Acoustic reflex on newborns: the influence of the 226 and 1,000 Hz probes

A influência da sonda de 226 e 1.000 Hz no registro do reflexo acústico em neonatos

ABSTRACT

Purpose: To analyze the occurrence of acoustic reflex and its threshold on newborns using the 226 and 1,000 Hz probes. Methods: Thirty-six newborns with “PASS” results in newborn hearing screening and tympanogram with one or two peaks for both probe tones were included. Group I comprised 20 full-term newborns without risk indicator for hearing loss, and Group II comprised 16 newborns with at least one risk indicator. The study about ipsilateral acoustic reflex thresholds was conducted in 500, 1,000, 2,000, and 4,000 Hz. Results: The groups presented the acoustic reflex thresholds between 50 and 100 dB for both probe tones. In the comparison between the probes, there were differences in all frequencies evaluated in Group I, with the lowest threshold mean for the 1,000 Hz probe. In Group II, differences were detected at 2,000 Hz. The mean acoustic reflex thresholds were similar in both groups for the 226 Hz probe. There was a difference for the 1,000 Hz probe in all tested frequencies. The percentage of response was higher in both groups for the 1,000 Hz probe. The kappa test showed extremely poor agreement in the comparison of results between both probes. Conclusion: The occurrence of acoustic reflex was higher in newborns and its thresholds were lower with the 1,000 Hz probe both for healthy newborns and for newborns at risk.

RESUMO

Objetivo: Verificar a ocorrência e o limiar do reflexo acústico em neonatos utilizando a sonda de 226 e 1.000 Hz. Métodos: Trinta e seis neonatos com resultado “PASS” na triagem auditiva neonatal e timpanogramas com um ou dois picos para os dois tons de sonda. O Grupo I foi composto por 20 neonatos a termo sem indicador de risco para deficiência auditiva e o Grupo II, por 16 neonatos com pelo menos um indicador de risco. A pesquisa dos limiares do reflexo acústico ipsilateral foi realizada em 500, 1.000, 2.000 e 4.000 Hz. Resultados: Os grupos apresentaram limiar do reflexo acústico entre 50 e 100 dB para ambos os tons de sonda. Na comparação entre as sondas, houve diferença em todas as frequências avaliadas no Grupo I, com a média de limiares menor com a sonda de 1.000 Hz. No Grupo II, foi detectada diferença em 2.000 Hz. Com a sonda de 226 Hz, os limiares médios do reflexo acústico foram semelhantes nos dois grupos. Com a sonda de 1.000 Hz, houve diferença em todas as frequências avaliadas. A porcentagem de presença de resposta foi maior para ambos os grupos para a sonda de 1.000 Hz. O teste kappa revelou concordância extremamente pobre na comparação dos resultados entre as duas sondas. Conclusão: A ocorrência de reflexo acústico em neonatos foi maior e os limiares menores com a utilização da sonda de 1.000 Hz, tanto para neonatos saudáveis como para os neonatos de risco.

Study carried out at the Speech Language Pathology and Audiology Department, Dental School of Bauru, Universidade de São Paulo – USP – Bauru (SP), Brazil. (1) Speech Language Pathology and Audiology Department, Dental School of Bauru, Universidade de São Paulo – USP – Bauru (SP), Brazil. (2) Department of Orthodontics, Pediatric Dentistry and Public Health, Dental School of Bauru, Universidade de São Paulo – USP – Bauru (SP), Brazil. Financial support: Conselho Nacional de Desenvolvimento Científico e Tecnológico – CNPq. Conflict of interests: nothing to declare.
INTRODUCTION

Acoustic impedance tests are objective, non-invasive, safe, simple, and easy to conduct. They are widely used for hearing assessment[1-4]. These tests provide information regarding the mobility of the tympanosacular system by presenting the pressure in the external auditory canal (tympanometry) and the contraction of the stapedius muscle when stimulated by high-intensity sounds (acoustic reflex)[5,6].

A probe is placed in the external acoustic meatus to conduct acoustic impedance tests, thus causing hermetic seal, whereas one tone is continuously presented to the auditory system with varying pressure in the external acoustic meatus. At the point where the magnitude of the acoustic admittance is maximal, there is a pressure peak in tympanometry and, under these conditions, with the same probe tone, the acoustic reflex is analyzed[7-9]. The acoustic reflex is defined as the lowest stimulus intensity able to produce a detectable change in acoustic admittance[5,6,10]. The analysis of the presence or absence of Acoustic Stapedial Reflex Threshold (ASRT) may indicate a middle-ear dysfunction, significant cochlear hearing loss, retrocochlear pathology, or dysfunction in the facial nerve[11-14]. The results of the acoustic reflex also help to interpret the audiological findings after verifying the compatibility between diagnostic tests[15]. This procedure has diagnostic value in all age groups. In the pediatric population, due to the anatomical changes in the first two years of life, such as the increasing size of the auditory canal, changes in the orientation of the tympanic membrane, decreasing mass in the middle ear, and others[16,17], the mass component is prevalent in the middle ear; therefore, the resonance frequency tends to be low when compared to normal adults[18]. Therefore, this must be considered for choosing the frequency probe tone used to generate the acoustic reflex, thus preventing mistaken interpretations regarding the results of the examination.

Despite its clinical important, the acoustic stapedius reflex test has not been widely used among infants (0–6 months old)[10]. Nowadays, there is no regulation for the research of acoustic reflex threshold in this population[19]; some studies have tried to establish levels of ASRT for healthy newborns, by using different pure tones in broadband noise (BBN). Therefore, it is necessary to perform studies verifying the occurrence and threshold of acoustic reflex among newborns with different probe tones to generate the stimulus. The objective of this study is to verify the occurrence of ASRT in newborns using the 226 and 1,000 Hz probes.

METHODS

The study was approved by the Human Research Ethics Committee (Registration Number 078/2007). All parents and people in charge signed the informed consent.

The medical history of each newborn was analyzed, and parents or people in charge were interviewed in order to verify issues related to pre-, peri-, and post-natal history.

The analyses were conducted in the maternity ward of a public hospital.

Sample selection

Inclusion criteria

Newborns from both genders who obtained the “PASS” result in the newborn hearing screening and tympanogram, with one or two peaks for the 226 and 1,000 Hz probe tone, were included. These patterns indicate the normal function of the middle ear for the newborn population.

Sample

Thirty-six newborns participated in the study and were subdivided in two groups:

- Group I: 20 newborns (mean age of 36 hours) who were born at a term without history of pre-, peri-, or post-natal intercurrences and without risk indicator for hearing impairment[20];
- Group II: 16 newborns (mean age of 32 days) who presented at least one risk criterion for hearing impairment[20]. They were all in the Neonatal Intensive-Care Unit.

Transient-Evoked Otoacoustic Emissions

The equipment Madsen AccuScrenPRO, with nonlinear acoustic click stimulus at 73 dB SPL (maximum sound pressure level at 85 dB SPL) and frequency rate of 1.4 to 4 kHz, was used. To analyze the presence of response, the software used a binomial statistical test that calculates the probability of an emission having been recorded in a succession of sampling points, ranging from 6 to 12 ms after the end of the stimulus, presenting the results “PASS” or “REFER”.

Acoustic immittance measures

After the newborn hearing threshold, acoustic immittance measures were taken, and the ear was chosen randomly. That is, in the first newborn, the test was performed in the right ear, and, in the second one, in the left year, and so on and so forth.

The tympanometry and the analysis of the ipsilateral reflex threshold frequencies 500, 1,000, 2,000, and 4,000 Hz were performed with the equipment Madsen Otoflex 100 middle ear analyzer (GN Otometrics).

Data analysis

The descriptive analysis of the quantitative and qualitative variables (mean, median, standard deviation, maximum value, minimum value, 5th percentile, 95th percentile) was used, and, in the inferential analysis, the t-test was used to compare Groups I and II. The paired t-test was used to compare the results obtained with the 226 Hz probe and those obtained from the 1,000 Hz probe in the same group. The Pearson’s correlation test was used to measure the correlation between the results obtained from the 226 and 1,000 Hz probes and...
the agreement coefficient (kappa) to observe the concordance between the obtained results with the 226 and the 1,000 Hz probes. In all of the analyses, the adopted significance level was 5% (p≤0.05).

RESULTS

Table 1 presents the values regarding the descriptive analysis of the results found for the acoustic reflex thresholds obtained with the 226 and 1,000 Hz probes for Groups I and II.

Figures 1 and 2 show the percentage of presence and absence of responses obtained in the acoustic reflex study with the 226 and 1,000 Hz probes, respectively, for Groups I and II. Tables 2 and 3 provide the observed concordance (%) and the Kappa coefficient for this analysis in Groups I and II, respectively.

Tables 4 and 5 shows the comparison between the 226 and the 1,000 Hz probes (Table 4) and between groups (Table 5) for the acoustic reflex thresholds obtained in the frequencies of 500 to 4,000 Hz.

DISCUSSION

Nowadays, the regulation regarding data on ASRT with the 1,000 Hz probe tone is lacking. In the literature studied, only a few studies focused on analyzing the acoustic reflex threshold with the 1,000 Hz probe. In 2007, authors\(^{(21)}\) analyzed the data regarding ASRT in 267 ears, using 1,000 Hz pure tone stimuli, and the acoustic reflex threshold was considered as the lowest intensity in which there was a change in the admittance of at least 0.02 mmho. Based on this criterion, they found out that 94% of the ears had a mean acoustic

Table 1. Descriptive data (median, mean, standard deviation, minimum and maximum value, 5\(^{th}\), and 95\(^{th}\) percentile) regarding the acoustic reflex thresholds obtained in the 500 to 4,000 Hz frequencies of newborns in Groups II and II, conducted with 226 and 1,000 Hz probe tones

<table>
<thead>
<tr>
<th>Frequency (Hz)</th>
<th>500</th>
<th>1,000</th>
<th>2,000</th>
<th>4,000</th>
<th>500</th>
<th>1,000</th>
<th>2,000</th>
<th>4,000</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median</td>
<td>90</td>
<td>90</td>
<td>80</td>
<td>80</td>
<td>90</td>
<td>85</td>
<td>85</td>
<td>77</td>
</tr>
<tr>
<td>Mean</td>
<td>85.5</td>
<td>87.5</td>
<td>79.4</td>
<td>79.5</td>
<td>71.4</td>
<td>74.2</td>
<td>62.9</td>
<td>60.0</td>
</tr>
<tr>
<td>Standard deviation</td>
<td>9.9</td>
<td>12.8</td>
<td>11.5</td>
<td>11.2</td>
<td>11.4</td>
<td>12.9</td>
<td>9.3</td>
<td>10.0</td>
</tr>
<tr>
<td>Minimum</td>
<td>60</td>
<td>65</td>
<td>50</td>
<td>55</td>
<td>50</td>
<td>50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>Maximum</td>
<td>95</td>
<td>100</td>
<td>100</td>
<td>95</td>
<td>95</td>
<td>95</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td>5th percentile</td>
<td>65</td>
<td>65</td>
<td>65</td>
<td>65</td>
<td>65</td>
<td>65</td>
<td>65</td>
<td>65</td>
</tr>
<tr>
<td>95th percentile</td>
<td>95</td>
<td>100</td>
<td>100</td>
<td>95</td>
<td>95</td>
<td>85</td>
<td>90</td>
<td>85</td>
</tr>
</tbody>
</table>

Figure 1. Data regarding the percentage of presence and absence of acoustic reflex in the 500, 1,000, 2,000, and 4,000 Hz frequencies in newborns from Group I
Table 2. Kappa coefficient for the agreement between the probe tones (226 and 1,000 Hz) in newborns from Group I

<table>
<thead>
<tr>
<th>Frequency (Hz)</th>
<th>500</th>
<th>1,000</th>
<th>2,000</th>
<th>4,000</th>
<th>Kappa</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,000 Hz x 226 Hz probes</td>
<td>62.86</td>
<td>51.85</td>
<td>62.96</td>
<td>74.07</td>
<td>-0.07</td>
</tr>
</tbody>
</table>

Table 3. Kappa coefficient for the agreement between probe tones (226 and 1,000 Hz) in newborns from Group II

<table>
<thead>
<tr>
<th>Frequency (Hz)</th>
<th>500</th>
<th>1,000</th>
<th>2,000</th>
<th>4,000</th>
<th>Kappa</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,000 Hz x 226 Hz probes</td>
<td>58.62</td>
<td>48.28</td>
<td>48.28</td>
<td>55.17</td>
<td>0.22</td>
</tr>
</tbody>
</table>

Table 4. Comparison between the 226 and 1,000 Hz probes for acoustic reflex thresholds obtained in the 500–4,000 Hz frequencies considering Groups I and II

<table>
<thead>
<tr>
<th>Frequency (Hz)</th>
<th>500</th>
<th>1,000</th>
<th>2,000</th>
<th>4,000</th>
</tr>
</thead>
<tbody>
<tr>
<td>p-value</td>
<td>0.003*</td>
<td>0.002*</td>
<td>0.001*</td>
<td>0.000*</td>
</tr>
</tbody>
</table>

*Statistically significant (p<0.05)

Table 5. Comparison between Groups I and II for the reflex thresholds obtained with the 226 and 1,000 Hz probes in frequencies 500 to 4,000 Hz

<table>
<thead>
<tr>
<th>Frequency (Hz)</th>
<th>500</th>
<th>1,000</th>
<th>2,000</th>
<th>4,000</th>
</tr>
</thead>
<tbody>
<tr>
<td>226 Hz probe</td>
<td>0.850</td>
<td>0.995</td>
<td>0.206</td>
<td>0.321</td>
</tr>
<tr>
<td>1,000 Hz probe</td>
<td>0.831</td>
<td>0.010*</td>
<td>0.006*</td>
<td>0.002*</td>
</tr>
</tbody>
</table>

*Statistically significant (p<0.05)
reflex threshold of 93 dB and 90% between 80 and 105 dB HL. Similarly, however, with pure tone stimuli of 2 kHz and broadband noise, a study\(^{22}\) detected the presence of acoustic reflex (AR) in 97.6% of 42 normal newborns, with mean ASRT of 76.2 (±7.9) for the 2 kHz frequency and 64.9 (±7.8) for the broadband noise, respectively. Even though the results of these studies provide useful clinical information, they are restricted to the analysis of ASRT in only one frequency, or with broadband stimulus. A study from 2005\(^{23}\) analyzed the changes in the contralateral acoustic reflex of children aged 6 weeks and adults, using a probe with 250 and 8,000 Hz signals and broadband noise. Changes in AR were similar, above 1,000 Hz among children and adults; however, values lower than 1,000 Hz among children were less reliable. The reflexes were detected more successfully among infants when the correlation was calculated in 1,000–8,000 Hz, and among adults, from 250 to 2,000 Hz.

The ipsilateral reflex threshold was analyzed\(^{14}\) in the 1,000, 2,000, and 4,000 Hz frequencies, with the 226 Hz probe, and it was observed that for the female gender and the right ear, the mean dB (±standard deviation) was 96.43 (±10.62), 93.29 (±9.40), and 91.85 (±7.77); whereas for the male gender and the left ear, it was 92.67 (±9.0), 92.38 (±8.89), and 91.67 (±8.86). For the male gender and the right ear, the mean dB was 94.71 (±8.54), 93.06 (±10.59), and 92.18 (±7.18); whereas for the female gender and the left ear, it was 94.25 (±9.77), 94.13 (±9.76), and 95.47 (±6.82). There was no significant difference between gender and frequency. A study\(^{24}\) observed newborns and the association between Transient-Evoked Otoacoustic Emission (TEOAE) and changes in acoustic immittance with a 226 Hz probe. For that, the authors assessed 20 newborns with absent TEOAEs (experimental group) and 101 with TEOAEs (study group). The mean reflex threshold was 98.57 (±14.52) for the 500 Hz frequency, 102.50 (±9.64) for the 1,000 Hz frequency, 103.33 (±6.83) for the 2,000 Hz frequency, and 111.0 (±10.84) for the 4,000 Hz frequency in the group with absent TEOAEs; and 92.83 (±9.04) for the 500 Hz frequency, 93.48 (±8.48) for the 1,000 Hz frequency, 93.88 (±9.48) for the 2,000 Hz frequency, and 93.21 (±9.99) for the 4,000 Hz frequency in the group with TEOAEs.

In this study, for newborns in Group I, the mean ASRT obtained with the 226 Hz probe was lower (Table 1) in all frequencies in comparison to that found by other studies\(^{14,24}\). With the 1,000 Hz probe, the average was 74.2 dB for the 1,000 Hz frequency and 62.9 dB for 2,000 Hz (Table 1). A study\(^{21}\) found a mean of 93 dB for the 1,000 Hz and two other studies\(^{10,22}\) found means of 73.05 and 76.2 dB for 1,000 and 2,000 Hz, respectively. In the these studies, no other frequencies were analyzed. As in this study, another analysis\(^{30}\) has recently observed the ASRTs in 73 newborns with integrity of the external ciliated cells and observed the presence of AR in all the newborns. Values ranged between minimum and maximum: right ear — between 85 and 110 dB for 500 Hz and 80 and 110 dB for 1,000 to 4,000 Hz frequencies in ipsi- and contralateral measurements; left ear — between 85 and 110 dB for 500, 1,000, and 2,000 Hz frequencies and between 90 and 110 dB for 4,000 Hz in the contralateral measure. For the ipsilateral measurement, values between 80 and 110 dB were observed for the 1,000 and 2,000 Hz frequencies. These ASRT values are higher than the ones found in this study, which ranged from 55 to 95 dB for the 500 Hz frequency and 50 to 95 dB for the 1,000 to 4,000 Hz frequencies (Table 1).

Generally, the ASRT in Groups I and II varied between 50 dB (minimum value or 5th percentile) and 100 dB for both probe tones (226 and 1,000 Hz). The occurrence of acoustic reflex at 50, 55, or 60 dB was higher with the 1,000 Hz probe. Lower ASRTs were also found\(^{10,21,22}\) and were justified by the small size of the external auditory canal of newborns by vibration of the wall of the canal after a sound stimulus. However, it is worth to mention that the minimum intensity for the analysis of AR in the equipment is 50 dB NPS.

Regarding the acoustic reflex thresholds obtained among the newborns in Group II, literature has only one study\(^{25}\) with the same subject of investigation. In that study, the authors analyzed the Brainstem Auditory Evoked Potentials (BAEP), the TEOAEs, and the acoustic reflex among 53 children presenting risk indicators for hearing impairment. They observed that, from the ears presenting BAEP thresholds lower than 30 dB nHL, 91% had TEOAEs, which suggests psychoacoustic thresholds within normal patterns; out of these, 78% had acoustic reflex. Therefore, the authors proposed that the analysis of TEOAE acoustic reflex could be an efficient newborn hearing screening protocol.

In the comparison between probes (Table 4), it was possible to verify the differences in all of the frequencies assessed in the group of healthy newborns (Group I), and the mean of thresholds was lower with the 1,000 Hz probe. In Group II, a difference was detected between probes only in the 2,000 Hz frequency. However, the small sample size for the 226 Hz probe may have influenced the analysis of this variable. After comparing the groups (Table 5), the mean acoustic reflex thresholds found with the 226 Hz probe in the frequencies of 500 to 4,000 Hz were similar in both studied groups. With the 1,000 Hz probe, there was a difference between the 1,000, 2,000, and 4,000 Hz frequencies; the group of healthy newborns (Table 1) presented lower acoustic reflex thresholds. It is believed that the highest acoustic reflex thresholds for newborns at risk were due to the immaturity of the structures in the external and middle ear, caused by the size and weight of the newborn, as well as auditory pathways, since most newborns in this group presented prematurity as a risk indicator. Some authors studied the neurological maturation of auditory pathways\(^{26-28}\) and reported that it starts in the 6th month of intrauterine life, when the myelination of the peripheral region takes place, until the 6th month of postpartum life, and is completed around 18 months after birth — when the myelination of the brainstem is carried out. So, since most newborns in Group II were premature, it is natural that the maturation of the auditory pathways may be occurring at different times for Groups I and II. It is only possible to detect such a difference in the results obtained by using the 1,000 Hz probe in the ASRT research.
The analysis of the data presented in Figures 1 and 2, regarding the occurrence of acoustic reflex in Groups I and II, respectively, clearly demonstrates the difference between the findings of both the probes. In the analysis of acoustic reflex, studying thresholds in healthy newborns (Figures 1 and 2), the specificity was also 100% for the 1,000, 2,000, and 4,000 Hz with the 1,000 Hz probe. The occurrence (100%) in the presence of AR in Group I is higher than that found in two studies — 97.6% (22) and 91.3% (10). On the other hand, the percentage of the presence of response in the group with risk indicators (Group II) for the 1,000 Hz probe (Figure 2) ranged from 79% (500 Hz) to 93% (4,000 Hz). It is important to mention that the inclusion criteria involved newborns who had otoacoustic emissions, based on the assumption that, in general, the presence of TEOAEs suggests not only the normality in the cochlear amplifier mechanism, but also normal middle ear function, or close to normal. However, since there are reports in literature mentioning that conductive impairment may otoacoustic emissions, based on the assumption that, in general, the presence of TEOAEs suggests not only the normality in the cochlear amplifier mechanism, but also normal middle ear function, or close to normal. However, since there are reports in literature mentioning that conductive impairment may not influence the record of TEOAEs, or that resulted in reduced level of response (19, 29), only newborns who presented TEOAEs, tympanogram with one or two peaks for both probe tones were included in the study sample. These patterns are indicators of normal middle ear function for the newborn population. Therefore, the absence of acoustic reflex in some newborns from Group II may suggest the existence of new-born retardation in the cochlear function in Group I newborns. But what is the presence of AR in Group I is higher than that found in two studies — 97.6% (22) and 91.3% (10). On the other hand, the percentage of the presence of response in the group with risk indicators (Group II) for the 1,000 Hz probe (Figure 2) ranged from 79% (500 Hz) to 93% (4,000 Hz). It is important to mention that the inclusion criteria involved newborns who had otoacoustic emissions, based on the assumption that, in general, the presence of TEOAEs suggests not only the normality in the cochlear amplifier mechanism, but also normal middle ear function, or close to normal. However, since there are reports in literature mentioning that conductive impairment may not influence the record of TEOAEs, or that resulted in reduced level of response (19, 29), only newborns who presented TEOAEs, tympanogram with one or two peaks for both probe tones were included in the study sample. These patterns are indicators of normal middle ear function for the newborn population. Therefore, the absence of acoustic reflex in some newborns from Group II may suggest the existence of newborns with retrocochlear changes in the sample, as these presented risk indicators for hearing impairment, according to the Joint Committee on Infant Hearing (20).

The discrepancy between the results obtained with the 226 and 1,000 Hz probes in the acoustic reflex analysis was proved by the kapa test (Tables 2 and 3). In this test, the agreement between the responses obtained from both probes was assessed. The values revealed extremely poor agreement (0.8 to 1 is considered as almost perfect agreement), that is, in many cases, the analysis of ASRTs revealed absence of response when conducted with the 226 Hz probe, and the presence of response when conducted with the 1,000 Hz probe.

The results found in this study show that the analysis of the stapedius acoustic reflex in newborns from both groups was more efficient with the 1,000 Hz probe.

CONCLUSION

Based on the presented results, it is possible to state that the occurrence of acoustic reflex in newborns was higher, whereas thresholds were lower, when the analysis was conducted with the 1,000 Hz probe in comparison to the 226 Hz probe, both for healthy and at risk newborns.

ACKNOWLEDGEMENTS

We would like to thank the National Council for Scientific and Technological Research (CNPq) for the support.

REFERENCES