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‘Standard’ versus ‘nose reference’ electrode placement for measuring oVEMPs with air-conducted sound: test-retest reliability and preliminary patient results

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ABSTRACT

Objectives: This study compared two electrode placements (‘standard’ versus ‘nose reference’ placement) for measuring oVEMPs, elicited by air-conducted 500 Hz tone bursts. The test-retest reliability of both positions was evaluated and additionally both electrode placements were applied on a group of vestibular patients.

Methods: Eighteen healthy volunteers (range of 20-25 years) participated in the first part and were retested after one week for evaluation of the test-retest reliability. Eleven patients (range of 41-74 years) with a variety of vestibular pathologies were tested once.

Results: In the normal group, the nose reference electrode placement resulted in significantly larger peak-to-peak amplitudes ($p<0.001$), shorter n10 ($p=0.001$) and p15 ($p<0.001$) latencies and smaller 95% prediction intervals for the Inter-Ocular Ratio (IOR) ([−67.90, 67.90] for the standard position versus [−32.15, 32.15] for the nose reference position). Furthermore, an excellent amplitude and IOR test-retest reliability was observed with the nose reference configuration, as shown by the intraclass correlation coefficient (ICC), the coefficient of variation of the method error ($CV_{ME}$) and the minimal detectable differences (MDD). In the patient group, the same significant amplitude difference was found. Moreover, three patients presented with absent oVEMPs when recorded with the standard placement, whereas the nose reference placement could evoke a detectable oVEMP response.

Conclusions: This study demonstrated that a nose reference electrode position results in larger oVEMP amplitudes and achieves a better reliability for the most important clinical parameters (amplitude and IOR). Our patient data substantiate the possible clinical benefit of this position, but further systematic patient verification is required.

Significance: The nose reference electrode position facilitates the detection of generally very small oVEMP responses and shows a high test-retest reliability, showing promising potential for future use in the vestibular clinic.
KEYWORDS
Vestibular
Electrode placement
ocular Vestibular Evoked Myogenic Potential (oVEMP)
Test-retest reliability
Air-conducted stimuli

HIGHLIGHTS

1. The nose reference configuration evokes larger oVEMP amplitudes than the standard electrode configuration.
2. The nose reference configuration is able to record reliable responses in a normal and patient group.
3. A 100% response rate can be obtained with air-conducted sound stimuli using the nose reference configuration.

1. INTRODUCTION
Vestibular evoked myogenic potentials, abbreviated as ‘VEMPs’, were first described by Colebatch et al. (1992). VEMPs can be recorded in two ways: from tonically contracted cervical muscles (termed ‘collic’ or ‘cervical’ VEMPs (cVEMPs)) and from extra-ocular muscles (termed ‘ocular’ VEMPs (oVEMPs)). Both types are short-latency myogenic responses and can be elicited by air-conducted sound stimuli (ACS), bone-conducted vibrations (BCV), and galvanic stimuli. The cVEMP response to ACS is a manifestation of
the vestibulo-collic reflex, initiated by excitation of the saccule and the inferior branch of the vestibular nerve (Rosengren et al., 2010) and recorded from the sternocleidomastoid muscles in the neck (Colebatch et al., 1994). The oVEMP response results from the vestibulo-ocular reflex and is recorded from the extra-ocular muscles, in particular the inferior oblique (IO) muscle (Rosengren et al., 2005, Iwasaki et al., 2007, Todd et al., 2007). The oVEMP was discovered more recently by placement of surface electrodes underneath the eyes (Curthoys et al., 2006, Chihara et al., 2007, Iwasaki et al., 2007, Todd et al., 2007, Welgampola et al., 2009) and the specific origin of the response was unclear until lately. For this reason, the oVEMP is not widely implemented in clinical practice yet. The eVEMP to ACS, on the other hand, is well characterized today and therefore commonly used in vestibular clinics for assessment of the saccular function.

Inducing oVEMPs requires presentation of loud and abrupt sound stimuli. It is believed that subsequent excitation of the afferents of the otolith organ will travel across the vestibular nerve and project to the vestibular nuclei in the brainstem. These nuclei are connected to the motor neurons in the nucleus of the oculomotor nerve by the crossing longitudinal medial fascicle. Consequently, the IO and inferior rectus muscle (IR) contralateral to the stimulus side will be activated and the response can be measured with surface electrodes underneath the eyes. The oVEMP response consists of a biphasic waveform: first, a negative peak (n10 or N1) is seen around 10 ms, followed by a positive peak (p15 or P1) around 15 ms (Todd et al., 2007, Chihara et al., 2009, Welgampola et al., 2009).

The origin of the oVEMP response is a topic much debated between several researchers. However, most of the literature supports the hypothesis that oVEMPs (to air- or bone-conducted stimuli) reflect predominantly utricular and superior vestibular nerve function (Suzuki et al., 1969, Uchino et al., 1996, Isu et al., 2000, Iwasaki et al., 2009, Manzari et al.,

The reliability of the oVEMP test in diagnosing vestibular malfunctions is still under question, in particular for oVEMPs evoked by ACS. Ocular vestibular evoked myogenic potentials elicited by BCV, delivered by a minishaker (4810, Brüel & Kjaëér) or a reflex tendon hammer at the forehead (Fz position) or the mastoid, were found to provide large amplitudes and high response rates (in the order of 90% to 100%). On the other hand, lower amplitudes and lower response rates were observed for ACS-evoked oVEMPs, ranging from 72% to 100% (Cheng et al., 2009, Nguyen et al., 2010, Wang et al., 2010, Rosengren et al., 2011, Kantner et al., 2012). These findings are unfortunate because ACS transducers are readily available in most clinical centers, in contrast with the more expensive minishaker.

An attempt to increase the ACS-evoked oVEMP response rate and amplitude was presented by Sandhu et al. (2013), who shifted the active electrode beneath the eye towards the lateral canthus (presumably more on the IO muscle) and the reference electrode close to the medial canthus on the nose (on the IO muscle tendon) in a so-called belly-tendon montage. With this new montage, larger oVEMP amplitudes were found compared to the ‘standard’ electrode position, i.e. active and reference electrode both centrally beneath the eye. A recent study of Vanspauwen et al. (2016) also reported higher oVEMP n10-p15 peak-to-peak amplitudes and smaller confidence intervals for left-right amplitude differences (inter-ocular ratio (IOR)) with this electrode position, using bone-conducted sound stimuli delivered by a minishaker at Fz position. Since the oVEMP response is very small and therefore difficult to detect, this new electrode position seems to be promising for clinical use. However, further research is needed regarding the test-retest reliability and its clinical application with ACS. Therefore, the aim of the present study was to compare this position with the ‘standard’ electrode configuration in a
normal population and in a group of patients, using ACS. Furthermore, the test-retest reliability of both electrode placements was evaluated.

2. METHODS

2.1. Subjects

The study was carried out at the Department of Speech-Language Pathology and Audiology of the University of Pretoria (South Africa). Ethical approval was obtained from the institutional review board of the university before data collection commenced. Eighteen volunteers with normal hearing and without a history of vertigo or balance problems participated in the study (18 females, ranging in age from 20 to 25 years old with a mean age of 21.4 and a standard deviation of 1.4 years). All healthy volunteers were retested after a time interval of one week in order to evaluate test-retest reliability of the results obtained with both electrode configurations. Furthermore, eleven patients with a variety of vestibular symptoms and pathologies (7 males, 4 females, ranging in age from 41 to 74 years old with a mean age of 53.09 and a standard deviation of 10.08) were tested once. Table 1 shows an overview of relevant patient information and describes the tympanometry, cVEMP and caloric test results and the suspected diagnosis (where possible). Diagnosis was supported by an extensive interview of the patient, using the mnemonic tool ‘SOSTONED’ (Wuyts et al., 2016).

Due to the significant age difference ($p < 0.001$) as well as differences in gender diversity and sample size between the normal group and the patient group, no valid comparison of these groups could be made. Consequently, the results for both electrode positions were compared within each group.

2.2. oVEMP procedure
Tympanometry (226 Hz oto-admittance) was conducted preceding the oVEMP measurements in order to rule out any conductive component. The oVEMP measurements were performed using a Bio-Logic® Navigator® Pro system. Air-conducted sound stimuli were delivered by insert earphones (3M EARTONE®, 3A). The selection of the stimulus and recording parameters was based on recent literature and a preliminary investigation performed at the Department of Speech, Language and Hearing Sciences of the University of Ghent (Belgium) on a group of 17 young and healthy volunteers.

2.2.1. Stimulus and recording parameters

Air-conducted 500 Hz tone bursts with an intensity of 95 dB normalized HL, a stimulus rate of 5.1 Hz and a total of 150 stimulus repetitions were used to elicit the oVEMP responses. Stimuli were delivered monaurally and responses were recorded unilaterally. Furthermore, we chose for a rise/plateau/fall time of 1 – 4 – 1 ms and a band-pass filter of 1 – 500 Hz based on our own research and existing literature (Murnane et al., 2011, Wang et al., 2013, Kantner et al., 2014). The subjects were lying in supine position in a soundproof booth. They were asked to maintain an upward gaze during the measurement by fixating a stationary marked sign on the ceiling of the booth. Since the amplitude of the response increases with an upward gaze of 30 degrees or more (Rosengren et al., 2005, Iwasaki et al., 2008, Chou et al., 2009, Govender et al., 2009, Welgampola et al., 2009, Huang et al., 2012), the fixation point was placed in such a way that the subjects were looking up over an angle of 30°. Self-adhesive Ag/AgCl (Ambu®, Bluesensor) electrodes were used to record the oVEMP responses. The skin was prepared with alcohol wipes and Nuprep gel® prior to the positioning of the electrodes, in order to keep the impedances below 5 kΩ. Two electrode configurations were used to measure oVEMPs. The ‘standard’ electrode position is shown in Figure 4-1a and can be described as follows: the active electrodes were placed on the orbital margin under the center of the eye, the reference electrodes 2 cm beneath the active electrodes and the ground
electrode was applied on the chin (Rosengren et al., 2010). As shown in Figure 21b, the ‘nose reference’ electrode configuration, corresponding with the ‘belly-tendon montage’ of Sandhu et al. (2013) and the ‘nose position’ of Vanspauwen et al. (2016), uses a more lateral placement of the active electrodes (just below the lateral canthus and presumably more on the IO muscle). The reference electrodes were placed close to the medial canthus on the nose (Sandhu et al., 2013). Depending on the facial features of the individual, the inter-electrode distance ranged between 3 and 4 cm. Each oVEMP measurement consisted of two runs to assure reproducibility of the response.

2.2.2. Response parameters

The following parameters were analyzed in the current study: the absolute latencies of n10 and p15 (ms), the peak-to-peak amplitude (µV) and the inter-ocular ratio (IOR) (%). The IOR \( \text{IOR} = \frac{\text{left} \, \mu\text{V} - \text{right} \, \mu\text{V}}{\text{left} \, \mu\text{V} + \text{right} \, \mu\text{V}} \times 100, \% \) was calculated for the peak-to-peak amplitude and is based on the Jongkees formula for asymmetry calculations in vestibular testing (Jongkees et al., 1962).

2.3. cVEMP procedure

In the patient group, cVEMPs were obtained with the same equipment and similar stimulus and response parameters. Air-conducted 500 Hz tone bursts were presented through insert earphones with a stimulation rate of 5.1 Hz and an intensity of 95 dBlHL. Stimuli were delivered monaurally and recorded unilaterally, and a rise/plateau/fall time of 2 – 0 – 2 ms was used. Active electrodes were applied on the upper part of the sternocleidomastoid muscles, the reference electrode was placed on the sternum beneath the inter-clavicular ligament and the common electrode on the chin. Contraction of the sternocleidomastoid muscle was achieved by lifting and rotating the head to the non-stimulus side in supine position.
2.4. Statistical analysis

Statistical analysis was performed using SPSS software (IBM Corp. Released 2013. IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp.). Tests of normality (Kolmogorov-Smirnov, Shapiro-Wilk, QQ-plots) were applied and revealed a deviation from the normal distribution for the latency and amplitude parameters in the normal group as well as the patient group. Further analysis was performed using parametric and non-parametric tests, which delivered highly similar results. The reported \( p \)-values in the results section originate from non-parametric (Wilcoxon signed-rank) tests. For all analyses \( p < 0.05 \) was used as criterion of statistical significance.

After ruling out significant differences between the first and second run of the oVEMP measurements, all analyses were performed on the mean of run 1 and run 2. The Wilcoxon signed-rank test was used to detect possible differences between data obtained from the left and right side, and for making the comparison between both electrode positions on the data of the first test session. Subsequently, mean values and standard deviations were determined for all response parameters.

The test-retest reliability was evaluated using non-parametric tests and specific reliability index parameters. The reliability analysis was conducted on the data obtained for the left and right side separately as well as on the mean of both sides. Firstly, Wilcoxon signed-rank tests were carried out to reveal possible differences between the two test sessions. Secondly, the Intraclass Correlation Coefficient (ICC) was calculated. This coefficient demonstrates the amount of relative consistency and reproducibility between quantitative measurements from different test sessions. A maximum ICC value of 1.00 reflects perfect test-retest reliability.

The ICC values were interpreted following the categorization of Versino et al. (2001): a value above 0.75 implied excellent reliability, a value between 0.40 and 0.75 indicated fair-to-good reliability and a value below 0.40 indicated poor reliability. A 2-way random effect model
(absolute agreement) average measures ICC was applied, in which $p < 0.05$ was considered as criterion for statistical significance. Since only healthy subjects participated in the test-retest reliability evaluation, the between-subject variability is estimated to be low. This might cause the ICC value to be misleadingly low, resulting in an invalid ICC when the significance limit of $p = 0.05$ is exceeded. Therefore, the Method Error (ME) was determined, which is not affected by a lack of between-subject variability. The ME describes the test-retest reliability in terms of the percentage of variation between trials. The calculation uses the standard deviation of the difference scores ($SD_{diff}$) between test and retest session, using the following formula: $\text{ME} = \frac{SD_{diff}}{\sqrt{2}}$ (Portney, 2008). Because the ME must be interpreted relative to the mean values of the response parameters, conversion to a percentage is required, denominated as coefficient of variation of the method error ($\text{CV}_{\text{ME}}$): $\text{CV}_{\text{ME}} = \frac{2\text{ME}}{(X_1 + X_2)} \times 100$. This enables a valid comparison of the test-retest reliability between different response parameters, with lower $\text{CV}_{\text{ME}}$ values representing higher reliability. Finally, the Standard Error of Measurement (SEM) was calculated as third reliability parameter. This parameter is predominantly used for clinical purposes and provides a reference for evaluating test scores over time. It was determined as an indicator of absolute reliability, estimating the band of confidence around an individual’s raw score. The following formula was used for the calculation of the SEM: $\text{SEM} = \text{SD} \sqrt{1-\text{ICC}}$ (Weir, 2005), with SD representing the standard deviation of the response values between subjects. From this measurement, confidence intervals ensuring 95% accuracy were calculated as $\pm 1.96$ SEM. Based on the SEM, the Minimal Detectable Differences were calculated using the following formula: $\text{MD} = 1.96 \times \text{SEM} \times \sqrt{2}$ (Portney, 2008). The MDD describe the required amount of change in an outcome measure to represent a true test-retest difference, to enable a correct interpretation of these differences as improvement or worsening of the results.

3. RESULTS
3.1. Standard versus nose

The oVEMP response was present in all eighteen healthy subjects on both the left and the right side, resulting in a response rate of 100%. Statistical analysis (Wilcoxon) demonstrated a significant effect of the electrode position on the n10 ($p = 0.001$) and p15 ($p < 0.001$) latency: significantly shorter latencies were obtained in the nose reference position compared to the standard position (*Figure 32*). For the peak-to-peak amplitude, again a significant effect of the electrode position was revealed ($p < 0.001$): significantly higher amplitudes were obtained with the nose reference position relative to the standard position (*Figure 43*). The oVEMP responses of a single test person are displayed in *Figure 54*, where the obvious difference in amplitude between standard and nose reference position can be observed. Inter-ocular ratios were calculated for the amplitude. No significant difference between the mean IOR and 0 (one-sample t-test) could be found for both electrode positions. Consequently, the 95% PI for the IOR were calculated as [-1.96*SD, 1.96*SD], resulting in [-68, 68] for the standard position and [-32, 32] for the nose reference position. Thus, the nose reference position evoked the smallest confidence intervals for the IOR of the amplitude. This difference in dispersion of results is also clearly visible in the right boxplots of *Figure 43*.

*Table 2* shows an overview of the mean values and standard deviations of the n10 latency, p15 latency, peak-to-peak amplitude and IOR in the two different test conditions (standard and nose reference position). The values are displayed for each ear separately and additionally for the mean of both sides.

3.2. Test-retest reliability

All eighteen healthy volunteers were retested after one week and oVEMP responses could again be obtained for all subjects, resulting in a response rate of 100%. Examination of the difference scores (Wilcoxon signed-rank test) between the two test sessions could not
demonstrate any significant between-trial differences for both standard and nose reference position.

Table 3 shows the different reliability measures for the amplitude and IOR in both electrode positions, i.e. the mean differences between two test trials (Mean diff) with their standard deviation (SDdiff), the intraclass correlation coefficient (ICC) with the coefficient of variation of the method error (CVME), and the standard error of measurement (SEM) with the minimal detectable differences (MDD). The ICC values of the n10-p15 peak-to-peak amplitude were higher than 0.75 for both standard and nose reference position, indicating an excellent reliability. The highest value was obtained with the nose reference position (0.98 vs. 0.93). In addition to this, the lowest CVME value and thus the best reliability was observed when using the nose reference position (26.86 vs. 12.36). Significantly lower ICC values were found for the IOR, with the lowest value in the nose reference position (0.52 vs. 0.34). However, IOR CVME values are comparable between electrode positions (2.43 vs. 3.00). For both the amplitude and the IOR, the MDD values were lower with the nose reference position compared to the standard position (7.19 vs. 5.55 and 40.81 vs. 32.36), demonstrating a higher reliability of the nose reference position.

3.3. Patient group

The numerical results of the patient group are shown in Table 4 for the standard and nose reference electrode position. Figure 6 and Figure 7 provide a graphical visualization of the trends for the latency and amplitude parameters within the patient group. Since several patients presented with a unilaterally absent oVEMP response, statistical analysis (Wilcoxon signed-rank test) was performed on the right and left ear separately.

No significant differences were found for the n10 and p15 latencies between the standard and nose reference position for both ears. In contrast, the peak-to-peak amplitude was significantly larger when the nose reference position was used compared to the standard
position, for both the left ear ($p = 0.028$) and the right ear ($p = 0.028$). Furthermore, patient 6 showed bilaterally absent oVEMP responses when the standard position was applied, whereas the nose reference position could still evoke a detectable response on the right side. Likewise, patient 8 and 11 presented with unilaterally absent responses (on the right side) when the standard position was used, but bilaterally present oVEMPs were found with the nose reference position.

4. DISCUSSION

This study aimed to compare the widely implemented standard electrode configuration with the recently introduced nose reference position for oVEMP measurements, with the use of AC sound stimuli. In a second part, the test-retest reliability of both electrode placements was evaluated and the results were compared with other oVEMP studies (listed in Table 5). In a third part, both electrode placements were applied on a group of vestibular patients.

4.1. Standard versus nose reference

4.1.1. Amplitude

The majority of researchers are currently using the standard electrode position, where the active electrodes are placed on the inferior rectus (IR) muscle and the reference electrodes approximately 2 cm beneath the active electrodes (Rosengren et al., 2010). However, measurements of the single motor unit activity of human IR and IO muscles demonstrated that the oVEMP response mainly results from excitation of the IO muscle, whatever type of stimulation mode was used (Weber et al., 2012). Therefore, repositioning of the active electrodes more laterally on the IO muscle seemed favorable over the standard infra-orbital midline position. Sandhu et al. (2013) recommended a more medial placement of the reference electrodes to increase the distance between the active and reference electrode. This
was based on research of Piker et al. (2011), who found a significantly larger oVEMP amplitude when the reference electrode was shifted to the chin. Later on, Zuniga et al. (2014) observed the same trend in patients with superior canal dehiscence and normal controls. According to Piker et al. (2011), there is a considerable risk of obtaining equal electromyographic (EMG) responses in the active and reference electrode if they are placed in too close proximity of another, resulting in an absent oVEMP response (this approach is termed ‘reference contamination’). Consequently, the reference electrodes in the current study were placed on the IO muscle tendon (between the medial canthus of the eye and the nostril) for the measurements with the nose reference electrode position. Our results confirmed the findings of abovementioned authors: significantly larger amplitudes were obtained with this position relative to the standard position (Figures 4 & 5). More specifically, the mean n10-p15 peak-to-peak amplitude doubled in size with use of the nose reference position (from 11.9 (7.40 µV) to 22.7 (13.33 µV)). We hypothesize that this results from the combined effect of a more selective representation of IO activity in the active electrode, and less reference contamination due to a larger inter-electrode distance. This can be explained by recent studies of Sandhu et al. (2013), Vanspauwen et al. (2016) and Govender et al. (2016). Sandhu et al. (2013) investigated the effect of shifting the active and reference electrodes to several locations relative to the standard position. When the reference was kept in the standard position, the amplitude was largest when the active was shifted towards the lateral canthus, supposedly closer to the IO muscle belly. However, the replacement did not lead to a statistically significant difference with the standard position. When also the reference electrode was shifted towards the medial canthus (IO tendon) to obtain a larger distance between active and reference, the amplitude increased further and differed significantly from the amplitude measured with the standard configuration. Vanspauwen et al. (2016) also compared different electrode positions, but with use of bone-conduction stimuli delivered by a
minishaker at Fz position. Moving the reference electrode to the sternum (with the standard position as ‘reference position’) resulted in a 72% increase of the oVEMP amplitude. Application of the nose reference position led to a further increase of the amplitude (121% increase relative to the standard position), but the difference with the sternum configuration was not statistically significant. Both studies indicate a greater impact of the inter-electrode distance on the oVEMP amplitude than the more lateral positioning of the active electrode. However, more recent observations of Govender et al. (2016) show that the latter should not be underestimated. The authors compared 5 active electrode locations around the eye, including the standard position (M) and two more lateral ones (ML and L). The largest response amplitudes were observed using the ML electrode, which corresponds with the active electrode as placed in the nose (reference) position. Moreover, the ML position showed a more linear response to gaze angle than the standard (M) position, suggesting a higher sensitivity of the this mediolateral location for detecting IO muscle activity.

In contrast with the cVEMP responses, an oVEMP response is generally small and therefore more difficult to detect. This increases the risk of false negative responses and possible misdiagnosis. The nose reference position elicited remarkably larger oVEMP responses than the standard position, which might help the clinician to detect an oVEMP response more readily and take doubts away. However, an important note must be made here regarding reflex purity. As already stated by Sandhu et al. (2013), shifting the active and reference electrodes to the nose reference configuration may lead to a closer placement to other extra-ocular muscle bellies (such as the medial and lateral rectus), which may also be activated by the stimulus (Govender et al., 2011). This can be a plausible explanation for the doubling of the standard deviation (from 7.40 to 13.33 µV) when using this position, and must be considered as a possible disadvantage of this electrode montage.

4.1.2. Latency
The electrode position also had a significant effect on the latency parameters: shorter n10 and p15 latencies were obtained with the nose reference position relative to the standard position (Figure 32). In contrast, Sandhu et al. (2013) and Vanspauwen et al. (2016) found no significant difference for the n10 and p15 latency between standard and nose reference position. Our latency values measured with the standard position were slightly closer to 10 and 15 ms, values favored in literature. However, the latency difference stays within 1-2 ms (Table 2), which may be statistically significant but is in our opinion clinically irrelevant since there is still no risk of obfuscation of the n10 peak by a stimulus artefact.

4.1.3. IOR

The IOR (%) was calculated to examine possible left-right differences in amplitude. The mean IOR did not significantly differ from zero, indicating that the left-right differences are negligible. The 95% prediction intervals were significantly smaller with the nose reference position [-32, 32] than the standard position [-68, 68]. This is clinically of great importance, since abnormalities will be detected sooner with the nose reference position: a left-right difference will be considered abnormal from an IOR of 32%, whereas with the standard position a difference of 68% is needed to make the same conclusion. Moreover, the results are comparable to the values obtained in our vestibular lab for the unilateral weakness (UW) of the caloric test: [-17.5, 17.5] % (Maes et al., 2011) and for the IOR of the cVEMP: [-29.9, 29.9] % and [-34.5, 34.5] % (Vanspauwen, 2009, Maes et al., 2011).

Although the smaller IOR values that were obtained with the nose reference configuration might be clinically beneficial, it must be noted that crosstalk from the other eye might have contributed to this significant IOR difference. Since the active electrode is moved laterally in this configuration, it might capture activity of the lateral rectus muscle. Govender et al. (2011) revealed that the pathways underlying the responses of the lateral rectus muscle have a more bilateral nature, possibly resulting in a reduction of the left-right differences.
4.2. Comparison with recent literature

Table 5 presents an overview of recent oVEMP studies for comparison with our numerical results. We only included studies where the stimulus intensity and type of transducer was comparable with our settings. Latency values of the current study are comparable with the latencies found in literature, ranging from 9.4 to 11.1 ms for N1 and from 14.2 to 17.6 ms for P1, and this for both standard and nose reference position. Furthermore, our (n10-p15 peak-to-peak) amplitudes measured with the standard position are slightly higher than the studies where AC sound was used (ranging from 6.5 to 8.2 µV). Only two of the listed studies applied the nose reference position. Sandhu et al. (2013) measured the amplitude from baseline to the n10 peak, making comparison with our values impossible. However, we are able to compare the relative increase in amplitude, which is in line with our findings: there was a twofold increase of the mean amplitude with the nose reference versus standard position. The same can be concluded about the study of Vanspauwen et al. (2016) where bone-conducted stimuli were used, which are known to evoke even larger oVEMP amplitudes (Cheng et al., 2009, Weber et al., 2015).

4.3. Test-retest reliability

Paired analysis did not show significant differences between the first and second test session for any response parameter and for both electrode configurations. Furthermore, we calculated different categories of reliability measures. The intraclass correlation coefficient (ICC) showed an excellent reliability (ICC > 0.75) for the amplitude in both electrode positions, but with a slightly better result for the nose reference position. In contrast, the ICC values for the IOR were generally low (ranging from poor to fair-to-good reliability). This finding can possibly be attributed to a lack of between-subject variability, since we only tested normal
individuals. As explained earlier, this can lead to misleadingly low ICC values. To avoid misinterpretation, we also calculated the coefficient of variation of the method error (CV_{ME}), which is not influenced by a lack of between-subject variability. Based on these calculations, the reliability of the IOR is very good in both positions (CV_{ME} close to zero). The CV_{ME} values for the amplitude indicate a better reliability of the nose reference position. Likewise, the MDD values for amplitude and IOR favor the nose reference position.

Two other studies investigated the test-retest reliability of the oVEMP responses, elicited by air-conducted 500 Hz tone bursts and using the standard electrode configuration. Nguyen et al. (2010) also found an excellent reliability for the amplitude (ICC = 0.79) and a fair-to-good reliability (ICC = 0.50) for the IOR. Slightly higher values were found in the study of Piker et al. (2011) for the amplitude (ICC = 0.87) and IOR (ICC = 0.61). The same argumentation as above can be applied for the fair-to-good reliability findings in both studies. Unfortunately the CV_{ME} was not calculated in their study, making a valid comparison with our values impossible.

4.4. Patient group

Although a variety of vestibular deficits was observed in the patient group, statistical analysis of the results was useful to verify the trends visible in Figure 65 and Figure 76. Ideally, a large group of patients with isolated utricular damage should be tested, but this is clinically rather rare.

For the p15 and n10 latencies, no significant differences were seen between the standard and nose reference configuration (Figure 65), as opposed to the significant differences found in the healthy subjects. This might be attributed to the age difference between both groups: the mean age was significantly higher and the age range was wider in the patient group.

In contrast, the amplitude difference (Figure 76) was in line with the findings in the normal group: the nose reference configuration evoked significantly larger amplitudes than the
standard configuration. Moreover, in three patients the responses were considered absent with the standard position, whereas the nose reference position could still evoke a detectable oVEMP. Thus, these patients would be misdiagnosed with ‘no residual utricular function’ when the standard position was applied. Consequently, the nose reference position seems to be more sensitive in recording oVEMP responses. Whether the placement is also more sensitive in detecting utricular and superior nerve deficits, remains to be elucidated. Obviously, oVEMP results should be interpreted together with the outcome from the standard vestibular assessment and the subjective complaints of the patient to form a plausible diagnosis, but oVEMP results could give valuable complementary information on the function of the vestibular system. At this stage, only the unilateral centrifugation test (Buytaert et al., 2010, Schonfeld et al., 2011) is able to objectively evaluate utricular function, but the required equipment and experience is not eligible for most clinical centers. The oVEMP test with the nose reference position is therefore promising for clinical use, but more patient verification is required.

4.5. Shortcomings and future research

A total of 18 healthy subjects were tested in this study, which is insufficient to collect representative normative data for the nose reference position. Subjects of different age groups should be tested and the sample size should be larger. Furthermore, the gender diversity of the study group was not ideal (18 females). In a study of Sung et al. (2011), the oVEMP responses of 10 males and 10 females were compared for various stimulation modes. They observed significantly larger amplitudes in males compared to females, regardless of the stimulation mode. This may be attributed to variance in the muscle bulk between males and females. Based on these findings, it is likely that the amplitude values will be higher than in the current study. However, a more recent study of Versino et al. (2015) did not reveal a gender effect on the oVEMP responses. Another drawback of the present study was our
inability to make a valid comparison between the normal group and the patient group. Therefore, we want to stress the importance of age- and gender-matched controls in future research.

Despite of the abovementioned shortcomings, the advantages of the investigated ‘nose reference’ electrode placement are promising for the future.

5. CONCLUSION

This study demonstrated that the nose reference position generally evokes larger oVEMP amplitudes and subsequently leads to an increased sensitivity. In addition, the nose reference position shows a better reliability than the widely used standard position. These results were obtained with AC sound presented through insert earphones, which are readily available in most clinical centers. Consequently, it may be a promising placement for implementation in clinical practice after further verification in a larger patient population.

CONFLICT OF INTEREST

None.

ACKNOWLEDGMENTS

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REFERENCES


Vanspauwen R, Wuyts FL, Krijger S, Maes L. Comparison of different electrode configurations for the oVEMP with bone-conducted vibration. Ear Hear. 2016 [accepted for publication].


FIGURE LEGENDS

Figure 1a: Standard electrode configuration. Red circle = recording area of active electrode; green circle = recording area of reference electrode; blue circle = recording area of ground electrode.

Figure 2b: Nose reference electrode configuration. Red circle = recording area of active electrode; green circle = recording area of reference electrode; blue circle = recording area of ground electrode.

Figure 3: Boxplots of the latency of n10 and p15 (ms) in the standard and nose reference electrode position.

Figure 4: Boxplots of the peak-to-peak amplitude (µV) and the Inter-ocular Ratio (IOR) of the amplitude (%) in the standard and nose reference electrode position. The outlier above the IOR boxplot for the nose reference position may be explained by the tympanometric results of
this subject. Although a normal type A tympanogram (according to the Liden-Jerger
classification) was observed on both sides, the compliance was considerably lower in the left
ear (0.5 cc versus 1.1 cc). This may have resulted in a less efficient transmission of the
oVEMP stimulus to the utricle, ultimately leading to smaller oVEMP amplitudes on this side.

**Figure 5A**: oVEMP responses of a healthy subject with the standard (upper trace) and nose
reference electrode position (lower trace).

**Figure 6A**: Line plots of the latency of n10 and p15 (ms) in the standard and nose reference
electrode position. The numbers correspond with the patients as presented in Table 1. R =
right ear, L = left ear.

**Figure 7A**: Line plots of the peak-to-peak amplitude (µV) in the standard and nose reference
electrode position. The numbers correspond with the patients as presented in Table 1. R =
right ear, L = left ear.
Table 1: Patient information: sex, age in years, tympanogram according to the Jerger classification (Jerger 1970, cited by Srirreddy et al. 2002), cVEMP, caloric test and suspected diagnosis.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Sex</th>
<th>Age (y)</th>
<th>Tympanogram</th>
<th>cVEMP</th>
<th>Caloric test</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>46</td>
<td>Type A (L,R)</td>
<td>Normal (L,R)</td>
<td>Normal (L,R)</td>
<td>Utricular dysfunction (R)</td>
</tr>
<tr>
<td>2</td>
<td>F</td>
<td>53</td>
<td>Type A (L,R)</td>
<td>Normal (R), absent (L)</td>
<td>Normal (L,R)</td>
<td>Unknown</td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>49</td>
<td>Type A (L,R)</td>
<td>Normal (R), decreased (L)</td>
<td>Normal (R), hypofunction (L)</td>
<td>Peripheral polyneuropathy (due to head trauma) (L)</td>
</tr>
<tr>
<td>4</td>
<td>M</td>
<td>49</td>
<td>Type A (L,R)</td>
<td>Normal (R), absent (L)</td>
<td>Normal (L,R)</td>
<td>Auto-immune inner ear disease</td>
</tr>
<tr>
<td>5</td>
<td>F</td>
<td>66</td>
<td>Type C (R), type A (L)</td>
<td>Normal (R), absent (L)</td>
<td>Normal (R), hypofunction (L)</td>
<td>Vestibular neuritis (L)</td>
</tr>
<tr>
<td>6</td>
<td>M</td>
<td>59</td>
<td>Type A (L,R)</td>
<td>Normal (R), absent (L)</td>
<td>Normal (R), areflexia (L)</td>
<td>Vestibular schwannoma (L)</td>
</tr>
<tr>
<td>7</td>
<td>M</td>
<td>54</td>
<td>Type A (L,R)</td>
<td>Increased (R), normal (L)</td>
<td>Normal (R), hypofunction (L)</td>
<td>Vestibular neuritis (L), superior canal dehiscence (R)</td>
</tr>
<tr>
<td>8</td>
<td>F</td>
<td>74</td>
<td>Type A (L,R)</td>
<td>Normal (L,R)</td>
<td>Normal (L,R)</td>
<td>History of BPPV (L/R)</td>
</tr>
<tr>
<td>9</td>
<td>F</td>
<td>41</td>
<td>Type A (L,R)</td>
<td>Normal (L,R)</td>
<td>Normal (L,R)</td>
<td>Vestibular migraine</td>
</tr>
<tr>
<td>10</td>
<td>M</td>
<td>52</td>
<td>Type A (L,R)</td>
<td>Normal (L), absent (R)</td>
<td>Normal (L), areflexia (R)</td>
<td>Bilateral Meniere’s Disease, labyrinthectomy (R)</td>
</tr>
<tr>
<td>11</td>
<td>M</td>
<td>41</td>
<td>Type A (L,R)</td>
<td>Normal (L,R)</td>
<td>Normal (L,R)</td>
<td>Utricular dysfunction (R)</td>
</tr>
</tbody>
</table>

L = left ear; R = right ear; F = female; M = male
Table 2: Mean values and standard deviation (SD) of the n10 latency (ms), p15 latency (ms), peak-to-peak amplitude (µV) and inter-ocular ratio (IOR, %) in the two different electrode positions.

<table>
<thead>
<tr>
<th>Electrode position</th>
<th>Ear</th>
<th>Parameter</th>
<th>Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Left</td>
<td>Latency n10</td>
<td>9.67 (0.52)</td>
</tr>
<tr>
<td></td>
<td>Right</td>
<td>Latency n10</td>
<td>9.57 (0.47)</td>
</tr>
<tr>
<td></td>
<td>Mean (L+R)</td>
<td>Latency n10</td>
<td>9.62 (0.45)</td>
</tr>
<tr>
<td></td>
<td>IOR</td>
<td>Amplitude</td>
<td>-20.07 (28.33)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Electrode position</th>
<th>Ear</th>
<th>Parameter</th>
<th>Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Left</td>
<td>Latency n10</td>
<td>9.47 (0.54)</td>
</tr>
<tr>
<td></td>
<td>Right</td>
<td>Latency n10</td>
<td>9.38 (0.33)</td>
</tr>
<tr>
<td></td>
<td>Mean (L+R)</td>
<td>Latency n10</td>
<td>9.42 (0.39)</td>
</tr>
<tr>
<td></td>
<td>IOR</td>
<td>Amplitude</td>
<td>-6.39 (15.17)</td>
</tr>
</tbody>
</table>
Table 3: Values of reliability parameters for amplitude and IOR in standard and nose reference position.

<table>
<thead>
<tr>
<th>Reliability parameters</th>
<th>Standard position</th>
<th>Nose reference position</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Amplitude</td>
<td>IOR</td>
</tr>
<tr>
<td>Mean diff</td>
<td>-1.11</td>
<td>-6.92</td>
</tr>
<tr>
<td>SDdiff</td>
<td>4.73</td>
<td>24.08</td>
</tr>
<tr>
<td>ICC</td>
<td>0.93</td>
<td>0.52</td>
</tr>
<tr>
<td>CVME</td>
<td>26.86</td>
<td>2.43</td>
</tr>
<tr>
<td>SEM</td>
<td>2.59</td>
<td>14.72</td>
</tr>
<tr>
<td>MDD</td>
<td>7.19</td>
<td>40.81</td>
</tr>
</tbody>
</table>

Mean diff = mean differences; SDdiff = standard deviation of mean difference; ICC = intraclass correlation coefficient; CVME = coefficient of variation of the method error; SEM = standard error of the measurement; MDD = minimal detectable differences.
Table 4: Patient data for the n10 latency (ms), p15 latency (ms), peak-to-peak amplitude (µV) and IOR of the amplitude (%) in the standard and nose reference electrode position.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Electrode position</th>
<th>Lat n10_L (ms)</th>
<th>Lat n10_R (ms)</th>
<th>Lat p15_L (ms)</th>
<th>Lat p15_R (ms)</th>
<th>Ampl_L (µV)</th>
<th>Ampl_R (µV)</th>
<th>IOR Ampl (%)</th>
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<tbody>
<tr>
<td>1</td>
<td>S</td>
<td>10.03</td>
<td>NR</td>
<td>13.15</td>
<td>NR</td>
<td>11.98</td>
<td>NR</td>
<td>100</td>
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<tr>
<td></td>
<td>N</td>
<td>10.03</td>
<td>NR</td>
<td>13.36</td>
<td>NR</td>
<td>18.19</td>
<td>NR</td>
<td>100</td>
</tr>
<tr>
<td>2</td>
<td>S</td>
<td>NR</td>
<td>10.24</td>
<td>NR</td>
<td>16.90</td>
<td>NR</td>
<td>8.77</td>
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<tr>
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<td>N</td>
<td>NR</td>
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<td>NR</td>
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<td>NR</td>
<td>10.11</td>
<td>100</td>
</tr>
<tr>
<td>3</td>
<td>S</td>
<td>11.28</td>
<td>10.65</td>
<td>17.31</td>
<td>17.73</td>
<td>5.04</td>
<td>4.00</td>
<td>-11.57</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>11.69</td>
<td>11.07</td>
<td>17.53</td>
<td>18.35</td>
<td>6.09</td>
<td>6.41</td>
<td>2.56</td>
</tr>
<tr>
<td>4</td>
<td>S</td>
<td>NR</td>
<td>11.69</td>
<td>NR</td>
<td>18.35</td>
<td>NR</td>
<td>3.98</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>NR</td>
<td>11.28</td>
<td>NR</td>
<td>18.35</td>
<td>NR</td>
<td>4.19</td>
<td>100</td>
</tr>
<tr>
<td>5</td>
<td>S</td>
<td>NR</td>
<td>10.03</td>
<td>NR</td>
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<td>NR</td>
<td>5.94</td>
<td>100</td>
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<tr>
<td></td>
<td>N</td>
<td>NR</td>
<td>9.61</td>
<td>NR</td>
<td>15.86</td>
<td>NR</td>
<td>8.04</td>
<td>100</td>
</tr>
<tr>
<td>6</td>
<td>S</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>NR</td>
<td>10.65</td>
<td>NR</td>
<td>14.82</td>
<td>NR</td>
<td>4.49</td>
<td>100</td>
</tr>
<tr>
<td>7</td>
<td>S</td>
<td>NR</td>
<td>10.65</td>
<td>NR</td>
<td>18.15</td>
<td>NR</td>
<td>51.98</td>
<td>100</td>
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<tr>
<td></td>
<td>N</td>
<td>NR</td>
<td>10.24</td>
<td>NR</td>
<td>17.73</td>
<td>NR</td>
<td>74.68</td>
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</tr>
<tr>
<td>8</td>
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<td>9.82</td>
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<td>NR</td>
<td>100</td>
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<td>10.24</td>
<td>14.82</td>
<td>14.19</td>
<td>5.74</td>
<td>4.27</td>
<td>-14.74</td>
</tr>
<tr>
<td>9</td>
<td>S</td>
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<td>3.86</td>
<td>5.39</td>
<td>16.50</td>
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<td>NR</td>
<td>3.07</td>
<td>NR</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>11.69</td>
<td>NR</td>
<td>15.86</td>
<td>NR</td>
<td>5.64</td>
<td>NR</td>
<td>100</td>
</tr>
<tr>
<td>11</td>
<td>S</td>
<td>9.6</td>
<td>NR</td>
<td>12.9</td>
<td>NR</td>
<td>18.8</td>
<td>NR</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>10.8</td>
<td>9.2</td>
<td>13.2</td>
<td>12.5</td>
<td>23.9</td>
<td>5.3</td>
<td>63.70</td>
</tr>
</tbody>
</table>

S = standard electrode placement; N = nose reference electrode placement; Lat n10_L = n10 latency of the left side; Lat n10_R = n10 latency of the right side; Lat p15_L = p15 latency of the left side; Lat p15_R = p15 latency of the right side; Ampl_L = peak-to-peak amplitude of the left side; Ampl_R = peak-to-peak amplitude of the right side; IOR Ampl = Interocular Ratio of the amplitude; NR = no response
Table 5: Comparative studies for oVEMPs (evoked by a 500 Hz tone burst stimulus).

<table>
<thead>
<tr>
<th>Study</th>
<th>AC/BC</th>
<th>Ears (n)</th>
<th>Electrode configuration</th>
<th>Response rate (%)</th>
<th>Age (years)</th>
<th>Latency N1 (ms)</th>
<th>Latency P1 (ms)</th>
<th>N1 Amplitude (µV)</th>
<th>N1-P1 Amplitude (µV)</th>
<th>IOR (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Current study</td>
<td>AC</td>
<td>38</td>
<td>Standard</td>
<td>100</td>
<td>21 (20-25)</td>
<td>9.6 (0.5)</td>
<td>15.1 (1.0)</td>
<td>11.9 (7.4)</td>
<td>20.1 (28.3)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Nose reference ('belly-tendon')</td>
<td></td>
<td>100</td>
<td>9.4 (0.4)</td>
<td>14.7 (1.0)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Wang et al. (2009)</td>
<td>AC</td>
<td>40</td>
<td>Standard</td>
<td>95</td>
<td>28 (22-33)</td>
<td>11.1 (0.7)</td>
<td>15.9 (1.0)</td>
<td>6.5 (2.9)</td>
<td></td>
<td>22.7 (13.3)</td>
</tr>
<tr>
<td>Nguyen et al. (2010)</td>
<td>AC</td>
<td>106</td>
<td>Standard</td>
<td>35</td>
<td>20-70</td>
<td>10.3 (0.5)</td>
<td>15.5 (0.8)</td>
<td>4.2 (2.4)</td>
<td></td>
<td>8.2 (6.3)</td>
</tr>
<tr>
<td>Piker et al. (2011)</td>
<td>AC</td>
<td>58</td>
<td>Standard</td>
<td>100</td>
<td>34 (18-49)</td>
<td>12.5 (0.9)</td>
<td>17.6 (1.1)</td>
<td>5.1 (3.1)</td>
<td></td>
<td>13.0 (10.0)</td>
</tr>
<tr>
<td>Sandhu et al. (2013)</td>
<td>AC</td>
<td>16</td>
<td>Standard</td>
<td>100</td>
<td>26 (22-36)</td>
<td>10.7 (0.9)</td>
<td>3.1 (2.1)</td>
<td>15.8 (6.3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Nose reference ('nose')</td>
<td></td>
<td></td>
<td></td>
<td>5.7 (3.4)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vanspauwen et al. (2016)</td>
<td>BC</td>
<td>28</td>
<td>Standard</td>
<td>100</td>
<td>23 (2.6)</td>
<td>10.7 (0.7)</td>
<td>14.9 (0.7)</td>
<td>15.8 (6.3)</td>
<td></td>
<td>27.1 (12.2)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Sternum</td>
<td>100</td>
<td></td>
<td>10.3 (0.6)</td>
<td>15.4 (1.1)</td>
<td>27.1 (12.2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Nose reference ('nose')</td>
<td>100</td>
<td></td>
<td>10.3 (0.4)</td>
<td>14.2 (0.5)</td>
<td>35.2 (19.1)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

AC = air conduction; BC = bone conduction
‘Standard’ versus ‘nose reference’ electrode placement for measuring oVEMPs with air-conducted sound: test-retest reliability and preliminary patient results

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Latency of n10 and p15: standard vs. nose