EFFECT OF AGE ON SPECTRAL DISTRIBUTION OF CLICK AND TONEBURST EVOKED OTOACOUSTIC EMISSIONS IN INFANTS

Chandni Mallik

Register No: 10AUD006

A Dissertation Submitted in Part Fulfillment of the Degree of

Masters of Science (Audiology)

University of Mysore

Mysore

ALL INDIA INSTITUTE OF SPEECH AND HEARING

MANASAGANGOTHRI, MYSORE - 570006

May – 2012

Dedicated To

My Family

Mummy, Papa,

Di and Bhai

Certificate

This is to certify that this dissertation entitled ***'Effect of Age on Spectral Distribution of Click and Toneburst Evoked Otoacoustic Emissions in Infants'*** is a bonafide work in part fulfillment for the degree of Master of Science (Audiology) of the student with Registration No. 10AUD006. This has been carried out under the guidance of a faculty of this institute and has not been submitted earlier to any other University for the award of any other Diploma or Degree.

Mysore,

June, 2012

Dr. S. R. Savithri

Director

All India Institute of Speech and Hearing

Manasagangothri,

Mysore - 570006

Certificate

This is to certify that this dissertation entitled ***'Effect of Age on Spectral Distribution of Click and Toneburst Evoked Otoacoustic Emissions in Infants'*** has been prepared under my guidance and has not been submitted earlier to any other University for the award of any other Diploma or Degree.

Mysore,

June, 2012

Ms. Mamatha N. M.

Guide

Lecturer in Audiology

Department of Audiology

All India Institute of Speech and Hearing

Manasagangothri,

Mysore - 570006

Declaration

This dissertation entitled ***'Effect of Age on Spectral Distribution of Click and Toneburst Evoked Otoacoustic Emissions in Infants'*** is the result of my own study under the guidance of Ms. Mamatha N. M., Lecturer in Audiology, Department of Audiology, All India Institute of Speech and Hearing, Mysore, and has not been submitted earlier to any other University for the award of any other Diploma or Degree.

Mysore, **Register No:** 10AUD006

June, 2012

Acknowledgement

*Words are not enough to thank my guide, **Ms. Mamatha. N. M. ,** for her wonderful patience. I express my sincere & heartfelt gratitude to my guide. Madam, I thank u for rendering me constant support and guidance throughout & I sincerely thank you for having trust & confidence on me. Your words and your guidance will remain with me throughout my life.*

*I would like to thank **Prof. S. R. Savithri**, Director, All India Institute of Speech and Hearing, to permitting me to carry out this study.*

***Mummy** and **Papa**, you both are so very special to me. You have blessed me with such a wonderful life. You are my biggest strength…. I am deeply indebted to you for your motivation, guidance and prayers. I feel so blessed to be called as your daughter.*

*I would also like to take the opportunity to thank my dearest and most lovable **bro** and **di,** who are always by my side. Love u both….*

*My heartfelt thanks to **Dr. Animesh Barman**, Head of the Department, Audiology, for allowing me using the test equipments for the study.*

*I whole heartedly thank **Dr. M. S. Vasanthalakshmi** and **Mr. Santhosh C. D.** for their help and guidance regarding statistical analysis. Thank you so much madam for all your valuable suggestions, which has helped me during my study. Your extra patience and dedication to work has always encouraged me. You have always been there to clear all my doubts regarding statistics, be it day or night…. Thank you once again madam.*

*My sincere thanks to **Mr. Vijay Kumar Narne** and **Mr. Kishore Tanniru** for sharing their valuable knowledge. Thank you sir, for adding on my knowledge regarding OAEs….*

*I thank all **my teachers** for motivating me and immensifying my knowledge.*

*Thanks to **Mr. Srikar,** for helping me out in finding infants for my study and also to lend a hand for my data collection.*

*A very special thanks to **Ms. Anisha,** for her sincere efforts to give finishing touch to my study with her expertise. I would be ever thankful to u….*

*Thanks to **Mr. Ratul** for helping me throughtout my study. You really were of a real great help to me Ratul. I thank you once again for helping me with my references and all the other helps you have rendered to me.*

*Thanks to **Ms. Garvita, Ms. Deepthi, Ms. Kruthika, Ms. Apoorva, Ms. Aparna** , **Ms. Spoorthi.** Thank you for the support you guys have rendered to me. It was a real nice time with you people. Every moment spent with you will be cherished forever.*

*I thank **Mr. Narendranath** for helping me out in my data collection. I thank you for supporting me and lending your helping hand when I required…. Thanks.*

*Thanks to all the **library staffs**, who have helped in searching and issuing books and other study materials.*

*I also like to thank **Mr. Suhas** for helping me find the articles required for my study. Thank you so much….*

*I thank all the co-operative **parents of the infants** taken for my study, without which the work would not have been done.*

*Thanks to all my lovely **classmates,** for making my days wonderful both on-campus and off-campus. Thanks guys….!!*

*Last but not least, I thank **almighty GOD** to have gifted me with all such wonderful people around who make my life real beautiful.*

Table of Contents

List of Tables

List of Figures

Chapter 1

INTRODUCTION

Otoacoustic emissions (OAEs) are sounds that originate in the cochlea and propagate through the middle ear and into the ear canal, where they can be measured using a sensitive microphone. OAEs were predicted by Gold in 1948 and its existence was first demonstrated experimentally by Kemp in 1978. Because of its relative simplicity, better sensitivity and objectivity of the technique, the OAEs are a promising means for monitoring cochlear function. OAEs are the property of healthy normal functioning cochlea, generated by active frequency selective, non-linear elements within the partition, the critical components being the outer hair cells (Kemp, 1988). There are 4 types of otoacoustic emissions. They are as follows:

- Spontaneous otoacoustic emissions (SOAEs) - Sounds emitted without an acoustic stimulus i.e., spontaneously.
- Transient evoked otoacoustic emissions (TEOAEs) - Sounds emitted in response to acoustic stimuli of very short duration; usually clicks, tone-bursts can also be used.
- Distortion product otoacoustic emissions (DPOAEs) - Sounds emitted in response to the presentation of 2 simultaneous tones of different frequencies, and
- Stimulus-frequency otoacoustic emissions (SFOAEs) - Sounds emitted in response to a continuous tone.

Evoked otoacoustic emissions (EOAEs) are easy to perform, quick, non-invasive and cost effective with high sensitivity. The primary purpose of otoacoustic emission (OAE) tests is, to determine cochlear status, specifically the outer hair cell function. This information can be used to (1) screen for hearing, particularly in neonates, infants, or individuals with developmental disabilities, (2) partially estimate hearing sensitivity within a limited range, (3) differentiate between the sensory and neural components of sensorineural hearing loss, and (4) test for functional (feigned) hearing loss. The development of EOAEs has gained much interest because they are used as a valid and relatively quick test to assess cochlear integrity in the very youngest subjects.

Transient evoked OAEs (TEOAEs or TrOAEs) are frequency dispersive responses following a brief acoustic stimulus such as click or tone burst (Kemp, 1978; Norton & Neely, 1987). A click stimulus covers a broad frequency range, whereas, a toneburst is a brief duration pure tone stimulus. The TEOAE recording has proved to be the most sensitive among the OAE techniques and provides frequency specific information from 500 Hz up to 5 kHz and have decreasing latencies with increasing stimulus frequencies. The evoked response from a click covers the frequency range up to around 4 kHz, while a toneburst elicits a response from the region that has the same frequency as the pure tone. TEOAEs exhibits compressive non-linearity.

Click evoked otoacoustic emissions (CEOAEs) have been widely used to assess the functioning of cochlear outer hair cells. The click stimulus has a broad spectrum, and hence consequently it stimulates broad frequency region of the cochlea in a single measurement. Lutman, Mason, Sheppard, and Gibbin (1989) opined that, the presence of CEOAEs are a powerful indicator of normal hearing. Therefore it has been applied as a general tool in universal neonatal hearing screening (UNHS) programs.

Toneburst evoked OAEs (TBOAEs) include narrow bandwidth tone stimuli, which has stimulus energy concentrated on a particular area of the basilar membrane and elicits a more frequency-specific cochlear response. TBOAEs are more prominent than CEOAEs and at the frequencies of spontaneous otoacoustic emissions, prominent peaks in both click and tone burst evoked otoacoustic emissions were present (Probst, Coats, Martin & Lonsburry, 1986).

An advantage of toneburst stimulus is that more energy can be introduced in a specific frequency range compared to click, which is a more frequency dispersive stimuli. CEOAEs were difficult to detect in 20% of ears, demonstrating a broadband pattern. A broadband stimulus may not be ideal for clinical and screening purposes. Rather a frequency specific stimulus such as relatively long duration toneburst may be necessary to obtain the highest possible incidence of normal ears (Probst, Coats, Martin & Lonsbury-Martin, 1986).

There have been few studies done on the effects of age on evoked emissions. It was suggested that, age related changes must be understood, if EOAEs are to be used as a clinical tool (Norton & Widen, 1990). The anatomy of the infant's ear is different from that of the adult's ear. These differences include changes in the external and middle ear acoustics, developmental changes in the cochlear mechanics, and age-associated cochlear changes due to normal wear and tear. There are also differences in the size, shape, and tissues of neonates and infant ear canals compared to adult ears. The tympanic membrane is more horizontal in neonates than adults (Anson & Donaldson, 1981).

As mentioned by Merchant, Horton and Voss (2010), during the first 6 - months of life there is an increase in size of the ear canal diameter, ear canal length and the middle ear cavities. There is also a change in the orientation of the tympanic membrane. Tightening of the ossicular joints connecting the ossicles, formation of the bony ear canal wall and a decrease in the overall mass of the middle ear because of changes in bone density and loss of mesenchyme are also seen. Due to these anatomical differences between the infant's ear and adult's ear, the EOAE are robust in normal infant's ear than in normal adult's ear (Norton & Widen, 1990). And also the neonate's TEOAE has higher amplitude than that measured in adults, and the energy is present over a wider range of frequencies than that from the adults.

As reported by Moleti, et al. (2008), latency of CEOAEs in infants is 2–3 ms shorter compared to adults in the frequency range of 1.5–2.5 kHz. This could be explained by a reduced middle ear forward transmission, as a similar latency decrease was found with a 10 dB increase of stimulus intensity (Sisto & Moleti, 2007). Another explanation for the differences in OAE characteristics could be an immature cochlea. Although the histological development of the human cochlea seems completed at about 26 weeks post conception, there are still ultrastructural processes which are incomplete. For example, the strength of coupling of stereocilia to the tectorial membrane can change the properties of inner ear tissues, e.g., stiffness of the basilar membrane, and thereby change OAE characteristics (Pujol & Lavigne-Rebillard, 1995).

Need for the study

Infants are more responsive to signal in the frequencies below 4 kHz compared to high frequencies. The behavioral response of infants for frequency specific sounds shift towards higher frequency region as they mature. Behavioral testing of children and infants is a cornerstone of pediatric audiology (Madell, 1988).

The estimation of the hearing sensitivity of infants using behavioral response is a difficult task. The disadvantage of behavioral observation audiometry (BOA) is that, it is difficult to estimate tester bias. Also the responses of infants and young children extinct quickly without reinforcement, and a wide variety of responses are noted in youngsters. To deal with these difficult to test subjects, the objective tests such as acoustic immittance, OAEs and auditory evoked responses have been developed.

The OAE results indicate a greater sensitivity in detecting early cochlear damage. In a study done by Kok, Zanten and Brocaar (1992), it was found that, newborns failing the EOAE screen in the first 24 hours after birth can pass, if retested 1 day later, simply because of growth of EOAE strength. Otoacoustic emissions are an essential part of the audiologic evaluation and are routinely used in the pediatric population to verify behavioral responses and to obtain additional frequency specific information. In addition, they are routinely used in newborn hearing screening programs across the world to validate auditory thresholds obtained via other techniques, and to assess the cochlear contribution to hearing.

Researchers have advocated the use of OAEs as an excellent tool for diagnosing hearing loss in infants and children. Lutman, Mason, Sheppard, and Gibbin (1989) opined that, the presence of CEOAEs is a powerful indicator of normal hearing. An advantage of CEOAE is that all accessible parts of cochlea are tested simultaneously. To obtain a response from a restricted band width, the click stimulus can be replaced by a toneburst stimulus.

TEOAEs can also be evoked using toneburst (TBOAEs) which have narrower bandwidth and energy concentration around the center frequency of toneburst. Fourier analysis of TBOAEs indicates their spectral composition similar to that of the evoking toneburst (Norton & Neely, 1987; Stover & Norton, 1993). Although tone burst stimuli have greater frequency specificity compared to click stimuli, TBOAEs have not been routinely used in pediatric populations.

TBOAE is a useful method for investigating cochlear function at specific frequency ranges in neonates. Further studies of TBOAE time-frequency analysis and measurements in newborns are needed. Very few studies have been carried out using TBOAEs for hearing assessment in neonates and young children. It is suggested that lower frequency TBOAEs may elicit better and more robust OAE response than CEOAEs in the lower frequency region, and thus assist in reducing the high referral rate found in traditional CEOAE in neonatal hearing screening programs. The emissions in response to toneburst are quite frequency specific and are often prominent than CEOAEs.

The sensory habituation of infants to a puretone was studied by Bridger (1961). He showed that changing the frequency of the puretone led to enhanced startle response after habituation to a particular tone. This indicated that infants can discriminate between different frequencies. Such changes in the behavioral responses of infants for frequency specific sounds shift towards the higher frequency region. Thus, it suggests that there could be some amount of re-organization within the auditory structures, especially in the cochlea. Hence the present study has been designed to study, the TEOAE responses at different frequency bands which could highlight the relationship between the behavioral & physiological changes.

Various studies have been carried out in infants and adults on CEOAEs and TBOAEs. As to the research and clinical application of TBOAEs, studies have been mainly focused on adult population. Whereas, there are very limited number of studies which have been done on infants. Hence there is a need to study the utility and efficacy of TBOAEs among infants. Also, there are very limited studies on the spectral information of TEOAEs as the age advances. The information regarding the comparison between CEOAEs and TBOAEs in infants is very limited. Hence the present study was undertaken.

Aim of the study

The aims of the present study were:

1) To investigate the influence of age on click and toneburst evoked OAEs, and to investigate the pattern of frequency shifts with age.
2) To study the sensitivity of click & toneburst-evoked OAEs and to monitor the frequency specific maturational changes in the cochlea.

Chapter 2

REVIEW OF LITERATURE

The concept of extraneous acoustic energy reflected externally from the cochlea was theorized by Gold as early as 1948. However, Kemp (1978) was the first to identify these reflected energies as otoacoustic emissions (OAEs) in the human external ear canal. Kemp's (1978) description of OAEs has significantly changed the understanding of cochlear processing and has influenced clinical audiology in various ways. Ever since OAEs were first defined by Kemp (1978), there has been growing interest in their clinical application to identify the presence of hearing loss (Bonfils & Uzeil, 1989; Kemp et al., 1987; Bray & Kemp, 1987; Collect, 1991; Harris, 1990; Martin et al., 1990a; Martin et al., 1990b; Lonsbury-Martin & Martin, 1990; Nelson & Kimberley, 1992). In general, the results of these investigators showed that the evoked otoacoustic emissions (EOAEs) are present in majority of normal hearing subjects and are absent in subjects with hearing loss once it exceeds 30 to 50 dB HL.

OAE measurements are noninvasive and quick to perform and they can appear spontaneously, but for clinical purposes they are evoked by transient stimuli (TEOAEs) or they are also measured using two-tonal stimulations which are termed as intermodulation products (DPOAEs). TEOAEs and DPOAEs represent objective measurements of the active micromechanical function of the outer hair cells in the inner ear. These two are now becoming a part of routine clinical investigation of hearing both in infants and adults. Although infants tend to show OAE response patterns that are inherently physiologically noisier than noted with adults, OAEs has tremendous application in hearing screening programs (Northern & Downs, 2006).

According to the Joint Committee on Infant Hearing (JCIH), use of OAE is a mandatory component of the audiologic test battery.

2.1 Effect of age on evoked otoacoustic emissions

There have been few studies done on the effects of age on evoked emissions. It was suggested that, age related changes must be understood, if EOAEs are to be used as a clinical tool (Norton & Widen, 1990). The anatomy of the infant’s ear is different from that of the adult’s ear. These differences include changes in the external and middle ear acoustics, developmental changes in the cochlear mechanics, and age-associated cochlear changes due to normal wear and tear. There are also differences in the size, shape, and tissues of neonates and infant ear canals compared to adult ears. The tympanic membrane is more horizontal in neonates than adults (Anson & Donaldson, 1981).

As mentioned by Merchant et al. (2010), during the first 6 months of life there is an increase in size of the ear canal diameter, ear canal length and the middle ear cavities. There is also a change in the orientation of the tympanic membrane. Tightening of the ossicular joints connecting the ossicles, formation of the bony ear-canal wall and a decrease in the overall mass of the middle ear because of changes in bone density and loss of mesenchyme are also seen. Due to these anatomical differences between the infant’s ear and adult’s ear, the EOAE are robust in normal infant’s ear than in normal adult’s ear (Norton & Widen, 1990). And also the neonate’s TEOAE has higher amplitude than that measured in adults, and the energy is present over a wider range of frequencies than that from the adults.

Transient evoked otoacoustic emissions (TEOAEs) are frequency dispersive responses arising in the cochlea on presentation of a brief acoustic stimulus such as a click or a tone burst that can be measured in the external ear canal (Kemp, 1978; Norton & Neely, 1987). TEOAEs are measurable in all individuals with normal middle ear and normal cochlea (Kemp, 1978; Johnson & Elberling, 1982; Probst, 1986). Since OAEs are found to be existing in almost all normal ears, it is considered to be a sensitive tool to detect even minor changes in the hearing status which lead to absence of OAEs (Norton & Neely, 1987; Bonfils & Piron, 1988).

It is known that over 80 % of healthy newborns have an open aqueduct of cochlea (Wlodyka, 1978). This leads to increased CSF pressure to the cochlea. As the cochlea matures, the pressure is reduced and therefore there is improvement seen in TEOAE responses. Phillip (1979) has stated that the CSF pressure at 1 day after birth is almost double of that of at 3 days age. Chuang, Gerber and Thronton (1993) have opined that the increase in amplitude of TEOAE responses is more for short latency, high frequencies. The author could not postulate any middle ear or external ear related changes that would have an effect on the change in TEOAE responses. Only reason that was opined is the maturational process. Salomon, Anthonisen, Groth and Thomsen (1992) stated that the changes in TEOAE responses were nowhere related to effusion or any other structural developments of the middle ear. Rather it depends upon the amount of oxygenation to the cochlea. Their findings were supported by Sohmer and Freeman (1993) who found that there is a reduced concentration of oxygen in cochlea in-utero. Soon after birth, as normal breathing starts, there is an increase in oxygen level in neonatal blood.

A conclusion was made by Thronton, Kimm, Kennedy and Cafarelli-Dees (1993) that, TEOAE responses can be adversely affected by middle ear pressure differences and external ear obstruction in neonates but other factors such as maturation and oxygenation to cochlear hair cells also contribute to it. Vohr, White, Maxon and Johnson (1993) in their study have found that most of the neonates have debris in the external ear canal. They obtained TEOAE responses before and after cleaning of the external ear canal. They found that there was an increase in TEOAE responses after cleaning the debris. The results of TEOAE in cleaned ears were identical to that of ears with no obstruction. Therefore, it was concluded that only factor that contributes to the changes in TEOAE response is an external obstruction. However, they found that middle ear pressure and presence of fluid play little role.

In adults, though the occurrence of TEOAEs has been found to be 100%, it is slightly lower in neonates and infants. Bonfils, Dumont, Marie, Francois and Narcy (1990) measured TEOAEs in neonates ranging in age from 2 hours to 4 days, and it was found that 98% of the ears tested had presence of emissions. There was found to be no significant difference in the occurrence of emissions between 1 and 4 days postpartum, but the occurrence of OAEs increased within the first 24 hours. The incidence of OAEs in neonates and infants are same as in adults. Kok, et al. (1992) and Vohr et al. (1993) observed a 100% increase in the ears with emissions when the ears were first tested 3 to 51 hours after birth and was repeated at least 24 hours later. Audiological screening with TEOAE recording showed an overall 100% sensitivity and 99.02% specificity (Capua, Felice, Costantini, Bagnoli & Passali, 2003). Kemp (1978) from his initial reports on TEOAEs noted that energy in the frequency region near 1500 Hz dominated the response to acoustic click stimuli and that the response amplitude was related to the stimulus magnitude in a complex way.

Robust response amplitude is a salient property of TEOAEs obtained from healthy newborns. The data in literature indicate that the neonatal TEOAE responses, with respect to the adult ones, are characterized by a large signal amplitude (Norton et.al., 2000; Probst et.al., 1989); and a wider and more uniform spectrum, shifted occasionally toward higher frequencies (Norton, 1993). TEOAE amplitude in infants increases from 1 to 9 months of age, where as the amplitude decreases in older children aged 4-13 years (Norton & Widen, 1990; Widen, 1997). As the child develops, the level and the frequency content of OAEs change. In newborns, the TEOAE levels are higher than that in adults (Prieve, 2007). In the same study, TEOAE levels were found to be higher in case of infants in comparison to that in older children and young adults

These changes in the OAE levels across age are frequency dependant, with no significant decrease for frequencies 1500 Hz and higher (Prieve, et al., 1997a, 1997b). Among preterm infants, the TEOAE level increases with the postconceptional age (Smurzynski, 1994). Further, few authors (Hancur, 1999; Welch, Greville, Thorne and Purdy, 1996) have found that the TEOAE levels increase between birth and 4 weeks of age. Zanten et al. (1995) observed that TEOAE amplitude reached maximum values for infants at a postconceptional age of 47 weeks.

The response amplitude of OAEs recorded from neonates is up to 10 dB stronger than those in the adults (Kemp, Ryan & Bray, 1990; Norton & Widen, 1990). Contrary to these observations, Johnsen, Bagi and Elberling (1983), Elberling, Parbo, Johnsen and Bagi (1985) and Johnsen, Parbo and Elberling (1989) found no such differences; they reported similar values of magnitude in both adults and neonates. Kok et al. (1993) reported that the magnitude of TEOAE grows idiosyncratically with age during the first few days of life of normal newborns. They also reported that the median response amplitude for infants is approximately 20 dB SPL, and that the response reproducibility is directly related to the response amplitude. Prieve et al. (1997) stated that, the largest changes in the TEOAE amplitude occurs between birth and 4 years of age. The average TEOAE response amplitude to default stimuli (80 dB) declines from 20 dB to approximately 15 dB for preschool children and to 10 dB for adults. Several authors have reported that the TEOAE amplitude reduces with age (Kok et.al, 1992; Norton et.al, 1990, 2000).

The TEOAE measures proposed for clinical measurements include response reproducibility and signal-to-noise ratio (SNR) as a function of frequency. A 50% or greater reproducibility indicates the presence of an emission in the frequency region of interest (Kemp et al., 1990). They also found that reproducibility of 50% or greater is usually associated with a SNR of 3 dB or more. However, the relationship between the reproducibility and SNR measures is complex. Costa, Almeida and Sampaio (2009) analyzed TEOAE reproducibility, amplitude, and SNR on premature infants. The results showed that TEOAEs were present in 71% of the infants. And the frequency of 3 kHz presented a better performance in the average reproducibility, amplitude and SNR. Kapoor and Panda (2006) analyzed the TEOAE amplitude, wave reproducibility and the SNR in neonates (0-1 month), infants (1 month- 1 year), and children (1-6 years). The results showed that the mean amplitude in the neonates was significantly higher than that in infants or children, and the wave reproducibility was constant across the age. The mean SNR for all neonates, infants and children were well above 3 dB at frequencies 1.5 k, 2 k, 3 k and 4 kHz. The results also showed that the neonates had the lowest SNR ranging between 3.47 to 9.62 dB, whereas the infants showed the highest SNR ranging between 6.13 to 13.11 dB. Kei, et al. (1997) found a significant difference in SNR across sex, with females showing a higher mean

SNR and also the right ear was found to have higher values in reproducibility and response level than the left ear.

Neonatal TEOAEs have been reported to reveal a boost in the energy in the high frequency region when compared with responses obtained from adults with normal hearing (Kemp et.al., 1990; Lafreniere et.al., 1991; Norton & Widen, 1990; Smurzynski, 1994). As Norton & Widen (1990) noted, the increase may be due to obvious differences in the size and shape of the middle and outer ear systems and their effects on the resonance characteristics of the ear, rather than changes in the cochlear mechanics. The frequency spectra of OAEs in neonates and infants are of a wider frequency range than in older children or adults, with more power at the high frequencies. In studies done on neonates, a frequency range of 0.7-5 kHz is reported (Kemp, et al, 1990; Uziel & Piron, 1991). Collet (1991) documented a similar lowering of the dominant frequency in the emission spectrum as a function of age in a subject group with ages ranging from 6 weeks to 83 years.

Since the analysis of TEOAE is reproducible, diagnostically accurate, easy to perform and non-invasive, TEOAEs are presently the method of choice for neonatal audiological screening both in the general population as well as in the high-risk infants (Probst et.al., 1991). TEOAEs as mentioned earlier, can be measured following the presentation of a brief acoustic stimulation using a click (CEOAEs) or toneburst (TBOAEs) stimuli.

2.2 Click- evoked otoacoustic emissions (CEOAEs)

Most research works on TEOAEs have been performed using click stimulus. There have been many studies showing that the CEOAEs can be recorded in most normally hearing adults & more recently in normal newborns and in babies admitted to a special care unit (Stevens et al., 1989). CEOAEs may be recorded in neonates at much lower gestational ages (Cope & Lutman, 1993).

Stevens et al. (1990) recorded CEOAEs in 30 normal newborns and showed that 96% produced an emission at the highest level tested of 41 dB nHL. These results prove high degree of confidence that can be placed in the test when using it to detect hearing impairments. Uziel and Piron (1991) recorded CEOAEs in neonates ranging in age from a few days to 2 months after birth. The results showed that, in neonates, the emissions were stronger and covered a wider frequency range than those from the adults. They further stated that, in about 70% of the ears, a wide range of activity was observed in the OAE spectra between 0.7 kHz up to 5 kHz, with several narrowband peaks. The peaks of maximum energy were displaced towards high frequencies when compared to the adult OAE spectra.

The consistency of CEOAE response waveforms has been demonstrated by Johnsen and Elberling (1982), who performed retest measurements in the same ear at 4-5 week intervals and found the response pattern to be almost unchanged. The CEOAEs of neonates appear to change with increasing age.

2.3 Toneborst- evoked otoacoustic emissions (TBOAEs)

The incidence of TBOAEs is strongly dependent on the frequency of the stimulus. Probst, et al. (1986), in their study on normal hearing individuals, reported a greater percentage of subjects demonstrating emissions in response to 1.5 kHz than to 0.5 kHz tonebursts. Hauser, Probst and Lohle (1991) reported similar observations. Concerning the prevalence of TBOAEs, different studies have reported different findings. Liu, Song, Liu, and Zhao, X. Y. (1996) noted a 100% prevalence rate for 1 kHz TBOAE in 35 normal hearing adults. Similarly, Chan and McPherson (2000) found that 1 kHz tone stimulus with high stimulus level could elicit TBOAEs in all normal hearing adults tested. However, Probst et al. (1986) reported that not all the tested adult ears responded to all tone burst stimuli (ranging from 0.5 to 3 kHz). They found the percentages of detected emissions for stimuli at 0.5, 1, 1.5 and 3 kHz were 36%, 82%, 100% and 93%, respectively.

As there is no standard protocol for TBOAE measurements, results presented in the literature are generally not directly comparable. A variety of different recording instruments, stimulus levels, stimulus center frequencies, stimulus rates, number of averages, and analysis windowing parameters have been employed.

Previous studies have utilized widely differing stimuli, ranging from very short clicks to longer duration tonebursts (TBs). It was found that the stimulus parameters influenced the incidence of evoked otoacoustic emissions. If evoked otoacoustic emissions are to be utilized eventually in clinical diagnostic testing, it is important to know the efficacy of each stimulus type in eliciting emissions (Probst, Coats & Lonsbury-Martin, 1986). They carried out a study on normal hearing young adults using spontaneous, click- and toneburst- evoked otoacoustic emissions. It was found that toneburst-evoked emissions were often more prominent than click-evoked emissions and no spontaneous emissions were detected. Highly similar peaks were present in the spectra of toneburst-evoked emissions within the range of toneburst spectra.

The magnitude and the spectral characteristics of the TBOAE appear primarily to be dependent on the frequency of the evoking stimulus (Cope & Lutman, 1993). Wit and Ritsma (1979), documented that the higher the frequency of the stimulus the smaller the response, even when the stimuli of different frequency are presented at the same sensation level. Elberling, et al. (1985) have indicated that the TBOAE could be predicted accurately by convolving the CEOAE with the waveform of the stimulus. TBOAEs at 1 kHz were more robust than CEOAEs in terms of emission response level and signal-to-noise ratio (SNR) at both 1 and 1.5 kHz frequency bands. The prevalence rate for CEOAE and TBOAE responses in these two frequency bands was significantly different (Zhang et.al. 2007).

A robust 1 kHz component of a CEOAE response is reported to likely be contaminated by low frequency noise in a normal hearing individual (Norton et.al, 2000; Welch et.al., 1996). To resolve this, tone burst evoked OAEs (TBOAE) would be a promising supplement to the conventional CEOAE screening technique, since tone burst stimuli can elicit a more frequency-specific response (Hall, 2000; Kemp, et al., 1990). 1 kHz TBOAEs could be recorded clearly in healthy ears, and their test-retest reliability is comparable to that of CEOAEs (Chan et al., 2000; McPherson et al., 2006).

From the review of literature it is evident that, most of the research has been carried out using click stimuli, and also the studies have been concentrated on adult population. Very few studies have been carried out using toneburst stimuli for hearing screening and hearing assessment. It is also evident from the review of literature that very limited studies have been done on the spectral information of TEOAEs as the age advances. The information regarding the comparison between CEOAEs and TBOAEs in infants is limited. Hence the present study was undertaken.

Chapter 3

METHOD

The pattern of frequency shifts with age for click and toneburst transient evoked otoacoustic emissions (TEOAEs) was studied in the present study. The relationship between click and toneburst evoked otoacoustic emissions (CEOAEs & TBOAEs respectively) in infants was also compared. To investigate these, the following method was employed:

Subjects

The study was conducted on forty infants (80 ears) in the age range of 0- 12 months. The forty infants were further categorized into four subgroups based on their age as shown in the table 3.1.

Table 3.1: *Age range for the four subgroups and number of infants in each subgroup.*

Groups	Age range	Number of infants
Group I	0 - 4 months	10
Group II	4 - 6 months	10
Group III	6 - 8 months	10
Group IV	8 - 12 months	10

***Subject Selection criteria*:** All the 40 infants, who came for hearing evaluation after they got discharged from hospital, were evaluated.

The infants considered in the study had to meet the following criteria:

- Normal otoscopic findings.

- No middle ear pathology and middle ear infections as reported by ENT specialist.
- Normal birth history.
- Healthy with no symptoms of cold or ear discharge at the time of assessment.
- No complaint and prior history of any high risk factors.
- No complaint and history of any neurological symptoms.
- No other otological history as reported by parents.
- Age appropriate response at minimum levels in behavioral observation audiometry.
- Normal outer hair cell functioning, ensured by recording Transient Evoked Otoacoustic Emissions (TEOAEs).
- Normal hearing sensitivity, ensured by recording Auditory Brainstem Response (ABR).

Instrumentation

The following instruments were used for the study:

- An otoscope to observe the status of the external auditory canal and tympanic membrane.
- A calibrated Grason Stadler Inc. Tympstar middle ear analyzer (GSI Tympstar, version 2) to carry out tympanometry and acoustic reflexometry.
- A calibrated two-channel diagnostic audiometer Madsen Orbiter-922 (version 2) with impedance matched loudspeakers, to present the stimuli for Behavioral Observation Audiometry (BOA) or Visual Reinforcement Audiometry (VRA).
- A personal computer based Intelligent Hearing Systems (IHS) Smart EP version 3.94 evoked potential system to record ABR.

- A calibrated otoacoustic emission system ILO version 6 software (Otodynamics Ltd., UK), to record TEOAEs for both clicks and the tone burst stimuli.

Test environment

The testing was carried out in a sound treated room with noise levels within permissible limits as per ANSI S3.1 (1991). The test room was comfortable enough for the infants in terms of light and temperature.

Test procedure

Case history

Detailed information about the history of prenatal, natal, and post natal medical conditions were collected for all the infants. A detailed report regarding the auditory behaviour of the infant at home for various environmental sounds like calling bell, voices from TV or radio, pressure cooker, whistle, name call, repetitive babbling etc., were obtained from the parents or caregivers.

High Risk Register (HRR)

Medical reports were reviewed to make sure that all the infants were devoid of various risk factors and other medical conditions. This was done by administering the modified High Risk Register developed by Anitha and Yathiraj (2001) to rule out the high risk factors in infants.

Otoscopic examination

Visual examination of the ear canal and the tympanic membrane of infants were done using a hand held otoscope. This was done to rule out the presence of wax, foreign bodies in the ear canal and / or external and middle ear pathologies.

Tympanometry

Tympanograms were obtained using 1000 Hz probe tone frequency for infants till the age of 6 months and 226 Hz probe tone frequency for infants above 6 months of age. The pressure was swept from +200 to -400 daPa with a positive to negative sweep with the pump speed of 200 daPa / sec. The probe intensity was 85 dB SPL for 226 Hz and 75dB SPL for 1000 Hz probe tone frequency.

Acoustic reflex measurement

Ipsilateral acoustic reflex thresholds (ARTs) were measured for pure tone stimuli of 500 Hz, 1 kHz, 2 kHz and 4 kHz using 226 Hz and 1 kHz probe tone frequencies. ART for 1 kHz was not recorded when 1 kHz probe tone was used as it might interact with the reflex activator signal frequency causing artifacts (Wilson & Margolis, 2001).

ARTs were determined using an ascending technique by increasing the intensity of stimulus in 5 dB steps from 60 dB HL as the starting intensity, until a significant change in the acoustic-admittance occurred immediately after the stimulus. The minimum intensity at which the repeatable change in the acoustic-admittance value is observed by taking the criterion as 0.03 mmhos was considered as acoustic reflex threshold (ART).

Behavioral Observation Audiometry (BOA)

The behavioral responses (eg., startle, eye widening, a grimace, cessation or initiation of sucking, crying, arousal from light sleep etc.) for the auditory stimulus were observed for warble tones from 500 Hz to 4 kHz separated in octaves and for speech stimuli. It was carried out in free field and in a double-room situation. The infants were seated comfortably on the parents / caregivers lap at a distance of 1 meter from the loudspeakers and at an azimuth of 45º in the observation room. One clinician was present in the observation room to draw the attention of the infant to the midline and to observe the unconditioned responses. The other clinician was in the test room, and presented the test stimuli sequentially with the initiation level being decided below the level at which the infant is expected to exhibit some kind of auditory behaviour, as reported by the parents/caregivers. The lowest levels of presentation of each of the stimuli, at which the infant exhibited some kind of auditory behaviours, were noted down.

Visual reinforcement audiometry (VRA)

VRA was carried out in sound treated double room situation. The computer that was used to present the stimuli was housed in the tester room and the sound field speaker system was in the patient room. The infants were seated comfortably on the caregivers lap at a distance of 1 meter from the loudspeakers and at an azimuth of 45º. The examiner was seated in front of the child. Reinforcement was provided through the LCD TV monitor placed next to the loudspeakers which delivered the test stimuli. The infant's attention was drawn to the midline by the examiner. The response was based on operant conditioning techniques, and the infant's natural response to sound that is a head turn in the direction of the sound source was rewarded. Head turn towards the stimuli or the LCD TV monitor, was considered as a response only when it occurred within 3 seconds of the stimulus presentation. Thresholds were obtained for the warble tones at octave frequencies from 250 Hz to 8 kHz and also for speech stimuli.

Auditory Brainstem Response (ABR)

ABR was recorded using IHS Smart EP system, version 3.94. Initially, the electrode sites were cleaned with the help of skin preparing gel. Electrodes were placed on the recording sites along with conduction paste and were fixed with the help of surgical tape. It was ensured that, the absolute electrode impedance was less than 5 kΩ and inter electrode impedance was within 2 kΩ. The parameters used for recording ABR are shown in tables 3.2 and 3.3.

Table 3.2. *Stimulus parameters used to record ABR*

Stimulus parameters	Transducers	Insert earphones (ER-3A)
	Stimulus type	Clicks
	Number of sweeps	1500
	Intensity	30 dB nHL and 50 dB nHL
	Repitition rate	11.1/sec
	Stimulus polarity	Rarefaction

Table 3.3. *Acquisition parameters used to record ABR*

Acquisition parameters	Analysis time	15 msecs
	Filter settings	30-3000 Hz
	Notch filter	On
	Artifact rejection	30 µV
	Number of channels	Single
	Electrode placement	
	Inverting	Test ear mastoid
	Non-inverting	High forehead (Fz)
	Ground	Non-test ear

The latency of wave V was considered for threshold estimation. If the ABR wave V was clearly seen at 30dB nHL, the infants were considered to have normal hearing sensitivity.

Transient evoked otoacoustic emissions (TEOAEs)

TEOAEs were recorded using ILO- V6 instrument. Infants were tested during natural or sedated sleep or in quiet condition. The probe was inserted gently into the ear canal with an appropriate sized probe tip so as to give a flat stimulus spectrum across the frequency range. Probe fit was ensured for adequate fitting of the probe in the ear canal. An erroneous position of the probe can result in the absence of a response because of the presence of external noise.

Both click and tone burst-evoked OAEs were recorded. The evoked response for click stimulus included the frequency range from 500 Hz to 6 kHz (Zwicker & Schloth, 1984; Kemp et al., 1986), and the evoked response for toneburst was recorded individually at 500 Hz, 1 kHz, 2 kHz and 4 kHz. The stimulus was clear, with a positive and negative deflection followed by a flat line. The stimulus spectrum was smooth, with a rounded curve. A dip in the stimulus spectrum measured at the probe may be caused by standing waves in the ear canal. With some infants, if a slightly rounded peak was visible in the 2000-3000 Hz range, the probe was refitted until the best possible fit was obtained. The protocol used for recording click and tone burst evoked TEOAEs are shown in table 3.4.

Table 3.4: *Parameters used to record click and toneburst evoked TEOAEs.*

Stimulus parameters	Stimulus type	Click and Toneburst
	Stimulus intensity	80 dBpk SPL
	Stimulus rate	50 stimulus/sec
	No. of stimulus	260
	Stimulus polarity	Non-linear
Acquisition parameters	Amplification	100-10,000 times
	Filter settings	High pass filtered at 300 or 400 Hz

The stimuli for clicks are trains of 4 biphasic clicks of 80 µsecs in the non-linear position. The non-linear protocol includes 3 clicks of positive polarity followed by a 4th click of an inverse polarity with a relative magnitude of 9.5dB (3times) higher than the corresponding positive clicks (Kemp et al., 1986; Bray, 1989). The non-linear protocol removes the stimulus artifacts of linear nature which can be misinterpreted as TEOAE response. In order to ensure a significant artifact rejection, the first 2.5 ms of the recording was eliminated.

The preset stimulus was repeated 260 times and the "delayed" cochlear responses in the ear canal are acquired and accumulated in a memory bank in order to enhance the detection of the small cochlear signals against the background noise.

On termination of the test, the OAE response data that is obtained is averaged and an OAE waveform is displayed in the time domain. A Fast Fourier Transform (FFT) is performed on the OAE response spectrum and the OAE and noise energy are displayed in a frequency spectrum (Hall, 1986; Norcia, Sato, Shinn, & Mertus, 1986; Hall, 1992). The spectrum was displayed as a half-octave histogram, which is standard.

The responses were considered as emissions based on the signal-to-noise ratio (SNR) and reproducibility. The overall SNR of greater than or equal to +3 dB and the reproducibility of greater than 50% were considered (Dijk & Wit, 1987) for the presence of otoacoustic emissions to determine normal outer hair cell functioning.

Analysis

To arrive at the goal, the data collected was tabulated and analysed using appropriate statistical procedures using SPSS (Version 20). The values were obtained for various parameters (absolute amplitude and SNR) of CEOAEs and TBOAEs for the frequencies - 1 kHz, 2 kHz and 4 kHz across four age groups. The data obtained for various parameters (absolute amplitude and SNR) of CEOAEs and TBOAEs for 1 kHz, 2 kHz and 4 kHz across age groups were compared using appropriate statistical analysis.

Chapter 4

RESULTS AND DISCUSSION

The present study was conducted with the aim of investigating the influence of age on click evoked otoacoustic emissions (CEOAEs) and toneburst evoked otoacoustic emissions (TBOAEs). The comparison of spectral distribution of energy between CEOAEs and TBOAEs and the pattern of frequency shift with age was also investigated. To accomplish these aims, the absolute amplitude and the signal-to-noise ratio (SNR) of CEOAEs at the frequency bands of 1 kHz, 2 kHz, and 4 kHz, and of TBOAEs at the frequency 1 kHz, 2 kHz, and 4 kHz were measured. Comparison of absolute amplitude and SNR for different frequency bands of CEOAEs and TBOAEs, across age groups were also carried out. To analyse the data, Statistical Package for the Social Science (SPSS) version 20 software was used. The following statistical tools were used to analyse the obtained data:

- Descriptive statistics to calculate the mean and standard deviation (SD) for various parameters (absolute amplitude and SNR) of CEOAEs and TBOAEs for the frequencies - 1 kHz, 2 kHz and 4 kHz, across different age groups.
- Mixed analysis of variance (Mixed ANOVA) was done to study the main effect of age, stimulus and frequency on the absolute amplitude and SNR for CEOAEs and TBOAEs.
- Multivariate analysis of variance (MANOVA) was done to study the effect of age groups on each click frequencies (1 kHz, 2 kHz & 4 kHz) on amplitude and SNR.
- Duncan’s Post Hoc analysis was done to study between which two age groups, the amplitude and SNR differed significantly.

- Repeated measure ANOVA was done to study the effect of different frequencies on each age group.
- Bonferroni's Post Hoc analysis was done to study between which two frequencies, the amplitude and SNR differed significantly.
- Paired sample t-test was done to study the effects of age on CEOAEs and TBOAEs for the frequencies - 1 kHz, 2 kHz and 4 kHz. And also within age between CEOAEs and TBOAEs at each frequency.

The results obtained from the different statistical tools are discussed below for both CEOAEs and TBOAEs across different age groups.

4.1. Absolute amplitude and SNR for different frequencies of CEOAEs across age groups

The mean, SD and range values for absolute amplitude and SNR for click evoked OAEs at 1 kHz, 2 kHz and 4 kHz, across different age groups are depicted in the table 4.1 and the same is depicted in the figure 4.1.

Table 4.1. *Mean SD and Range for absolute amplitude and SNR for CE OAEs obtained at 1 kHz, 2 kHz and 4 kHz across age groups*

Age groups (in months)	**Frequencies**																	
	1 kHz						**2 kHz**						**4 kHz**					
	Absolute amplitude (dB SPL)			**SNR (dB)**			**Absolute amplitude (dB SPL)**			**SNR (dB)**			**Absolute amplitude (dB SPL)**			**SNR (dB)**		
	Mean N = 20	**SD**	**range**	**Mean N = 20**	**SD**	**range**	**Mean N = 20**	**SD**	**range**	**Mean N = 20**	**SD**	**range**	**Mean N = 20**	**SD**	**Range**	**Mean N = 20**	**SD**	**Range**
0 – 4	6.04	3.25	4.51-7.56	5.39	2.37	4.28-6.50	11.00	2.97	9.61-12.39	10.07	4.45	7.99-12.15	15.83	5.25	13.37-18.29	15.20	6.77	12.03-18.37
4 – 6	7.45	1.07	6.95-7.95	8.18	2.80	6.86-9.49	10.87	2.42	9.74-12.00	8.81	2.90	7.45-10.16	12.15	2.14	11.15-13.15	11.96	2.00	11.02-12.90
6 – 8	9.18	4.14	7.24-11.12	9.43	5.01	7.08-11.78	14.18	4.15	12.23-16.12	13.19	6.80	10.00-16.37	15.93	5.87	13.17-18.68	15.39	7.43	11.90-18.87
8 - 12	7.56	1.02	7.08-8.03	11.48	1.59	10.73-12.22	8.40	0.95	7.90-8.84	13.70	1.48	13.01-14.39	8.90	0.92	8.47-9.33	14.57	1.63	13.81-15.33

N= Number of ears

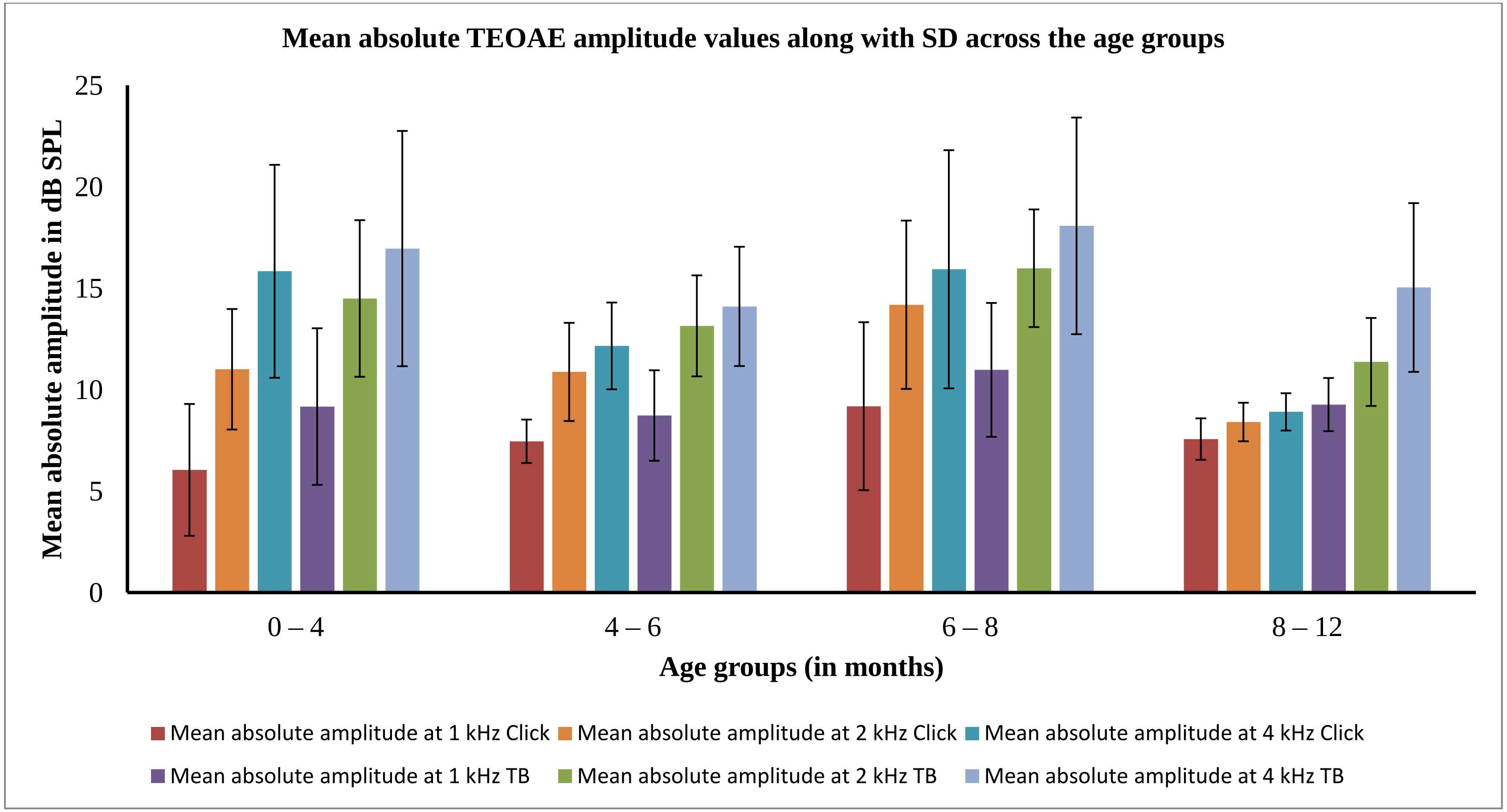

Figure 4.1. Mean, SD and range values for absolute amplitude at 1 kHz, 2 kHz, and 4 kHz for click and tone burst stimuli across different age group.

- It can be inferred from table 4.1 and figure 4.1 that, at 1 kHz, the absolute amplitude and SNR increases with increase in age. But such a trend was not observed for 2 kHz and 4 kHz. The absolute amplitude and SNR was more at 2 kHz and 4 kHz compared to 1 kHz across all the age groups. However, the mean absolute amplitude in the eldest age group was least for all the click frequency bands.
- A possible explanation for higher absolute amplitude at 2 kHz and 4 kHz is that the resonance frequency of the ear canal is of high frequency. Thus at higher frequencies the amplitude is more. As the age increases, the resonant frequency of the ear canal shifts towards the mid frequencies. This would have resulted in an increase in the amplitude at 1 kHz with increase in the age.
- The reduction in amplitude at other frequencies with increasing age could be attributed to the fact that, with the increase in age, the ear canal volume increases and because the SPL measured inside the cavity is inversely proportional to the volume, this could have led to the decrease in amplitude with increase in age.

Similar findings were stated by Smaurzynski (1994).It was found that the click evoked TEOAE levels increases with the post conceptional age. The TEOAE levels were found to decrease with increasing age (Engdahl, Arnesen & Mair, 1994; Glattke, Pafitis, Cummiskey & Herer, 1995; Norton et al., 1990; Nozza & Sabo, 1992; Prieve, et al., 1997; Spektor, Leonard, Kim, Jung & Smurzynski, 1991), which is also evident from the present study, where, least TEOAE amplitudes were obtained for 8-12 months age group at all the frequencies.

Further, Zanten et al. (1995) observed that TEOAE amplitude for click stimuli reached maximum values for infants at a post conceptional age of 47 weeks. However, in the present study, there was a reduction in amplitude found at 8-12 months age group but the maximum value reached by 6-8 months.

Kapoor and Panda (2006) analyzed SNR values for TEOAEs in neonates (0-1 month) and infants (1-12 months). The results showed that the neonates had the lowest SNR ranging between 3.47 to 9.62 dB whereas the infants showed the highest SNR values ranging between 6.13 to 13.11 dB. These findings support the present study where similar results were found.

The results of the present study is in accordance with the study done by Prieve, Hancur-Bucci and Peterson (2009), who screened neonates in the age from birth to one month. They measured overall TEOAE levels and levels in half-octave frequency bands centred at 1.5, 2, 3 and 4 kHz. They found that TEOAE levels in infants significantly increased from birth to 1 month of age across all frequencies tested. It was also found that the increase in TEOAE levels was frequency dependant. The amplitude was highest at 4 kHz.

Shi, et al. (2010) recorded TEOAEs from neonates and analyzed the SNR values. Results showed that, lowest SNRs were obtained at low frequency (0.8 kHz) for neonates and at higher frequency (4 kHz) for younger adults. These findings were similar to that obtained from the present study, where, lowest SNR values were obtained at the low frequency (1 kHz) across all age groups.

Similarly, with respect to the SNR values, the results of the present study are in consonance with Prieve (2007). In this study, the SNR of TEOAEs in babies, at the frequencies- 1 kHz, 1.5 kHz, 2 kHz, 3 kHz, and 4 kHz were measured and it was found that as the stimulus frequency increased, the mean SNR values also increased.

4.2. Absolute amplitude and SNR for different frequencies of TBOAEs across age groups

The mean, SD and range values for absolute amplitude and SNR for toneburst stimuli, at 1 kHz, 2 kHz and 4 kHz, across different age groups are depicted in the table 4.2 and also in the figure 4.2.

Table 4.2. *Mean SD and range for amplitude and SNR for TB frequencies obtained at 1 kHz, 2 kHz and 4 kHz across age groups*

Age groups (in mths)	Frequencies																	
	1 kHz						2 kHz						4 kHz					
	Absolute amplitude (dB SPL)			SNR (dB)			Absolute amplitude (dB SPL)			SNR (dB)			Absolute amplitude (dB SPL)			SNR (dB)		
	Mean N = 20	SD	range	Mean N = 20	SD	range	Mean N = 20	SD	range	Mean N = 20	SD	range	Mean N = 20	SD	range	Mean N = 20	SD	Range
0 – 4	9.16	3.86	7.35-10.96	10.58	5.55	7.97-13.18	14.49	3.86	12.68-16.29	19.17	4.97	16.84-21.50	16.95	5.80	14.23-19.66	15.91	5.96	13.12-18.70
4 – 6	8.72	2.23	7.67-9.76	12.90	4.59	10.75-15.04	13.14	2.49	11.97-14.30	13.26	3.33	11.70-14.82	14.10	2.94	12.72-15.47	13.86	3.15	12.38-15.33
6 – 8	10.97	3.30	9.42-12.52	14.70	6.09	11.85-17.55	15.98	2.90	14.62-17.34	18.75	4.86	16.47-21.02	18.07	5.34	15.57-20.57	21.56	3.33	19.99-23.12
8 - 12	9.26	1.31	8.64-9.87	10.23	1.43	9.55-10.90	11.36	2.17	10.34-12.37	13.22	2.91	11.85-14.58	15.03	4.16	13.08-16.97	17.64	3.98	15.78-19.50

N = Number of ears

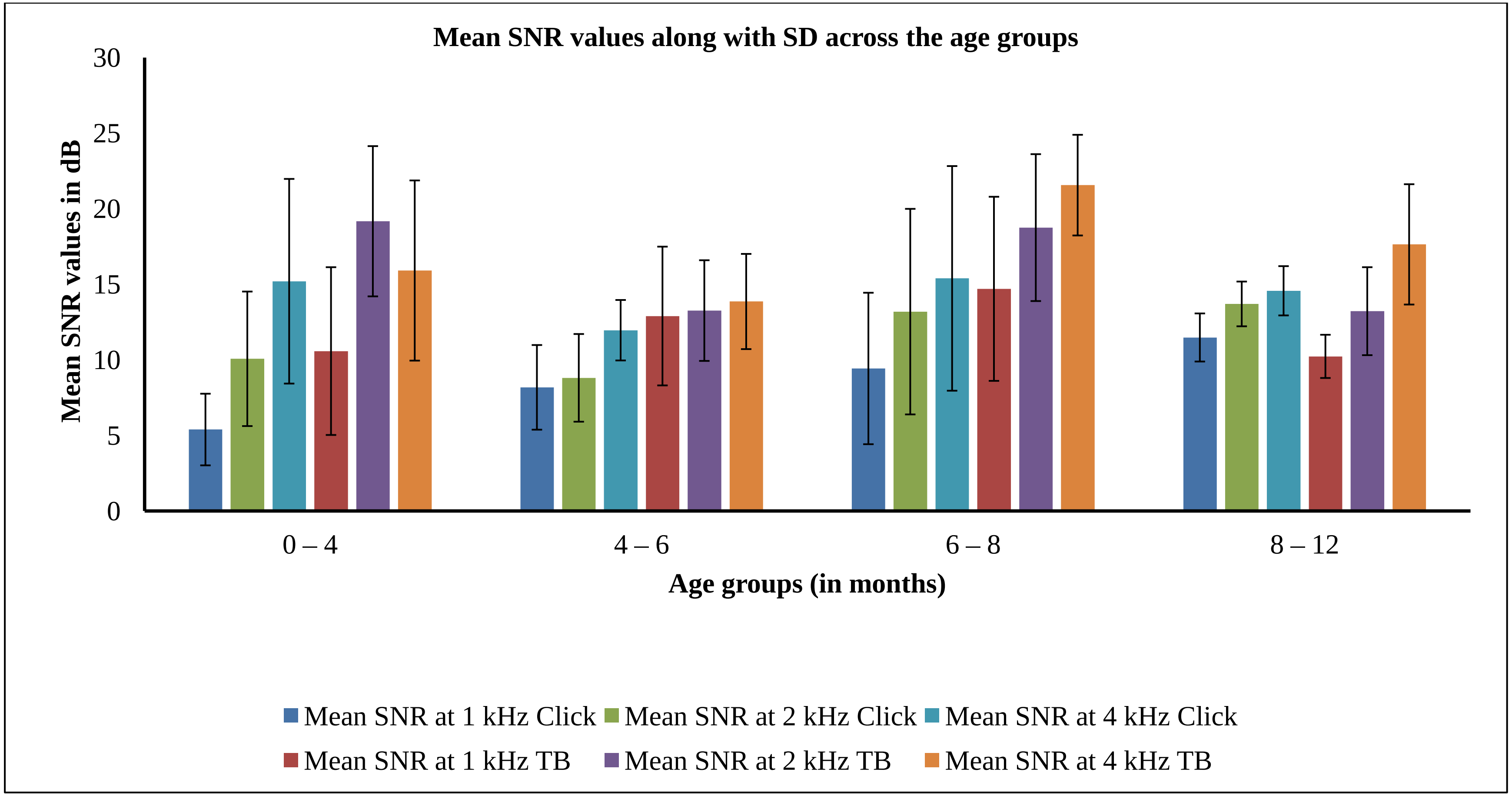

Figure 4.2: Mean, SD and range values for SNR at 1 kHz, 2 kHz, and 4 kHz for click and tone burst stimuli across different age groups

As it is evident from table 4.2 as well as from the figure 4.2, there was no definite trend followed for absolute amplitude across the age groups for different toneburst frequencies. However, the absolute amplitude was found to be higher at 2 kHz and 4 kHz compared to l kHz toneburst frequency. When SNR values for different toneburst stimulus frequencies were considered, similar findings were obtained. Very little has been studied in literature about the effect of age in TBOAEs.

Zhang et al, (2008) investigated the characteristics (amplitude and SNR) of the 1 kHz TBOAE response in neonates. The results showed that, the mean TBOAE response at 1 kHz was significantly higher than that of 2 kHz ($P < 0.05$), whereas, the SNR at 1 kHz was significantly lower than that at 1.5 kHz and 2 kHz frequency bands. The main reason for these findings relates to the greater noise in the 1 kHz frequency region, which may mask out the weaker TBOAE response. And also, a default low frequency filter setting in the ILO system may also reduce the noise levels at lower frequencies, and also reduces the OAE response (Prieve et al, 1996; Hussain, Gorga, Neely, Keefe, & Peters, 1998). The mean TBOAE responses obtained in the present study did not correlate with the results obtained from Zhang et al, (2008) study. Whereas, in the present study, the SNR values at 1 kHz were in accordance with Zhang et al, (2008) study.

4.3 Comparison of absolute amplitude and SNR across age groups for different frequencies of CEOAEs and TBOAEs

Mixed ANOVA was carried out to see the significant interaction across age groups, frequencies and stimuli for amplitude and SNR measures separately. The results of mixed ANOVA that were obtained for amplitude revealed the following:

- A high statistically significant difference was found between the age groups [$F(3, 76) = 13.24$, $p < 0.05$].
- Also a high statistically significant difference was found between different frequencies [$F(2, 152) = 88.89$, $p < 0.05$].
- A high statistically significant difference was found between the two different stimuli [$F(1, 76) = 105.7$, $p < 0.05$] for all the four age groups.
- A high statistically significant difference was found between frequencies and age groups [$F(6, 152) = 9.09$, $p < 0.05$].
- A high statistically significant difference was found between age groups and stimuli [$F(3, 76) = 1.53$, $p < 0.05$].
- A high statistically significant difference was found between stimuli and frequencies [$F(2, 152) = 8.37$, $p < 0.05$].
- Also a statistically different significance was also found between the stimuli, frequencies and age groups [$F(6, 152) = 3.52$, $p < 0.05$].

The results that were obtained from mixed ANOVA for the SNR are as follows:

- A high statistically significant difference was found between the age groups [$F(3, 76) = 5.70$, $p < 0.05$].
- Also a high statistically significant difference was found between different frequencies [$F(2, 152) = 55.04$, $p < 0.05$].
- A high statistically significant difference was found between the two different stimuli [$F(1, 76) = 95.8$, $p < 0.05$] for all the four age groups.
- A high statistically significant difference was found between frequencies and age groups [$F(6, 152) = 4.44$, $p < 0.05$].

- A high statistically significant difference was found between age groups and stimuli [$F (3, 76) = 4.35, p < 0.05$].
- A high statistically significant difference was found between stimuli and frequencies [$F (2, 152) = 10.04, p < 0.05$].
- Also a statistically different significance was also found between the stimuli, frequencies and age groups [$F (6, 152) = 8.41, p < 0.05$].

4.3.1 Effect of age on amplitude at each click frequency

As mixed ANOVA showed significant interaction across click frequencies on amplitude measures, to see the effect of age on amplitude, multivariate analysis of variance (MANOVA) was done at each click frequencies. The results showed the following.

- There was significant difference in amplitude [$F (3, 76) = 4.40, p < 0.05$] across age groups for 1000 Hz click frequency
- There was significant difference in amplitude [$F (3, 76) = 13.61, p < 0.05$] across age groups for 2000 Hz click frequency
- There was significant difference in amplitude [$F (3,76) = 13.39, p <0.05$] across age groups for 4000 Hz click frequency

As MANOVA showed significant difference between age groups for 1000 Hz, 2000 Hz and 4000 Hz click frequency on amplitude, further analysis using the Duncan post hoc analysis test was done to see between which two age groups, amplitude differ significantly. The results of Duncan post hoc test for each click frequency are shown in table 4.2

Table 4.3: *Results of Duncan post hoc analysis comparing absolute amplitude at different age groups for different click frequencies (1 kHz, 2 kHz & 4 kHz)*

Click frequencies	Age groups (In months)	0-4 months (group I)	4-6 months (group II)	6-8 months (group III)	8-12 months (group IV)
1 kHz	0-4		> 0.05	< 0.05*	> 0.05
	4-6	> 0.05		> 0.05	> 0.05
	6-8	< 0.05*	>0.05		> 0.05
	8-12	> 0.05	> 0.05	> 0.05	
2 kHz	0-4		> 0.05	< 0.05*	< 0.05*
	4-6	> 0.05		< 0.05*	< 0.05*
	6-8	< 0.05*	< 0.05*		< 0.05*
	8-12	< 0.05*	< 0.05*	< 0.05*	
4 kHz	0-4		< 0.05*	>0.05	< 0.05*
	4-6	< 0.05*		< 0.05*	> 0.05
	6-8	> 0.05	< 0.05*		< 0.05*
	8-12	< 0.05*	>0.05	< 0.05*	

Note. * Indicates statistically significant difference

- As depicted from the table 4.3, the results of the present study showed that, when 1000 Hz click was used, there was age effect seen with significant difference between group I and group III.
- When 2 kHz click was used, there was age effect where group I significantly differed from group III and group IV. Similarly group III significantly differed from all the age groups however group II significantly differed from all the age groups except from group I.

- At 4 kHz click frequency, group I was statistically significantly different from all the other age groups except group III. Similarly group II showed significant difference from all the age groups except from group IV.

According to Norton et al., (2000), there was no much difference in the TEOAE levels for infants born between 28 and 40 weeks of GA and those between birth and 28 days after birth. A significant group effect was observed at 2 kHz. Post hoc multiple comparisons were carried out, and the results showed that there was no consistent significant difference observed when each group was compared with the other two groups. This study supports the findings of the present study where similar results were obtained.

4.3.2 Effect of age on SNR at each click frequency

As mixed ANOVA showed significant interaction across click frequencies on SNR measures, to see the effect of age on SNR, multivariate analysis of variance (MANOVA) was done at each click frequencies. The results showed the following.

- There was significant difference in SNR [$F(3, 76) = 12.56$, $p < 0.05$] across age groups for 1000 Hz click frequency
- There was significant difference in SNR [$F(3, 76) = 5.89$, $p < 0.05$] across age groups for 2000 Hz click frequency
- There was no significant difference in SNR [$F(3,76) = 1.86$, $p > 0.05$] across age groups for 4000 Hz click frequency

As MANOVA showed significant difference between age groups for 1000 Hz, 2000 Hz and 4000 Hz click frequency on SNR, further analysis using the Duncan post hoc analysis test was done to see between which two age groups, SNR differ significantly. The results of Duncan post hoc test for each click frequency are shown in table 4.4.

Table 4.4: Results of *Duncan's Post Hoc analysis comparing SNR at different age groups for different click frequencies (1 kHz, 2 kHz & 4 kHz).*

Click frequencies	**Age groups (In months)**	**0-4 months**	**4-6 months**	**6-8 months**	**8-12 months**
1 kHz	**0-4**		**< 0.05***	**< 0.05***	**< 0.05***
	4-6	**< 0.05***		> 0.05	**< 0.05***
	6-8	**< 0.05***	> 0.05		> 0.05
	8-12	**< 0.05***	**< 0.05***	> 0.05	
2 kHz	**0-4**		> 0.05	> 0.05	> 0.05
	4-6	> 0.05		**< 0.05***	**< 0.05***
	6-8	> 0.05	**< 0.05***		> 0.05
	8-12	> 0.05	**< 0.05***	> 0.05	
4 kHz	**0-4**		> 0.05	> 0.05	> 0.05
	4-6	> 0.05		> 0.05	> 0.05
	6-8	> 0.05	> 0.05		> 0.05
	8-12	> 0.05	> 0.05	> 0.05	

Note. * Indicates statistically significant difference

- For the SNR values at 1 kHz click frequency, the group I was found to be significantly different from the other age groups, as it is depicted from the table 4.4. And the group II was significantly different from group I and IV. Whereas, the group III was significantly different from only group I.

- For the SNR values at 2 kHz, group I showed no statistically significant difference between the age groups. Group II was statistically significant from group III and IV.
- The SNR values at 4 kHz click frequency did not show any significant difference across the age groups.

4.3.3 Effect of age on amplitude at each tone burst frequency

To see the effect of age on amplitude, multivariate analysis of variance (MANOVA) was done at each tone burst frequencies. The results showed the following:

- There was no significant difference in amplitude [$F(3, 76) = 2.41$, $p > 0.05$] across age groups for 1000 Hz tone burst frequency
- There was significant difference in amplitude [$F(3, 76) = 9.03$, $p < 0.05$] across age groups for 2000 Hz tone burst frequency
- There was significant difference in amplitude [$F(3,76) = 2.94$, $p > 0.05$] across age groups for 4000 Hz tone burst frequency

As MANOVA showed significant difference between age groups for 1000 Hz, 2000 Hz and 4000 Hz tone burst frequency on amplitude, further analysis using the Duncan post hoc analysis test was done to see between which two age groups, amplitude differ significantly. The results of Duncan post hoc test for each tone burst frequency are shown in table 4.5.

Table 4.5: *Results of Duncan post hoc analysis comparing absolute amplitude at different age groups for different toneburst frequencies (1 kHz, 2 kHz & 4 kHz)*

Toneburst frequencies	**Age groups (In months)**	**0-4 months**	**4-6 months**	**6-8 months**	**8-12 months**
1 kHz	**0-4**		>0.05	>0.05	>0.05
	4-6	>0.05		>0.05	>0.05
	6-8	>0.05	>0.05		>0.05
	8-12	>0.05	>0.05	>0.05	
2 kHz	**0-4**		> 0.05	**< 0.05***	> 0.05
	4-6	> 0.05		**< 0.05***	> 0.05
	6-8	**< 0.05***	**< 0.05***		**< 0.05***
	8-12	**< 0.05***	> 0.05	**< 0.05***	
4 kHz	**0-4**		> 0.05	> 0.05	> 0.05
	4-6	> 0.05		**< 0.05***	> 0.05
	6-8	> 0.05	**< 0.05***		> 0.05
	8-12	> 0.05	> 0.05	> 0.05	

Note. * Indicates statistically significant difference

- It is apparent that, not at all instances, the difference is statistically significant but in few instances. As inferred from the table 4.5, the absolute amplitude values for 1 kHz toneburst frequency showed statistically no significant difference across the different age groups.
- When 2 kHz toneburst frequency was considered, the absolute amplitude values at 0-4 months and 4-6 months age groups were significantly different from 6-8 months groups only. Whereas 6-8 months group was found to be statistically

significant from all the other age groups, and also the 8-12 months group was significantly different from the other groups except from the 4-6 months age group.

- For 4 kHz frequency, the 4-6 months age group was significantly different from 6-8 months group and the 6-8 months age group was significantly different from 4-6 months group. But for the other age groups (0-4 months & 8-12 months), statistically no significant difference was observed across age groups.

4.3.4 Effect of age on SNR at each tone burst frequency

To see the effect of age on amplitude, multivariate analysis of variance (MANOVA) was done at each tone burst frequencies. The results showed the following:

- There was significant difference in SNR [$F (3, 76) = 3.86$, $p < 0.05$] across age groups for 1000 Hz tone burst frequency
- There was significant difference in SNR [$F (3, 76) = 12.86$, $p < 0.05$] across age groups for 2000 Hz tone burst frequency
- There was significant difference in SNR [$F (3,76) = 11.75$, $p < 0.05$] across age groups for 4000 Hz tone burst frequency

As MANOVA showed significant difference between age groups for 1000 Hz, 2000 Hz and 4000 Hz tone burst frequency on SNR, further analysis using the Duncan post hoc analysis test was done to see between which two age groups, SNR differ significantly. The results of Duncan post hoc test for each tone burst frequency are shown in table 4.6.

Table 4.6: *Results of Duncan post hoc analysis comparing SNR at different age groups for different toneburst frequencies (1 kHz, 2 kHz & 4 kHz).*

Toneburst frequencies	**Age groups (In months)**	**0-4 months**	**4-6 months**	**6-8 months**	**8-12 months**
1 kHz	**0-4**		> 0.05	**< 0.05***	> 0.05
	4-6	> 0.05		>0.05	> 0.05
	6-8	**< 0.05***	> 0.05		**< 0.05***
	8-12	> 0.05	> 0.05	**< 0.05***	
2 kHz	**0-4**		**< 0.05***	> 0.05	**< 0.05***
	4-6	**< 0.05***		**< 0.05***	> 0.05
	6-8	> 0.05	**< 0.05***		**< 0.05***
	8-12	**< 0.05***	> 0.05	**< 0.05***	
4 kHz	**0-4**		> 0.05	**< 0.05***	> 0.05
	4-6	> 0.05		**< 0.05***	**< 0.05***
	6-8	**< 0.05***	**< 0.05***		**< 0.05***
	8-12	> 0.05	**< 0.05***	**< 0.05***	

Note. * Indicates statistically significant difference

- As inferred from the table 4.6, the SNR values for the group III differed significantly from all the age groups except group II. Whereas, group II group showed no significant difference with the other age groups.
- When 2 kHz toneburst frequency was considered, the SNR values for group I was statistically significant from group II and group IV. And the group II showed statistically significant difference from all the other age groups except from group IV. Similarly, group III was statistically significant from all the other age groups

except from group I. The 8-12 months age group was statistically significant from all the other age groups except from 4-6 months group.

- For 4 kHz frequency, the SNR values for group III were found to be significantly different from all the age groups. The group II and IV showed statistically significant difference from all the other age groups except for Group I.

Zhang et al, (2008) in his study found that, the age of the neonate (mean test age: 2.54 days) had no effect on the mean response and SNR at each frequency band (1 kHz, 1.5 kHz and 2 kHz). The findings were in support with the present study, but not for all the frequencies.

4.3.5 Effect of click frequency on amplitude at each age group

As mixed ANOVA showed significant interaction across frequencies, to see the effect of age on amplitude, repeated measure analysis of variance (ANOVA) was done at each age group. The results showed the following:

- There was significant difference in amplitude [$F(2, 38) = 29.14$, $p < 0.05$] across click frequencies in group I.
- There was significant difference in amplitude [$F(2, 38) = 46.49$, $p < 0.05$] across click frequencies in group II.
- There was significant difference in amplitude [$F(2, 38) = 24.29$, $p < 0.05$] across click frequencies in group III.
- There was significant difference in amplitude [$F(2, 38) = 37.23$, $p < 0.05$] across click frequencies in group IV.

As repeated measure ANOVA showed significant difference between click frequencies across age groups on amplitude, further analysis using the Bonferroni's multiple pair wise comparison was done to see between which two click frequencies, amplitude differ significantly. The results of Bonferroni's multiple pair wise comparison test for each age group are shown in table 4.7.

Table. 4.7: *Results of Bonferroni's Post Hoc analysis comparing absolute amplitude at different click frequencies (1 kHz, 2 kHz & 4 kHz)*

Age groups (in months)	**Click frequencies (Hz)**	**1 kHz**	**2 kHz**	**4 kHz**
0 – 4	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		**< 0.05****
	4 kHz	**< 0.05****	**< 0.05****	
4 – 6	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		>0.05
	4 kHz	**< 0.05****	>0.05	
6 – 8	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		>0.05
	4 kHz	**< 0.05****	>0.05	
8 – 12	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		**< 0.05****
	4 kHz	**< 0.05****	**< 0.05****	

Note: **Indicates high statistically significant difference

- As depicted from the table 4.7, the results of the present study showed that, for 0-4 months and 8-12 months age group, the absolute amplitude for the click stimulus of 1 kHz, 2 kHz and 4 kHz was significantly different from the other frequencies.

- Whereas, 4-6 months age group showed statistically significant difference for 1 kHz and 2 kHz, but 4 kHz was significantly different only from 1 kHz, and for 6-8 months group, only 1 kHz was significantly different from the other frequencies.

4.3.6 Effect of click frequency on SNR at each age group

To see the effect of age on amplitude, repeated measure analysis of variance (ANOVA) was done at each age group. The results showed the following:

- There was significant difference in SNR [F (2, 38) = 26.61, $p < 0.05$] across click frequencies in group I.
- There was significant difference in SNR [F (2, 38) = 22.10, $p < 0.05$] across click frequencies in group II.
- There was significant difference in SNR [F (2, 38) = 8.17, $p < 0.05$] across click frequencies in group III.
- There was significant difference in SNR [F (2, 38) = 20.02 , $p < 0.05$] across click frequencies in group IV.

As repeated measure ANOVA showed significant difference between click frequencies across age groups on SNR, further analysis using the Bonferroni's multiple pair wise comparison was done to see between which two click frequencies, SNR differ significantly. The results of Bonferroni's multiple pair wise comparison test for each age group are shown in table 4.8.

Table. 4.8: *Results of Bonferroni's Post Hoc analysis comparing SNR at different click frequencies (1 kHz, 2 kHz & 4 kHz)*

Age groups (in months)	Click frequencies (Hz)	1 kHz	2 kHz	4 kHz
0 – 4	1 kHz		< 0.05**	< 0.05**
	2 kHz	< 0.05**		< 0.05*
	4 kHz	< 0.05**	< 0.05*	
4– 6	1 kHz		> 0.05	< 0.05**
	2 kHz	>0.05		< 0.05**
	4 kHz	< 0.05**	< 0.05**	
6 – 8	1 kHz		< 0.05*	< 0.05**
	2 kHz	< 0.05*		>0.05
	4 kHz	< 0.05**	>0.05	
8 – 12	1 kHz		< 0.05**	< 0.05**
	2 kHz	< 0.05**		>0.05
	4 kHz	< 0.05**	>0.05	

Note.*Indicates statistically significant difference

** Indicates high statistically significant difference

- The SNR values for 0-4 months age group was significantly different across all the frequencies as it is inferred from the table 4.8.
- For 4-6 months age group, the SNR values at 1 kHz and 2 kHz were significantly different only from the 4 kHz click frequency.

- For 6-8 months and 8-12 months age group, the SNR values at 1 kHz were found to be statistically significantly from the other frequencies. Whereas, 2 kHz and 4 kHz was significantly different only from 1 kHz click frequency.

4.3.7 Effect of tone burst frequency on amplitude at each age group

To see the effect of age on amplitude, repeated measure analysis of variance (ANOVA) was done at each age group. The results showed the following:

- There was significant difference in amplitude [$F(2, 38) = 21.15$, $p < 0.05$] across tone burst frequencies in group I.
- There was significant difference in amplitude [$F(2, 38) = 64.73$, $p < 0.05$] across tone burst frequencies in group II.
- There was significant difference in amplitude [$F(2, 38) = 24.61$, $p < 0.05$] across tone burst frequencies in group III.
- There was significant difference in amplitude [$F(2, 38) = 52.04$, $p < 0.05$] across tone burst frequencies in group IV.

As repeated measure ANOVA showed significant difference between tone burst frequencies across age groups on amplitude, further analysis using the Bonferroni's multiple pair wise comparison was done to see between which two tone burst frequencies, amplitude differ significantly. The results of Bonferroni's multiple pair wise comparison test for each age group are shown in table 4.9.

Table. 4.9: *Results of Bonferroni's Post Hoc analysis comparing absolute amplitude at different toneburst frequencies (1 kHz, 2 kHz & 4 kHz)*

Age groups (in months)	Toneburst frequencies (Hz)	1 kHz	2 kHz	4 kHz
0 – 4	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		>0.05
	4 kHz	**< 0.05****	>0.05	
4 - 6	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		**< 0.05****
	4 kHz	**< 0.05****	>0.05	
6 – 8	**1 kHz**		**< 0.05***	**< 0.05****
	2 kHz	**< 0.05***		>0.05
	4 kHz	**< 0.05****	>0.05	
8 – 12	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		**< 0.05****
	4 kHz	**< 0.05****	**< 0.05****	

Note.*Indicates statistically significant difference

** Indicates high statistically significant difference

- As interpreted from the table 4.9, the absolute amplitude for the 0-4 months age group at 2 kHz toneburst frequency showed that there was no statistically significant difference with the 4 kHz frequency and vice versa. There was a statistically significant difference between 2 kHz and 1 kHz, and 1 kHz and 4 kHz.

- In the 4-6 months age group, the absolute amplitude at all the tone burst frequencies was statistically significant, except for the 4 kHz which did not have a significant difference with 2 kHz.
- Similarly, the 4 kHz tone burst frequency showed statistically no significant difference with 2 kHz frequency and vice versa at 6-8 months age range. There was statistically significant difference between 2 kHz and 1 kHz, and 4 kHz and 1 kHz.
- Whereas, 8-12 months age group showed statistically significant difference across all the tone burst frequencies.

4.3.8 Effect of tone burst frequency on SNR at each age group

To see the effect of age on SNR, repeated measure analysis of variance (ANOVA) was done at each age group. The results showed the following:

- There was significant difference in SNR [$F(2, 38) = 24.34$, $p < 0.05$] across click frequencies in group I.
- There was no significant difference in SNR [$F(2, 38) = 1.39$, $p > 0.05$] across click frequencies in group II.
- There was significant difference in SNR [$F(2, 38) = 14.61$, $p < 0.05$] across click frequencies in group III.
- There was significant difference in SNR [$F(2, 38) = 58.33$, $p < 0.05$] across click frequencies in group IV.

As repeated measure ANOVA showed significant difference between tone burst frequencies across age groups on SNR, further analysis using the Bonferroni's multiple pair wise comparison was done to see between which two tone burst frequencies, SNR differ significantly. The results of Bonferroni's multiple pair wise comparison test for each age group are shown in table 4.10.

Table. 4.10: *Results of Bonferroni's Post Hoc analysis comparing SNR at different toneburst frequencies (1 kHz, 2 kHz & 4 kHz)*

Age groups (in months)	**Toneburst frequencies (Hz)**	**1 kHz**	**2 kHz**	**4 kHz**
0 – 4	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		**< 0.05***
	4 kHz	**< 0.05****	**< 0.05***	
4– 6	**1 kHz**		>0.05	>0.05
	2 kHz	>0.05		>0.05
	4 kHz	>0.05	>0.05	
6 – 8	**1 kHz**		**< 0.05****	**< 0.05***
	2 kHz	**< 0.05****		**< 0.05****
	4 kHz	**< 0.05***	**< 0.05****	
8 – 12	**1 kHz**		**< 0.05****	**< 0.05****
	2 kHz	**< 0.05****		**< 0.05****
	4 kHz	**< 0.05****	**< 0.05****	

Note.*Indicates statistically significant difference

** Indicates high statistically significant difference

As it can be inferred from the table 4.10 that, all the age groups, except 4-6 months age group, showed a statistically significant difference across all the toneburst frequencies tested.

4.4.1. Comparison of absolute amplitude across click and tone burst stimuli at each age group for each frequency

As mixed ANOVA showed significant interaction across stimuli, in order to compare the absolute amplitude of click frequencies (1 kHz, 2 kHz & 4 kHz) and tone burst frequencies (1 kHz, 2 kHz & 4 kHz) at each age group for each of the frequencies, paired sample t-test was done. The results are shown in the table 4.11.

Table 4.11. *Results of paired sampled t-test for absolute amplitude, comparing across the stimuli for each frequency at each age group*

Age groups (months)	Parameters	Df	t-value	Sig (2 – tailed)
0 – 4	Absolute amplitude for 1 kHz click - Absolute amplitude for 1 kHz TB	19	2.699	.014*
	Absolute amplitude for 2 kHz click - Absolute amplitude for 2 kHz TB	19	4.676	.000**
	Absolute amplitude for 4 kHz click - Absolute amplitude for 4 kHz TB	19	0.934	.362
4 – 6	Absolute amplitude for 1 kHz click - Absolute amplitude for 1 kHz TB	19	3.548	.002**
	Absolute amplitude for 2 kHz click - Absolute amplitude for 2 kHz TB	19	2.999	.007**
	Absolute amplitude for 4 kHz click - Absolute amplitude for 4 kHz TB	19	2.873	.010*
6 – 8	Absolute amplitude for 1 kHz click - Absolute amplitude for 1 kHz TB	19	2.430	.025*
	Absolute amplitude for 2 kHz click	19	1.694	.107

	- Absolute amplitude for 2 kHz TB			
	Absolute amplitude for 4 kHz click - Absolute amplitude for 4 kHz TB	19	1.499	.150
8 – 12	Absolute amplitude for 1 kHz click - Absolute amplitude for 1 kHz TB	19	4.468	.000**
	Absolute amplitude for 2 kHz click - Absolute amplitude for 2 kHz TB	19	6.415	.000**
	Absolute amplitude for 4 kHz click - Absolute amplitude for 4 kHz TB	19	7.040	.000**

**Indicates significant difference*

***Indicates high significant difference*

- As it can be inferred from the table 4.11, for 0-4 months age group, the comparison of the absolute amplitude at 1 kHz and 2 kHz click and toneburst stimuli was found to be statistically significant. Whereas, for the comparison of 4 kHz click and toneburst stimulus showed statistically no significant difference.
- For 6-8 months age group, statistically significant difference was observed only for the 1 kHz click and toneburst comparison. Statistically no significant difference was obtained for 2 kHz and 4 kHz frequency.
- Whereas, for 4-6 and 8-12 months age group, all the frequencies showed a statistically significant difference.

4.4.2. Comparison of SNR across click and toneburst stimuli at each age group for each frequency

To compare the SNR of click frequencies (1 kHz, 2 kHz & 4 kHz) and toneburst frequencies (1 kHz, 2 kHz & 4 kHz) at each age group for each of the frequencies, paired sample t-test was done. The results are shown in the table 4.12.

Table 4.12. *Results of paired sampled t-test for SNR, comparing across the stimuli for each frequency at each age group.*

Age groups (months)	**Parameters**	**df**	**t-value**	**Sig (2 – tailed)**
0 – 4	SNR for 1 kHz click - SNR for 1 kHz TB	19	3.960	.001**
	SNR for 2 kHz click - SNR for 2 kHz TB	19	5.954	.000**
	SNR for 4 kHz click - SNR for 4 kHz TB	19	0.342	.736
4 – 6	SNR for 1 kHz click - SNR for 1 kHz TB	19	3.949	.001**
	SNR for 2 kHz click - SNR for 2 kHz TB	19	5.021	.000**
	SNR for 4 kHz click - SNR for 4 kHz TB	19	1.987	.062
6 – 8	SNR for 1 kHz click - SNR for 1 kHz TB	19	3.143	.005**
	SNR for 2 kHz click - SNR for 2 kHz TB	19	4.878	.000**
	SNR for 4 kHz click - SNR for 4 kHz TB	19	3.202	.005**
8 – 12	SNR for 1 kHz click - SNR for 1 kHz TB	19	2.403	.027*
	SNR for 2 kHz click - SNR for 2 kHz TB	19	0.612	.547
	SNR for 4 kHz click - SNR for 4 kHz TB	19	3.692	.002**

**Indicates significant difference ($p < 0.05$)*

***Indicates high significant difference ($p < 0.05$)*

- For 0-4 months and 4-6 months age group, the comparison of SNR values between click and toneburst stimuli at 1 kHz and 2 kHz were statistically significantly different ($p < 0.05$), except for 4 kHz.
- For 6-8 months age group, the comparison of SNR values between click and toneburst stimuli was found to be statistically significantly different ($p < 0.05$) at all the frequencies.
- Whereas, for 8-12 months age group, except at 2 kHz frequency, all the other frequencies showed a statistically significant difference ($p < 0.05$).

Chapter 5

SUMMARY AND CONCLUSION

Otoacoustic emissions (OAEs) are sounds that originate in the cochlea and propagate through the middle ear and into the ear canal, where they can be measured using a sensitive microphone. These emissions are generated either spontaneously or in response to acoustic stimulation. One among the evoked OAEs is the transient evoked otoacoustic emissions (TEOAEs) which are frequency dispersive responses following a brief acoustic stimulus such as a click or a tone burst. The evoked response from a click covers the frequency range up to around 4 kHz, while a toneburst elicits a response from the region that has the same frequency as the pure tone. TEOAEs exhibits compressive non-linearity. Click evoked otoacoustic emissions (CEOAEs) have a broad spectrum, and hence consequently it stimulates broad frequency region of the cochlea in a single measurement. Tone burst evoked otoacoustic emissions (TBOAEs) includes narrow bandwidth tone stimuli, which has stimulus energy concentrated on a particular area of the basilar membrane and elicits a more frequency-specific cochlear response. Although tone burst stimuli has greater frequency specificity compared to click stimuli, TBOAEs have not been routinely used in pediatric populations.

Further studies of TBOAE time-frequency analysis and measurements in newborns are needed. Very few studies have been carried out using TBOAEs for hearing assessment in neonates and young children. The TBOAE responses are more prominent than CEOAEs. Various studies have been carried out in infants and adults on CEOAEs and TBOAEs. As to the research and clinical application of TBOAEs, studies have been mainly focused on adult population. Whereas, there are very limited studies on infants. Hence there is a need to study the utility and efficacy of TBOAEs among infants. Also, there are very limited studies on the spectral information of TEOAEs as the age advances. The information regarding the comparison between CEOAEs & TBOAEs in infants is very limited. Hence the present study was undertaken.

The aims of the present study were:

3) To investigate the influence of age on click and toneburst evoked OAEs, and to investigate the pattern of frequency shifts with age.
4) To study the sensitivity of click & toneburst-evoked OAEs and to monitor the frequency specific maturational changes in the cochlea.

To accomplish these aims, TEOAE amplitude and SNR were recorded on a total of 40 infants (80 ears) in the age group of 0 – 12 months using click stimuli at the frequencies- l kHz, 2 kHz and 4 kHz, and toneburst stimuli at the frequencies- l kHz, 2 kHz and 4 kHz.

The data collected was tabulated and analysed using appropriate statistical procedures using SPSS (Version 20). The following statistical tools were used to analyse the obtained data:

- Descriptive statistics was done to calculate the mean and standard deviation for absolute amplitude and SNR of CEOAEs and TBOAEs for the frequencies - 1 kHz, 2 kHz and 4 kHz across different age groups.
- Mixed ANOVA was carried out to study the main effects of age, stimulus and frequency on CEOAEs and TBOAEs absolute amplitude and SNR.

- Multivariant analysis of variance (MANOVA) was done to study the effect of age groups on each click frequencies (1 kHz, 2 kHz & 4 kHz) on amplitude and SNR.
- Duncan's Post Hoc analysis was done to study between which two age groups, the amplitude and SNR differed significantly.
- Repeated measure ANOVA was done to study the effect of different frequencies on each age group.
- Bonferroni's Post Hoc analysis was carried out to study the age effects by performing pair wise comparison independently.
- Paired sample t-test was carried out to study the effects of age on CEOAEs and TBOAEs for the frequencies - 1 kHz, 2 kHz and 4 kHz.

The results of the present studies can be summarized as follows:

5.1 Absolute amplitude and SNR for different frequencies of CEOAEs across age groups shows

- An increase in the mean absolute amplitude and SNR values as the frequency of the click stimulus increased.
- The mean absolute amplitude for 1 kHz click frequency, across the age groups was found to increase with the increasing age, except for 8-12 months age group. The mean absolute amplitude at 2 kHz click frequency did not show an increasing trend with the increasing age. The highest mean SNR value was obtained for 6-8 months age group, whereas, least value was obtained for 8-12 months age group. And for the 4 kHz click frequency, least mean absolute amplitude was obtained for 8-12 months age group.
- The mean SNR values showed an increasing trend across different frequencies of click stimulus as well as across the age groups.

5.2 Absolute amplitude and SNR for different frequencies of TBOAEs across age groups

- The mean absolute amplitude and SNR values increases as the frequency of the toneburst stimulus increased.
- Whereas, across the age groups, no trend was observed for the mean absolute amplitude and SNR values.

5.2.1 ***Effect of age on amplitude at each click frequency***

The absolute amplitude at 2 kHz showed a statistically significant difference across all the age groups. Whereas, the absolute amplitude at 1 kHz and 4 kHz showed, significant difference for few age groups only.

5.2.2 ***Effect of age on SNR at each click frequency***

- Only the 0-4 month's age group showed statistically significant difference across all the age groups for the SNR values at 1 kHz click frequency.
- For 2 kHz frequency, all the age groups other than 0-4 months group, showed a statistically significant difference.
- Whereas, the SNR values at 4 kHz showed no statistically significant difference across all the age groups.

5.2.3 ***Effect of age on amplitude at each tone burst frequency***

- The absolute amplitude at 1 kHz toneburst stimulus showed statistically no significant difference across all the age groups.

- For the absolute amplitude at 1 kHz toneburst stimulus, the lower age groups (0-4 months & 4-6 months) were statistically significant from the higher age groups (6-8 months & 8-12 months).
- Whereas, the absolute amplitude at 4 kHz toneburst stimulus, statistically significant difference was found only between 4-6 months and 6-8 months age group.

5.2.4 *Effect of age on SNR at each tone burst frequency*

- The SNR at 1 kHz toneburst stimulus, showed statistically no significant difference for 4-6 months age group. Whereas, the other age groups showed statistically significant difference with few age groups and no statistically significant difference with others.
- For the SNR at 2 kHz toneburst stimulus, 0-4 months group was significant different from 4-6 months and 8-12 months age group, 4-6 months group was significant different from 0-4 and 6-8 months age group, 6-8 months age group was significant different from 4-6 and 8-12 months age group, and the 8-12 months age group was significant different from 0-4 and 6-8 months age group.
- The SNR for 4 kHz toneburst stimulus, was statistically significantly different for 6-8 months age group. Whereas, the other age groups showed statistically significant difference with few age groups and no statistically significant difference with others.

5.2.5 *Effect of click frequency on amplitude at each age group*

- The absolute amplitude for 0-4 months and 8-12 months age group was found to be statistically significantly different across all the click frequencies.

- For 4-6 and 6-8 months age group, the absolute amplitude at 2 kHz showed statistically no significant difference from 4 kHz and vice versa.

5.2.6 *Effect of click frequency on SNR at each age group*

- Only the 0-4 months age group showed statistically significant SNR values across all the click frequencies tested.
- Whereas, for the other age groups, few frequencies showed significant difference and few did not show significant difference.

5.2.7 *Effect of tone burst frequency on amplitude at each age group*

The absolute amplitude only for 8-12 months age group showed statistically significant difference across all the toneburst frequencies.

5.2.8 *Effect of toneburst frequency on SNR at each age group*

All the age groups, except 4-6 months age group, showed a statistically significant difference for SNR, across all the toneburst frequencies tested.

5.3 *Comparison of absolute amplitude across click and toneburst stimuli at each age group for each frequency*

Only for 4-6 months and 8-12 months age group, the comparison of the absolute amplitude values between click and toneburst stimuli was found to be statistically significantly different ($p < 0.05$) at all the frequencies.

5.4 *Comparison of SNR across click and toneburst stimuli at each age group for each frequency*

Only for 6-8 months age group, the comparison of SNR values between click and toneburst stimuli was found to be statistically significantly different ($p < 0.05$) at all the frequencies.

Conclusion

From the present study it can be concluded that, TBOAEs showed a stronger response level than CEOAEs. Hence, to obtain a more frequency specific response, a toneburst stimulus can be used instead of click stimulus. A significant difference across the age groups was found for both CEOAEs and TBOAEs. The response in infants is more robust and it contains more high frequency energy. As the mean SNR value is higher at higher frequencies for both click and toneburst stimulus, while evaluating the response in infants not only the amplitude of the response should be considered but also the SNR values.

Future directions

- The obtained values of the present study can be used as a clinical tool for screening and diagnostic purposes in the pediatric populations.
- Toneburst stimuli can be used instead of click stimuli to obtain a more frequency specific response in both pediatric and adult populations.
- As the prevalence of TBOAEs are higher than compared to CEOAEs in infants, it can be included in the screening and diagnostic test battery.

REFERENCES

American National Standards Institute. (1991). *American National Standard maximum permissible ambient noise levels for Audiometric Test room.* (ANSI S3.1-1991). New York: American National Standards Institute.

Anitha, T. (2002). Auditory *Learning Manual for English Speaking Hearing Impaired Children.* Unpublished Masters Dissertation. University of Mysore.

Anson, B. J., & Donaldson, J. A. (1981). In *Surgical anatomy of the temporal bone*. (pp. 283). Philadelphia: Saunders, W. B.

Bonfils, P., & Uziel, A. (1989). Clinical applications of evoked acoustic emissions: Results in normally hearing and hearing impaired subjects. *Annals of Otology, Rhinology and Laryngology, 98,* 326-331.

Bonfils, P., Dumont, A., Marie, P., Francois, M., & Narcy, P. (1990). Evoked otoacoustic emissions in newborn hearing screening. *Laryngoscope, 100,* 186-189.

Bridger, O. (1961). In Northern, J. L. & Downs, M. P. (2002). *Hearing in Children, (5th edn.).* (pp. 268). Lippincott: Williams & Wilkins.

Bray, P., & Kemp, D. T. (1987). An advanced cochlear echo technique suitable for infant screening. *British Journal of Audiology, 21,* 191-204.

Capua, et al. (2003). Newborn hearing screening by transient evoked otoacoustic emissions: analysis of response as a function of risk factors. *Acta otorhinolaryngology, 23,* 16-20.

Chan, R. H., & McPherson, B. (2000). Test-retest reliability of toneburst evoked otoacoustic emissions. *Acta Otolaryngologica, 120,* 825-834.

Chuang, S. W., Gerber, S. E., & Thronton, A. R. D. (1993). Evoked Otoacoustic Emissions in Preterm Infants. *International Journal of Pediatric Otorhinolaryngology, 26,* 39-45.

Collet, L., Gartner, M., Moulin, A., & Morgon, A. (1990). Age related changes in evoked otoacoustic emissions. *Annals of otology, Rhinology and Laryngology, 99,* 993-997.

Cope, Y., & Lutman, M. E. (1993). In Mccormick, B. (2004). Otoacoustic emission. *Paediatric audiology 0.5 years, (2nd edn.).* (pp. 261- 265). San Diego: Singular Publishing Co.

Costa, J. M. D., Almeida, V. F., & Sampaio, A. L. L. (2009). Transient and distortion product evoked emissions in premature infants. *International Archives Otorhinolaryngology, 13 (3),* 309-316.

Elbergling, C., Parbo, J., Johnson, N. J., & Bagi, P. (1985). Evoked acoustic emissions clinical application. *Acta otolarygologica, Supple, 421,* 77-85.

Engdhal, B., Arnesen, A. R., & Mair, I. W. S. (1994). Otoacoustic emission in the first year of life. *Scandinavian Audiology, 23,* 195-200.

Glattke, T. J., Pafitis, I. A., Cummiskey, C., & Herer, G. R. (1995). Identification of hearing loss in children and adults using measures of transient otoacoustic emission reproducibility. *American Journal of Audiology, 4,* 71-87.

Hancur, C. (1999). *Transient evoked otoacoustic emissions in the neonatal period.* Unpublished Masters Thesis. Syracuse University.

Hauser, R., Probst, R., & Cohle, E. (1991). Click and tone burst evoked otoacoustic emissions in normally hearing ears and in ears with high frequency sensory neural hearing loss. *European Archives of Oto rhino laryngology, 248,* 345-352.

Hussain, D. M., Gorga, M. P., Neely, S. T., Keefe, D. H., & Peters, J. (1998). Transient evoked otoacoustic emissions in patients with normal hearing and in patients with sensory neural hearing loss. *Ear and Hearing, 19,* 434-449.

Jayaradha, S. (2000). *Comparison of signals-to-of-clicks and tone bursts evoked otoacoustic emissions in normal and in patients with different magnitude of hearing loss.* Unpublished Independent Project, University of Mysore.

Johnsen, N. J., Bagi, P. & Elberling, C. (1983). Evoked acoustic emission from the human ear. *Scandinavian Audiology, 12,* 17-24.

Johnsen, N. J., Parbo, J. & Elberling, C (1989). Evoked acoustic emission from the human ear. *Scandinavian Audiology, 18,* 59-62.

Johnsen, N. J., Parbo, J. & Elberling, C. (1982). Evoked acoustic emission from the human ear: Normative data in young adults and influence of posture. *Scandinavian Audiology, 11,* 69-77.

Kapoor, R., Panda, N. K. (2006). Transient evoked otoacoustic emissions. *Indian Journal of Pediatrics, 73 (4),* 283-286.

Kemp, D. T. (1978). Stimulated acoustic emissions from the human auditory system. *Journal of the Acoustical Society of America, 64,* 1386-1391.

Kemp, D. T., Bray, P., Alexander, L., & Brown, A. M. (1986). Acoustic emission cochleography : Practical aspects. *Scandinavian audiology, (supple: 25),* 71-95.

Kemp, D. T. (1988). Developments in cochlear mechanics and techniques for non-invasive evaluation. *Advances in Audiology, 5,* 27-45.

Kemp, D. T., Ryan, S., & Bray, P. (1990). A guide to the effective use of otoacoustic emissions. *Ear and Hearing, 11*, 93-105.

Kok, M. R., Zanten, G. A., & Brocaar, M. P. (1992). Growth of evoked otoacoustic emissions during the first days postpartum. *Audiology, 31*, 140-149.

Kok, M. R., Zanten, G. A., Brocaar, M. P., & Wallenburg, H. C. S. (1993). Click-evoked otoacoustic emissions in 1036 ears of healthy newborn. *Audiology, 32*, 213-223.

Kei, J., McPherson, B., Smyth, V., Latham, S., & Loscher, J. (1997) Transient evoked otoacoustic emission in infants: Effects of gender, ear asymmetry and activity status. *Audiology, 36,* 61-71.

Lafreniere, D., Jung, M. D., Smurzynski, J., Leonard, G., Kim, D. O., & Sasek, J. (1991). Distortion product and click evoked otoacoustic emissions in healthy newborns. *Archives of Otolaryngology, Head and Neck Surgery, 117,* 1382-1389.

Liu, B., Song, B. B., Liu, C., & Zhao, X. Y. (1996). A research for basic properties of several OAEs in normal hearing subjects. *Chinese Archives of otolaryngology head neck surgery, 3,* 3-8.

Lutman, M. E., Mason, S. M., Sheppard, S., & Gibbin, K. P. (1989). Differential diagnostic potential of otoacoustic emissions: a case study. *Audiology, 28,* 205-210.

Madell, J. R. (1988). Identification and treatment of very young children with hearing loss. *Infants and Young Children, 1*(2), 20-30.

McPherson, B., Li, S. F., Shi, B. X., Tang, J. L., & Wong, B. Y. (2006). Neonatal hearing screening: evaluation of tone-burst and click evoked otoacoustic emission test criteria. *Ear and Hearing,* 256-262.

Martin, F. N. (1990). The pediatric patient. In *Introduction to Audiology. (*pp. 395). New Jersy: Prentice-Hall Inc.

Martin, G. K., Ohlms, L. A., Franklin, D. J., Harris, F. P., & Lonsbury-Martin, B. L. (1990). Distortion Product Emissions in Humans: Influence of Sensorineural Hearing Loss. *Annals of Otology, Rhinology and Laryngology, 99,* 30-42.

Merchant, G. R., Horton, N. J., & Voss, S. E. (2010). Normative reflectance and transmittance measurements on healthy newborn and 1-month-old infants. *Ear and Hearing, 31,* 746-754.

Moleti, A., Sisto, R., Paglialonga, A., Siblla, F., Anteunis, L., Parazzini, M., & Tognola, G. (2008). Transient evoked otoacoustic emission latency and estimates of cochlear tuning in pre-term neonates. *Journal of Acoustical Society of America, 124,* 2984-2994.

Nelson, D. A. & Kimberley, B. P. (1992). Distortion product emissions and auditory sensitivity in human ears with normal hearing and cochlear hearing loss. *Journal of Speech and Hearing Research, 35,* 1141-1159.

Northern, J. L., & Downs, M. P. (2006). *Hearing in Children (6th edn.).* (pp. 233-235). Lippincott: Williams and Wilkins.

Norton, S. J., & Neely, S. T. (1987). Tone-burst evoked otoacoustic emissions from normal-hearing subjects. *Journal of the Acoustical Society of America, 81,* 1860-1872.

Norton, S. J., & Widen, J. E. (1990). Evoked otoacoustic emissions in normal-hearing infants and children: Emerging data and issues. *Ear and Hearing, 11,* 121-127.

Norton, S. J. (1993). Application of transient evoked otoacoustic emissions to pediatric populations. *Ear and Hearing, 14,* 64-73.

Norton, S. J., Gorga, M. P., Widen, J. E., Folsom, R. C., Sininger, Y., & Cone-Wesson, B. (2000). Identification of neonatal hearing impairment: evaluation of transient evoked otoacoustic emission, distortion product otoacoustic emission, and auditory brain stem response test performance. *Ear and Hearing, 21,* 508-528.

Nozza, R. J, & Sabo D. L. (1992). Transiently evoked OAE for screening school-aged children. *The Hearing Journal, 45,* 29-31.

Phillip, A. G. S. (1979). Non-invasive Monitoring of Intracranial Pressure: A New Approach for Neonatal Clinical Pharmacology Symposium on Pharmacology. *Clinical Perinatology, 6*, 123-137.

Prieve, B. A., Fitzgenald, T. S., & Schuttle, L. E. (1997a). Basic characteristics of click-evoked otoacoustic emission in infants and children. *Journal of Acoustical Society of America, 102,* 2860-2870.

Prieve, B. A., Fitzgenald, T. S. & Schuttle, L. E. (1997b). Basic Characteristics of DPOAE in infants and children. *Journal of Acoustical Society of America, 102,* 2871-2879.

Probst. R., (1991). A review of otoacoustic emission. *Journal of Acoustic Society of America, 89,* 2027-2067.

Probst, R., Coats, A. C., Martin, G. K., & Lonsbury-Martin, B. L. (1986). Spontaneous, click and toneburst-evoked otoacoustic emissions from nornal ears. *Hearing Research, 21,* 261-275.

Probst, R., Antonelli, C., & Pierren, C. (1989). Methods and preliminary results of distortion product otoacoustic emissions in normal and pathological ears. Abstracts. Second International Conference on Cochlear Mechanics and Otoacoustic Emissions, Rome, Italy.

Pujol, R., & Lavigne-Rebillard, M. (1995). Early stages of innervations and sensory dell differentiation in the human fetal organ of corti. *Acta Oto-Laryngologica (Supple: 423),* 43-50.

Salomon, G., Anthonisen, B., Groth, J., & Thomsen, P. P. (1992). In Bess, F. H., & Hall, J. W. (Eds.). Screening Children for Auditory Functions. (pp. 191-206). Nashville: Wilkerson Center Press.

Shi, J. F., Wang, N. Y., Yuan, J. J., Li, L., & Zhang, J. (2010). Comparison of transient evoked otoacoustic emissions in new born and frequency specific approach. *Zhonghua Er Bi Yan Hou. Jing wai ke za zhi, 45*(3), 206-211.

Sisto, R., & Moleti, A. (2007). Transient evoked otoacoustic emission latency and cochlear tuning at different stimulus levels. *Journal of Acoustical Society of America, 122,* 2183-2190.

Smurzynski. J. (1994). Longitudinal measurements of distortion product and click evoked otoacoustic emissions of pre-term infants: Preliminary results. *Ear and Hearing, 15,* 210-223.

Sohmer, H., & Freeman, S. (1993). Hypoxia Induced Hearing Loss in Animal Models of the Fetus in-utero. *Hearing Research, 55,* 92-97.

Speak, Z., Leonard, J., Kim, D. O., Jung, M. D. & Smurzynski, J. (1991). Otoacoustic emission in normal and Hearing Impairment Children and Normal Adults. *Laryngoscope, 101,* 965-976.

Stevens, J. C., Webb, H. D., Hutchinson, J., Connel, J., Smith, M. F., & Buffin, J. T. (1990). Click Evoked otoacoustic emissions in neonatal screening. *Ear and Hearing, 11,* 128-133.

Stevens, J. C., Webb, H. D., Hutchinson, J., Connel, J., Smith, M. F., & Buffin, J. T. (1989). Click evoked otoacoustic emissions compared with brainstem electric response. *Archives of Diseases in Childhood, 64,* 1105-1111.

Stover, L., & Norton, S. (1993). The effects of aging on otoacoustic emissions. *Journal of Acoustical Society of America, 94,* 2670–2681.

Thronton, A. R. D., Kimm, L., Kennedy, C. R., & Cafarelli-Dees, D. (1993). External and Middle Ear Factors Affecting Evoked Otoacoustic Emissions in Neonates. *British Journal of Audiology, 27,* 319-327.

Uziel, A., & Piron, J. P. (1991). Evoked otoacoustic emission from normal newborn and babies admitted to an intensive care baby unit. *Acta otolaryngology, (supple: 482),* 85-91.

Zanten, B. G. A., Kok, M. R., Brocaar, M. P., & Sauer, P. J. J. (1995). The click-evoked oto-acoustic emission, c-EOAE, in preterm-born infants in the post conceptional age range between 30 and 68 weeks. *International Journal of Pediatric Otorhinolaryngology. (Suppl, 32),* 187-197.

Vohr, B. R., White, K. R., Maxon, A. B., & Johnson, M. J. (1993). Factors Affecting the Interpretation of Transient Evoked Otoacoustic Emission Results in Neonatal Hearing Screening. *Seminars in Hearing, 14*(1), 57-72.

Welch, D., Greville, K. A., Thorne, P. R., & Purdy, S. C. (1996). Influence of acquisition parameters on the measurement of click evoked otoacoustic emissions in neonates in a hospital environment. *Audiology, 35,* 143-157.

Widen, J. E. (1997). Evoked otoacoustic emissions in evaluating children. In Robinette, M. S., & Glattke, T. J. (Eds.). *Otoacoustic emissions: clinical applications (pp. 271-306).* New York: Thieme.

Wit, H. P., & Ritsma, R. J. (1979), stimulated acoustic emissions from the human ear. *Journal of Acoustical Society of America 66,* 911-913.

Wlodyka, J. (1978). Studies on Cochlear Aqueduct Patency. *Annals of Otolaryngology,* 87, 22-28.

Zhang, V. W., & McPhersan, B. (2008). Tone burst evoked otoacoustic emissions in neonates; normative data. *BMC ear, nose and throat disorders, 8,* 171-176.

Zhang, V. W. McPherson, B., Shi, B. X., Tango, J. L. F., & Wong, B. Y. K. (2007). Neonatal hearing screening: A combined click evoked and tone burst otoacoustic emissions approach. *International journal of Paediatric Otorhinolaryngology, 72,* 351-360.