Transient Evoked Otoacoustic Emission in Neonates with and without Vernix

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Abstract- Transient evoked otoacoustic emissions (TEOAEs) are not necessarily recordable in all healthy neonates, before discharge, due to transient conditions (such as vernix in ear canals). To determine the possibility and feasibility of screening of very young newborns before discharge from hospital (JCIH, 2007), it is important to know how vernix in external ear during this time affects screening results and how it should be managed. After establishing an interobserver agreement index , the presence of varying degrees of vernix among newborn and their TEOAE finding was studied in 112 full term medically stable newborns before 72 hours from birth or before discharge (whichever was early), and after 15 days from the time of first screen. Vernix was found in 52.2% ear canals of neonates from birth to 72 hours, which dropped to only 15.2% of non-occluding vernix after 15 days. Prevalence of occluding external canal vernix is 35.4% in well newborns 24 hours or younger and 29.4% in infants aged 48 to 72 hours. Since vernix in ear canal did not drop significantly before 72 hours of age, it is recommended that the screening may be undertaken after 15 days to overcome the problem of vernix. It was observed that 73 out of the total 79 ears that failed TEOAE screening had vernix. The presence of vernix significantly (X^2=123.086, p<0.05) influenced the pass rate on TEOAE screening. 19.6% of ears had vernix and passed TEOAE while 32.6% had vernix and failed the screen. After 15 days 98.7% of previously referred ears passed the screen. Cleaning of vernix has been observed to improve pass rates from 76% to 91% (Chang et al, 1993) and 58.5% to 69% (Doyle et al, 2000), yet it is not practiced while screening. To overcome the problem of vernix it is also recommended that the reproductive and child health guidelines to include a system for clearing the ear as are for eyes.

Index Terms- Transient Evoked Otoacoustic Emission, prevalence, false positive, Vernix Caseosa

I. INTRODUCTION

This article guides a stepwise walkthrough by Experts for writing a successful journal or a research paper starting from inception of ideas till their publications. Research papers are highly recognized in scholar fraternity and form a core part of PhD curriculum. Research scholars publish their research work in leading journals to complete their grades. In addition, the published research work also provides a big weight-age to get admissions in reputed varsity. Now, here we enlist the proven steps to publish the research paper in a journal.

UNCRPD (2006), article on health (25b, p. 14), emphasized the need for "early identification and intervention as appropriate, and services designed to minimize and prevent further disabilities". In an attempt to optimize infant hearing screening, JCIH (2007) recommends that all well baby nursery should "provide 1 hearing screening and, when necessary, a repeat screening no later than at the time of discharge from the hospital, using the same technology both times." Newborns are often discharged at the age of about 48 hours. At this age, TEOAEs are not necessarily recordable in all healthy neonates due to transient conditions causing temporary conductive hearing loss.

Vernix Caseosa is one among the most common causes for temporary conductive hearing loss in infants. It is a fatty (neutral lipid) residue of amniotic fluid found on the neonate's skin immediately after birth. The majority of the newborns have Vernix in ear canal shortly after birth (Prieve, 2007). McLellan and Webb (1961) encountered presence of vernix in 47% of a series of healthy neonates. Cavanaugh (1987) found that the eardrum was obscured in 56% of newborn less than 24 hours old, but only 19% had obscured eardrum by age 3 days. Kenner and Lott (2007) reported that otoscopy is not included in the examination of the newborn period since the ear canal is filled with vernix, amniotic debris and blood, which clears in approximately 60% of the term infants by 1 week of age but may persist for weeks. It may get lodged in the probe while recording OAE, thereby obstructing or attenuating the signal. Hall (2000, p. 226) reported that Vernix, which is not soluble in water and cannot be removed by simple irrigation of the external ear canal, clearly must be reckoned with if OAEs are to be recorded from neonates.

Literature for the prevalence of vernix is based on studies in the white race. Therefore, a preliminary study is needed to know the presence of vernix in Indian population. To determine the possibility and feasibility of screening of very young newborns before discharge from hospital (JCIH, 2007), it is important to know how vernix in external ear during this time affects screening results. This study investigated the relationship between external ear factors i.e., vernix Caseosa and hearing screening results by TEOAEs at two different time in neonates with vernix and after clearance of it. The objectives of this study are as follows: (1.) To study the Presence of vernix in neonatal ear canal, (2.) To study TEOAE in ear with vernix and without vernix before 72 hours from birth or before discharge (whichever being early), (3.) To

study TEOAE of referred ears(in 1st screening) with vernix and without vernix after 15 days from 1st screening, (4.) To study the false positive TEOAE on 1st screening in ear with vernix.

I. Method

A purposive sample of 112 neonates from SSKM Hospital in Kolkata were included in the study. The neonate's age ranged from 10 hours to 72 hours. Informed written consent was obtained from a parent of the newborn before testing. Otoscopic examination and TEOAE testing was done in a quite isolated room. Two experiments were conducted.

In experiment I the inter observer agreement index for quantifying vernix was obtained. (Rangasayee, 1986).A Keeler pocket otoscope with 2.5X magnifying lens was used to examine external ear canal of neonates for vernix. Vernix was quantified on a three category rating scale (Chang et al, 1993; Doyle et al, 1997, 2000; Prieve et al, 2009) as clear (It had very small amount of vernix along the edge of the canal i.e., 25% or less), partially occluding (anything between 25% debris and occlusion, & the tympanic membrane was not commonly observed in this condition), or occluding (white debris with no pockets of darkness along the edges). Absent indicated that vernix was not present in the ear canal.

Audiologist observed the otorhinolaringologist otoscopic examination of 10 full term medically stable newborn. The audiologist and otorhinolaryngologist individually observed ear canal of 20 neonates and interobserver agreement was obtained.

In experiment II, TEOAE were studied in neonates with and without vernix, before 72 hours from birth or before discharge (whichever was early), and after 15 days from first screen.TEOAE testing results were recorded as pass or refer.A total of 238 ears (119 neonates) 119 neonates were studied in stage 1, of which 7 did not follow up in stage 2 hence 112 neonates were included in the study. The neonates selected were full term medically stable newborns of the age range from birth to 72 hours or before discharge (whichever was early), who did not have any risk indicator (JCIH, 2007) and were delivered normally or by a caesarean section. Consent was taken from parents/guardians before including the neonates in the study. The same Keeler Pocket Otoscope with deluxe, medic luxe specula of no. 1 and 2 were used for vernix assessment and GSI Audio Screener plus was used for TEOAE screening. The clicks were presented at 50 Hz, and levels was 80 dB pSPL. The frequency range was 250 Hz to 5 KHz and included 5 half octave bands. The passing criterion followed for the TEOAE screening was of NIH study. It included that. At least 3-dB SNR at each half octave frequency band centered 1 KHz and 1.5 KHz and of 6 dB at each of the half-octave frequency bands centered at 2, 3, and 4 kHz. To pass the screening this SNR was required at 4 out of 5 octave band (Norton et al, 2000). Two stage same technology protocol was used.

In stage 1, ear canals of neonates were first observed using an otoscope, followed by which, TEOAE screening was done on each ear. Those ears with "TEOAE- refer" finding in stage 1 were subsequently re-examined by otoscope and rescreened for TEOAE after 15 days in stage 2.

The neonates were accompanied by a family member/ guardian or a nurse, to a quiet room. Both the tests were administered when the neonates were asleep to avoid movements, or quite with minimal movements, in quite environment to reduce the effect of noise and with auto calibrated instrument. Results were explained to parents in both stages. On failing the second screen, parents were counseled and guided for detailed audiological evaluation and intervention.

II. RESULTS

The interobserver agreement index was calculated using the following formula (cited in Rangasayee, 1986) and it was found to be 86.05%. As the agreement index was more than 80% so the agreement between the two observers was good.

$$\text{Agreement index} = \frac{\text{Agreed score (37)}}{\text{Agreed score (37) + difference Score (6)}} \times 100 = 86.05\%$$

Prevalence of external ear canal vernix.

Prevalence of vernix in the study was 52.2 %(117). Prevalence of occluding vernix was 30.8 %(69), partially occluding was 12.9% (29) and clear ear canal was 8.5% (19). In the second stage, after 15 days, out of the 52.5% neonates who had vernix, only 15.2 % had vernix but not occluding (clear). The decrease in the vernix was statistically significant after 15 days. The prevalence of completely occluding ear canal remains well above 29% even after 72 hours from birth, however partial occlusion of ear canal was more in neonates 48 to 72 old (13.6%) than in neonates younger than 24 hours.

TEOAE finding in 1st stage.

In the study, 64.5% (145) of the total 224 ears passed the screening while 35.5% (79) failed. Figure 2 reflects, out of the 35.5% (79 of 224) ears which failed, 74.7% (59 of 79) had completely occluding vernix; and out of the 64.5% which passed, 69.7% (101 of 145) did not have vernix. Therefore, with the increasing degree of vernix occlusion, the refer rate increased and the pass rate decreased. Among the 52.2% (117 of 224) who had vernix of various degrees, 62.4% (73) ears failed the TEOAE testing. 82.2% of these failed ears had completely occluding vernix. The presence of vernix significantly (X^2 =123.086, p<0.05) influenced the pass rate on TEOAE screening (Table 1). Therefore the initial refer rate of TEOAE in ears with vernix was 62.4% and that in ears without vernix was 5.6%.

Table I: Pearson Chi-Square value.

	Value	df	P-Value (95% confidence interval)
Pearson's chi square for Vernix in stage 1 and 2	25.005(a)	3	.000*
Effect of vernix on TEOAE	123.086(b)	3	.000

(a) 3 cells (37.5%) have expected count less than 5. The minimum expected count is 1.10. *p<0.05; (b) 0 cells (.0%) have expected count less than 5. The minimum expected count is 6.70.

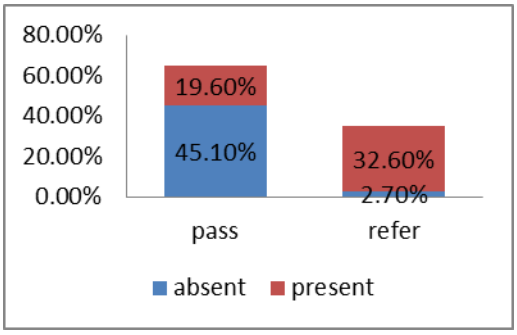


Figure I: The number of ears with and without vernix, which passed and failed the first and second TEOAE screening.

TEOAE findings in 2nd stage. The 79 ears that failed TEOAE screening in the first stage were followed up after 15 days. On repeat TEOAE screening, after the otoscopic examination, Out of the 79 ears that failed TEOAE in first screening, 84.8% did not have vernix and 15.2% still had clear ear canal (less than 25% vernix in canal). (Figure III).On second screening, out of the 79 ears, 83.4% ears without vernix passed, 15.2% ears with vernix passed but 1.3% without vernix failed.

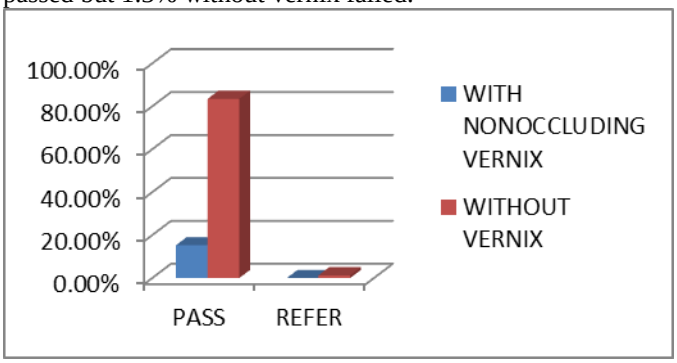


Figure III: TEOAE finding of ear with vernix and without vernix in stage 2

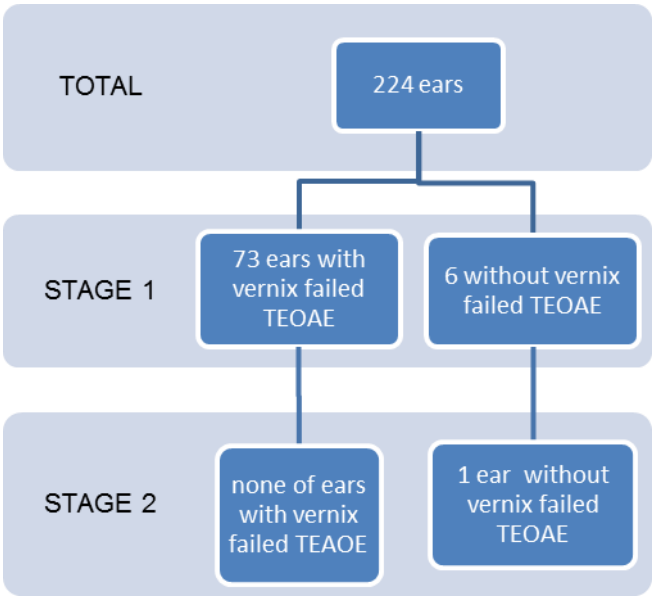


Figure IV: TEOAE fails in ears without and with vernix, in stage one and two.

False positive TEOAE finding.

Table II shows 78 out of the 79 ear that failed in first stage but subsequently passed in the second stage, constituted the false positive rate of TEOAE, i.e., 34.8%. Of these false positive TEOAE, 93.6% were those who had some amount of vernix. This high false positive result affects the specificity.

Table II: Performance

	(?) Hearing Loss Present	Hearing Loss Absent	Total
TEOAE Pass	0(false negative)	145 (true negative)	145
TEOAE Refer	1	78 (false positive)	79
Total	1	223	224

DISCUSSION

If hearing screening in well newborns is to be performed before hospital discharge, most infants will be tested before they reach 48 hrs.of age. Very few authors have studied the prevalence of vernix in this age group of neonates. Cavanaugh (1987) (cited in Thornton et al, 1993) studied 81 healthy neonates, reported vernix obscured ear canal in 56% ear of neonates < 24 hours, 24% ears of neonates aged 24 to 48 hrs.and 19% ears of neonates aged 48 to 72 hrs. Balkony et al (1978) (cited in Thornton et al, 1993) observed infants≤ 24hrs. Partial obstruction was found in all infants ≤ 24 hrs.after birth. Eavey (1993) studied 44 infants in NICU, reported vernix in 24 out of 88 ears. Levi et al (1997) tested 65 full term normal neonates, and found 15.4% (20/130 ears) that failed TEAOE had completely occluded ear canals. In the present study vernix was found present in 52.2 % of the ears, which was comparable to those observed by Cavanaugh (1987). Doyle et al (1997) tested 200 healthy newborns between the ages of 5 to 120 hrs.with mean age of 24.1 hrs. They observed 13% ear with completely occluding vernix and 32% non-occluding vernix. In the present study 30.8% had completely occluding 12.9% had partially occluding and 8.5% non-occluding vernix. Doyle et al reported 14.3% occluding vernix in < 24 hour old and 11.7% occluding vernix in > 24 hour old while in present study 35.4% of those ≤ 24 hours had completely occluding vernix and 22.2% of those 24 to 48 hours old had completely occluded vernix.

Doyle, Rodger, Fujikawa & Newman (2000) tested 200, 5 to 48 hrs.old healthy newborns, and reported 28% neonates had vernix that obscured ear canal. They reported reduced completely occluding as well as non-occluding vernix whereas in the present study the completely occluding vernix is found to reduce with age while partially occluding vernix is more in 48 to 72 hours old neonates than in neonates younger than 24 hours. This may be explained as a process of clearance of vernix, in which the completely occluding vernix might first reduce into partially occluding and subsequently resulting in a clear ear canal.

The majority of sites (n = 12 sites (57.13%)) report an average length of stay for a Vaginal delivery to be more than 24 h (between 24 and 48 h) (Kumar and Mohapatra, 2011). Thus out of the neonates included in this study, 84.8% were younger than or equal to 48 hours of age. In neonates younger than 24 hours, 37.2% had occluding vernix while 5.9% had non occluding vernix. In neonates 24 to 48 hours 34.7% had occluding vernix while 13.8% had non occluding vernix. However the occluding vernix was completely absent after 15 days and non-occluding

vernix was present only in 15% (12/79) of the followed up neonates. This reduction in the prevalence of vernix in neonates after 15 days was significant. Thus vernix was still prevalent in neonates 48 hours old which could potentially increase the refer rates, while it was cleared and did not interfere in TEOAE findings after its clearance (i.e., after 15 days).

Thornton et al (1993) reported that transient conditions like obstruction of external ear canal by debris lowers the success rate of recording TEOAE. Similar finding was observed in the present study since the ears with vernix has a higher initial refer rate (i.e., 32.6%) than the ears without vernix (i.e., 2.7%). It was also observed that since vernix was more prevalent in the right ear hence the TEOAE refer rate was higher in the right ear. The TEOAE refer rates were different for different degree of occlusion by vernix in the ear canal. Significantly higher refer rates were observed for completely occluding vernix than partially occluding vernix as compared with clear ear canals, as seen in figure II. All the ears with non-occluding vernix passed the TEOAE screening in the second stage. Hence, it can be stated that the partial occlusion and complete occlusion of vernix significantly influenced the TEOAE finding. However clear external auditory canal with non-occluding vernix did not influence the TEOAE findings.

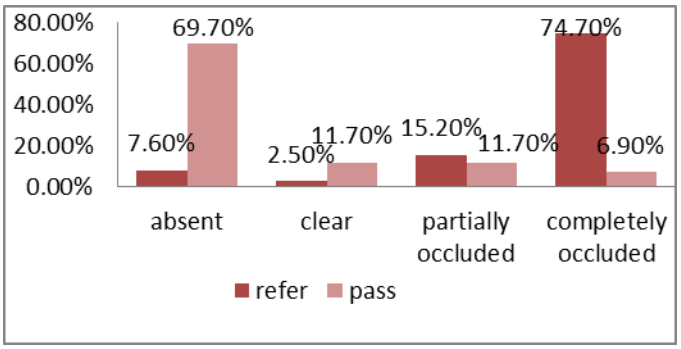


Figure II: Comparison of percentage of ear canals with different degree of vernix and the TEOAE findings in them in stage 1.

Chang et al (1993) suggested that cleaning the ear canal may raise the pass rate at earlier ages. Levi et al (1997) reported that TEOAE were obtained at first session in only 58.5% in both ears and in 30.8% in one ear only. They also reported that all ears from which TEOAE could not be recorded initially were occluded with vernix. In the present study 32.6% (73/224) had varying degree of occlusion, failed TEOAE screening while only 2.7% (6) ears without vernix failed TEOAE. Hence there is a difference in TEOAE finding in ears with vernix and without vernix.

Doyle et al (1997) used an ear curette; Doyle et al (2000) used a Baron number 3 or number 5 suction device and Levi et al used gentle suction under microscope was performed by an ENT specialist for cleaning of vernix. A standard procedure for cleaning of vernix was however neither described nor mentioned. Till now no consensus has been reached on the question whether the vernix should be cleaned or should be left to be expelled naturally. The WHO recommended intervention for improving maternal and newborn health (2009) and WHO Guideline on basic newborn resuscitation (2012) do not include cleaning of ear canal. Indian literature by AIIMS on care of baby at birth also does not include cleaning of ear canal. Doyle et al (2000) recommended for permitting time for external canal vernix to be naturally expelled. However this may increase time between follow up screen and may cause maternal anxiety for even the false positive OAEs. Considering a screening program utilizing TEOAE in neonatal units which discharge newborns within 48 hours, Levi et al (1997) reported that "the need of screening during the first 48 hours more than compensates for the added cost of otoscopic examination and cleaning by a specialist".

Levi et al (1997) found an increased pass rate of 78.5% in both ears and 21.5% in one ear. In agreement to them, in the present study, all ears with vernix which did not pass TEOAE in the first screen, passed it on second screening. All but 1 of the 6 ears without vernix, which did not pass in first screen, passed TEOAE in the second screen. Hence it was found that there is difference in refer rate of TEOAE finding between 1st and 2nd screening in ears with vernix and also in ears without vernix. This contention is supported by Doyle et al (1997, 2000) and Levi et al (1997).

It is reported in a previous study (Ng et al., 2004) that scheduling screening after day 20 would minimize false positive rate of the DPOAE screening. In this study, all infants involved underwent the second TEOAE screening at 15 days. The current study revealed significant increase in TEAOE pass in second screening, after the clearance of the vernix in ear canal i.e., from 64.7% to 98.7%

Vernix most frequently occurs during the first week of life, and may cause a false positive referral to TEOAE screening. False positive referrals observed in the present study was, 34.8%, which was much higher as compared to 15% observed by Chang et al (1993) and 20% in both ears by Levi et al (1997). The increase in the OAE pass rates after clearance of vernix reported by various researchers can be compared in table 4.

In order to determine the optimal timing of screening to be performed, the following factors should be taken into account.Firstly, the primary purpose of the UNHS was to detect infants with hearing loss as soon as possible. Secondly, positive ("refer") screens cause substantial parental concern and anxiety and most parents can only feel relieved after diagnostic audiological assessment (Poulakis, Barker, & Wake, 2003). Hence the high false positive should be minimized in order to avoid the unnecessary anxiety in parents. This can be done by providing screening after the clearance of vernix.

Table 4: Research findings on increase in pass rates of TEOAE after cleaning vernix.

S.No.	STUDY	TEOAE Pass Rate Before vernix cleaning	TEOAE Pass Rate After vernix cleaning
1	Chang et al (1993) studied 41 full term infants of age 43 hrs.	76%	91%
2	Levi et al (1997) studied 65 full term normal neonates	58.5% both ear & 30.8% one ear	78.5% both ear & 21.5% one ear
3	McNellis & Klein (1997) studied 50 healthy newborn, &	61% at first screen	98% at fourth screen

	repeated TEOAE screening 4 times		
4	Doyle et al (1997) studied 200 healthy new-born's, of 5 to 48 hrs.	79%	84%
5	Doyle et al (2000) studied 200 new-borns of age 5 to 48 hrs.	12.5% (14 ears) with vernix passed	51% (57 ears) passed after cleaning

I. SUMMARY & CONCLUSION

The presence of varying degrees of vernix among newborn were studied before discharge and 15 days from the time of first screen. It is recommended that by making guidelines for cleaning of ear as for eyes, the effect of vernix on hearing screening by TEOAE can be reduced. . Further studies are needed to standardize a procedure for safe cleaning of vernix from ear canal of neonates. Since the occurrence of vernix significantly reduces after 15 days, hence screening can be done 15 days after the first screen, resulting in low false positive value.

Further studies should investigate whether the high initial refer rate is attributable to vernix or middle ear effusion. Further research should continue studying the effects of vernix on other physiological hearing screening test like DPOAE, Since Doyle et al (2000) refute recommending cleaning procedure due to the risk involved, it would be useful to device a noninvasive medical methods (ear drops) for cleaning.. The factors for this variation in prevalence of vernix based on birth method needs to be studied further.

Appendix 1: ***Risk indicators associated with permanent congenital, delayed-onset, or progressive hearing loss in childhood.***

Risk indicators that are marked with a "§" are of greater concern for delayed-onset hearing loss.

1. Caregiver concern§ regarding hearing, speech, language, or developmental delay.
2. Family history§ of permanent childhood hearing loss.
3. Neonatal intensive care of more than 5 days or any of the following regardless of length of stay: ECMO,§ assisted ventilation, exposure to ototoxic medications (gentimycin and tobramycin) or loop diuretics (furosemide/Lasix), and hyperbilirubinemia that requires exchange transfusion.
4. In utero infections, such as CMV,§ herpes, rubella, syphilis, and toxoplasmosis.
5. Craniofacial anomalies, including those that involve the pinna, ear canal, ear tags, ear pits, and temporal
6. Physical findings, such as white forelock, that are associated with a syndrome known to include a sensorineural or permanent conductive hearing loss.
7. Syndromes associated with hearing loss or progressive or late-onset hearing loss,§ such as neurofibromatosis, osteopetrosis, and Usher syndrome; other frequently identified syndromes include Waardenburg, Alport, Pendred, and Jervell and Lange-Nielson.
8. Neurodegenerative disorders,§ such as Hunter syndrome, or sensory motor neuropathies, such as Friedreich ataxia and Charcot-Marie-Tooth syndrome.
9. Culture-positive postnatal infections associated with sensorineural hearing loss,§ including confirmed bacterial and viral (especially herpes viruses and varicella) meningitis.
10. Head trauma, especially basal skull/temporal bone fracture§ that requires hospitalization.
11. Chemotherapy. §

Appendix II: CONSENT FORM

Date-

Your signature / thumb impression on this form indicates that you have understood to your satisfaction, the information regarding participation in the research and agree your child participation.

I, Mr/Mrs _____, Father/ Mother/ Guardian of _____ have been explained clearly the aim method and benefit of this test. I understand that my participation in the study is entirely voluntary and I have also been informed that I can withdraw from it at any time without losing on any benefit/ treatment for which my child is entitled. I also understand that this test is safe and will not cause any harm to my child.

Clinician signature

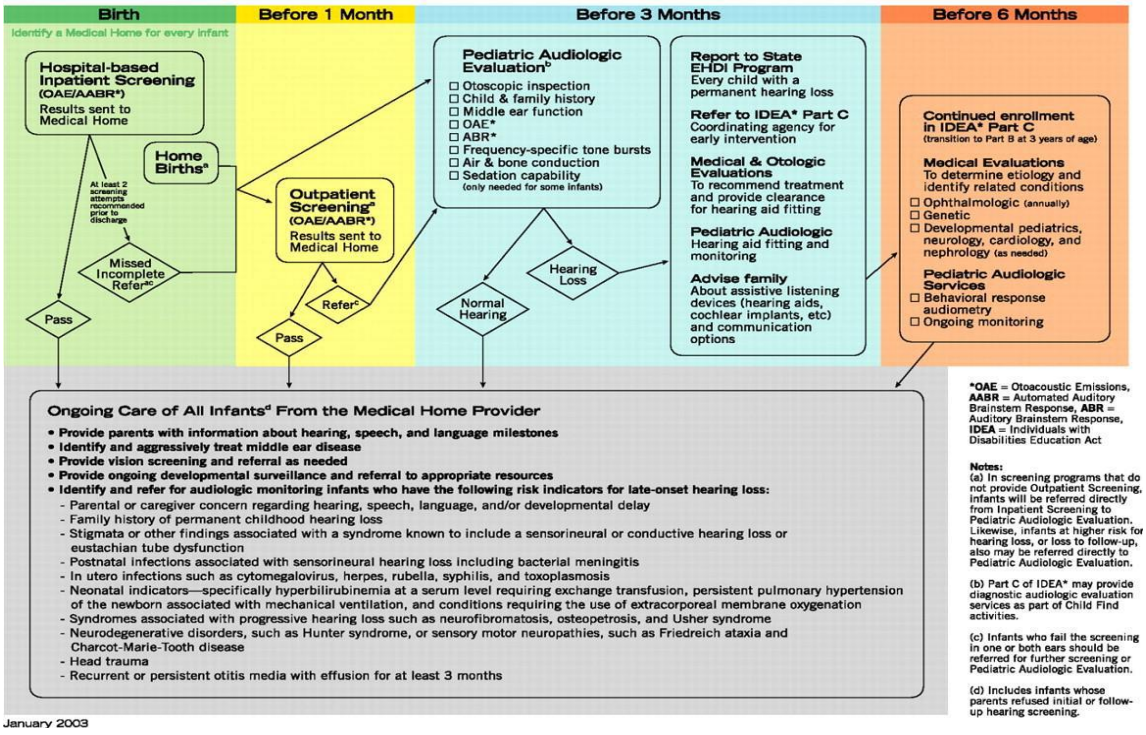
Parent/guardian signature

Date - Place-

Appendix III: algorithm for hearing screening. Available at: http://www.medicalhomeinfo.org/screening/Screen%20Materials/Algorithm.pdf

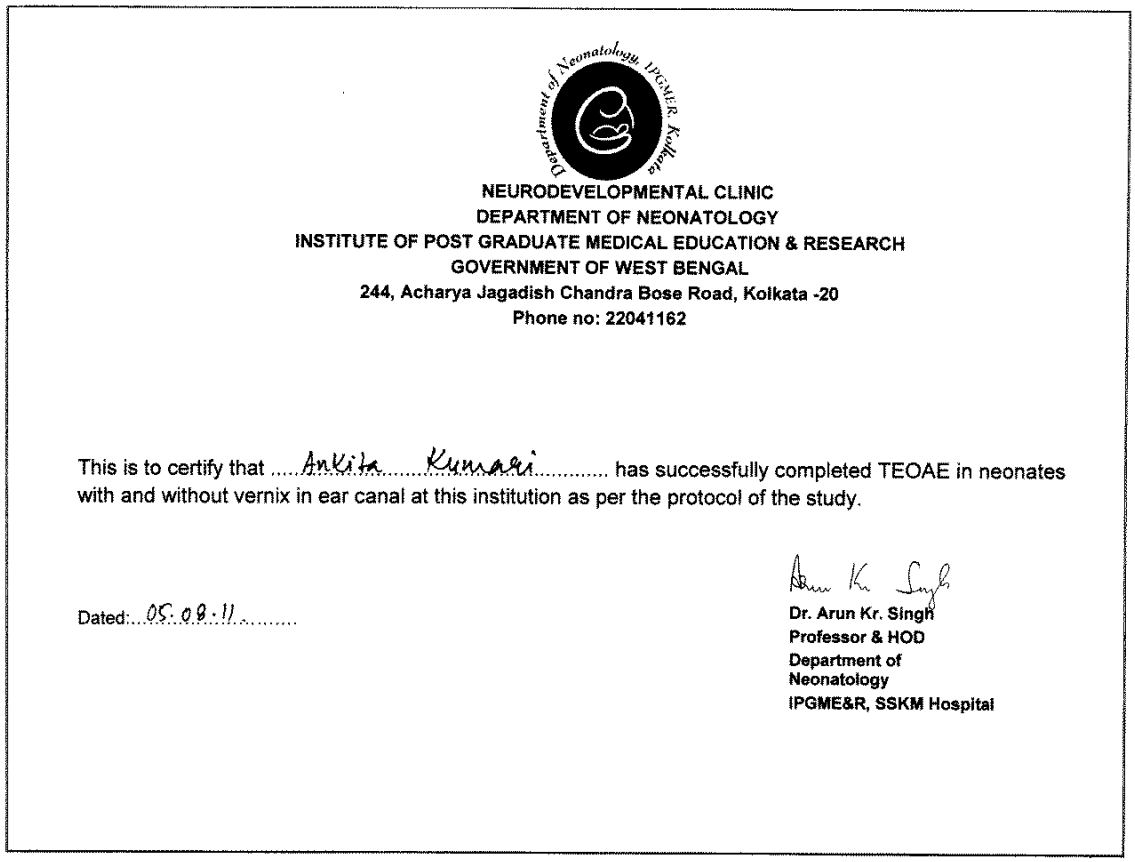
Universal Newborn Hearing Screening, Diagnosis, and Intervention

Guidelines for Pediatric Medical Home Providers



January 2003

Appendix IV.-CERTIFICATE OF DATA COLLECTION



NEURODEVELOPMENTAL CLINIC
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This is to certify that ....Ankita Kumari..... has successfully completed TEOAE in neonates with and without vernix in ear canal at this institution as per the protocol of the study.

Dated:...05.08.11.....

Dr. Arun Kr. Singh
Professor & HOD
Department of Neonatology
IPGME&R, SSKM Hospital

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