DP I/O function thresholds are measured at a range of decreasing or increasing intensities for each frequency pair (Hall, 2000). Therefore, DP I/O functions yield more indicators of cochlear function (e.g., the absolute amplitudes, DP I/O function thresholds and dynamic range of DP I/O function) than the DPgram. Changes in these indicators over time

could potentially be used to monitor ototoxicity. However, similar to the DPgram, currently no criteria for DP I/O function measures exist for use in the monitoring of ototoxicity. Therefore, further research is required to establish criteria for DP I/O function measures that indicate clinically significant change in individuals receiving aminoglycoside treatment.

Previous research has not tested the suggestion that DP I/O function is more sensitive than DPgram absolute amplitudes in detecting abnormal cochlear function due to ototoxicity (at the time of the current study). This suggestion is, in fact, not supported by the current study, because DPgram absolute amplitudes were able to detect cochlear changes due to ototoxicity earlier and in more ears than the DP I/O function thresholds. The difference between the results of the current study and Halls' (2000) suggestion that the DP I/O function should be a more sensitive test than the DPgram test to detect abnormal cochlear function, could possibly be due to measurement errors. The possibility of measurement errors could account for the outcome of the current study not supporting Hall's suggestion. Thus, a similar study with strict measurement criteria comparing the ability of the two DP measures to detect deterioration in cochlear function needs to be conducted with a larger sample, to contextualise the findings of the current study and to evaluate Hall's suggestion. The measurement criteria ought to include that all DP I/O function measures have to be replicable and therefore, have to be measured at least twice in the same recording session; a lower noise floor should be regarded as acceptable before recording is stopped, e.g., < - 20 dB SPL; stringent calibration procedures as specified by Dreisbach and Siegel (2001) should be

University of Cape Town

employed to minimise calibration artefacts. Such research could be beneficial to the efficient clinical monitoring and early detection of ototoxicity, if the DP measure most sensitive to change in cochlear function could be determined.

Another possible reason for the difference between Hall's suggestion and the current research outcome could be that the lowest stimulus intensity used in the current study was L_2 = 45 dB SPL due to equipment limitations. DP I/O function thresholds below this level could not be monitored, and any deterioration in DP I/O function thresholds due to aminoglycoside exposure that occurred at stimulus intensities with L_2 lower than 45 dB SPL could have been concealed. Therefore, future research with a wider stimulus range, e.g., L_2 between 20 and 70 dB SPL, is recommended in order to monitor DP I/O function thresholds at lower intensities. The use of lower stimulus intensities might enable the researcher to evaluate deterioration of cochlear function with DP I/O function thresholds more accurately, and this could result in an outcome that is different from the current study when comparing the DPgram and DP I/O function threshold.

University of Cape Town

The different measures of cochlear function, i.e., pure tone testing, DPgram and DP I/O function thresholds, address different aspects of the ototoxic effect. The results of the pure tone testing reflect sensorineural hearing loss, which is perceived by the individual as "poorer" hearing, which occurs after OHC damage has occurred. Thus, a smaller number of participants had sensorineural hearing loss relative to the number that had abnormal or

decreased DPOAE measures which signals hair cell damage ahead of perceptible hearing loss.

DP I/O function slopes

Participants with normal hearing as per pure tone thresholds

In the current study, the slopes of DP I/O functions at 818, 1636, 2454 and 3284 Hz in those participants exposed to ototoxic drugs, but who did not demonstrate hearing loss, were predominantly non-linear for the L_2 intensity range of 55 to 65 dB SPL. Hence, those participants exposed to aminoglycosides who did not develop ototoxic hearing loss had DP I/O function slopes and therefore, cochlear function similar to that of normal ears in the Kummer et al. and Dorn et al. studies in the low frequencies. However, the current study found linear DP I/O function slopes in the high frequencies at 4102 and 4919 Hz at most stimulus intensities, thus, suggesting that cochlear function did no longer demonstrate compressive behaviour at the high frequencies. The implication may be that the OHCs in the basal area of the cochlea responsible for processing high frequencies may have been affected by the ototoxic exposure. This change would be consistent with the literature on ototoxicity, i.e., that high frequency OHCs are damaged before those responsible for processing low frequencies (Barclay & Begg, 1994; Dreisbach & Siegel, 2001; Fausti et al., 1984). Kummer et al. and Dorn et al. reported compressive DP I/O function slopes at these moderate intensities. A reason for this discrepancy could be that the participants of the current study were exposed to aminoglycosides during the study, whereas Kummer et al. and Dorn et al.'s normal-hearing participants were not.

University of Cape Town

The frequencies of DP I/O function slopes that indicate abnormal cochlear function (4102 and 4919 Hz) are similar to the DPgram absolute amplitudes frequencies affected. This agreement increases the validity for using DP I/O function slopes in evaluating cochlear function.

However, current information available on DP I/O function slopes is limited. For example, Stover et al. (1996) reported varied overall shapes of DPOAE I/O function slopes with a fixed 10 dB stimulus level difference in 103 normal-hearing participants. The shapes included: non-monotonic (compressive), saturated (no increase in DP amplitude with increase in stimulus intensity) and continual growth for even the highest stimulus levels. Therefore, it can be deduced that variability does exist in DP I/O function slopes in normally functioning cochleas when a stimulus level difference of 10 dB is used. Stover et al.'s results differ from the non-linear DP I/O function slopes of Kummer et al. (1998) and Dorn et al., and the linear DP I/O function slopes of Dreisbach and Siegel (2001). The discrepancy in results could arise from the differences in intensity between L_1 and L_2 used in three of the four studies. Kummer et al. used the formula $L_1 = 0.42L_2 + 39$ to calculate the $L_1 - L_2$ difference and Dreisbach and Siegel employed an L_1-L_2 difference of 15 dB. It is clear that large-sample investigations, with systematic control of variables, e.g., stimulus intensity and $L_1 - L_2$ difference, on the DP I/O function slopes are needed to establish normative data. Until there are norms, it may not be feasible to use DP I/O function slopes clinically.

University of Cape Town

According to Dreisbach and Siegel (2001), discrepancies in DP I/O function results may possibly be ascribed to calibration artefacts, which could influence all DPOAE amplitude levels. "These artefacts can produce differences in the levels of (otoacoustic) emissions as large as 20 dB" (p. 2467). The authors suggested that employing a consistent definition for the input level to the ear (i.e., the eardrum SPL) could improve the consistency of DPOAE levels when compared with the conventional calibration procedure utilising the DPOAE probe in the participant's ear. (Refer to Dreisbach and Siegel for a detailed description of calibration artefacts). Thus, a limitation of the current study was that a stringent calibration procedure was not employed, and could have resulted in calibration artefacts influencing the results.

Participants who developed hearing loss as per pure tone thresholds

The current study found linear DP I/O function slopes at 4919 Hz at 40 and 55 to 65 dB SPL in those participants exposed to ototoxicity, who developed hearing loss. This finding indicates a lack of compressive function of the cochlea at the highest frequency tested. This result is similar to the findings of Kummer et al. (1998) in their 15 participants with hearing loss, and that of Dorn et al. (2001) in 84 ears (50 participants). However, at the remaining frequencies 818, 1636, 2454, 3284 and 4102 Hz, the non-linear DP I/O function slopes found in the current study is in direct contrast with the results of Kummer et al. and Dorn et al. They reported linear DP I/O function slopes, indicating non-compressive cochlear function in all participants with hearing loss for the intensity range of L_2 between 30/40 and 60/70 dB SPL at all

University of Cape Town

frequencies. In contrast, the current study reports non-linear slopes for low frequencies, but linear slopes at the highest frequency (4919 Hz). Although this finding is not consistent with other research on DP I/O function slopes in individuals with hearing loss at least the highest frequency DP I/O function slope reflected loss of cochlear compressive function. This finding is in line with the theory that aminoglycosides affect the high frequencies first. Therefore, one would expect DP I/O function slopes to reflect abnormal (compressive) cochlear function for the high frequencies prior to abnormal low frequencies.

A possible reason for the different results between the current and the Kummer et al. (1998) and Dorn et al. (2001) studies could be that ototoxic exposure predisposes OHCs in the basal area to be affected first and, thus, the high frequencies may be differentially affected relative to the lower frequencies at the beginning of an ototoxic hearing loss. While Kummer et al. and Dorn et al. reported on sensorineural hearing loss, it was not ototoxic in origin. In addition, calibration artefacts may have contributed to the discrepancy in findings. Also, the results of the current study are based on three participants who developed hearing loss, whereas the Kummer et al. study included 15 participants with hearing loss, and Dorn et al. had 50 participants.

Although DP I/O function slopes may be used to obtain information on cochlear functioning at high vs. low frequencies, clinical use of the DP I/O function slopes await the development of normative data.

University of Cape Town

As DP I/O function slopes seemed to detect abnormal cochlear function before conventional pure tone audiometry and DPgram absolute amplitudes, the slope information may have potential as an indicator of risk for hearing loss due to aminoglycosides. The limited time frame for the current study did not allow for abnormal DPOAEs at high frequencies to be monitored further to determine whether the DP I/O function slope indication of decreased non-linearity, i.e., reduced cochlear compression at high frequencies in 1) those participants who had normal hearing was associated with later onset of hearing loss; and in 2) those participants who had hearing loss whether abnormal cochlear function, i.e., reduced cochlear compression manifested in the lower frequencies in the long term. As ASHA (1994) has suggested, the effects of ototoxicity may manifest up to six months post termination of aminoglycoside administration. Therefore, research is needed to determine whether abnormal cochlear function, as indicated by linear DP I/O function slopes at moderate stimulus intensity levels, translate into sensorineural hearing loss with exposure to aminoglycosides.

University of Cape Town

DP I/O function stimulus parameters

DP I/O function thresholds for high frequencies (3284, 4102 and 4919 Hz) were found to identify more cochlear abnormalities than the low frequencies (818, 1636 and 2454 Hz) in the current study, which is similar to previous research findings in animals (Mills, Loos & Henley, 1999) and humans (Stavroulaki et al., 2002). The frequency range of DP I/O function thresholds that are affected is similar to the DPgram absolute amplitudes frequencies

that reflected abnormal cochlear function. This similarity indicates consistency with regard to the pattern of cochlear abnormalities detected by DPOAE measures.

These DP I/O function threshold results are similar to the DPgram absolute amplitudes and DP I/O function slope information of this study with reference to the frequency range affected. Therefore, this agreement increases the validity for using high frequency DP I/O function thresholds in evaluating cochlear function.

Clinically it may be useful to include only these DP I/O function threshold frequencies (3284, 4102 and 4919 Hz) when monitoring ototoxicity with individuals with no prior problems of cochlear function. Using fewer frequencies will reduce the overall test time of the DP I/O function. The possibility of using fewer frequencies (i.e., only high frequencies ≥ 3284 Hz) needs to be researched with a larger sample to determine which frequencies best identify ototoxicity. However, even if fewer frequencies are used, and test time is reduced, the DP I/O function test would still take at least three times as long as the DPgram (if three frequencies are used), and therefore, it may still not be a clinically viable test in routine ototoxicity monitoring. Thus, further research needs to be conducted with the DP I/O function to determine whether the number of test frequencies can be limited, without compromising its potential sensitivity to detect ototoxicity.

University of Cape Town

The maximum DP I/O function thresholds were between 60 and 75 dB SPL Hz over the eight-week monitoring period. These thresholds indicate that only at 4102 and 4919 Hz would it be necessary to use the stimulus intensity of 75 dB SPL. If the stimulus intensity range can be decreased the test time for DP I/O functions could be reduced. A reduction in test time could contribute to making the DP I/O function a clinically feasible measure that could be used to monitor ototoxicity in individuals receiving aminoglycosides. Further research with a larger sample than the current study needs to be conducted to obtain more representative results. The current study only included 17 ears, and therefore, these results may not be truly representative of the population with TB receiving aminoglycosides.

The minimum DP I/O function threshold at all frequencies was ≤ 45 dB SPL. Due to the fact that the lowest stimulus intensity used in the current study was L_2 = 45 dB SPL, deterioration of DP I/O function thresholds could not be monitored below 45 dB SPL due to equipment limitations. Some studies have found DP I/O function thresholds at ≤ 20 dB SPL for individuals with normal hearing, as well as those with hearing impairment (Dorn et al., 2001; Kummer et al, 1998). Monitoring the deterioration or progression of these DP I/O function thresholds over time has yet to be researched. A recommendation would be for future research to include lower stimulus intensities in the determining of DP I/O function thresholds, e.g., L_2 = 20 dB SPL, in order to monitor DP I/O function threshold deterioration over time with exposure to ototoxic medication.

University of Cape Town

Findings of the current study suggest that the DPgram test may be the most sensitive test to monitor ototoxicity. However, the measurement of DPOAEs is confounded by the presence of outer and middle ear problems (Hall, 2000). Thus, when including the DPgram test in the monitoring of ototoxicity, it is essential that tympanometry be used concurrently, which has financial implications for such a monitoring protocol. Most pure tone audiometry set-ups have to have tympanometry as well to conduct routine hearing tests. In the case of a hearing loss exceeding 40 dB HL the use of DPOAEs are precluded (Konrad-Martin et al., 2005), and pure tone audiometry will be the test of choice in this population with pre-existing hearing loss. The presence of noise (internal and external) can influence accurate OAE measures (Robinette & Glattke, 2002), which could be problematic with individuals suffering from TB who breathe heavily. Therefore, it is important to minimise noise sources when using OAEs as an monitoring tool for ototoxicity. The concurrent use of tympanometry with the DPgram test (provided that noise is kept to a minimum) may prove to be a sensitive battery for the clinical monitoring of ototoxicity. If individuals with cochlear damage are identified early enough before speech perception is affected, the cost to the government may be less for management of a person with a permanent hearing loss.

In a country like South Africa, with limited resources, OAE equipment is not always readily available. Where it is not possible to conduct OAE testing as part of a protocol for monitoring ototoxicity, conventional pure tone audiometry can be used as an alternative monitoring tool. When hearing loss is detected with pure tone audiometry, it must be kept in mind that if DPOAEs were used,

University of Cape Town

abnormal cochlear function could have been detected at least two weeks prior to detection with pure tone audiometry. Therefore, immediate medical intervention will be required in terms of altering or ceasing the ototoxic medication, in order to prevent permanent hearing loss.

Conclusion

Tuberculosis in South Africa has been declared a national health priority by the Minister of Health (30 July, 2004), because the disease is reaching epidemic proportions in certain areas (e.g., Western Cape). Due to multi-drug resistance, many individuals with TB receive ototoxic medication as part of their treatment, which can significantly affect their hearing. Thus, audiologists should devote sufficient attention to the hearing needs of individuals with TB, especially those receiving ototoxic medication as part of their treatment.

University of Cape Town

While different methods are available to the audiologist to monitor ototoxicity, which include conventional pure tone audiometry, high frequency audiometry and OAEs, this study found that DPgram absolute amplitudes appear to detect abnormal cochlear function due to exposure to ototoxic medication at least two weeks before conventional pure tone audiometry. Therefore, it can be recommended that the measurement of DPgram absolute amplitudes be the preferred method of testing to monitor ototoxicity in individuals with normal hearing and middle ear function at the baseline test. Apart from being able to detect cochlear abnormality earliest, this recommendation also stems from the fact that the DPgram is a quick and objective method for detecting abnormal cochlear function (Hall, 2000), which makes it an ideal test to use with sick

individuals, e.g., patients with TB receiving ototoxic medication, who may not be able to actively participate in a test like pure tone audiometry (Konrad-Martin et al., 2005).

Constraints regarding the clinical application of DPOAEs, including DPgram absolute amplitudes, DP I/O function thresholds and slope information, are the lack of norms and criteria to indicate cochlear damage due to exposure to aminoglycosides. Therefore, further research is needed before these measures can be meaningful in a clinical setting in order to detect ototoxicity.

As healthcare professionals, it is of the utmost importance that audiologists preserve hearing ability in individuals with TB, thereby enhancing their quality of life. It is thus, crucial to have an evidence-based practice approach in monitoring the cochlear function of individuals who receive ototoxic medication to provide the most efficient audiological care.

University of Cape Town

References

Abdala, C., & Sininger, Y. (1996). The development of cochlear frequency resolution in the human auditory system. *Ear and Hearing, 17(5)*, 374-385.

American Speech-Language-Hearing Association. (1994, March). Guidelines for the audiological management of individuals receiving cochleotoxic drug therapy. *ASHA, 36* (Suppl. 12), pp. 11-19.

Aran, J., Erre, J., Da Costa, D.L., Debbarh, I. & Dulon, D. (1999). Acute and chronic effects of aminoglycosides on cochlear hair cells. *Annals of the New York Academy of Sciences, 884,* 60-68.

Arnold, D.J., Lonsbury-Martin, B.L. & Martin, G.K. (1999). High-frequency hearing influences lower-frequency distortion-product otoacoustic emissions. *Archives of Otolaryngology, 125,* 215-222.

Arslan, E., Orzan, E. & Santarelli, R. (1999). Global problem of drug-induced hearing loss. *Annals of the New York Academy of Sciences, 884*, 1-14.

Barclay, M. & Begg, E. (1994). Aminoglycoside toxicity and relation to drug regimen. *Adverse Drug React. Toxicol. Rev., 13* (4), 207-234.

University of Cape Town

Beattie, R.C., & Bleech, J. (2000). Effects of sample size on the reliability of noise floor and DPOAE. *British Journal of Audiology, 34 (5).* 305-309.

Bennett, C. (1996). The Aminoglycosides. Primary Care Update on Obstetrics and Gynaecology, 3 (6), 186-191.

Berlin, C. (Ed.) (1998). *Otoacoustic emissions: Basic Science and Clinical Applications.* London: Singular Publishing.

Berninger, E., Karlsson, K., Hellgren, U. & Eskilsson, G. (1995). Magnitude changes in transient evoked otoacoustic emissions and high-level 2f1-f2 distortion products in man during quinine administration. *Scandinavian Audiology, 24* (1), 27-32.

Bess, F. & Humes, L. (1995). *Audiology: the fundamentals.* Baltimore: Williams and Wilkins

Brownell, W. E. (1990). Outer hair cell electromotility and otoacoustic emissions. *Ear and Hearing, 11,* 89-92.

Bykhovskaya, Y., Mengesha, E., Wang, D., Yang, H., Estivill, X., Shohat, M., & Fischel-Ghodsian, N. (2004). Phenotype of non-syndromic deafness associated with the mitochondrial A1555G mutation is modulated by

University of Cape Town

mitochondrial RNA modifying enzymes MTO1 and GTPBP3. *Molecular Genetics and Metabolism, article in press.*

Campbell, K.C.M., & Durrant, J. (1993). Audiologic monitoring for ototoxicity. *The Otolaryngologic Clinics of North America, 26 (5),* 903-914.

Cohen, S.M., Labadie, R.F., Dietrich, M.S., & Haynes, D.S. (2004). Quality of life in hearing-impaired adults: The role of cochlear implants and hearing aids. *Otolaryngology – Head and Neck Surgery, 131 (4),* 413-421.

Department of Health (1999). *The management of multidrug-resistant tuberculosis in South Africa* (2nd ed.). Pretoria: Department of Health.

Dorn, P.A., Konrad-Martin, D., Neely, S.T., Keefe, D.H., Cyr, E., & Gorga, M.P. (2001). Distortion product otoacoustic emission input/output functions in normal-hearing and hearing-impaired human ears. *Journal of the Acoustical Society of America, 110(6),* 3119-3131.

Dreisbach, L. & Siegel, J. (2001). Distortion-product otoacoustic emissions measured at high frequencies in humans. *Journal of the Acoustical Society of America, 110* (5), 2456-2468.

Dreschler, W. A., Van der Hulst, R. J., Tange, R. A. & Urbanus, N. A. M. (1985). The role of high frequency audiometry in early detection of ototoxicity. *Audiology, 24,* 387-395.

University of Cape Town

Durrant, J. D. & Lovrinic, J. H. (1995). *Bases of hearing science* (3rd ed.). Baltimore: Williams & Wilkins.

Dye, C., Espinal, M.A., Watt, C.J., Mbiaga, C., & Williams, B.G. (2002). Worldwide incidence of multi-drug resistant tuberculosis. *The Journal of Infectious Diseases, 185,* 1197-1202.

Fätkenhauer, G., Taelman, H., Lepage, P., Schwenk, A. & Wenzel, R. (1999). The return of tuberculosis. *Diagnostic Microbiology of Infectious Diseases, 34,* 139-146.

Fausti, S.A., Frey, R.H., Henry, J.A., Olson, D.J., & Shaffer, H.I. (1993). High-frequency testing techniques and instrumentation for early detection of ototoxicity. *Journal of Rehabilitative Research and Development, 30(3),* 333-341.

Fausti, S.A., Rappaport, B.Z., Schechter, M.A., Frey, B.S., Ward, T.T. & Brummett, R.E. (1984). Detection of aminoglycoside ototoxicity by high frequency auditory evaluation: Selected case studies. *American Journal of Otolaryngology, 5* (3), 177-182.

Franklin, D.J., McCoy, M.J., Martin, G.K., & Lonsbury-Martin, B.L. (1992). Test-retest reliability of distortion-product and transiently evoked otoacoustic emissions. *Ear and Hearing, 13,* 417-429.

University of Cape Town

Frieden, T., Sterling, T., Pablos-Mendez, A., Kilburn, J. Cauthen, M. & Dooley, S. (1993). The Emergence of Drug-Resistant Tuberculosis in New York City. The *New England Journal of Medicine, 328* (8), 521-526.

Fourie, B. (n.d.) The burden of tuberculosis in South Africa. Retrieved July 16, 2004, from http://www.sahealthinfo.org/ tb/tbburden.htm.

Gaskill, S. & Brown, A. (1993). Comparing the level of the acoustic distortion product 2f1-f2 with behavioural threshold audiograms from normal-hearing and hearing-impaired ears. *British Journal of Audiology, 27,* 397-407.

Gelfand, S. (1997). *Essentials of Audiology*. New York: Thieme

GSI 38 Manual (2003). Madison: Grason-Stadler

Hall, J. W. (2000). *Handbook of Otoacoustic Emissions.* California: Singular Publishing Group

Harris, F. P. (1990). Distortion-Product otoacoustic emissions in humans with high frequency sensorineural hearing loss. *Journal of Speech and Hearing Research, 3,* 594-600.

University of Cape Town

Harris, F.P., Lonsbury-Martin, B.L., Stagner, B.B., Coats, A.C., & Martin, G.K. (1989). Acoustic distortion products in humans: Systematic changes in amplitude as a function of f_2/f_1 ratio. *Journal of the Acoustical Society of America, 85 (1),* 220-229.

Hashino, E., Tinitan, E. & Salvi, R. (1995). Base-to-apex gradient of cell proliferation in the chick cochlea following kanamycin-induced hair cell loss. *Hearing Research, 88,* 156-198.

Henley, C.M., Weatherly, R.A., Martin, G.K., & Lonsbury-Martin, B. (1996). Sensitive developmental periods for kanamycin ototoxicity effects on distortion-product otoacoustic emissions. *Hearing Research, 98,* 93-103.

Herselman, S. (2002). 'An epidemic within an epidemic': the case of multidrug-resistant tuberculosis. Anthropology Southern Africa, 25 (3 & 4).

Hotz, M., Harris, F. & Probst, R. (1994). Otoacoustic emissions: An approach for monitoring aminoglycoside-induced ototoxicity. *Laryngoscope, 104,* 1130-1134.

Howell, D.C. (1999). *Fundamental statistics for the behavioural sciences (4th ed.).* California: Brooks/Cole Publishing.

University of Cape Town

ILO OAE instrument user manual (1992). Hertfordshire, U.K.: Otodynamics Ltd.

Jaramillo, E. (1999). Encompassing treatment with prevention: the path for a lasting control of tuberculosis. *Social Science and Medicine, 49,* 393-404.

Kakigi, A., Hirakawa, H., Harel, N., Mount, R. & Harrison, R. (1998). Basal cochlear lesions result in increased amplitude of otoacoustic emissions. *Audiology & Neuro-otology, 3*, 361-372.

Katz, J. (2002). (ed.) *Handbook of Audiology. 5th ed.* Lippincott Williams & Wilkins: Philadelphia

Katzenellenbogen, J.M., Joubert, G., & Abdool Karim, S.S. (1997). *Epidemiology: A manual for South Africa.* Oxford University Press: Cape Town.

Kemp, D. (1978). Stimulated acoustic emissions from within the human auditory system. *Journal of the Acoustical Society of America, 64* (5), 1386-91.

Khan, M., Kinsara, A., Osoba, A., Wali, S., Samman, Y. & Memish, Z. (2001). Increasing resistance of M. tuberculosis to anti-TB drugs in Saudi Arabia. *International Journal of Antimicrobial Agents, 17,* 415-418.

University of Cape Town

Kironde, S. (2000). Tuberculosis (Chapter 17). *South African Health Review 2000.* Durban: The Press Gang.

Kon, K., Inagaki, M., & Kaga, M. (2000). Developmental changes of distortion product and transient evoked otoacoustic emissions in different age groups. *Brain and Development, 22,* 41-46.

Konrad-Martin, D., Helt, W.J., Reavis, K.M., Gordon, J.S., Coleman, L.L., Bratt, G.W., & Fausti, S.A. (2005). Ototoxicity: Early detection and monitoring. *The ASHA Leader, 10 (7).*

Kucers, A. & Bennett, N. (1987). *The use of antibiotics: a comprehensive review with clinical emphasis.* London: William Heinemann Medical Books.

Kummer, P., Janssen, T., & Arnold, W. (1998). The level and growth behavior of the 2 *f*1-*f*2 distortion product otoacoustic emission and its relationship to auditory sensitivity in normal hearing and cochlear hearing loss. *Journal of the Acoustical Society of America, 103 (6),* 3431-3444.

Le Calvez, S., Avan, P., Gilain, L., & Romand, R. (1998). CD1 hearing-impaired mice. I: Distortion product otoacoustic emission levels, cochlear function and morphology. *Hearing Research, 120,* 37-50.

University of Cape Town

Lerner, S.A., & Matz, G.J. (1979). Suggestions for monitoring patients during treatment with aminoglycoside antibiotics. *Otolaryngology, Head and Neck Surgery, 87,* 222-228.

Lonsbury-Martin, B., Martin, G., Probst, R., & Coats, A. (1987). Acoustic distortion products in rabbit ear canal. I. Basic features and physiological vulnerability. *Hearing Research, 28,* 173-189.

Lonsbury-Martin, B.L., & Martin, G.K. (1990). The clinical utility of distortion product otoacoustic emissions. *Ear and Hearing, 15,* 199-209.

Lopez-Gonzalez, M.A., Delgado, F., & Lucas, M. (1999). Aminoglycosides activate oxygen metabolites production in the cochlea of mature and developing rats. *Hearing Research, 136,* 165-168.

Marean, G.C., Cunningham, D., Burt, J.M., Beecher, M.D., & Rubel, E.W. (1995). Regenerated hair cells in the European starling: Are they more resistant to kanamycin than original hair cells? *Hearing Research, 82,* 267-276.

Medical Research Council of South Africa (2002). *Guidelines on ethics for medical research: General Principles.* 4th ed.

University of Cape Town

Mills, C. D., Loos, B. M., & Henley, C. (1999). Increased susceptibility of male rats to kanamycin-induced cochleotoxicity. *Hearing Research, 128*, 75-79.

Mills, D. & Rubel, E. (1996). Development of the cochlear amplifier. *Journal of the Acoustical Society of America, 100 (1)*, 428-441.

Miyakita, T., Ueda, A., Zusho, H., & Kudoh, Y. (2002). Self-evaluation scores of hearing difficulties and quality of life components among retired workers with noise-related hearing loss. *Journal of Sound and Vibration, 250 (1)*, 119-128.

Moore, R.D., Smith, C.R., & Lietman, P.S. (1984). Risk factors for the development of auditory toxicity in patients receiving aminoglycosides. *Journal of Infectious Diseases, 149 (1),* 23-30.

Mukherjee, J., Rich, M., Socci, A., Joseph, K., Alcántara Virú, F., Shin, S., Furin, J., Becerra, M., Barry, D., Yong Kim, J., Bayona, J., Farmer, P., Smith Fawzi, M. & Seung, K. (2004). Programmes and principles in treatment of multidrug-resistant tuberculosis. *Lancet, 363*, 474-481.

Mulrow, C.D., Aguilar, C., Endicott, J.E., Velez, R., Tuley, M.R., Charlip, W.S., & Hill, J.A. (1990). Association between hearing impairment and the quality of life of elderly individuals. *Journal of American Geriatrics Society, 38,* 45-50.

University of Cape Town

Mulheran, M., & Degg, C. (1997). Comparison of distortion product OAE generation between a patient group requiring frequent gentamicin therapy and control participants. *British Journal of Audiology, 31*, 5-9.

Nicol, K., Hackney, C., Evans, E., & Pratt, S. (1992). Behavioural evidence for recovery of auditory function in guinea pigs following kanamycin administration. *Hearing Research, 61,* 117-131.

Ozturan, O., & Lam, S. (1998). The effect of hemodialysis on hearing using pure tone audiometry and distortion-product otoacoustic emissions. *Journal for Oto-Rhino-Laryngology and its Related Specialities, 60 (6),* 306-313.

PAWC (2003). *TB Control Programme: Annual report 2003.* Directorate: HIV/AIDS/STI/TB.

Popelka, G.R., Osterhammel, P.A., Nielsen, L.H., & Rasmussen, A.N. (1993). Growth of distortion product otoacoustic emissions with primary-tone level in humans. *Hearing Research, 71 (1-2),* 12-22.

Probst, R., Harris, F.P., & Hauser, R. (1993). Clinical monitoring using otoacoustic emissions. *British Journal of Audiology, 27, (2),* 85-90.

University of Cape Town

Probst, R., Lonsbury-Martin, G., & Martin, G. (1991). A review of otoacoustic emissions. *Journal of the Acoustical Society of America, 89(5),* 2027-2067.

Robinette, M.S., & Glattke, T.J. (Eds.) (2002). *Otoacoustic emissions: Clinical applications. 2nd ed.* New York: Thieme.

Roland, P.S. (2004). New developments in our understanding of ototoxicity. *ENT-Ear, Nose & Throat Journal (4),* 15-17.

Ruben, R. J. (2000). Redefining the survival of the fittest: Communication disorders in the 21st century. *The Laryngoscope, 110,* 241-245.

Ruggero, M.A., Rich, N.C., Recio, A., Narayan, S.S., & Robles, L. (1997). Basilar membrane responses to tones at the base of the chinchilla cochlea. *Journal of the Acoustical Society of America, 101.* 2151-2163.

Satoh, Y., Kanzaki, J., O-Uchi, T., & Yoshihara, S. (1998). Age-related changes in transiently evoked otoacoustic emissions and distortion product otoacoustic emissions in normal-hearing ears. *Auris Nasus Larynx, 25,* 121-130.

Sha, S. & Schacht, J. (1997). Prevention of aminoglycoside-induced hearing loss. *Keio Journal of Medicine, 46 (3),* 115-119.

University of Cape Town

Sha, S., Taylor, R., Forge, A., Schact, T. (2001). Differential vulnerability of basal and apical hair cells is based on intrinsic susceptibility to free radicals. *Hearing Research, 155,* 1-8.

Siegel, J. (1994). Ear canal standing waves and high frequency sound calibration using otoacoustic emission probes. *Journal of the Acoustical Society of America, 95,* 2589-2597.

Siegel, S., & Castellan, N.J., (1988). *Nonparametric statistics for the behavioural sciences.* New York: McGraw-Hill.

Song, B., Sha, S., & Schacht, J. (1998). Iron chelators protect from aminoglycoside-induced cochleo- and vestibulotoxicity. *Free radical Biology and Medicine, 25* (2), 189-195.

Sprinthall, R. C. (1987). *Basic statistical analysis.* New Jersey: Prentice-Hall

Stach, B. (2003). (Ed.). *Comprehensive dictionary of audiology: Illustrated. 2nd ed.* Delmar Learning: New York.

Stavroulaki, P., Apostolopoulos, N., Dinoupoulo, D., Vossinakis, I., Tsakanikos, M., & Douniadakis, D. (1999). Otoacoustic emissions: An approach for monitoring aminoglycoside induced ototoxicity in children. *International Journal of Paediatric Otorhinolaryngology, 50,* 177-184.

University of Cape Town

Stavroulaki, P., Vossinakis, I., Dinopoulou, D., Doudounakis, S., Adamopoulus, G., & Apostopoulus, N. (2002). Otoacoustic emissions for monitoring aminoglycoside-induced ototoxicity in children with cystic fibrosis. *Archives of Otolaryngology- Head & Neck Surgery, 128(2)*, 150-155.

Stover, L. J., Gorga, M. P., Neely, S. T., & Montoya, D. (1996). Toward optimising the clinical utility of distortion product otoacoustic emission measurements. *Journal of the Acoustical Society of America, 100*, 968-977.

Suckfüll, M., Schneeweiß, S., Dreher, A., & Schorn, K. (1996). Evaluation of TEOAE and DPOAE measurements for the assessment of auditory thresholds in sensorineural hearing loss. *Acta Otolaryngologica, 116,* 528-533.

Tabachnick, B.G., & Fidell, L.S. (2001). *Using multivariate statistics. 4th ed.* Boston: Allyn and Bacon.

Tlumak, A.I., & Kilney, P.R. (2001). Parameters that affect the measurement of otoacoustic emissions. *Current Opinion in Otolaryngology and Head and Neck Surgery, 9,* 279-283.

University of Cape Town

Trautwein, P., Hofstetter, P., Wang, J., Salvi, R., & Nostrant, A. (1996). Selective inner hair cell loss does not alter distortion product otoacoustic emissions. *Hearing Research, 96,* 71-82.

Van Rie, A., Warren, R., Richardson, M., Gie, R., Enarson, D., Beyers, N., & Van Helden, P. (2000). Classification of drug-resistant tuberculosis in an epidemic area. *Lancet, 356,* 22-25.

Vasquez, R., & Mattucci, K.F. (2003). A proposed protocol for monitoring ototoxicity in patients who take cochleo- or vestibulotoxic drugs. *Ear, Nose and Throat Journal, 82(3)*, 181-184.

Vaughan, N.E., Fausti, S.A., Celius, S., Phillips, D., Helt, W., & Henry, J.A. (2002). An efficient test protocol for identification of a limited, sensitive frequency test range for early detection of ototoxicity. *Journal of Rehabilitation Research and Development, 39(5)*, 567-574.

Voogt, G. (1987). Early detection of ototoxicity by high frequency audiometry – a case study. *South African Journal of Communication Disorders, 34,* 67-70.

Voogt, G., & Schoeman, H. (1996). Ototoxicity of aminoglycoside drugs in tuberculosis treatment. *South African Journal of Communication Disorders, 43,* 3-6.

University of Cape Town

Wallenstein, S., Zucker, C., & Fleiss, J. (1980). Some statistical methods useful in circulation research. *Circulation Research, 47 (1)*, 1-9.

Wang, S., Bian, Q., Liu, Z., Feng, Y., Lian, N., Hui, C., Hu, C., Dong, Y., & Cai, Z. (1999). Capability of serum to convert streptomycin to cytotoxin in patients with aminoglycoside-induced hearing loss. *Hearing Research, 137,* 1-7.

Weyer, K. (1997). *Tuberculosis.* Accessed online at: http://www.hst.org.za/sahr/97/chap22/htm

Wu, W., Sha, S., McLaren, J.D., Kawamoto, K., Raphael, Y., & Schacht, J. (2001). Aminoglycoside ototoxicity in adult CBA, C57BL and BALB mice and the Sprague-Dawley rat. *Hearing Research, 158,* 165-178.

Zhao, F., & Stephens, D. (1999). Test-retest variability of distortion-product otoacoustic emissions in human ears with normal hearing. *Scandinavian Audiology, 28,* 171-178.

Zwarenstein, M., Schoeman, J.H., Vundule, C., Lombard, C.J., & Tatley, M. (1998). Randomised controlled trial of self-supervised and directly observed treatment of tuberculosis. *Lancet, 352,* 1340-43.

University of Cape Town

Appendix A-1

English Subject Information Sheet

Streptomycin is one of the drugs that are administered to tuberculosis (TB) sufferers. Your doctor has decided that your TB can best be treated by streptomycin. Streptomycin is known to be damaging to the inner ear, and, thus, can cause damage to your hearing. Because you can lose your hearing it is important to check your hearing ability and detect any hearing loss as soon as possible.

The aim of this study is to determine whether a test called otoacoustic emissions test can detect ear damage sooner than the regular hearing test. If you agree to participate in the study you will be required to undergo one hearing test before you begin treatment with streptomycin, as well as every 2 weeks for a two-month period afterwards. It will thus, be able to monitor your ears and hearing for that period. If any changes in hearing are detected, you, as well as your doctor, will be notified immediately, so that the doctor, together with you, can make decisions regarding your treatment. All the tests will be done in the Audiology section at Brooklyn Chest Hospital.

University of Cape Town

Before the start of each test I will give you instructions and explain the test to you. If you are unsure of what to do, you can ask a question at any stage. The whole test procedure will take approximately 40 minutes per session. None of these tests are harmful to you in any way. The 4 tests are as follows:

(a) I will look into your ear with a light.

(b) Then I will place a probe into your ear that will measure how well your middle ear is working. You will not have to do anything but remain quiet for the test duration.

(c) Then your hearing will be tested. After the earphones have been placed on your ears you will be alone in the sound-treated room. You will still be able to communicate with me, though. You will hear sounds through the earphones and one ear at a time will be tested. Most of the sounds will be very soft, but you will have to raise your hand

whenever you hear the sound. The aim of this test is to see what the softest sounds are that you can hear. None of the above tests will be uncomfortable.

(d) After the regular hearing test you will get a break of 5 minutes. Then otoacoustic emission tests will be done, which involves putting a probe into your ear, which might be mildly uncomfortable, but not painful. The probe sends sounds into the ear and then measures the responses of the inner ear. You do not have to do anything but remain quiet for the duration of the test.

All the information obtained will remain confidential and form part of your hospital record, because you will not be identified in any publications from this study. You are free to withdraw from the study at any stage, without your normal treatment being affected in any way. The overall results of the study will be made known to the superintendent of the hospital.

A benefit from taking part in this study is that if you begin to show signs of hearing loss you and your doctor will be advised in order to attempt to minimize the effects of ear damage caused by streptomycin/kanamycin. There are no risks in participation in the study.

If you have any questions, you are free to ask them now or at any time during the study.

Researcher:	Lucretia Petersen
Contact telephone number:	(021) 404 6074
Address:	Division of Communication Sciences and Disorders
	E47 Old Main Building
	Groote Schuur Hospital
	Observatory
	7925

University of Cape Town

Appendix A2

Afrikaans Subject Information Sheet

Streptomycin is een van die medikasies wat gebruik word om tuberkulose (TB) te behandel. U dokter het besluit dat u TB die beste met streptomycin behandel kan word. Dit is bekend dat streptomycin skadelik is vir die binne-oor en 'n moontlike gehoorverlies kan veroorsaak. Omdat u 'n gehoorverlies kan opdoen is dit belangrik om u gehoor to monitor en enige gehoorverlies so gou as moontlik op te spoor.

Die doel van hierdie studie is om te bepaal of 'n toets genaamd otoakoestiese emissies oorskade vroeër kan optel as die gewone gehoortoets. As u instem om aan die studie deel te neem, sal u 4 tipe oortoetse moet ondergaan voordat u begin met streptomycin behandeling, of so gou as moontlik daarna, sowel as elke 2 weke vir die volgende 2 maande. Hiermee sal dit moontlik wees om u ore, sowel as u gehoor, te monitor. Indien enige verandering in u ore of gehoor bespeur word, sal u, sowel as u dokter, onmiddellik in kennis gestel word sodat julle besluite aangaande u behandeling kan maak. Al die toetse sal in die Oudiologie-afdeling van Brooklyn Chest hospital geskied.

University of Cape Town

Voor die aanvang van enige toets sal ek die nodige instruksies verskaf en die toets aan u verduidelik. Indien u onseker is oor enigiets kan u enige tyd 'n vraag stel. Die hele toetsprosedure sal ongeveer 40 minute duur per sessie. Geeneen van die toetse is skadelik vir u nie. Die 4 toetse is as volg:

(a) Terwyl u op 'n gemaklike stoel sit, sal ek met 'n lig in u oor kyk.

(b) Hierna sal ek 'n proppie in u oor plaas wat gaan meet hoe goed u middeloor werk. Behalwe om stil te sit, hoef u niks te doen gedurende die toets nie.

(c) Hierna volg die gehoortoets. Nadat die oorfone op u ore geplaas is, sal u alleen in die klankbehandelde kamer wees. U sal nog steeds in staat wees om met my te kommunikeer. U sal klanke deur die oorfone hoor, in een oor op 'n slag. Meeste van die klanke gaan baie sag wees, maar u moet u hand opsteek elke keer as u die klank hoor.

Die doel van die toets is om te bepaal wat die sagste klanke is wat u kan hoor. Geeneen van die bogenoemde toetse is ongemaklik nie.

(d) Na die gewone gehoortoets sal u 'n rustyd van ongeveer 5 minute hê. Dan sal die otoakoestiese emissie toets gedoen word. Dit behels dat 'n proppie in u oor geplaas word, wat dalk ongemaklik kan wees, maar definitief nie seer nie. Die proppie stuur klanke in die oor in, en meet die reaksies van die binne-oor. U hoef geensins iets te doen vir die duur van die toets nie, behalwe stilsit.

Al die inligting wat ingewin is, sal vertroulik bly en deel van u hospitaalrekords vorm, want u sal nie geïdentifiseer word in enige publikasie wat moontlik mag spruit uit hierdie publikasie nie. U is vry om enige tyd van die studie te onttrek, sonder dat u normal behandeling enigsins geaffekteer sal word. Die algehele resultate van die studie sal aan die superintendent van die hospital bekend gemaak word.

'n Voordeel verbonde aan deelname aan die studie is dat indien u enigsins tekens van gehoorverlies of oorskade sou toon, u en u dokter daaroor ingelig sal word, sodat die moontlike effek van streptomycin op u ore en gehoor geminimaliseer kan word. Daar is geen risikos verbonde aan deelname aan die studie nie. Indien u enige vrae het, kan u dit nou, of enige tyd gedurende die studie stel.

Navorser: Lucretia Petersen

Kontaktelefoonnommer: (021) 404 6074

Adres: Afdeling van Kommunikasiewetenskappe en –Afwykings

E47 Ou Hoofgebou

Groote Schuur Hospitaal

Observatory

7925

University of Cape Town

Appendix A-3

Xhosa Subject Information Sheet

Istreptomycin yenye yamachiza athi anikwe abantu abagula ngesifo sephepha (iTuberculosis okanye iTB). Ugqirha wakho ugqibe entweni yokuba istreptomycin lelona chiza elinokunyanga iTB yakho. Istreptomycin yaziwa ngokudala umonakalo phakathi endlebeni, ngoko ke iyakwazi ukubangela iindlebe zingeva kakuhle. Kuba unokwazi ukuphelelwa kukuva kweendlebe, kubalulekile ukuba uxilongise ukuva kwendlebe zakho ukwenzela zifunyanwe ngokukhawuleza iingxaki zokuva ezinokuthi zibekho.

Injongo yoluphando kukuqonda ukuba uvavanyo olwaziwa njenge 'otoacoustic emissions test' luwubona msinyane umonakalo womphakathi wendlebe kunezinye indlela zokuvavanya zesiqhelo. Kwi otoacoustic emissions test kufakwa iprobhu kwindlebe yakho. Oko kungakwenza uzive ungemnandanga nje kancinci kodwa akusayi kubabuhlungu. Iprobhu le ithumela iizandi phakathi endlebeni ize ichaze ngokusebenza nangesimo somphakathi wendlebe. Ngelixesha lovavanyo akukho nto oyakuthi uyenze ngaphandle kokuhlala uzolile lude lugqitywe uvavanyo.

Phambi kokuba ufumane istreptomycin olu vavanyo lulandelayo luyakwenziwa kwigumbi elikhuselekileyo kwingxolo apha kwisibhedlele sase Brooklyn Chest:

(a) Ndizokusebenzisa isibane ndijonge endlebeni yakho.

(b) Ndilandele ngokufaka iprobhu endlebeni yakho ukuqinisekisa ukuba umbindi wendlebe yakho usebenza kakuhle. Akukho nto ozakufuneka uyenzile ngaphandle koku hlala uthe cwaka de luphele uvavanyo.

(c) Ze kuxilongwe ukuva kweendlebe zakho. Emveni kokuba iiearphones zibekwe ezindlebeni zakho uyakusala wedwa kweli gumbi likhuseleke engxolweni. Kodwa uzokwazi ukuthetha nam. Uzakuva izandi ziphuma kwiiearphones, kuzokuxilongwa indlebe enye ngexesha. Iizandi ezininzi ziyakuvakalela ezantsi (azingxoli), kodwa kufuneka uphakamise isandla sakho nanini na usiva isandi. Injongo yolu vavanyo

University of Cape Town

kukuqonda ezona zandi zisezantsi okwazi ukuziva. Olu vavanyo alisayikwenza uzive ungemnandanga.

(d) Emva kolu vavanyo lwesiqhelo uyakuthabatha ikhefu yemizuzu emihlanu (5). Emveni koku i 'otoacoustic emissions test' iyakuthi yenziwe njengokuba besekucacisiwe apha ngentla.

Phambi kokuba uvavanyo ngalunye luqale ndizakukunika imiqathango ndiye ndikucacisele ngovavanyo. Ukuba awuqinisekanga ngokumele ukwenze ungabuza imibuzo nanini na. Uvavanyo lulonke luyakuthatha imizuzu eyi-40 ngeseshini. Alukho uvavanyo oluyakuthi lubeyingozi kuwe nangaluphi na uhlobo.

Emveni kokuba ufumene istreptomycin uvavanyo oluchazwe ku (a), (c) naku (d) luyakuphindwa qho emva kweeveki ezimbini kwi thuba elingageenyanga ezimbini ukwenzela kubonwe naziphi inguqu ezibangelwa zistreptomycin ezinothi zenzeke kukuva kweendlebe zakho. Xa na kungathi kubekho iingxaki ezibonwayo wena nogqirha wakho niyakuthi naziswe ngazo ngoko-nangoko.

Yonke inkcazelo eyakuthi ifunyanwe iyakuhlala iyimfihlelo kuba igama lakho alisayi kuvezwa kwiziphumo zolu phando ngaphandle kwemvume yakho. Uvumelekile ukuba urhoxe kolu phando nanini na, oku akuyi kuchaphazela ukufumana kwakho unyango nakanjani na. Iziphumo zolu phando ziyakwaziswa umphathi (usuperintendent) wesibhedlele. Awusayi kuchazwa ngegama nakweziphi izinto ezishicelelweyo ngolu phando.

Ubuhle bokuthabatha inxaxheba kolu phando lolokuba nayiphina ingxaki yokuva onokuthi ubenayo iyakubhaqwa ngexesha. Akukho ngozi ngokuthabatha inxaxheba kolu phando.

Imibuzo onayo ungayibuza ngoku okanye nanini na ngelixesha lophando.

Umphandi: Lucretia Petersen

Inombolo yemfono-mfono: (021) 404 6074

Idilesi: Division of Communication Sciences and Disorders
E47 Old Main Building, Groote Schuur Hospital
Observatory, 7925

University of Cape Town

Appendix A4

English Subject Consent Form

I hereby acknowledge that the purpose and procedures of the study has been fully explained to me. I also understand what is expected of me. I am aware that I can withdraw from the study at any stage without my treatment being affected. There was an opportunity for me to ask questions.

I hereby give my consent to participate in the study. I also understand that my test results can be shared with myself, as well as my doctor, if it is necessary. I am also aware that the overall results of the study will be made known to the superintendent of the hospital.

Signed: _____(Subject)

_____(Researcher)

Date: _____

University of Cape Town

Appendix A-5

Afrikaans Subject Consent Form

Hiermee gee ek te kenne dat die doel en die prosedure ten volle aan my verduidelik is. Ek verstaan wat van my verwag word. Ek is die geleentheid gebied om vrae te stel. Ek is bewus daarvan dat ek myself ter enige tyd van die studie kan onttrek, sonder dat my behandeling enigsins geaffekteer sal word.

Hiermee stem ek in om deel te neem aan die studie. Ek gee my toestemming dat my toetsresultate met my dokter gedeel kan word, indien nodig. Ek is ook bewus daarvan dat die algehele toetsresultate aan die hospitaal se superintendent bekend gemaak sal word.

University of Cape Town

Geteken: _____(Proefpersoon)

_____ (Navorser)

Datum: _____

Appendix A-6

Xhosa Subject Consent Form

Ndiyavuma ukuba yonke into emalunga nenjongo neendlela zokuqhuba oluphando ndiyicaciselwe kakuhle. Kwaye ndiyakuqonda konke okulindelwe kum. Ndiyazi ukuba ndingarhoxa koluphando nanini na. Ndilinikiwe ithuba lokubuza imibuzo.

Ndinikeza ngemvume yokuthabatha inxaxheba kolu phando. Ndiyaqonda ukuba iziphumo zovavanyo ziyokwaziwa ndim nangugqirha wam ukuba oko kunyanzelekile.

Ndiyaqonda kwakhona ukuba zonke iziphumo zovavanyo ziyakwaziswa umphathi (usuperintendent) wesibhedlele, kodwa andisayi kuchazwa ngegama nakweziphi na izinto ezishicelelweyo ngoluphando.

Sayina: _____ (Umthathi-nxaxheba)

_____ (Umphandi)

Umhla: _____

University of Cape Town

Appendix B1.

Distribution of mean pure tone thresholds in all participants over the eight-week period

	Mean dB SPL (*SD*)					Skewness					Kurtosis				
Week	0	2	4	6	8	0	2	4	6	8	0	2	4	6	8
Frequency (Hz)															
250	10(*1*)	11(*2*)	12(*5*)	15(*12*)	18(*19*)	1.37	1.87	2.62	3.44	3.16	-0.15	1.67	5.83	12.25	10.40
500	10(*0*)	10(*0*)	11(*4*)	14(*11*)	18(*18*)		.	2.41	3.74	3.25	.	.	5.20	14.00	10.82
1000	10(*0*)	10(*0*)	10(*0*)	12(*4*)	16(*17*)	4.12	.	.	3.74	3.46	17.00	.	.	14.00	12.00
2000	10(*1*)	10(*1*)	10(*0*)	13(*7*)	16(*16*)	2.61	4.12	.	3.74	3.33	5.44	17.00	.	14.00	11.30
4000	10(*1*)	11(*2*)	11(*4*)	17(*19*)	20(*23*)	2.24	1.22	0.73	3.27	3.02	4.58	0.87	-1.59	11.33	9.73
8000	12(*1*)	12(*2*)	19(*4*)	21(*19*)	21(*23*)	3.14	2.20	2.24	2.94	2.44	9.80	4.87	4.37	9.21	6.29

University of Cape Town

Appendix B2.

Distribution of DPgram absolute amplitudes in all participants over the eight-week period

		Mean dB SPL (*SD*)					Skewness					Kurtosis				
	Week	0	2	4	6	8	0	2	4	6	8	0	2	4	6	8
Frequency (Hz)	Norm range															
818	1.1 – 18.7	12.9(4)	12.6(4)	12.5(4)	12.4(5)	7.0(3)	-0.53	-0.34	-0.32	-1.26	-0.01	-0.46	-0.82	-1.22	1.99	0.70
1038	-1.2 – 22.8	13.3(4)	13.3(4)	13.6(4)	12.7(5)	7.9(4)	-1.06	-0.56	-0.18	0.15	-1.21	0.65	-0.79	-0.54	-1.26	1.87
1306	1.5 – 22.8	13.7(3)	12.3(5)	12.7(3)	11.9(5)	7.1(3)	-0.60	-0.12	-0.25	0.14	-2.22	-0.34	-0.72	-0.65	-1.21	6.66
1636	0.0 – 16.3	13.0(5)	11.7(7)	12.0(5)	11.2(6)	6.7(4)	-0.97	-0.22	-0.15	-1.01	-0.96	0.95	-0.74	-1.38	2.00	0.15
2063	0.4 – 21.1	10.5(6)	9.5(5)	8.9(6)	8.4(7)	3.6(9)	-0.19	-0.98	-0.75	-1.77	-1.78	-0.97	0.98	0.33	3.90	2.21
2600	1.3 – 21.0	9.1(6)	8.3(4)	7.1(3)	6.0(5)	2.3(8)	-0.63	-0.62	-0.77	-0.61	-0.97	-0.07	-0.63	0.91	-0.35	0.47
3284	1.1 – 19.8	7.2(6)	5.2(6)	5.8(4)	3.0(8)	-0.6(8)	0.90	0.02	0.15	0.28	0.60	1.29	-0.18	-0.25	0.25	0.71
4126	7.2 – 22.2	10.3(4)	9.6(5)	8.6(6)	6.1(9)	2.3(9)	-0.34	-0.57	-0.30	-0.20	-1.07	-0.79	-1.08	-1.39	-1.35	0.94
5200	0.0 – 20.6	5.7(8)	5.1(8)	2.9(9)	0.8(10)	-2.0(9)	0.60	0.70	0.09	-0.32	0.57	-0.20	-1.01	-0.39	0.60	-1.00

University of Cape Town

Appendix C. Statistical comparison of noise floor over time as measured during DPgram measurement

		Mean noise dB SPL (*SD*)					Friedman's ANOVA		
	Week	0	2	4	6	8	*F*	*df*	*p*
Frequency (Hz)									
818		-3 (2)	-5 (2)	-4 (4)	-2 (4)	-4 (4)	5.9	4	.209
1038		-8 (3)	-9 (2)	-9 (3)	-9 (4)	-8 (4)	0.8	4	.938
1306		-7 (3)	-9 (2)	-9 (3)	-7 (3)	-8 (2)	5.2	4	.271
1636		-11 (2)	-12 (2)	-11 (2)	-11 (2)	-12 (1)	2.2	4	.691
2063		-12 (3)	-11 (2)	-12 (2)	-11 (2)	-12 (2)	3.3	4	.509
2600		-12 (2)	-11 (2)	-12 (3)	-11 (2)	-12 (2)	3.5	4	.482
3284		-13 (1)	-13 (1)	-13 (2)	-14 (3)	-13 (3)	1.3	4	.864
4126		-10 (4)	-12 (2)	-14 (1)	-12 (3)	-13 (1)	2.6	4	.626
5200		-9 (3)	-10 (2)	-11 (2)	-9 (2)	-10 (3)	3.1	4	.541

University of Cape Town

Appendix D. Statistical comparison of noise floor over time as measured during DP I/O function measurement

		Mean dB SPL (*SD*)					Friedman's ANOVA		
	Week	0	2	4	6	8	F	*df*	*p*
Frequency (Hz)	L_2 intensity (dB SPL)								
818	75	-6 (1)	-4 (4)	-5 (4)	-3 (5)	-4 (6)	3.2	4	.53
	70	-6 (1)	-5 (4)	-5 (4)	-5 (5)	-5 (6)	0.4	4	.98
	65	-6 (1)	-6 (5)	-6 (4)	-4 (6)	-6 (6)	5.1	4	.28
	60	-7 (3)	-5 (5)	-5 (4)	-4 (6)	-4 (6)	3.4	4	.49
	55	-6 (2)	-5 (4)	-6 (4)	-4 (5)	-5 (6)	3.8	4	.43
	50	-7 (2)	-6 (4)	-5 (4)	-4 (6)	-5 (7)	3.8	4	.43
	45	-8 (3)	-6 (4)	-6 (3)	-5 (7)	-5 (6)	4.0	4	.40
1636	75	-9 (4)	-11 (1)	-11 (2)	-10 (2)	-10 (2)	2.8	4	.59
	70	-9 (4)	-10 (3)	-11 (1)	-10 (3)	-10 (3)	0.4	4	.98
	65	-10 (2)	-11 (3)	-11 (2)	-11 (2)	-10 (3)	1.9	4	.74
	60	-12 (1)	-11 (2)	-11 (2)	-12 (2)	-10 (3)	2.8	4	.58
	55	-11 (3)	-11 (2)	-11 (2)	-11 (2)	-10 (3)	4.2	4	.38
	50	-11 (3)	-12 (2)	-11 (3)	-11 (3)	-11 (2)	1.5	4	.82
	45	-11 (2)	-11 (2)	-12 (2)	-11 (2)	-12 (2)	1.8	4	.77
2454	75	-12 (3)	-11 (2)	-10 (2)	-10 (2)	-10 (2)	3.8	4	.43
	70	-11 (4)	-11 (2)	-12 (1)	-12 (2)	-11 (3)	3.6	4	.46
	65	-11 (2)	-11 (1)	-12 (2)	-12 (1)	-12 (2)	2.3	4	.68
	60	-12 (2)	-12 (2)	-12 (1)	-13 (1)	-12 (2)	1.7	4	.78
	55	-12 (2)	-12 (2)	-13 (2)	-13 (3)	-11 (2)	3.7	4	.44
	50	-12 (2)	-13 (2)	-13 (2)	-14 (2)	-12 (2)	4.6	4	.32
	45	-13 (3)	-13 (2)	-13 (2)	-14 (2)	-12 (2)	5.9	4	.20
3284	75	-12 (3)	-10 (2)	-12 (2)	-11 (2)	-11 (2)	9.3	4	.05
	70	-12 (4)	-10 (1)	-12 (1)	-13 (1)	-11 (3)	11.0	4	.02
	65	-13 (2)	-11 (1)	-12 (1)	-12 (2)	-11 (2)	4.9	4	.29
	60	-13 (1)	-12 (3)	-13 (2)	-13 (2)	-13 (2)	2.2	4	.69
	55	-13. (1)	-12 (2)	-14 (3)	-13 (2)	-13 (3)	1.5	4	.82
	50	-14 (3)	-12 (2)	-13 (1)	-12 (4)	-13 (3)	2.0	4	.72
	45	-13 (1)	-12 (2)	-13 (2)	-12 (2)	-13 (4)	3.4	4	.49

University of Cape Town

Appendix D. (cont.)

4102	75	-11 (2)	-12 (2)	-11 (2)	-11 (3)	-11 (2)	0.9	4	.92
	70	-11 (4)	-12 (2)	-12 (1)	-12 (2)	-12 (3)	1.3	4	.86
	65	-12 (3)	-11 (3)	-12 (2)	-11 (2)	-13 (2)	4.6	4	.32
	60	-11 (2)	-11 (2)	-12 (2)	-12 (3)	-12 (2)	1.6	4	.80
	55	-13 (1)	-13 (1)	-12 (2)	-11 (2)	-13 (2)	4.9	4	.29
	50	-13 (2)	-12 (1)	-12 (2)	-13 (3)	-13 (2)	5.1	4	.27
	45	-14 (1)	-13 (2)	-13 (2)	-12 (2)	-13 (2)	7.8	4	.10
4919	75	-8 (3)	-9 (3)	-11 (3)	-8 (2)	-11 (4)	10.1	4	.03
	70	-10 (3)	-11 (2)	-11 (2)	-10 (2)	-12 (2)	8.1	4	.08
	65	-12 (1)	-12 (1)	-12 (3)	-11 (2)	-13 (1)	4.5	4	.34
	60	-14 (2)	-14 (2)	-14 (2)	-14 (2)	-15 (2)	1.5	4	.82
	55	-11 (3)	-11 (4)	-12 (3)	-11 (3)	-14 (3)	7.4	4	.11
	50	-12 (2)	-13 (2)	-12 (2)	-12 (2)	-14 (2)	7.7	4	.10
	45	-13 (2)	-13 (2)	-13 (2)	-13 (2)	-14 (1)	2.2	4	.70

Note: Bonferroni correction: $\alpha = .016$

University of Cape Town

Appendix E1. DP I/O function slopes at Week 8 in Participant A who developed hearing loss (as per pure tone thresholds)

	DP I/O function slopes (dB/dB)						
	Current study						Previous research (Dorn et al., 2001; Kummer et al., 1998)
L2 Stimulus intensity level dB SPL	Frequency (Hz)						Frequency (Hz)
	818	1636	2454	3284	4102	4919	1000 to 6000
40	Lin (1.1)	Nlin (-1.0)	Lin (0.6)	Nlin (-0.6)	Nlin (0.4)	Lin (4.5)	Nlin
45	Lin (1.1)	Nlin (-0.3)	Lin (1.6)	Lin (2.1)	Lin (1)	Nlin (-1.7)	Nlin
50	Nlin (0.4)	Lin (1.4)	Lin (0.6)	Lin (0.9)	Nlin (0.1)	Nlin (0.5)	Nlin
55	Lin (0.6)	Lin (1.3)	Lin (0.8)	Lin (0.9)	Nlin (-0.3)	Lin (0.6)	Nlin
60	Nlin (0.4)	Lin (0.6)	Nlin (-0.1)	Lin (0.6)	Nlin (-0.2)	Lin (1.2)	Lin
65	Nlin (0.1)	Nlin (0.4)	Nlin (0.4)	Nlin (0.4)	Lin (2.8)	Lin (0.8)	Lin
% Nlin at each frequency	50	50	33	33	66	33	
Overall % Nlin	44						

Note: Lin = Linear slope with range > 0.5 dB/dB (Ruggero et al., 1997)

Nlin = Non-linear slope with range ≤ 0.5 dB/dB (Ruggero et al., 1997)

University of Cape Town

Appendix E2. DP I/O function slopes at Week 8 in Participant B who developed hearing loss (as per pure tone thresholds)

	DP I/O function slopes (dB/dB)						
	Current study						Previous research (Dorn et al., 2001; Kummer et al., 1998)
Stimulus intensity level dB SPL	Frequency (Hz)						Frequency (Hz)
	818	1636	2454	3284	4102	4919	1000 to 6000
40	Lin (1.5)	Nlin (0.4)	Lin (1.1)	Lin (0.7)	Lin (0.6)	Lin (0.7)	Nlin
45	Lin (1.1)	Nlin (-0.1)	Lin (0.7)	Lin (0.6)	Nlin (0.5)	Nlin (0.3)	Nlin
50	Lin (0.8)	Lin (1.6)	Lin (1.1)	Nlin (0.4)	Nlin (0.3)	Nlin (0.2)	Nlin
55	Nlin (0.3)	Lin (1.0)	Lin (1.1)	Nlin (0.2)	Lin (0.6)	Nlin (0.3)	Nlin
60	Nlin (-0.4)	Nlin (0.3)	Lin (0.9)	Lin (0.7)	Nlin (0.5)	Nlin (0.5)	Lin
65	Nlin (-0.5)	Nlin (-0.2)	Nlin (0.2)	Lin (0.9)	Lin (0.8)	Lin (0.7)	Lin
% Nlin at each frequency	50	66	16	33	50	66	
Overall % Nlin	47						

Note: Lin = Linear slope with range > 0.5 dB/dB (Ruggero et al., 1997)

Nlin = Non-linear slope with range ≤ 0.5 dB/dB (Ruggero et al., 1997)

University of Cape Town

Appendix E3. DP I/O function slopes at Week 8 in Participant C who developed hearing loss (as per pure tone thresholds)

	DP I/O function slopes (dB/dB)						
	Current study						Previous research (Dorn et al., 2001; Kummer et al., 1998)
Stimulus intensity level dB SPL	Frequency (Hz)						Frequency (Hz)
	818	1636	2454	3284	4102	4919	1000 to 6000
40	Nlin (-0.6)	Nlin (-1.4)	Lin (0.9)	Nlin (-1.1)	Lin (0.8)	Lin (1.7)	Nlin*
45	Nlin (0.3)	Nlin (0.5)	Nlin (-0.7)	Nlin (-1.8)	Nlin (-2.4)	Lin (0.9)	Nlin*
50	Nlin (-0.3)	Lin (1.8)	Nlin (-0.2)	Lin (2.4)	Nlin (-1.0)	Nlin (-4.5)	Nlin*
55	Nlin (0.4)	Nlin (0.2)	Nlin (-0.6)	Lin (1.8)	Lin (1.2)	Lin (4.5)	Nlin*
60	Nlin (0.0)	Nlin (0.3)	Lin (1.4)	Nlin (-0.3)	Lin (1.3)	Nlin (0.4)	Lin*
65	Nlin (-0.4)	Nlin (-0.8)	Nlin (-0.3)	Nlin (-1.2)	Nlin (-0.4)	Nlin (0.3)	Lin*
% Nlin at each frequency							
Overall % Nlin							

Note: Lin = Linear slope with range > 0.5 dB/dB (Ruggero et al., 1997)

Nlin = Non-linear slope with range ≤ 0.5 dB/dB (Ruggero et al., 1997)

University of Cape Town