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Functional Enhancement of Auditory Activation through Multi-Site Stimulation
across the Isofrequency Dimension of the Inferior Colliculus

THESIS

Submitted in partial fulfillment of the requirements for the degree

DOCTOR OF PHILOSOPHY
(Ph.D.)

awarded at the University of Veterinary Medicine Hannover

by

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Tehran, Iran

Hannover, Germany 2014
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Dedicated

To my parents and my brothers

To my loving wife, Mona

Human beings are members of a whole.
In creation of one essence and soul.

If one member is afflicted with pain,
Other members uneasy will remain.

If you’ve no sympathy for human pain,
The name of human you cannot retain!

Saadi Shirazi
This thesis will be submitted in part for publication to the Journal of Neural Engineering under the title: Cortical responses to electrical stimulation using human auditory midbrain implants: response specificity. B Salamat, M Lenarz, HH Lim, N Alken, R Calixto, T Hartmann, T Rode, T Lenarz, A Kral.
ABSTRACT

Title: Functional Enhancement of Auditory Activation through Multi-Site Stimulation across the Isofrequency Dimension of the Inferior Colliculus

Author: Behrouz Salamat

Auditory midbrain implant (AMI) targets the inferior colliculus (IC) in patients with no implantable cochlea such as the patients with neurofibromatosis type 2 (NF2). The present study investigated a new version of the AMI which enables 3-dimensional (3D) stimulation across the IC. Using cats as an animal model, 3 human AMIs were implanted side by side in the IC. The inferior colliculus central nucleus (ICC) was electrically stimulated with different stimulation levels and the evoked local field potentials (LFP) were recorded from the primary auditory cortex (A1). The objectives of the current study were to determine the basic response properties based on the LFP to this stimulation (threshold, latency, dynamic range (DR)) and to compare this to the previous studies. Further, we wanted to investigate whether the IC stimulation provokes different cortical responses by varying the recording site. The present study demonstrates that the human AMI electrodes can selectively activate different neuronal populations in the ICs of the cat. Furthermore, the study indicates that the stimulation results in activation of partially overlapping neuronal populations, leading to responses at both ON-BF and OFF-BF recording positions in the cortex. In total, these data demonstrate some stimulation site specificity, but also a large overlap of the excitation pattern induced by midbrain stimulation.
ZUSAMMENFASSUNG

**Titel:** Funktionelle Optimierung der auditiven Aktivierung durch multifokale Stimulation über dem Isofrequenz-Areal des Colliculus inferior.

**Autor:** Behrouz Salamat

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ABBREVIATIONS

* \( p < 0.05 \)
*** \( p < 0.001 \)
3D 3-dimensional
+4 dB stimulation levels which were 4 dB above the MEA site thresholds
A area
A1 primary auditory cortex
ABI auditory brainstem implant
ABR auditory brainstem response
AMI auditory midbrain implant
ATH absolute threshold of hearing
AVCN anterior ventral cochlear nucleus
BF best frequency
C1 first LFP crossing of the threshold line (onset latency)
C2 second LFP crossing of the threshold line (offset latency)
Ca\(^{2+}\) calcium-ion
CAP central auditory prosthesis
CF characteristic frequency
Ch/CH channel/site
CI cochlear implant
CN cochlear nucleus
CNIC central nucleus of the IC
CNS central nervous system
CSD current source density
dB decibel
DBS deep brain stimulation
DCN dorsal cochlear nucleus
Di-I red stain
DNLL dorsal nucleus of the lateral lemniscus
DR dynamic range
DZ dorsal zone
F force
F frequency
FDA food and drug administration
FRM frequency response maps
Hz hertz
IC inferior colliculus
ICC central nucleus of the inferior colliculus
ILD interaural level difference
INLL intermediate nucleus of the lateral lemniscus
ITD interaural time difference
K\(^+\) potassium-ion
L\(_{dB}\) sound volume or intensity level in dB
LFP local field potential
LL lateral lemniscus
LSO lateral superior olive
MAD median absolute division
MAX maximum
MEA penetrating multi-site/channel electrode arrays
MGB medial geniculate body
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Definition</th>
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<tbody>
<tr>
<td>MIN</td>
<td>minimum</td>
</tr>
<tr>
<td>MNTB</td>
<td>medial nucleus of the trapezoid body</td>
</tr>
<tr>
<td>MSO</td>
<td>medial superior olive</td>
</tr>
<tr>
<td>MSS</td>
<td>multi-site electrical stimulation paradigm</td>
</tr>
<tr>
<td>n</td>
<td>sum of active sites</td>
</tr>
<tr>
<td>Na⁺</td>
<td>sodium-ion</td>
</tr>
<tr>
<td>NF1</td>
<td>neurofibromatosis type 1</td>
</tr>
<tr>
<td>NF2</td>
<td>neurofibromatosis type 2</td>
</tr>
<tr>
<td>NIDCD</td>
<td>national institute on deafness and other communication disorders</td>
</tr>
<tr>
<td>NS</td>
<td>not statistically significant</td>
</tr>
<tr>
<td>ON-BF</td>
<td>similar BF at the stimulation and recording electrodes</td>
</tr>
<tr>
<td>OFF-BF</td>
<td>3 octaves higher in recording position (A1) than in stimulation position (IC)</td>
</tr>
<tr>
<td>p</td>
<td>statistical probability</td>
</tr>
<tr>
<td>P</td>
<td>power</td>
</tr>
<tr>
<td>P₀</td>
<td>reference pressure</td>
</tr>
<tr>
<td>PABI</td>
<td>penetrating auditory brainstem implant</td>
</tr>
<tr>
<td>PSTH</td>
<td>post-stimulus time histograms</td>
</tr>
<tr>
<td>PVCN</td>
<td>posteroventral cochlear nuclei</td>
</tr>
<tr>
<td>$p_{WM}$</td>
<td>Wallis and Moore statistical significant probability</td>
</tr>
<tr>
<td>re</td>
<td>relative</td>
</tr>
<tr>
<td>S</td>
<td>saturation level</td>
</tr>
<tr>
<td>SD</td>
<td>standard deviation</td>
</tr>
<tr>
<td>SI</td>
<td>international system of units</td>
</tr>
<tr>
<td>SOC</td>
<td>superior olivary complex</td>
</tr>
<tr>
<td>SPL</td>
<td>sound pressure level</td>
</tr>
<tr>
<td>SSS</td>
<td>single-site electrical stimulation paradigm</td>
</tr>
<tr>
<td>T</td>
<td>threshold level</td>
</tr>
<tr>
<td>TRPA1</td>
<td>transient receptor potential cation channel, subfamily A, member 1</td>
</tr>
<tr>
<td>TDT</td>
<td>Tucker-Davis technology</td>
</tr>
<tr>
<td>VNLL</td>
<td>ventral nucleus of the lateral lemniscus</td>
</tr>
<tr>
<td>$Z_{WM}$</td>
<td>phase values of Wallis and Moore</td>
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1. INTRODUCTION

The cochlear implant (CI) has provided the neuroscientists and the physicians a tool to restore speech perception in a significant number of the patients with hearing impairment (Fig. 1). Despite the success of the cochlear implants, patients without an intact auditory nerve (as a consequence of a tumor, trauma, agenesis, etc.) or a non-implantable cochlea (severe ossification or congenital malformations) cannot benefit from this treatment (Lenarz T et al 2009; Lenarz M et al 2006; Lenarz T et al 2006).

In some of these patients the placement of a central auditory prosthesis (CAP) that bypasses the cochlea and the auditory nerve and directly stimulates the central auditory pathway will be the only remaining option.

The CAPs are divided based on their target of the stimulation in: 1) auditory brainstem implant (ABI) or penetrating auditory brainstem implant (PABI) which stimulates the cochlear nucleus in the brainstem (Fig. 1) and 2) auditory midbrain implant (AMI) which targets the inferior colliculus (IC) in the midbrain (Fig. 1, 2A). These devices could benefit
patients with acoustic neuromas such as patients with neurofibromatosis type 2 (NF2). In fact, initially, the ABIs were designed for patients with this genetic disorder.

Fig. 2A  The AMI array consists of 20 platinum ring sites mounted on a silicone carrier. The sites are spaced in intervals of 200 µm (center-to-center) where each site has a thickness of 100 µm and a surface area of about 126,000 µm² (Lenarz T et al 2006; Cochlear Ltd).

Fig. 2B  3-dimensional (3D) stimulation of the IC: Three AMIs were placed in the ICC. One stimulation electrode from each of these 3 AMIs was selected within the same ICC layer (marked as dark dots on the figure) utilizing ICC Frequency Response Maps and its best frequency (BF). These 3 selected positions were stimulated electrically. The responses in A1 were recorded with the help of MEA.

The recent surge in ABI implantation has been followed by varying success (Lim et al 2009). Particularly, only a small percentage of the NF2 patients have achieved moderate open set speech perception with the ABI; the majority of the patients make use of the implant to facilitate lip reading (Schwartz et al 2008). This suboptimal performance in the NF2 population has been attributed to tumor-related damage at the level of the cochlear nucleus (Colletti et al 2009). Consequently, at least in the NF2 patients, it was speculated that stimulation in the inferior colliculus central nucleus (ICC), distant from the damaged region, using an AMI might be more promising (Fig. 2A). One of the main reasons for
selecting ICC as a site for an auditory implant is its well-defined tonotopic organization (Lenarz M et al 2006; Schreiner and Langner 1997; Malmierca et al 1995; Stiebler and Ehret 1985; Serviere et al 1984; Merzenich and Reid 1974; Geniec and Morest 1971; Rose et al 1963). A prototype AMI was designed consisting of a single shank penetrating array (Fig. 2A). After performing animal studies to evaluate the electrophysiological function and the long term safety and stability of the device at our institution, a clinical study was performed in which NF2 patients were implanted with this device (Lenarz M et al 2006; Lenarz M et al 2007; Lim et al 2007c). However, initial results were suggestive that 3-dimensional (3D) stimulation of the IC might provide better performance in the future candidates (Fig. 2B).

Therefore, a new version of the AMI was developed enabling the stimulation across the 3-dimensional organization of the IC. In this study we performed acute experiments in anesthetized cats. Different locations in the ICC were stimulated with 3-shank AMIs and the activity (local field potential (LFP)) (Fig. 2B) of the primary auditory cortex (A1) was recorded with the use of penetrating multi-site electrode arrays (MEA).

The objectives of the current study were to determine the basic response properties to this stimulation (threshold, latency, dynamic range (DR)) and to compare this to previous studies. Furthermore, we wanted to investigate whether the IC stimulation provokes different cortical responses by varying the recording site.
1.1. Anatomy and Physiology of Hearing

This chapter will describe the anatomy of the outer, the middle and the inner ear (Fig. 3). Furthermore, the electrophysiology of the cochlea will be discussed.


1.1.1. Basic Knowledge

Sound waves are audible air pressure oscillations which are spread through a compressible medium (such as solid, liquid, or gas). Hereby, the sound waves displace the medium molecules with two important variables, namely, its frequency and its amplitude or intensity (Pickles 2008; Klinke et al 2005).

The resulting pressure change is called sound pressure \([N/m^2 = Pa]\). Other unit for the sound pressure is the sound pressure level (SPL), which is measured in decibels \([dB SPL]\)
\( L_{dB} = 20 \log_{10} \frac{P_x}{P_0} \) [dB]; (Sound Pressure / Reference Pressure \( P_0 = 2\times10^{-5} \) [N/m\(^2\)]) (Klinke et al 2005).

Sound volume or intensity level in dB SPL is the pressure difference between the compressed and the rarefied air volumes. This has been correlated to loudness. The frequency of the oscillation body determines the frequency of the sound waves [Hertz, Hz]. In figure 4, we can see a complex acoustic sound wave. After Fourier analysis of the wave, it is visible that the sound consists of 4 different tones. A simple tone has one single frequency. A complex acoustic sound is produced by adding the tones of different frequencies. Figure 4 shows production of a complex wave by adding the tones with the frequencies \(~\approx 150, \approx 300, \approx 450\) and \(~600\) Hz together (Pickles 2008; Klinke et al 2005).

The audibility of a sound wave depends on SPL and its frequency. If one examines the audibility of the individual frequencies, the result will be the absolute threshold of hearing (ATH). Figure 5 shows the ATH of humans (Klinke et al 2005).

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**Fig. 4**  
A) Complex acoustic sound waves. B) Two sine waves (tones) added together generate the complex wave seen in A. C) Wave Fourier analysis (Pickles 2008)

**Fig. 5**  
The range of normal human hearing: The ear is more sensitive at threshold of audibility curve; modified from Bartels and Bartels 1995.
In humans, at lower and higher frequencies, higher SPLs are needed to trigger auditory perception (Fig. 5). The normal hearing range of humans is between 20 Hz and 16 kHz. The ear consists of three parts: the outer ear, the middle ear, and the inner ear (Fig. 6) (Klinke et al 2005).

![The structure of the human ear: modified from Noback 1967.](image)

The section from the pinna to the eardrum is the outer ear. The eardrum defines the border to the middle ear. The eardrum and the ossicles (3 small bones) in the tympanic cavity are in the middle ear. The tympanic cavity is filled with air. The air pressure in the tympanic cavity equalizes via the Eustachian tube during the swallowing act. The area beyond the oval window is the inner ear. The inner ear consists of two parts: the vestibular apparatus and the cochlea. The outer ear and the middle ear conduct sound to the cochlea, which separates sounds with regard to frequency before they are transduced by the hair cells into a neural code in the fibers of the auditory nerve (Møller 2006a; Klinke et al 2005; Santi and Mancini 2005).
The description of the vestibular apparatus is omitted in this text; it is recommended for the reader to refer to appropriate text books.

1.1.2. Outer Ear
The function of the outer ear is to transmit the incoming sound waves to the tympanic membrane. The outer ear consists of the pinna (auricle) and the ear canal, which leads to the eardrum (tympanum) (Fig. 6). The pinna gathers sound waves from a wide area. All collected sound waves transfer via the ear canal to the eardrum. The pinna convolutions play an important role in the sound wave localization (Bear et al 2009; Møller 2006a; Klinke et al 2005; Pralong and Carlile 1994; Rice et al 1992; Middlebrooks et al 1989; Shaw 1974). The ear canal has a length of about 2.5 cm and a diameter of about 0.6 cm (Bear et al 2009; Møller 2006a; Klinke et al 2005).

1.1.3. Middle Ear
The principal function of the middle ear is acousto-mechanic transduction, namely, to conduct sound from the tympanic membrane to the inner ear and act as an impedance matching tool between the air-filled outer ear and the fluid-filled inner ear (Fig. 7). The tympanic cavity contains 3 small bones (malleus, incus and stapes). They are pivotally connected to each other and form the ossicular chain and serve to transmit sounds from the air-vibration (eardrum-vibration) to the fluid-filled labyrinth (cochlea). The stapes connects to the oval window which closes the fluid-filled inner ear. The objective of the ossicular chain is the transmission of the sound energy (from the eardrum-vibration) to the inner ear. However, there is a physical problem with this form of transmission; the acoustic impedance in the air is lower than the acoustic impedance in the fluid. In the case of impedance difference, as in this situation, by passing the sound wave from an air-filled channel to a liquid-filled medium, the majority (99%) of the sound wave energy is lost.
(Møller 2006a; Klinke et al 2005; Aibara et al 2001; Puria and Allen 1991; Zwislocki 1965; Wever and Lawrence 1954). One of the main tasks of the ossicles is impedance matching (Møller 2006a; Klinke et al 2005).

![Image](image.png)

**Fig. 7 The middle ear (Brodel 1946)**

The pressure is defined as the ratio of the force to the area over \( A \) which that force \( F \) is distributed to \( P = F/A \). The pressure on the oval window would be enlarged when the force is amplified and the area is reduced. The middle ear uses these two mechanisms to reach an impedance adjustment (Bear et al 2009; Pickles 2008):

1. Area differences

Since the area of the tympanic membrane is considerably larger than the stapes footplate on the oval window a higher pressure is created.
2. Force amplification

Malleus, incus and stapes; these 3 ossicles work together as lever arm, which causes amplification of the force on the oval window.

These two factors together cause that the sound wave pressure is enlarged on the oval window about 20 times more than by the eardrum. For protection from loud acoustic stimuli, 2 muscles, the tensor tympani muscle and the stapedius muscle, contract thus impairing the impedance adjustment (Bear et al 2009; Klinke et al 2005; Pang and Guinan 1997; Counter and Borg 1993; Simmons 1964; Wever and Vernon 1955).

1.1.4. Inner Ear

The function of the inner ear is mainly sound detection and balance (Fig. 8A). The inner ear is located in the temporal bone. It includes two sensory organs, the vestibular apparatus and the cochlea. Three spiral-shaped and hollow canals in the cochlea enable us to hear (Fig. 8B). These 3 canals are called the scala tympani, the scala media and the scala vestibuli (Fig. 8B) (Klinke et al 2005; Fawcett 1986).

Fig. 8A  The cochlea as a straight tube (Møller 2006a)

Fig. 8B  Cross-section of Guinea pig’s cochlea (Davis et al. 1953)
The scala tympani and the scala vestibuli are completed with the stapes footplate at the oval window and by a fine membrane on the round window. All scalae are filled with fluids; the scala media with endolymph, the 2 other scalae with perilymph (Bear et al 2009; Wangemann 2006; Klinke et al 2005).

The perilymph is similar in the composition to the one of the extracellular fluid with Na\(^+\) (140 mmol/l) and K\(^+\) (3 mmol/l) (Bear et al 2009; Møller 2006a; Klinke et al 2005; Wangemann and Schacht 1996).

The perilymph canals (scala tympani and scala vestibuli) communicate at the helicotrema to each other. The endolymphatic space is separated from the scala vestibuli via Reissner’s membrane and from the scala tympani via the basilar membrane. The endolymph is rich in potassium which is secreted from the stria vascularis. The endolymph is similar with respect to its ions to the intracellular fluid with K\(^+\) (145 mmol/l) and Na\(^+\) (1.5 mmol/l). The stria vascularis is metabolically active area on the lateral space of the cochlea (Bear et al 2009; Klinke et al 2005; Kuijpers and Bonting 1970; Kuijpers and Bonting 1969). It is responsible for the maintenance of K\(^+\) concentration of the endolymph, the ion transport, the K\(^+\) recycling, the pH regulation, and the water transport, and thus generating and maintaining the cochlear endolymph potential of +80 mV (Bear et al 2009; Klinke et al 2005; Wangemann 2006; Wangemann et al 2004; Marcus et al 2002; Salt et al 1987; Strekers et al 1984). This so-called endocochlear potential is integral to the hair cell transduction (Bear et al 2009; Møller 2006a; Klinke et al 2005; Santi and Mancini 2005; Wangemann et al 1995; Tasaki and Spyropoulos 1959).
1.1.4.1. The Organ of Corti

The organ of Corti is located on the basilar membrane and is covered by the tectorial membrane (Fig. 9A). This organ contains many different kinds of cells. The hair cells, so called because of the stereocilia (hair-like bundles) that are located on the apical surface of the cells, are sensory cells that are arranged in rows along the basilar membrane (Fig. 9A). The stereocilia protrude into the endolymph. There are approximately 80 stereocilia of different length on the top of each cell (Fig. 9B, C) (Møller 2006a; Klinke et al 2005; Legan et al 1997).

There is a fine elastic filament protein connection (“tip link”) between each stereocilia. Where the tip link connects to the stereocilia, ion channels are located which by opening cause transduction of the sound wave stimuli into receptor potential. There are two distinct
anatomical and functional types of hair cells; the outer hair (Fig. 9B) and the inner hair cells (Fig. 9C). The cochlea of the humans has about 12,000 outer hair cells which are positioned in 3–5 rows along the basilar membrane, and there are roughly 3,500 inner hair cells that are located in a single row (Møller 2006a; Glueckert et al 2005; Klinke et al 2005; Raphael and Altschuler 2003; Ulehlova et al 1987).

The tips of the longest stereocilia outer hair cells are connected to the tectorial membrane which is not the case for the stereocilia of the inner hair cells. The hair cells do not have axons by themselves. They form synaptic afferent (sensory) or efferent (activity coming from the central nervous system) connections with approximately 30000 neurons at their basal ends (Fig. 10) (Bear et al 2009; Klinke et al 2005; Warr 1992; Harrison and Howe 1974; Schuknecht 1960). They are innervated by the peripheral dendrites of the bipolar cells of the spiral ganglion whose together-bundled axons form the auditory portion of the vestibulocochlear nerve (eighth cranial nerve) (Bear et al 2009; Klinke et al 2005; Liberman et al 1990).

![Fig. 10  Innervation of the organ of Corti (Spoendlin 1974)](image)

The majority of the afferent axons (90%) ends on the inner hair cells and has myelin sheath (Fig. 10) (Klinke et al 2005; Brown 1987; Spoendlin 1972). A few afferent axons (10%)
innervate the outer hair cells (Fig. 10). They do not have myelin sheaths which probably do not play an important role to information transfer to brain. In contrast, the majority of efferent axons innervate the outer hair cells, and when activated from the central nervous system (CNS), they decrease sensitivity of the ear (Møller 2006a; Klinke et al 2005; Kandel et al 2000).

1.1.4.2. The Transduction Process

As a reaction to an acoustic stimulus the stapes moves back and forth, thereby, transferring the sound wave to the cochlear fluids and the cochlear membranes (Reissner’s, basilar and tectorial membrane). This causes the oscillation of the cochlear membranes (Fig. 11). The oscillated wave initiates a traveling wave in the cochlea (Klinke et al 2005; Békésy 1960; Békésy 1953). The traveling wave propagates from the base toward the cochlear apex. It grows in amplitude and slows in speed until a point of maximal displacement is reached. The maximal displacement point is determined by the sound frequency. The traveling wave has small amplitude but for each frequency between the stapes to the helicotrema there is a location where the traveling wave reaches larger amplitude (Purves et al 2008; Klinke et al 2005). One of the reasons for this phenomenon is that from the stapes to the helicotrema the basilar membrane stiffness is reduced 100-fold (Bear et al 2009; Klinke et al 2005; Emadi...
et al 2004; Békésy 1960; Békésy 1953). The stiffness and the oscillating mass of the basilar membrane determine together where the optimal oscillation position is localized for each stimuli frequency. The wave reaches its amplitude maximum where the stimulus frequency and the resonant frequency of the basilar membrane correspond. High frequencies reach their maximal amplitude near the stapes and low frequencies near the helicotrema (Fig. 11) (Bear et al 2009; Klinke et al 2005).

1.1.4.2.1. Outer Hair Cells in Sound Transduction

As mentioned previously, due to the sound wave stimulation, the stapes moves in the oval window, transferring the sound wave to the cochlear fluids. This leads to a traveling wave of the basilar membrane (Bear et al 2009; Békésy 1960). The traveling wave reaches its maximal amplitude, a vertical movement between the basilar and the tectorial membrane causes bending off the stereocilia of the outer hair cells (Fig. 12).

The tilt of stereocilia stretches the “tip link”, causing opening of the K⁺ channels in the stereocilia. The opening of the K⁺ channels causes the entry of K⁺ into the hair cell. It is interesting that the entry of K⁺ triggers depolarization of the hair cell (Bear et al 2009; Klinke et al 2005; Geleoc et al 1997; Hudspeth and Corey 1977), while the opening of the K⁺ channels hyperpolarizes most neurons. One reason is the high K⁺ concentration in the endolymph, which causes a K⁺ equilibrium.
potential of 0 mV, compared to the equilibrium potential of -80 mV in typical neurons. Another reason is the 80 mV endocochlear potential, which creates a 125 mV gradient across the stereocilica membranes. Prestin, potential-dependent protein, is located on the membrane of the outer hair cells. By depolarization of the cells, prestin changes its configuration and the hair cells length actively increases and decreases (Bear et al 2009; Klinke et al 2005; Zheny et al 2000a; Zheny et al 2000b; Holley et al 1992; Brownell et al 1985). This improves the frequency selectivity of the cochlea and the amplification of the traveling wave (Bear et al 2009; Klinke et al 2005; Libermann et al 2002; Frolenkov et al 1998).

1.1.4.2.2. Inner Hair Cells in Sound Transduction
The outer hair cells oscillations have the same oscillation frequencies as the stimuli wave. The above mentioned local amplification of the traveling wave causes indirect transport of the oscillation energy via endolymph motion to the inner hair cells stereocilia. Thereby, stereocilia bend off, their “tip link” stretches and opens the cation-selective channels. Then, $K^+$ flows in to the hair cells, which will cause depolarization of the cells.

Depolarization of the inner hair cells does not cause an active change in the length of the hair cells as with the outer hair cells (Bear et al 2009; Klinke et al 2005; Zheny et al 2000a; Brownell et al 1985) (prestin proteins are not located on the membrane of the inner hair cells). However, the inner hair cell depolarization leads to the opening of voltage dependent $Ca^{2+}$ channels, causing the release of...
Ca\textsuperscript{2+} in the axonal terminal of the cell (Fig. 13). As a result, the neurotransmitter glutamate will be released into the synaptic cleft (Bear \textit{et al} 2009; Klinke \textit{et al} 2005; Eybalin 1993; Ruel \textit{et al} 1999). Finally, as a consequence of the above steps, the post synaptic neuron (afferent nerve) will be depolarized and an action potential will be generated (Fig. 13) (Bear \textit{et al} 2009; Klinke \textit{et al} 2005; Hudspeth 1997; Hudspeth 1983).

1.1.5. Tonotopic Organization

As mentioned previously, the traveling wave generates its maximal amplitude in a specific location in the basilar membrane. This causes frequency- and position-dependent depolarization of the inner hair cells and the afferent nerve fibers. Each inner hair cell and its nerve fibers are optimized to specific frequency stimuli (sensitive at one frequency which is called the characteristic frequency (CF) or the best frequency (BF)). By the CF each auditory system neuron gives its greatest response. The more the stimulus frequency deviates from CF, the higher sound pressure is needed to stimulate the nerve fibers. The auditory nerve fibers in the auditory structure are organized in a systemic pattern based on their CFs which is called the tonotopic organization. The brain recognizes which fibers are activated in the tonotopic organization so that it interprets them as tone frequency (Bear \textit{et al} 2009; Pickles 2008; Klinke \textit{et al} 2005; Ruggero \textit{et al} 1997).

1.1.6. Auditory Pathway

Through the central auditory pathway, the auditory information is, among others, processed for sound localization. This is done by using the interaural time difference (ITD) and the interaural level difference (ILD) in the superior olivary complex (SOC) (Bear \textit{et al} 2009; Webster 1995; Oertel 1991; Yin and Chan 1990). The cochlear nerve is the starting point of the central auditory pathway leading to the cochlear nucleus (CN) in the brainstem (Fig. 14A) (Tan 2009). From that point, a complex of highly organized inter- and intra-
nuclear network connects the superior olivary complex (through the medial nucleus of the trapezoid body (MBTB)) in the medulla oblongata, the lateral lemniscus (LL) in the pons, the IC in midbrain, the medial geniculate body of the thalamus and the auditory cortex together (Fig. 14B) (Tan 2009; Frisina and Walton 2001; Aitkin 1990).

Most of auditory pathways’ nuclei consist of subnuclei. As an example the cochlear nucleus (CN) is comprised of 3 subnuclei, the dorsal (DCN), the anteroventral (AVCN) and the posteroventral cochlear nuclei (PVCN) (Santi and Mancini 2005; Paxinos and Watson 1998; Ramon y Cajal S 1995). The superior olivary complex (SOC) composed of several brainstem nuclei, the medial superior olive (MSO) and lateral superior olive (LSO) (Webster 1995). Three nuclei, namely, a dorsal (DNLL), an intermediate (INLL) and a ventral nucleus (VNLL) are embedded in the lateral lemniscus (Paxinos and Watson 1998; Ramon y Cajal S 1995). The IC consists of 3 subnuclei which are discussed in the following section more in detail.
1.1.6.1. Inferior Colliculus

The inferior colliculus (IC) in human is comparable to the cats’ IC and are 2 distinctive small rounded hemispherical protuberances on the dorsal aspect of the midbrain (Fig. 15) (Moore 1987; Morest and Oliver 1984). The ICs function as the principal auditory center and participate in the multisensory integration (Saint Marie et al. 1999; Friauf 1992; Aitkin et al 1994; Aitkin et al 1978). An important property of the IC is the ability to process sound input with complex temporal patterns (Tan 2009; Winer and Schreiner 2005). The IC divides into 3 subdivisions (Fig. 15): 1) a central nucleus (ICC) with mainly auditory function (Aitkin et al 1994; Aitkin et al 1978); 2) a lateral/external cortex with most likely a multisensory function (Aitkin 1978); and 3) a dorsal cortex.

Due to the large size of the nuclei of the IC, one might expect that these nuclei play an important role in the function of the auditory system (Winer and Schreiner 2005). The IC could be seen as a significant sound-processing center as it incorporates more neurons than all the other subcortical auditory nuclei together (Kulesza et al. 2002). Virtually, all parts

![Fig. 15](image-url)
of the cochlear nucleus send inputs to the IC (Fig. 14). There are also many connections between the nuclei of both ICs to each other (Winer and Schreiner 2005). Among auditory nuclei, the IC is the site that, essentially, all auditory brainstem data are converged and assimilated (Tan 2009; Thompson 2005). There are also some studies, suggestive that the IC functions as an integration site of sound localization as well (D’Angelo et al 2005; Ingham and McAlpine 2005; Loftus et al 2004; McAlpine and Palmer 2002; Ramachandran and May 2002). There seem also to be some evidence of the IC involvement in acoustico-motor behavior (e.g. auditory startle reflex, audiogenic seizures) and in nociception (Tan 2009; Winer and Schreiner 2005).

As mentioned before, this study has mainly focused on the auditory function part of the IC, the ICC. The ICC as known is a 3-dimensional structure which consists of two-dimensional, inhomogeneous isofrequency laminae (Oliver 2005; Brown et al 1997). One of the important properties of the ICC is its laminar structure (Fig. 15) (Oliver and Morest 1984). Oliver and Morest (1984) presented that the distance of these laminae from each other is about 150 µm (in the cat). Most neurons in the ICC are disk-shaped (Oliver 2005; Oliver and Morest 1984). The ICC cells are tonotopically organized from low frequencies at the dorsal part to high frequencies at the ventral part of the ICC (Malmierca et al 2008; Malmierca et al 1996; Friauf 1992; Saint Marie et al 1999; Semple and Aitkin 1979; Brown et al 1997; Schreiner and Langner 1997; Malmierca et al 1995; Stiebler and Ehret 1985; Serviere et al 1984; Merzenich and Reid 1974; Geniec and Morest 1971; Rose et al 1963). The ICC has excitatory and inhibitory ascended inputs from the most auditory nuclei (Merchán et al 2005; Cant and Benson 2003; Riqueleme et al 2001; Winer et al 1996; Oliver et al 1994; Glendenning et al 1992; Helfert et al 1989; Adams and Mugnaini 1984). This is suggestive that the ICC is an important center for all auditory information which is sent to the auditory cortex. As an example, the ICC receives information from the
SOC thus participating in sound localization (D’Angelo et al 2005; Ingham and McAlpine 2005; Loftus et al 2004; Ramachandran and May 2002; Webster WR 1995).

1.2. Conductive and Sensorineural Hearing Loss

The most frequent source of hearing deficits involves the structures that transmit and transduce sounds into neural impulses, essentially, the peripheral auditory system. The same characteristics that makes this system a delicate apparatus to discern sound, makes it also especially susceptible to damages resulting in peripheral hearing deficits. Hearing impairment in only one ear is characteristic of peripheral hearing loss, as damages at or above brainstem, even in the case of a unilateral insult, causes binaural hearing deficits due to the extensive bilateral neural connective framework of the central auditory pathways. Peripheral hearing losses are classified into conductive and sensorineural hearing deficits. Conductive hearing losses are those caused by insults to the structures of the outer or the middle ear. The sensorineural hearing impairments originate from injuries to the structures of the inner ear, such as the cochlear hair cells, or to the cochlear nerve. Though the signs of both conductive and sensorineural hearing losses are raised hearing thresholds on the damaged side, however, the diagnosis and the management of these two hearing impairments are different. Any occlusion of the ear canal due to, such as, cerumen (earwax), foreign bodies, ear canal tumors or congenital atresia of the auditory canal may lead to conductive hearing loss. Other causes are damages to the tympanic membrane, such as rupture, or damages or impairments of the ossicles and their functions, such as serous otitis media or arthritic ossification. On the contrary, congenital or environmental insults that lead to the hair cell injury or damage to the auditory nerve results in the sensorineural hearing loss, especially as both structures cannot regenerate. Managements of the conductive and the sensorineural hearing loss are different due to the underlying mechanism of the deficits. An external hearing device can be used to compensate for the
reduced conductive apparatus deficiency in the patients with conductive hearing loss. A hearing aid can be placed in the ear canal. It boosts the sound stimuli to compensate for the increased hearing thresholds. They are many forms of these hearing devices, however, the newer generations are smaller, more easily to wear and less visible to others. These new mini-devices consist of a microphone, a speaker and an amplifier. The management of sensorineural hearing loss due to the injuries to the hair cells or the auditory pathways, however, is more complex. Hearing aids are not most useful in this situation as sound intensification often cannot compensate for the inability of the hearing organ to generate or relay a neural impulse from the cochlea to the higher auditory centers. Its management would involve invasive placement of auditory implants such as CI, ABI and AMI, which are described in the following sections (Purves et al 2008; Middlebrooks et al 2005; Zeng 2004; Ramsden 2002; Rauschecker and Shannon 2002).

1.2.1. Neurofibromatosis Type II

Neurofibromatosis is a genetic disorder primarily characterized by the development of multiple tumors of the nerves and areas of skin hypo- or hyperpigmentation. There are 3 types of neurofibromatosis (Nutakki et al 2013; Monteiro et al 2012; Lu-Emerson and Evans 2009; Plotkin 2009):

1) Type 1 (NF1) which mostly consists of neurofibromas which may occur and cause many dependent neurological conditions and cutaneous and skeletal disfigurement (Nutakki et al 2013).

2) Type 2 (NF2) characterized by the development of vestibular schwannomas (acoustic neuromas), spinal cord schwannomas, meningiomas, and ependymomas, and juvenile cataracts, with a lack of cutaneous features (Monteiro et al 2012; Evans 2009).
3) Schwannomatosis which consist of schwannomas instead of neurofibromas. Multiple schwannomas may occur throughout the body or in isolated regions causing intense pain and neurological symptoms (Lu-Emerson and Plotkin 2009).

Of interest to this project is the neurofibromatosis type 2 (NF2). The most common tumors with NF2 are the vestibular schwannomas of the 8th cranial nerve (acoustic neuromas). NF2 is inherited in an autosomal dominant pattern. Incidence of NF2 is about 1 in 60000 (Evans 2009). It is believed that half of the new NF2 cases are new de-novo mutations.

Mutation/loss of the NF2 gene on the human chromosome 22 (22q12.2) causes the development of schwannomas, meningiomas, and ependymomas (Martuza and Eldridge 1988). Though essentially benign tumors, they have the potential of progression to malignancy in humans (Sekido 2011; McClatchey et al 1998; Evans et al 1992). Biallelic loss of NF2 gene that encodes the tumor suppressor protein Merlin causes abnormal proliferation of the Schwann cells and the development of schwannomas in the NF2 patients (Ammoun et al 2008).

Clinical features of NF2 are diverse. Ultimately, all of the NF2 patients develop schwannomas, typically affecting both vestibular nerves. This leads to tinnitus, unilateral or bilateral deafness, dizziness and/or imbalance (Monteiro et al 2012).

Non-acoustic symptoms include ophthalmic (reduced visual acuity and cataract), neurological (spinal cord lesions) and cutaneous (intracutaneous or subcutaneous skin lesions/nodules) symptoms. Diagnosis is made by clinical examination and neuroimaging studies. It is possible to diagnose patients prenatally (Evans 2009). NF2 management is difficult depending on the extent and the location of the tumors. Majority of the NF2
patients will face significant morbidity (Evans 2009), including loss of or damage to their auditory nerve during the tumor removal.

1.3. Auditory Implants

Two main categories of auditory implants (Fig. 1; see section 1.) have been used depending whether the patient has an intact/functional or non-intact/non-functional cochlear nerve.

1.3.1. Cochlear Implant

The National Institute on Deafness and other Communication Disorders (NIDCD) quotes the U.S. Food and Drug Administration (FDA) that more than 200,000 patients worldwide had undergone placement of a cochlear implant by the end of 2010 (NIDCD 2011). Since the introduction of single-electrode CI in 1976 (House 1976), CI has undergone significant modifications and advancements. It is meanwhile a small device consisting of several parts, including a peripherally placed microphone, a digital sound processor, a transmitter and receiver/stimulator and an electrode array. The electrode array is introduced through the round window into the cochlea (Fig. 16) (Purves et al 2008).

Fig. 16  *Cochlear implant (Lim 2009: Cochlear Ltd)*
It is surgically placed along the length of the tonotopically organized auditory nerve endings. The digital processor transforms sound into its spectral components. The additional electronics (transmitter, stimulator) use this data to activate the different contacts on the multisite stimulating electrode array. The placement of the electrode array enables electrical stimulation of the nerve fibers somewhat to mimic the spectral decomposition which is naturally achieved by the cochlea. Patients with acquired hair cell damage would benefit from the CI permitting speech recognition. However, there is ongoing debate about how much a CI can be beneficial in the patients with similar but congenital deficits when they have never been exposed to sounds which would be helpful for speech and language skills. Most children receive a cochlear device implanted in early childhood for early exposure to sounds that could promote speech comprehension. Additionally, intensive post-implant speech therapy is essential to optimize speech, language, and social development. Regardless of its imperfections as well as limitations on patient selection, with continued research and further advancement in its technology the CI has become the most successful neural prosthetic device to date (Purves et al 2008; Wilson and Dorman 2008; Møller 2006b; Middlebrooks et al 2005; Adams et al 2004; Zeng 2004; Ramsden 2002; Rauschecker and Shannon 2002).

1.3.2. Central Auditory Prosthesis

As described in the previous sections, patients with an intact auditory nerve and implantable cochlea might benefit from CI, however, patients with defects that involve the cochlear nerve or the cochlea itself cannot benefit from this modality (Lenarz T et al 2009; Lenarz M et al 2006). Recent research has culminated in the development of other devices that can bypass these structures and stimulate the central auditory pathway directly in such patients as with NF2. These central auditory prosthetics (CAP) are divided based on their stimulation target, namely, the auditory brainstem implant (ABI) or the penetrating
auditory brainstem implant (PABI) which target the cochlear nucleus in the brainstem and auditory midbrain implant (AMI) which stimulates the IC (Lenarz T et al 2009; Otto et al 2008; Lenarz M et al 2006).

### 1.3.2.1. Auditory Brainstem Implant

Using similar technology as by CI, but with a different target of stimulation, the surgically placed ABIs stimulate the cochlear nucleus (Fig. 17A, B). The device uses an electrode array placed on the surface of the cochlear nucleus stimulating the cochlear nucleus complex in the brainstem (Otto et al 2008; Otto et al 2002; Brackmann et al 1993; Shannon et al 1993; Moore 1987; Edgerton et al 1982; Moore and Osen 1979; Shannon et al 1997). The procedure is technically more demanding than the placement of the CI (Møller 2006b).

However, that is the only option for restoring of hearing sensations for patients with no auditory nerve. William House and his colleagues introduced the use of ABI for the first time (Møller 2006b; Brackmann et al 1993; Portillo et al 1993). Worldwide more than 700 patients have received the ABI. NF2 has been the most frequent indication for ABI placement (Otto et al 2008; Baser et al 2003) for the recipients.

Though this device helps in sound discrimination and recognition, and supports lip-reading (Otto et al 2008; Behr et al 2007; Jackson et al 2002; Lenarz M et al 2002; Nevison et al
2002; Otto et al 2002; Vincent et al 2002; Lenarz T et al 2001; Marangos et al 2000; Sollmann et al 2000), however, still most of the patients with NF2 acquire only dismal speech recognition (Otto et al 2008; Colletti and Shannon 2005a). It is postulated that one reason for this shortcoming might be that ABI with only surface electrodes fails to appropriately connect to the tonotopically organized cochlear nucleus (Otto et al 2008; Kuchta et al 2004). With development of the PABIs it was hoped to overcome this deficiency and to improve the selectivity of the stimulation and activate more targeted cluster of neurons (Fig. 17B). This micro-stimulation was hoped to reduce the threshold current levels and increase the range of pitch percepts, and, ultimately, enhance speech recognition. Insertion safety into the brainstem, safe long-term stimulation current levels (McCreery 2008; Otto et al 2008; McCreery et al 2000; McCreery et al 1997) and the concept that this micro-stimulation is able to produce highly selective tonotopic activation (Otto et al 2008; McCreery et al 1998) have been studied.

1.3.2.2. Auditory Midbrain Implant

Considering the current study, emphasis is given to discuss auditory midbrain implants (AMI) in more detail. Patient selection and the history of AMI as well as the summary of the results of the first 3 AMI-implanted patients are discussed (Fig. 18) (Lim et al 2009; Lenarz M et al 2007; Lim et al 2007c; Lenarz M et al 2006; Lenarz T et al 2006; Samii et al 2007).

Unlike to the CIs, the development and the advancements of the central auditory implants have been slower. In the case of the ABIs, the clinical success have been inconsistent (Colletti et al. 2009; Lim et al 2009; Grayeli et al 2008; Schwartz et al 2008; Behr et al. 2007; Colletti and Shannon 2005a; Otto et al 2002; Lenarz T et al 2001) and there have
only been limited studies directed toward knowledge how to obtain consistent and effective activation of higher auditory centers by triggering of the cochlear nucleus (Lim et al 2009).

![Fig. 18](image)

A) Standard deep brain stimulation (DBS) array and magnified image of the AMI array (B) for comparison (Lenarz T et al 2006; Lim et al 2009; Samii et al 2006; Cochlear Ltd) (C) AMI has like CI speech processor. It is in design and technique (to electrical stimulation) similar to CI (Lenarz T et al 2009)

It is also of interest that certain ABI recipients such as non-tumor patients with head trauma outperform the NF2 patients (Colletti et al 2009; Lim et al 2009; Colletti and Shannon 2005a). Furthermore, it has been shown that the same strategies and surface arrays used for the CI stimulation are suboptimal for the cochlear nucleus activation (Lim et al 2009; McCreery 2008; Schwartz et al 2008; Shivdasani et al 2008; Kuchta et al 2004).

Understanding these critical aspects is very crucial to optimize the performance of the CAP and the development of improved devices. More recently, these perspectives have directed the research towards the alternative types of CAPs, such as development of the PABIs and the AMIs, and as well as towards improved sound recognition and perception using animal models and clinical studies (Lim et al 2009; McCreery 2008; Otto SR et al 2008).

To achieve more targeted and frequency-specific stimulation of cochlear nucleus with the goal of improving speech perception in the NF2 patients, PABIs were developed
Though initially encouraging, as the PABI was able to achieve a wide range of pitch percepts across the implantation sites, still there were difficulties in targeting the proper sites as the results were not better compared to the performance of the current ABIs in the NF2 patients (Lim et al 2009; McCreery 2008; Otto et al 2008; Shivdasani et al 2008). Still this does not prove the inferiority of the PABIs compared to the surface ABIs. Due to the complexity of the cochlear nucleus neural structure (Lim et al 2009; Cant and Benson 2003; Young et al 1992; Young et al 1988; Moore and Osen 1979; Osen 1969) and the fact that higher auditory centers are activated differently depending on the location of the cochlear nucleus stimulation (Lim et al 2009; McCreery 2008; Shivdasani et al 2008), the goal has been to improve the target of stimulation. In addition, developing synergistic stimulation models targeting appropriate regions in the cochlear nucleus may benefit speech perception (Lim et al 2009; McCreery 2008; Shivdasani et al 2008).

Growing interest and research focusing on the auditory nerve and the cochlear stimulation for hearing restoration (Lim et al 2009; Wilson and Dorman 2008; Zeng 2004; Djourno and Eyries 1957; Djourno et al 1957a; Djourno et al 1957b; Andreev et al 1935) and the excel in research regarding deep brain stimulation (DBS) combined with the shortcomings and the limitations of the other hearing devices and implants (hearing aid, CI, ABI, PABI) have directed the more recent auditory prostatic development towards the development of the AMIs (Fig. 18A) (Lim et al 2009). Many studies have documented induction of sound sensations by stimulation of the central auditory system (Lim and Anderson 2006; Dobelle et al 1973; Penfield and Rasmussen 1950) and specially IC (Lim and Anderson 2007a; Lim and Anderson 2006; Simmons et al 1964).
In 2005, Colleti reported the first case of successful electrical stimulation of the IC in a patient with NF2 (Møller 2006b; Colleti et al 2005b). The AMI is a new generation hearing prosthesis with the goal to stimulate tonotopic well organized ICC (Fig. 18B) in profoundly hearing impaired patients who cannot benefit from cochlear implants (Lim et al 2007c; Lenarz M et al 2007; Lenarz M et al 2006; Lenarz T 2006). Auditory midbrain stimulation provides a wide range of level, spectral, and temporal cues. Though all of these aspects are essential for speech discernment, still their fusion appears to be insufficient to facilitate open set speech perception with the current stimulation modalities (Lim et al 2007c; Lenarz M et al 2006). One principal question was the selection of an auditory center in the midbrain that could be successfully stimulated. The selected structure needs to fulfill many criteria (Lim et al 2009; Lim and Anderson 2007a; Lim and Anderson 2006; Lenarz M et al 2006; Lenarz T et al 2006; Samii et al 2007): 1) it must have well-defined neuronal and tonotopic organization; 2) it should enable systematic spatial stimulation of different functional regions; 3) it should not be a remote center with more complex coding properties; and 4) it should be surgically accessible.

A center, somewhat fulfilling these criteria, is the ICC. The structure and neural properties of the IC and its nuclei have been discussed in the previous sections. As a relay center for almost all ascending auditory brainstem connections, (Casseday et al 2002) the ICC provides access to the pathways necessary for speech understanding (Lim et al 2009). The ICC consists of a laminated organization and has a well-defined tonotopic organization (Fig. 18A, B) (Lim et al 2009; Oliver 2005; Geniec and Morest 1971). As the ability of transmitting of a specific frequency information is essential for speech perception performance this tonotopic organization becomes a vital aspect (Lim et al 2009; Shannon et al 2004; Friesen et al 2001). Importantly, implantation of an electrode array in the IC is surgically feasible due to its anatomical location and access. Furthermore, its safe
deployment has been shown in various studies (Lim et al 2009; Green et al 2006; Wichmann and Delong 2006).

After selection of the IC for the device deployment, 2 stimulation methods have been used: 1) surface stimulation (termed IC implant) using a Med-El ABI array (Colletti et al 2007) and 2) penetrating stimulation (termed AMI) using a new cochlear DBS array (Lim et al 2009; Lim et al 2007c; Lenarz T et al 2006). In 2007, Colleti (Colletti et al 2007) demonstrated triggering of auditory sensation with surface IC stimulation. This has also been shown with the penetrating stimulation of the auditory midbrain with the AMI (Fig. 18) (Lim et al 2007c; Lenarz M et al 2006) which is the focus of the current research.

The AMI system consists of many parts with the crucial part being a single-shank multi-site array. This electrode array is adapted to the dimensions of the human IC aiming stimulation of the ICC’s tonotopically-organized layers (Lim et al 2009; Lenarz M 2006; Lenarz T 2006). This array has the benefit of being able to stimulate different frequency regions as well as of using materials and technologies known to be safe for the DBS in humans (Lim et al 2009; Lenarz M 2006; Lenarz T 2006). From Dacron mesh to the tip of the styllet, the AMI electrode array measures 6.4 mm and has a diameter of 0.4 mm (Fig. 18) (Lim et al 2009; Lenarz M 2006; Lenarz T 2006). The AMI array consists of 20 platinum ring sites mounted on a concentrically hollow silicone carrier (30 durometer hardness). The sites are spaced in intervals of 200 μm (center-to-center) where each site has a thickness of 100 μm and a surface area of about 126,000 is connected to a parylene-coated 25-μm thick wire made of 90% platinum and 10% iridium (Lim et al 2009; Lenarz M 2006; Lenarz T 2006).
The stiffness of the array is provided by a central stainless steel stylet, which enables insertion of the electrode array into the IC and is removed after the electrode array is in its final position in the midbrain, thus silicone carrier remains in the tissue (Lim et al 2009; Lenarz M 2006; Lenarz T 2006). To minimize the movement of the array after device deployment and to avoid overinsertion, a Dacron mesh is used to constraint the electrode onto the surface of the neural tissue (Lim et al 2009; Lenarz M 2006; Lenarz T 2006). The other parts of the AMI are analogous to the latest Nucleus CI system. The system has a behind-the-ear microphone and a processor that transmits the electromagnetic signals to a subcutaneous receiver-stimulator which is implanted in a bony bed on the skull near the craniotomy and is connected with a cable to the electrode array (Fig. 18C) (Lenarz T et al 2009; Lim et al 2009).

After feasibility and safety studies on animals and development of the surgical approach, clinical study was performed. Five volunteer NF2 subjects were implanted with the single shank AMI (Lenarz M et al 2007; Lim et al 2007c; Lenarz M et al 2006; lenraz T et al 2006; Samii et al 2006). The following is the summary of the results of the first 3 AMI implanted patients at our institution which has previously been published (Lenarz T et al 2009; Lim et al 2009; Lim HH et al 2008; Lim et al 2007c):

1. The AMI was safe and provided some hearing improvement. The AMIs could trigger acoustic sensations by all of the patients. All patients could profit from the implant in their daily life; they could hear environmental noise and the implant could help them to orientate themselves acoustically.

2. The results strongly depended on the location of the stimulation in the midbrain (the ICC was the best implant position). Patient, who was implanted in the ICC,
showed a better speech perception compared to the average NF2 patients with brainstem implants.

3. The stimulation of some areas in the IC induced strong adaptation (dorsal IC).

4. Still, regardless of the encouraging results, open set speech perception was not achieved with the current single-shank AMI and stimulation strategy. However, patients benefited from AMIs in association with lip reading.

5. The AMI's could be implanted safely in the IC of the patients.

6. There were no significant side-effects observed by the AMI stimulation. The only minor side-effects were temporary and reversible paresthesias which were resolved through the adjusting the stimulation program.

The current and the future studies at our center concentrate on:

1. How to temporally and spatially stimulate the ICC to achieve the desired auditory activation patterns that could lead to improved auditory perception?

2. How to stimulate the ICC to prevent adaptation?

3. Which effects multi-site stimulation of the ICC has on desired auditory perception (3D stimulation of the ICC, stimulation of the 3 sites on AMIs with different stimulation time delays (MSS))? 

4. How to improve the surgical approach?
2. MATERIALS AND METHODS

The experiments were performed on 6 anesthetized cats. All experiments were approved by the local state authorities and were performed in compliance with the guidelines of the European Community for the Care and Use of Laboratory Animals (EU VD 86/609/EEC) and the German law for protection of animals. Animal preparation, anesthesia, surgical procedures, electrical stimulation paradigm, and stimulation/recording setup were performed according to Calixto et al (2013; 2012) and are briefly reviewed here. Three human AMI devices (Cochlear Ltd., Lane Cove, Australia) were used in the present experiments (Fig. 2A, B). They consisted of a silicone carrier with 20 platinum mounted rings, space interval of 200 µm, ring thickness of 100 µm, surface area of about 126,000 µm², and impedances of 5–20 kΩ (at 1 kHz). The implants allowed us to stimulate 3 different IC regions at the same time (Fig. 2B).

2.1. Anesthesia and Surgery

2.1.1. Anesthesia

The animals were pre-medicated with medetomidine (0.1 mg/kg i.m.; Domitor®, Orion Pharma) and ketamine (10 mg/kg s.c.; Ketamin®, Gräub). General anesthesia was initiated with propofol (6 mg/kg i.v.; Propofol®, Braun) and was maintained with continuous intravenous administration of pentobarbital (3.6 to 7.2 mg/hr; Narcoren®, Meria). Local anesthesia was achieved with subcutaneous prilocaine 1% (1 to 4 ml per animal; Xylonest®, AstraZeneca). A warm water heating blanket controlled by a rectal temperature probe was used to keep the body temperature at 38° ± 0.5° C. The oxygen saturation was continuously monitored throughout the experiment via pulse oximetry. Heart rate and rhythm were monitored using electrocardiographic recordings (Alken 2009).
2.1.2. Surgery

After the induction of general anesthesia, the animals were placed in a stereotaxic frame (David Kopf Instruments, Tujunga, CA, USA). A craniotomy was performed to uncover the brain from the caudal end of the occipital lobe to the pseudosylvian sulcus of the temporal lobe. The IC was exposed using an occipital craniotomy beneath the tentorium. The tentorium was partially removed and when necessary a small portion of the cerebellum was aspirated to fully expose the IC. By this approach direct access was achieved to the surface of the IC. The penetration axis did not parallel the tonotopic axis of the IC completely, and therefore, this approach assured only a limited frequency range on the shanks of the probes. Nonetheless, this was the only possible approach as stereotactic placement of the AMI is impossible due to the bony tentorium in the cat, and the direct approach from dorsal requires removal of the cortex close to the investigated region (Calixto et al 2012a).

Three single-shank AMIs were inserted into the IC with the stylet manually under visual control through an operating microscope (Lenarz T et al 2006). The ICC was targeted with all 3 shanks. The AMI arrays were also marked with a fluorescent dye (3 mg Di-I per 100 μL acetone; Di-I: 1, 1-dioctadecyl-3, 3, 3′, 3-tetramethylindo-carbocyanine perchlorate; Molecular Probes, Eugene, OR, USA) to allow the investigators to recover the location of the arrays within the ICC through histological analysis (Lim and Anderson 2007a; DiCarlo et al 1996). However, histological reconstruction proved difficult as the final removal of the 3 AMIs affected the morphological integrity of the IC.

2.2. Placement of Electrode Arrays

Before placement of the cortical electrodes, normal hearing was verified by brainstem response audiometry (ABR). The verification of the probe placements was achieved by
acoustic-driven neural response patterns. The placement of the electrodes in the IC was verified by Frequency Response Maps (FRM) (Fig. 19) and post-stimulus time histograms (PSTH). The details on these analysis methods are presented in previous publications (Neuheiser et al 2010; Lenarz M et al 2006; Lim and Anderson 2006). For details on the FRM creation please refer to the next section.

The shift of latency at best frequency (BF) and sustained PSTHs in response to the broadband noise confirmed the placement within the ICC (which was not always the case for all electrodes) (Lim and Anderson 2006; Snyder et al 2004). Only the electrodes placed in the central nucleus were used for further experiments.

After the removal of the dura mater above the auditory cortex, a multi-site silicon substrate Neuronexus probe (Fig. 20A; 8 shanks, 32 channels [Ch], 200 μm apart, 32 recording sites, 413 μm2/site; NeuroNexus Technologies, Ann Arbor, MI, USA) could be placed into the A1 using a micro-manipulator with sites positioned in the main input layer (III/IV) using

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**Fig. 19  ICC Frequency Response Maps (ICC-FRM):** Recording of the acoustic responses from 3 sites on 3 AMI shanks (see Fig. 2B). ICC-FRM was used to confirm the proper AMI implantation position electrophysiologically. ICC has good tonotopic organization and BF shifting presented by FRM confirmed the positioning of the AMI in the ICC. By using this method for 3 AMIs it was possible to select 3 different sites, one from each AMI shanks, which have similar BF (marked on the plot with □, 1st AMI Ch1 BF = 1.15 kHz, 2nd AMI Ch10 BF = 1.15 kHz, 3rd AMI Ch12 BF = 1.56 kHz). The color scale represents the total Spikes: 0 (black) to 41 (white).
current source density (CSD) analysis (Kral et al 2000, Mitzdorf 1985, Muller-Preuss and Mitzdorf 1984) and FRM (Fig. 20B), corresponding to a mean penetration depth of 1248 µm ± 208 µm (standard deviation (SD)) from the pia. In the dorsoventral axis, positioning was aimed at the mid-distance from dorsal end of the posterior ectosylvian sulcus to the superior sylvian sulcus. This would correspond to the dorsal parts of the A1 (Imaizumi et al 2004), avoiding the dorsal zone (DZ, Mellott et al 2010). Additional information on how to perform the CSD analysis and to identify the layer III/IV is described in Calixto et al (2012).

Fig. 20A  Michigan probe: A1 penetration with the help of micro manipulator (~1300 µm vertically from surface of the pia). Penetration through the A1 was performed by using online CSD to identify the cortex layer III/IV. To match the BF of the A1 (recording position) with of the ICC-BF (stimulation position) or 3 octave higher, A1-FRM was used (see Fig. 20B).

Fig. 20B  A1 Frequency Response Maps (A1-FRM): Acoustic response recording form the A1 with the help of the MEA. BF shifting was used to confirm the MEA position on the A1 electrophysiologically. A1-FRM and its BF were also used to match the ICC AMI implantation with the MEA implantation. The color scale represents total Spikes: 0 (black) to 41 (white).

In the rostrocaudal direction, two recording positions were intended: one with matching the BF of the ICC electrodes (ON-BF) selected for the stimulation, and one that was about 3 octaves higher (3 mm rostral from the first one) (OFF-BF). Using a multi-shank
Neuronexus probe, this was restricted by the highly individual vascular map of the given animal.

After placement of both the IC and the cortex electrodes, brain at both sites was covered with agarose to prevent pulsations and drying. Recording was simultaneous from all cortical 32 sites. The position of the electrodes was functionally defined by measuring their receptive fields with the acoustic stimulation. All further recordings were obtained with open filters and only local field potentials were processed.

2.2.1. Frequency Response Maps: a Method for BF Measurement

To create the FRM, the acoustic stimulation was performed with pure tones of different frequencies (0-45 kHz) and stimulation levels (0-70 dB SPL), as is shown in figure 21.

![Technical pure tone stimulus representation as an example for production of the FRM.](image)

The recorded cortical responses were band pass-filtered (300–3,000 Hz) and spikes that exceeded 3.5 times the standard deviation of the background noise signal were detected.
Then, the spikes were binned into post-stimulus time histograms (1 ms bins (PSTH)). The driven spike rate was then calculated within a set PSTH window relative to the stimulus onset (A1 5–25 ms, ICC 5–65 ms); and the value (total spikes: 0 (black) to 41 (white)) was plotted for each stimulus to create the FRM (Neuheiser et al 2010). The BF could be measured as the centroid value at 10 dB SPL above the level where a noticeable and consistent response was observed (Neuheiser et al 2010). Details of the PSTH, FRM, and BF calculations have been published previously (Lim and Anderson 2006).

2.3. Electrical Stimulation Paradigm and Recording Setup

A computer interfaced with TDT System 3 hardware (Tucker-Davis Technology, Alachua, FL, USA) and a custom written software (Mathworks, Natick, MA, USA) were used for stimulation and recording.

In each animal, we identified one stimulation site per shank (through 3 AMIs) that showed similar frequency sensitivity with the help of FRM. Each site was consequently electrically stimulated with monopolar biphasic pulses (cathodic-leading, 200 µs/phase and ~500 ms interstimulus interval).

Each of the 3 stimulation electrodes was stimulated with different stimulation levels varying from 20 to 58 dB (in 2-dB steps relative to 1 µA, single site stimulation paradigm (SSS)). All stimuli were randomized within a total of 20 trials and were additionally complemented by 20 recordings of spontaneous activity to determine the baseline activity. The SSS was also used to study the adaption behavior. Time different LFPs characteristics were compared to each other to verify a possible adaptation.
2.4. Offline Data Analysis

Presented data are the results from the LFPs (Fig. 22) recorded from the Neuronexus probe (multi-electrode arrays, MEA). For this purpose, the unfiltered recorded signal was used. Such LFPs are dominated by low-frequency (<100 Hz) oscillations resulting from synchronous firing of many neurons relatively close to the electrode (Harrison et al 2004, Metting van Rijn et al 1986).

![Fig. 22 MEA sites LFP recording (Ch 1 to 32) of A1 responses to 2 different stimulation levels (48 dB and 58 dB re 1 µA) in ICC. Ch 1 to Ch 4 on MEA shank 1, Ch 5 to Ch 8 on MEA shank 2, Ch 9 to Ch 12 on MEA shank 3, Ch 13 to Ch 16 on MEA shank 4, Ch 17 to Ch 20 on MEA shank 5, Ch 21 to Ch 24 on MEA shank 6, Ch 25 to Ch 28 on MEA shank 7, and Ch 29 to Ch 32 on MEA shank 8. Plane 1 = [MEA Ch 1, 5, 9, 13, 17, 21, 25, 29], plane 2 = [MEA Ch 2, 6, 10, 14, 18, 22, 26, 30], plane 3 = [MEA Ch 3, 7, 11, 15, 19, 23, 27, 31] and plane 4 = [MEA Ch 4, 8, 12, 16, 20, 24, 28, 32].](image)

Electrical stimulation of the ICC can antidromically activate the neurons projecting from the A1 to the IC (Lim and Anderson 2007b), predominantly the layer V neurons (Neuheiser et al 2010). To eliminate the influence of the antidromic activation we focused the analysis on the LFP data. Stimulus artifacts were removed by blanking. All 20 sweeps were averaged and all analyses were performed on the averages.
From the data we determined the LFP duration, peak, latency, and temporal integral (area) using an automatic MatLab routine (Fig. 23). To determine the onset and the offset of LFPs, a threshold was calculated from the prestimulus time intervals (duration 15 ms in each trial). This threshold line (red line in Fig. 23) was the mean minus 6 times SD of the signal to prevent detecting recording noise. The LFP durations were defined as the time intervals between the first (C1, onset latency) or the second (C2, offset latency) LFP crossings of the threshold line. The LFP peak is the nadir of the curve (absolute maximum of the amplitude). The latency is defined peak latency of the LFP. The area of the LFP is the area under the LFP curve.

![Fig. 23 Peak and latency of the LFP are marked as a red star. The LFP peak is the nadir of the curve (absolute maximum of the amplitude). The latency is defined as the time interval between the stimulation onset and the LFP peak. The area of the LFP is the area under the curve where it crosses the 0 mV amplitude line. The LFP durations are the time intervals between time zero and the first (C1) or the second (C2) crossings of the threshold line (red line; marked black spheres). The threshold line is an arbitrary chosen line which is the mean of 15 ms impulsive activity minus its 6 standard deviation.](image)

The threshold level (T) was determined as minimum stimulation level at which the response could be visually determined in the response and with the increasing stimulus level further increased in the amplitude and decreased in the latency. The saturation level (S) was the current level at which the peak amplitude was maximal (maximum stimulation level restricted at 58dB). The dynamic range (DR) was consequently determined as follows: $DR = (S - T) \cdot 0.8$
For assessing cortical specificity of the responses, the difference of the best frequencies in the stimulating and recording electrodes in the ICC and the cortex were computed in octaves. Further, the responses were grouped based on the cortical BF in relation to the stimulation site into “ON-BF recordings” (± 0.5 octave) and “OFF-BF recordings” (>3 octaves difference) (Fig. 24). These groups were compared statistically. We decided to use 3 octaves, as the corresponding ~3 mm of cortical distance allows an acceptable distance between the recording sites with minimized contamination of far-fields between the sites. Corresponding to mapping studies performed with monopolar stimulation through a cochlear implant (Kral et al 2009), this distance would be off the hot spot determined with such stimulation.

![Fig. 24](image)

The stimulation positions of the sites in ICC as well as the recording positions in the A1 had a similar BF (ON-BF) or the recording positions in the A1 had a different BF (OFF-BF). The mean of octave cortical recording differences between the ON-BF and the OFF-BF supposed to be 3 octaves (MEA was modified from NeuroNexus Technologies 2006).

For assessing the ICC-stimulation specificity of the responses, we used the ON-BF experiments and compared to the OFF-BF groups. To evaluate whether stimulation of 3 different sites of the ICC would trigger different responses, median of the neural threshold activity and DR were measured from 32 cortical MEA sites and compared statistically to each other. The following comparisons of the 3 stimulated positions were performed using median comparisons with the Mann-Whitney rank sum statistic test: stimulation position 1
against stimulation position 2 (1<>2), 1 against 3 (1<>3) and 2 against 3 (2<>3). The data are presented in the tables.

To compare the amplitude and the latency relations, responses at the 32 MEA sites were normalized with regard to the thresholds, and the responses were compared at 4 dB above the threshold (+4 dB) (normalized stimulation protocol). This level was chosen to avoid large spread of excitation with the large currents. Also, it allowed including all recordings into the statistics. Furthermore, this level is comparable to the cochlear implant studies (with smaller DR of the responses).

Stimulation site dependence of the response in the A1 for each recording position, when stimulated with different AMI contacts, was examined by computing the differences between the response amplitudes and latencies using the following criteria:

1. LFPs were recorded from a MEA site which had a response to 2 or 3 ICC stimulation positions
2. The LFP crossed the threshold line

Using the Wallis and Moore test, these differences were then analyzed for statistical significance.
3. RESULTS

In 3 of the experiments, similar acoustic BFs could be found on each of the 3 AMI shanks (Table 1; Experiment 3, 4 and 5). In 2 experiments, matching BFs could not be found in the stimulation site (Experiment 1 and 2). As replacement of the AMI would result in significant damage to the IC, the AMI was left in place despite of this. In the last experiment, the AMI electrode did not deliver signals with sufficient quality to determine the FRMs in the IC.

<table>
<thead>
<tr>
<th>A) Experiment 1</th>
<th>B) Experiment 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1-BF = 19.8* ± 2.3kHz</td>
<td>A1-BF = 10.6* ± 0.4kHz</td>
</tr>
<tr>
<td>ICC-BF P1 = 13.4 kHz</td>
<td>ICC-BF P1 = 08.6 kHz</td>
</tr>
<tr>
<td>ICC-BF P2 = 03.6 kHz</td>
<td>ICC-BF P2 = 01.5 kHz</td>
</tr>
<tr>
<td>ICC-BF P3 = 09.0 kHz</td>
<td>ICC-BF P3 = 18.9 kHz</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>C) Experiment 3</th>
<th>D) Experiment 4</th>
<th>E) Experiment 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1-BF = 1.4* ± 0.3 kHz</td>
<td>A1-BF = 8.1* ± 0.7 kHz</td>
<td>A1-BF = 7.8* ± 1.4 kHz</td>
</tr>
<tr>
<td>ICC-BF P1 = 01.1 kHz</td>
<td>ICC-BF P1 = 01.1 kHz</td>
<td>ICC-BF P1 = 01.1 kHz</td>
</tr>
<tr>
<td>ICC-BF P2 = 01.2 kHz</td>
<td>ICC-BF P2 = 01.1 kHz</td>
<td>ICC-BF P2 = 01.1 kHz</td>
</tr>
<tr>
<td>ICC-BF P3 = 01.2 kHz</td>
<td>ICC-BF P3 = 01.5 kHz</td>
<td>ICC-BF P3 = 01.1 kHz</td>
</tr>
</tbody>
</table>

| F) Experiment 6 | | Table 1 |
|-----------------|-----------------|
| IC-unknown-BF-group | | Best frequency values for ICC and corresponding MEA sites recording in all 6 experiments. * Mean value of BF – MEA sites recording ± SD. |
| A1-BF = 8.7 ± 0.5 kHz | | |
| ICC-BF = unknown | | |

To assess the level dependence of the responses, stimulation of ICC with varying stimulation levels were used (Fig. 25, 26 A, B). With this method we were able to identify the thresholds and the saturation levels for the cortical responses to the AMI stimulation. The threshold and the saturation level values were used to calculate the DR. Level dependent stimulation of the ICC with increased cortical responses with increasing stimulation level was noted in all experiments. However, the maximum amplitude of the
responses was different for 3 different ICC stimulation positions (Fig. 25, 26A). These values also were dependent on the A1 32 MEA recording sites (Fig. 26B).

Fig. 25
Responses to SSS: Stimulations of ICC triggered different neural activities on A1 as shown by LFP. Marked * are LFPs which are 4 dB above the LFP-Threshold (LFP thresholds are marked as ▲). Position 1 demonstrated the lowest LFP threshold (26 dB). To better identify the LFP thresholds, the data are presented in zoomed-in views next to first analysis. First stimulation position BF = 1.15 kHz, 2nd position BF = 1.15 kHz, 3rd position BF = 1.56 kHz.
Fig. 26A
LFP-Area as a function of current level: Responses to SSS in 3 different stimulation positions in the ICC and their responses on MEA site 4. The LFP-Area enlarges by increasing the stimulation level differently for 3 different ICC stimulations. Unusual higher DR presets in position 2. Stars are stimulation levels plus 4 dB above the MEA site threshold.

Fig. 26B
Responses to SSS in position 2 (Fig. 26A) and their responses in A1 32 MEA sites. The LFP-Area enlarges by increasing the stimulation levels for all 32 sites; however, the maximum responses are different for 32 MEA site. The responses at 32 MEA sites were normalized at 4 dB above threshold.
3.1. Threshold and Dynamic Range

The response thresholds varied between 28 and 54 dB re 1 µA (Table 2A), when recorded in a total of 544 recording-stimulation positions. Interestingly, within each experiment, the threshold responses were different for the 3 different electrodes of the implant, even when the stimulation was performed in the same isofrequency lamina (Fig. 25, Table 2A). We observed that all stimulation positions (16 stimulation positions in 6 experiments) significantly differed with regard of the median of the evoked cortical threshold. The grand mean of the medians was 39.88 dB ± 7.71 dB (SD) (dB re 1 µA). Furthermore, the DR varied between 6 and 20.8 dB (Table 2B), whereas, in some configurations the DR could not be assessed due to the extreme threshold values. Medians of the DR as a function of the 3 stimulation positions in the IC differed significantly (Table 2B). The grand mean of the DR medians was 13.41 dB ± 5.66 dB (SD) (dB re 1 µA).

<table>
<thead>
<tr>
<th>LFP-Threshold [dB re 1 µA]</th>
<th>A</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Experiments</strong></td>
<td>1</td>
</tr>
<tr>
<td><strong>Positions</strong></td>
<td>1</td>
</tr>
<tr>
<td>Median</td>
<td>40</td>
</tr>
<tr>
<td>MAD</td>
<td>0</td>
</tr>
<tr>
<td>MIN</td>
<td>36</td>
</tr>
<tr>
<td>MAX</td>
<td>52</td>
</tr>
<tr>
<td>AVERAGE</td>
<td>40.68</td>
</tr>
<tr>
<td><strong>n</strong></td>
<td>32</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Dynamic Range [dB re 1 µA]</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Experiments</strong></td>
<td>1</td>
</tr>
<tr>
<td><strong>Positions</strong></td>
<td>1</td>
</tr>
<tr>
<td>Median</td>
<td>12.80</td>
</tr>
<tr>
<td>MAD</td>
<td>01.60</td>
</tr>
<tr>
<td>MIN</td>
<td>04.80</td>
</tr>
<tr>
<td>MAX</td>
<td>17.60</td>
</tr>
<tr>
<td>AVERAGE</td>
<td>13.31</td>
</tr>
<tr>
<td><strong>n</strong></td>
<td>32</td>
</tr>
</tbody>
</table>

Table 2
A) LFP-Threshold statistical values, measured as response to SSS (for the LFP-Threshold, data from 544 recorded position possibilities in A1 were used). B) DR statistical values, measured as response to SSS (for assessment of DR, data from only 541 positions from possible 544 recorded positions were used. The reason was that the thresholds on some of the MEA sites were larger or equal to 58 dB). Active sites (n) (LFPs which pass the threshold line), the median absolute division (MAD), minimum (MIN), maximum (MAX), the value of statistical probability (p), highly statistically significant (p < 0.001***).
3.2. LFP Characteristics and Distance Dependency between ICC and A1

Local field potentials are expected to depend on the relative position of the stimulating and recording electrodes within the tonotopic axis. In total, 224 recording-stimulation pairs could be compared. To quantify this relative distance, the BFs at recording and stimulation electrodes were determined and their relation was expressed in octaves ($\log_2[B_{A1}/B_{IC}]$). Two groups of data were attempted: ON-BF (similar BF at the stimulation and recording electrodes) and OFF-BF (~3 octaves higher in A1 than in IC) (Fig. 24). The data revealed that the BF in the ON-BF measurements was slightly underestimated, whereas the BF in OFF-BF was slightly overestimated (e.g. Fig. 27). Thirteen data points were considered outliers (Fig. 27A) and were discarded from statistical processing. In the remaining data (n=211), the 2 groups were well separated and we could compare them (LFP Characteristics; Fig. 23 section 2.4.) using Mann-Whitney rank sum test.

Both the LFP temporal integral (LFP-Area) as well as the peak amplitude of LFPs were larger in the ON-BF group (Fig. 27A, B and Fig. 28A, B; $p < 0.001$; Table 3). This was expected, given the strongest functional connection between the same best frequencies within the auditory pathway. Although the peak latency was not different in the 2 groups (Fig. 29A, B; $p = 0.154$; Table 4), the onset latency (C1 point) was shorter in the ON-BF group, as expected (Fig. 30A, B; $p = 0.005$; Table 4). Because the offset latency (C2 point) was significantly longer in the ON-BF group (Fig. 31A, B; $p < 0.001$; Table 5), it explains the larger temporal integral in the ON-BF group by demonstrating that the LFPs lasted longer in the ON-BF group. All this meets the assumptions for point-to-point projections within the auditory pathway and location-specific responses to stimulation in the IC.
Fig. 27A
LFP-Area -distance dependency between ICC and A1.

Fig. 28A
LFP-Peak -distance dependency between ICC and A1.

Fig. 27B
LFP-Area group comparisons; corresponding table 3 (-0.26 ≤ Log₂ (BFₐ/BFᵦ) ON-BF, n = 132 ≤ +1.46; +2.13 ≤ Log₂ (BFₐ/BFᵦ) OFF-BF, n = 79 ≤ +3.21). *** Highly statistically significant (p < 0.001).

Fig. 28B
LFP-Peak group comparisons; corresponding table 3 (-0.26 ≤ Log₂ (BFₐ/BFᵦ) ON-BF, n = 132 ≤ +1.46; +2.13 ≤ Log₂ (BFₐ/BFᵦ) OFF-BF, n = 79 ≤ +3.21). *** Highly statistically significant (p < 0.001).

Table 3
LFP-Characteristics group comparisons and their statistical values. Active sites (n) (LFPs which pass the threshold line), the median absolute division (MAD), minimum (MIN), maximum (MAX), the value of statistical probability (p), *** highly statistically significant (p < 0.001).

<table>
<thead>
<tr>
<th></th>
<th>LFP-Area [Vµs]</th>
<th>LFP-Peak [mV]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ON-BF</td>
<td>OFF-BF</td>
</tr>
<tr>
<td>Median</td>
<td>0.401</td>
<td>0.283</td>
</tr>
<tr>
<td>MAD</td>
<td>0.164</td>
<td>0.078</td>
</tr>
<tr>
<td>MIN</td>
<td>0.025</td>
<td>0.090</td>
</tr>
<tr>
<td>MAX</td>
<td>1.184</td>
<td>0.897</td>
</tr>
<tr>
<td>AVERAGE</td>
<td>0.423</td>
<td>0.303</td>
</tr>
<tr>
<td>n</td>
<td>132</td>
<td>79</td>
</tr>
<tr>
<td>p</td>
<td>&lt;0.001 (***</td>
<td>&lt;0.001 (***</td>
</tr>
</tbody>
</table>
Fig. 29A

Fig. 29B
LFP-Latency group comparisons; corresponding table 4 (-0.26 ≤ Log₂ (BF_A1/BF_IC) ON-BF, n = 132 ≤ +1.46; +2.13 ≤ Log₂ (BF_A1/BF_IC) OFF-BF, n = 79 ≤ +3.21). Not statistically significant differences (NS).

Fig. 30A
LFP-C1-distance dependency between ICC and A1.

Fig. 30B
LFP-C1 group comparisons; corresponding table 4 (-0.26 ≤ Log₂ (BF_A1/BF_IC) ON-BF, n = 132 ≤ +1.46; +2.13 ≤ Log₂ (BF_A1/BF_IC) OFF-BF, n = 79 ≤ +3.21). * Statistically significant (p < 0.05).

Table 4
LFP-Characteristics group comparisons and their statistical values. Active sites (n) (LFPs which pass the threshold line), the median absolute division (MAD), minimum (MIN), maximum (MAX), the value of statistical probability (p), * statistically significant (p < 0.05), not statistically significant differences (NS).

<table>
<thead>
<tr>
<th></th>
<th>LFP-Latency [ms]</th>
<th>LFP-C1 [ms]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ON-BF</td>
<td>OFF-BF</td>
</tr>
<tr>
<td>Median</td>
<td>12.676</td>
<td>13.310</td>
</tr>
<tr>
<td>MAD</td>
<td>01.373</td>
<td>00.860</td>
</tr>
<tr>
<td>MIN</td>
<td>09.136</td>
<td>09.747</td>
</tr>
<tr>
<td>AVERAGE</td>
<td>14.125</td>
<td>13.434</td>
</tr>
<tr>
<td>n</td>
<td>132</td>
<td>79</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>p</th>
<th>ON-BF</th>
<th>OFF-BF</th>
</tr>
</thead>
<tbody>
<tr>
<td>p</td>
<td>0.154 (NS)</td>
<td>0.005 (*)</td>
</tr>
</tbody>
</table>
Fig. 31A
*LFP-C2*-distance dependency between ICC and A1.

Fig. 31B
*LFP-C2* group comparisons; corresponding table 5 (-0.26 ≤ Log$_2$ (BF$_{A1}$/BF$_{IC}$)$_{ON}$; n = 132 ≤ +1.46; +2.13 ≤ Log$_2$ (BF$_{A1}$/BF$_{IC}$)$_{OFF}$; n = 79 ≤ +3.21). *** Highly statistically significant (p < 0.001).

Table 5
*LFP*-Characteristics group comparisons and their statistical values. Active sites (n) (LFPs which pass the threshold line), the median absolute division (MAD), minimum (MIN), maximum (MAX), the value of statistical probability (p). *** highly statistically significant (p < 0.001).

<table>
<thead>
<tr>
<th>Groups</th>
<th>ON-BF</th>
<th>OFF-BF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median</td>
<td>26.735</td>
<td>20.680</td>
</tr>
<tr>
<td>MAD</td>
<td>3.535</td>
<td>2.450</td>
</tr>
<tr>
<td>MIN</td>
<td>11.920</td>
<td>14.380</td>
</tr>
<tr>
<td>MAX</td>
<td>35.350</td>
<td>32.640</td>
</tr>
<tr>
<td>AVERAGE</td>
<td>25.331</td>
<td>20.712</td>
</tr>
<tr>
<td>n</td>
<td>132</td>
<td>79</td>
</tr>
<tr>
<td>p</td>
<td>&lt;0.001 (***)</td>
<td></td>
</tr>
</tbody>
</table>

However, the situation is more complex: the threshold was significantly higher in the ON-BF group than in the OFF-BF group when recorded in a total of 307 recording-stimulation positions (Fig. 32A, B; p < 0.001; Table 6) by 6 dB in median. This would demonstrate that stimulation at the threshold takes place off the position of the stimulating electrodes (distant neurons) and, only at the higher current levels, also activates close neurons. Because of the relation of the threshold and the maximum current level possible in the current sources, the DR of the neuronal responses could only be fully evaluated in a subset of the data. These were removed from statistical analysis. In the remaining data set (n = 278), the median of the ON-BF was 3.2 dB larger than in OFF-BF (Fig. 33A, B, p < 0.001; Table 6). These results have to be considered cautiously, as the range of the current source and the differences in threshold in the 2 groups have obviously limited the conclusion.
Fig. 32A
LFP-Threshold -distance dependencies between ICC and A1.

Fig. 33A
Dynamic Range -distance dependencies between ICC and A1. DR < 4 dB is not included.

Table 6
LFP-Threshold and Dynamic Range group comparisons and its statistical values. Active sites (n) (LFPs which pass the threshold line), the median absolute division (MAD), minimum (MIN), maximum (MAX), the value of statistical probability (p), *** Highly statistically significant (p < 0.001).

<table>
<thead>
<tr>
<th>Groups</th>
<th>LFP-Threshold [dB re 1 µA]</th>
<th>Dynamic Range [dB re 1 µA]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ON-BF</td>
<td>OFF-BF</td>
</tr>
<tr>
<td>Median</td>
<td>40.00</td>
<td>34.00</td>
</tr>
<tr>
<td>MAD</td>
<td>04.00</td>
<td>04.00</td>
</tr>
<tr>
<td>MIN</td>
<td>32.00</td>
<td>26.00</td>
</tr>
<tr>
<td>MAX</td>
<td>54.00</td>
<td>46.00</td>
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<tr>
<td>AVERAGE</td>
<td>42.77</td>
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<tr>
<td>n</td>
<td>173</td>
<td>154</td>
</tr>
<tr>
<td>p</td>
<td>&lt;0.001 (***</td>
<td>&lt;0.001 (***</td>
</tr>
</tbody>
</table>
3.3. Specificity on ICC Position

To investigate whether the stimulation site in the IC affects the response in A1, differences between the response amplitudes and latencies for each recording position were computed when the stimulation was with different AMI contacts. In total, stimulation with 3 different sites were compared where possible (Table 7A; B; 8A, B). The differences were in some comparisons relatively large (difference of medians in each experiment of up to 18 dB).

However, when the differences were tested for significance from zero in each animal using Wallis-Moore test (Phase-Frequency-statistical test, 5% level, compensation of multiple comparisons by Bonferoni procedure), the results did not show highly significant deviation from zero in any such comparisons. This indicates that the responses in the cortex do not systematically change when changing stimulation site in the IC.

<table>
<thead>
<tr>
<th>LFP-Threshold [dB re 1 μA]</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Experiments</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Positions’ differences</td>
<td>P1-P2</td>
<td>P1-P3</td>
</tr>
<tr>
<td>Median</td>
<td>0.016</td>
<td>0.008</td>
</tr>
<tr>
<td>MAD</td>
<td>0.016</td>
<td>0.008</td>
</tr>
<tr>
<td>MIN</td>
<td>0.016</td>
<td>0.008</td>
</tr>
<tr>
<td>MAX</td>
<td>0.016</td>
<td>0.008</td>
</tr>
<tr>
<td>AVERAGE</td>
<td>0.016</td>
<td>0.008</td>
</tr>
<tr>
<td>n</td>
<td>32</td>
<td>32</td>
</tr>
<tr>
<td>Z/M</td>
<td>0.432</td>
<td>0.293</td>
</tr>
<tr>
<td>P/M</td>
<td>0.666</td>
<td>0.767</td>
</tr>
</tbody>
</table>

Table 7

Statistical values of positions’ differences and Wallis and Moore test results in threshold (A) and peak amplitude (B). The phase values of Wallis and Moore (Z/M) and statistical significant probability value (P/M) of this test. Active sites (n) (the sum of number of MEA sites which can record the LFPs which pass the threshold line and also which had a response to 2 or 3 ICC stimulation positions; n>10: Wallis and Moore test not possible!), the median absolute division (MAD), minimum (MIN), maximum (MAX). * Low statistically significant, not statistically significant differences (NS).
### Table 8

Statistical values of positions’ differences and Wallis and Moore test results in peak latency (A) and onset latency (B). The phase values of Wallis and Moore (ZWM) and statistical significant probability value (pSM) of this test. Active sites (n) (the sum of number of MEA sites which can record the LFPs which pass the threshold line and also which had a response to 2 or 3 ICC stimulation positions; n>10: Wallis and Moore test not possible), the median absolute division (MAD), minimum (MIN), maximum (MAX). * Statistically significant, not statistically significant differences (NS).

### 3.4. Adaptation Behavior

SSS as a function of time and its LFP-Area function shows a substantial decrease of the LFP-Area over time and increase of the LFP thresholds with stimulation duration (Fig. 34).

This was a systematic finding in all experiments.
4. DISCUSSION

The present study investigated cortical response properties to the electrical midbrain stimulation in relation to the position of the recording electrodes in A1 and IC. It demonstrates that on one hand the responses that are at the corresponding portion within the tonotopic fields have larger amplitudes and smaller onset latencies; on the other hand we also found responses at the cortical positions 3 octaves higher than the place at which the stimulation electrode was positioned. This demonstrates a significant spread of excitation beyond the position of the stimulation electrode.

Methodological considerations

The present experiments reveal that a reproducible manual placement of the AMI is exceptionally difficult using the posterior approach. This corresponds to similar difficulties in human subjects (Lim et al 2007c; Samii et al 2006). In this approach the axis of the AMI is not parallel to the tonotopic axis, which implicates that the entire hearing range cannot be identified on a single AMI shank. Surprisingly, in the present study most electrodes were in low-frequency regions of the IC. This indicates not only a shallow penetration angle close to the surface of the IC, but also a too superficial placement of the electrodes. Although histological reconstructions were initially intended, positioning and extraction of the AMI lead to extensive tissue damage at the end of the experiment that precluded a histological analysis. This also indicates that the implantation of the AMI device may damage portions of the midbrain.

The present study concentrated on the LFPs because these are less likely affected by the antidromic electrical stimulation. The morphology of the LFP could well be compared to the electrical stimulation using a cochlear implant (Kral et al 2005) in the layers III/IV as
well as the evoked potentials from the past AMI studies on the guinea pigs (Lenarz M et al 2006).

The LFP response was dominated by a negative wave, corresponding to predominant inward (excitatory) currents evoked by the stimulation. Although mostly the orthodromic stimulation contributes to the LFPs, the antidromic spikes may elicit some LFP activity by axonal collaterals in the cortex. This effect will be, however, minor compared to the orthodromic stimulation. Also, due to the position of the stimulating electrodes in the central core of the IC, it is less likely to involve antidromic processes, as the corticofugal projection targets mainly the extralemniscal midbrain (Lim and Anderson 2007b).

Volume conduction must have affected the signals recorded in the cortex; nonetheless, stimulation site specificity was observed. This clearly demonstrates that despite the contribution of volume conduction the main components of the signals are active at or close to the recorded site. Further, previous mapping studies demonstrated that the field potentials recorded with microelectrodes are in fact more specific than generally assumed (Kral et al 2009).

**Threshold**

The here obtained stimulation threshold levels were between 28 and 54 dB, corresponding to 25.1 and 501.2 μA. Typically, the thresholds were at 30 or 32 dB, corresponding to 31.6 – 39.8 μA. These lowest stimulation currents correspond well to values observed for central stimulation strategies, like those observed in the brainstem implants (McCreery 2008) and the midbrain implants with other types of electrodes (Calixto et al 2013; Lim and Anderson 2006). This demonstrates the good physiological condition of the midbrain and the cortex during stimulation.
Dynamic range

Cortical responses demonstrated a DR that exceeds the DR for electrical stimulation in the cochlea (Tillein et al 2010; Raggio and Schreiner 1994). The reasons for this could be in the stimulation of extensive regions in the IC that converge on the same neuronal elements in the cortex. This effect could be responsible for the extended DR beyond the 6-10 dB typical for electrical stimulation (Kral et al 2006; Raggio and Schreiner 1994).

Also, previous studies demonstrated larger DR of midbrain stimulation in a rodent model. In a study by Lim and Anderson 2006, DR was evaluated by using 2 shanks (8 Chs per shank, silicon-substrate Michigan probe with 16 sputter-deposited iridium sites) for stimulating the ICC with less recording sites in the A1 as in the present study. Lim and Anderson calculated the DR by subtracting the stimulus levels corresponding to 75% and 25% of the maximum spike rate recorded on the A1 site in response to the stimulation site. However, they could not reach saturation rates in the A1. This resulted in lower overall estimated DRs. The ICC stimulation at thresholds were about 8 dB lower and DRs that were \( \geq 4 dB \) larger than in the cochlear implant stimulation (comp. Bierer and Middlebrooks 2002). The differences of our study compared to the study from Lim and Anderson were, among others, that we stimulated the ICC with human AMI, we calculated the DR for LFP responses (not for spike rate) in the A1, we were able to stimulate at higher levels (at 58 dB re 1 \( \mu A \)), and we recorded responses at more sites (32 MEA sites).

Additionally, an unusually high DR was observed when the peak amplitude was plotted as a function of the current level: for some stimulation sites, the DR exceeded 20 dB, much more than typical \(< 6 dB\) for cochlear implant stimulation (Kral et al 2006; Hartmann et al 1997). When comparing different recording sites, recorded simultaneously, a similarity of the DR was noted despite differences in the absolute amplitude. This indicates that what is
critical is the position of the stimulated electrode in the IC and not the position in the A1. However, a systematic investigation is required to resolve this issue.

In conclusion, the DR calculated for our study (13.41 ± 5.66 dB) was higher than the DR from the cochlear implant stimulation (comp. Kral et al 2006; Bierer and Middlebrooks 2002; Raggio and Schreiner 1994). Our DR results are in general in accordance with the results presented by Lim and Anderson (Lim and Anderson 2006), although, the present DR was even higher. In the present study, the stimulation electrode had much larger contacts, possibly explaining this difference.

**Stimulation level**

Stimulation of the ICC with varying stimulation levels were used in the present study. By that we were able to identify the threshold and absolute saturation levels and could perform the analysis at normalized stimulation levels (at 4 dB above threshold). Even in cases with similar BFs in the ICC, the thresholds of the cortical responses (as well as the DR) were significantly different. This demonstrates that stimulation of 3 different sites in a similar frequency region of the ICC can result in a site-specific response. Consequently, human AMIs can generate site-specific responses in the cortex. These were typical findings in all animals where such comparisons could be tested: when the median threshold was calculated from all recording positions in each animal, the thresholds were significantly different for the 3 stimulation positions. Previous AMI studies, with different materials and methods, further point to site-specific responses in the cortex (Lenarz M et al 2006). One interesting observation was the very small absolute deviation of the median, indicating that the variability of the threshold is small between the different cortical recording sites. That further indicates a large spread of excitation.
**Cortical recording position**

The specificity of the response at different cortical recording sites was evaluated by stimulating the same IC electrode. The LFP characteristics were evaluated as a function of distance between stimulation site in the ICC and recording site in the A1 (measured in octaves of the BF difference). If the stimulation in the ICC would be spatially restricted, responses at the ON-BF should be expected to have lower thresholds, higher DRs and larger amplitudes as well as larger temporal integrals and shorter latencies compared to the OFF-BF.

The thresholds were significantly lower at the OFF-BF positions when comparing to the ON-BF. This demonstrates that the cortical responses (LFP threshold) are recording-position independent (ON-BF or OFF-BF). However, it contradicts the above assumption of a spatially-restricted excitation and rather suggests a complex excitation-suppression pattern in the IC. It has been previously suggested that monopolar stimulation may generate a suppressive effect close to the stimulation electrode (Ranck 1975).

Furthermore, at 4 dB above the threshold the peak latency appeared the same at the ON-BF as well as the OFF-BF positions. But the temporal integration (LFP-Area) and the peak amplitude partially confirmed the above-mentioned expectation: they were significantly larger in the ON-BF group than in the OFF-BF group, and the onset latencies were significantly lower in the ON-BF positions than in the OFF-BF positions. All together the data indicate frequency-specific activation of the A1 through the AMIs, although more complex than expected.
Adaption behavior

Finally, the LFP amplitudes were compared as a function of the stimulation duration: the amplitudes consistently decreased throughout the 19 hours of experiment to approximately 30% of the original amplitude with the same type of stimulus at the same electrode. After the SSS has been interrupted (e.g., other stimulation paradigm not shown in the present study or in order to re-load the battery of current sources), the signals did not return to the original amplitude, indicating that the observed phenomenon corresponded to some kind of central habituation process. This adaptive behavior has also been seen in one of human AMI patients who was implanted in the dorsal IC region (Lim et al. 2008; Lim et al. 2007c). The cause of this adaptation is unclear. It is hypothesized that the rate and pattern of the stimulation may expand onto neurons located within the nonlemniscal IC region, an area which receives inputs from auditory and non-auditory centers (Winer 2005) and may be designed for stimulus specific adaptation or novelty detection (Antunes et al. 2010; Perez-Gonzalez et al. 2005). Nonetheless, this stimulus-specific adaptation has a very brief time constant, different from the present study. We used charge-balanced pulsatile stimulation well within the safety limits of the neuronal stimulation, and therefore neuronal damage appears an unlikely explanation.

However, decrease of the LFP-area and increase of the LFP thresholds with stimulation duration implicates that neural adaptation most likely occurs within the ICC. It would be interesting to analyze and examine stimulation and recording position dependency of this adaptive behavior for the LFP-characteristics and comparing to each stimulation positions in future studies.
Implications of the results

The use of the AMI is effective in stimulation of the midbrain, as also known from clinical trials with human subjects (Lim et al 2007c). The present study additionally demonstrates that it is feasible to stimulate the midbrain with all 3 probes. Nonetheless, the controlled positioning remains an issue to be resolved.

The DR observed was substantially larger than with the cochlear implant stimulation in cats (Kral et al 2006; Raggio and Schreiner 1994). As it appears unlikely that individual midbrain neurons have a larger DR for electrical stimulation than the auditory nerve neurons, the observed phenomenon is most likely the consequence of a convergence of activity from several excited midbrain neurons over large BF range. The difference most likely results from the higher neuronal density and the smaller distance from the electrode in the IC compared to the spiral ganglion.

Additionally, the current flow is very specific in the cochlea, including transversal and longitudinal currents that generate a very specific excitation profile (Kral et al 1998). In the midbrain, regions with inhibition and excitation may appear (Ranck 1975) that have not been described in the cochlear implant stimulation (Kral et al 2009; Snyder et al 2004; Bierer and Middlebrooks 2002; Kral et al 1998). Further, the DR was assessed using the LFPs which may additionally increase the DR. We evaluated the LFPs to minimize the influence of possible antidromic stimulation and therefore had to take this ambiguity into account.

The fact that the LFPs are similar in the ON- and OFF-BF is not straightforward; it could be related to stimulation of large population of neurons in the IC or to spread of excitation within the auditory cortex. The latter interpretation would be supported by the observation
of a difference in onset latency in favor of ON-BF positions. However, analysis of LFPs with the cochlear implant stimulation in the cat indicates that the hot spots are in the order of 3 mm in diameter (Kral et al 2009). The OFF-positions, particularly at 4 dB above threshold, should therefore show a clear amplitude gradient. This was not the case in the present study. In our interpretation the data indicate that monopolar stimulation activates large regions in the IC.

There is a partial discrepancy of the results shown; on one hand we observed some specificity to the stimulation site in the ICC, on the other hand also a non-specificity of the cortical responses in other measures. However, these results are not in contradiction. It is well known that the cathodic stimulation may lead to 2 different zones of interaction in the central nervous system: a region close to the electrode showing an “anodic surround” phenomenon and therefore not eliciting action potentials, and a 4 times larger peripheral area with excitatory responses (Ranck 1975). This specific pattern will differ for different stimulation sites in the ICC, leading to a large conjunct population of excited neurons, but a smaller region of disjunct populations that is inhibited with one stimulation site but excited with the other stimulation site. Further, axon diameter affects this relation too, and stimulation at the soma may yield a difference in the spatial organization of the excited region. We think the present result support such a heterogeneous excitation patterns in the ICC. One important aspect for further studies should include recordings within the ICC to investigate the patterns of stimulation.

The responses habituated in course of the experiment. This phenomenon corresponds to loudness adaptation observed in human subjects with the AMI electrodes (Lim et al 2007c). Such a behavior may reflect stimulus-specific adaptation observed in the cortex.
previously (Antunes et al 2010; Ulanovsky et al 2003), but most likely also other, non-
resolved neuronal fatigue mechanisms.

Previously, it has been suggested to use different electrodes in different portion of the
isofrequency plane to avoid this phenomenon (Lim et al 2007c). Here we suggest using
also more restricted stimulation configurations (bipolar or tripolar) to limit the spread of
excitation in the IC.
5. AUTHOR’S PEER REVIEWED ARTICLES

Investigation of a new electrode array technology for a central auditory prosthesis.
Calixto R, Salamat B, Rode T, Hartmann T, Volckaerts B, Ruther P, Lenarz T, Lim HH.

Abstract
Ongoing clinical studies on patients recently implanted with the auditory midbrain implant (AMI) into the inferior colliculus (IC) for hearing restoration have shown that these patients do not achieve performance levels comparable to cochlear implant patients. The AMI consists of a single-shank array (20 electrodes) for stimulation along the tonotopic axis of the IC. Recent findings suggest that one major limitation in AMI performance is the inability to sufficiently activate neurons across the three-dimensional (3-D) IC. Unfortunately, there are no currently available 3-D array technologies that can be used for clinical applications. More recently, there has been a new initiative by the European Commission to fund and develop 3-D chronic electrode arrays for science and clinical applications through the NeuroProbes project that can overcome the bulkiness and limited 3-D configurations of currently available array technologies. As part of the NeuroProbes initiative, we investigated whether their new array technology could be potentially used for future AMI patients. Since the NeuroProbes technology had not yet been tested for electrical stimulation in an in vivo animal preparation, we performed experiments in ketamine-anesthetized guinea pigs in which we inserted and stimulated a NeuroProbes array within the IC and recorded the corresponding neural activation within the auditory cortex. We used 2-D arrays for this initial feasibility study since they were already available and were sufficient to access the IC and also demonstrate effective activation of the central auditory system. Based on these encouraging results and the ability to develop customized 3-D arrays with the NeuroProbes technology, we can further investigate different stimulation patterns across the ICC to improve AMI performance.
Central auditory processing during chronic tinnitus as indexed by topographical maps of the mismatch negativity obtained with the multi-feature paradigm.


Abstract

This study aimed to compare the neural correlates of acoustic stimulus representation in the auditory sensory memory on an automatic basis between tinnitus subjects and normal hearing (NH) controls, using topographical maps of the MMNs obtained with the multi-feature paradigm. A new and faster paradigm was adopted to look for differences between 2 groups of subjects. Twenty-eight subjects with chronic subjective idiopathic tinnitus and 33 matched healthy controls were included in the study. Brain electrical activity mapping of multi-feature MMN paradigm was recorded from 32 surface scalp electrodes. Three MMN parameters for five deviants consisting frequency, intensity, duration, location and silent gap were compared between the two groups. The MMN amplitude, latency and area under the curve over a region of interest comprising: F3, F4, Fz, FC3, FC4, FCz, and Cz were computed to provide better signal to noise ratio. These three measures could differentiate the cognitive processing disturbances in tinnitus sufferers. The MMN topographic maps revealed significant differences in amplitude and area under the curve for frequency, duration and silent gap deviants in tinnitus subjects compared to NH controls. The current study provides electrophysiological evidence supporting the theory that the pre-attentive and automatic central auditory processing is impaired in individuals with chronic tinnitus. Considering the advantages offered by the MMN paradigm used here, these data might be a useful reference point for the assessment of sensory memory in tinnitus patients and it can be applied with reliability and success in treatment monitoring.
6. SCIENTIFIC GROUP WORK

The results of this thesis were from a collaborative work from a scientific group work:

**Idea to give birth and scientific supervisions:**  
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**Offline data analysis:**  
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This project was supported by the Cochlear GmbH and Georg-Christoph-Lichtenberg scholarship of the state of Lower Saxony, Germany.
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8. ACKNOWLEDGEMENTS

I would like to acknowledge the support and contribution of the following individuals

My Supervisors and Mentors:
Prof. Dr. Dr. Andrej Kral  
Prof. Dr. Simon Doclo  
Prof. Dr. Joachim Kurt Krauss  
Prof. Dr. Anaclet Ngezahayo
PD Dr. Minoo Lenarz  
Prof. Dr. Hubert H Lim  
Prof. Dr. Günter Reuter  
Prof. Prof. h.c. Dr. Thomas Lenarz

My Colleagues and Friends:
Dr. Pooyan Aliuos  
Ms. Nadine Alken  
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Dr. Roger Calixto  
Dr. Piera Ceschi  
Dipl.-Ing. Franziska Eckardt  
Dr. Dagmar Esser  
Prof. Dr. Beatrice Grummer  
Ms. Saskia Günter  
Ms. Tanja Hartmann  
Dr. Peter Hubka  
Dr. Souvik Kar
Dipl.-Ing. Nazia Khatir  
Mr. Rüdiger Land  
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Declaration

I herewith declare that I autonomously carried out the PhD-thesis entitled “Functional Enhancement of Auditory Activation through Multi-Site Stimulation across the Isofrequency Dimension of the Inferior Colliculus”.

No third party assistance has been used except who has been named on page 65. I did not receive any assistance in return for payment by any consulting agencies or any other person. No one received any kind of payment for direct or indirect assistance in correlation to the content of the submitted thesis.

I conducted the project at the following institutions:

Department of Otolaryngology
Institute of Audioneurotechnology
Hannover Medical University

The thesis has not been submitted elsewhere for an exam, as thesis or for evaluation in a similar context.

I hereby affirm the above statements to be complete and true to the best of my knowledge.

Hannover 07.01.2014

Dipl.-Ing. Behrouz Salamat