

**TEST RETEST RELIABILITY OF MASSETER VEMP BETWEEN TWO
ELECTRODE MONTAGES**

ANKITA

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**A Dissertation Submitted in Part Fulfillment of the Degree of Master of Science
(AUDIOLOGY)**

University of Mysore



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JUNE, 2022

CERTIFICATE

This is to certify that this dissertation entitled "**Test Retest Reliability of Masseter VEMP between two electrode Montages**" is a bonafide work submitted as part of fulfilment of the degree of Master of Science (Audiology) of the student with Registration Number **P01II24S023006**. This has been carried out under the guidance of a faculty of this institute and has not been submitted earlier to any other university for the award of any other Diploma or Degree.

8th Mysuru
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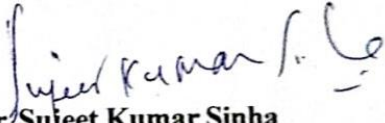

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CERTIFICATE

This is to certify that this dissertation entitled " **Test Retest Reliability of Masseter VEMP between two electrode Montages**" has been prepared under my supervision and guidance. It is also being certified that this dissertation has not been submitted earlier to any other university for the award of any other Diploma or Degree.

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DECLARATION

This is to certify that this dissertation entitled " **Test Retest Reliability of Masseter VEMP between two electrode Montages**" is the result of my own study under the guidance of Dr. Sujeet Kumar Sinha Associate Professor of Audiology, All India Institute of Speech and Hearing, Mysuru and has not been submitted earlier to any other university for the award of any other Diploma or Degree.

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ABSTRACT

Aim of the study : The aim of the study is to compare mVEMP response characteristics across two different electrode montages- zygomatic and Forehead and to evaluate their test retest reliability.

Method: A total of 30 healthy individuals were taken in the study (age range : 18-40 years). All the participants underwent mVEMP recording for both the montages. The same cohort group was retested once again after a gap of one week to assess the test retest reliability.

Result : All the participants had shown of mVEMP response for both the montages in both sets of data. In both sets, P1 and N1 latencies were prolonged for the forehead montage as compared to zygomatic montage. In the first set of data, no significant difference was found in P1-N1 rectified amplitude measure between the two montages. In the second set of data, higher P1-N1 amplitude measures were found for Forehead montage. Test retest reliability analysis good reliability for P1 latency and P1-N1 amplitude measures, moderate reliability for N1 latency for both the montages.

Conclusion: The zygomatic montage produced shorter P1 and N1 latencies, and higher amplitude measures were found for forehead montage but difference in amplitude measures were only found for one set of data. Both Zygomatic and forehead montage showed good-moderate test retest reliability , suggesting that either of the montage can be used , but the same montage should be used across repeated assessments.

CHAPTER -I

INTRODUCTION

The vestibular evoked myogenic potential has become a clinically promising neurophysiological tool used to assess the function of the vestibular system, specifically the otolith organs and vestibular nerve pathways. VEMPs are obtained from the saccule and utricle and their respective sacculo-ocular and otolith-ocular reflex pathways. VEMP is a short-latency myogenic response that can be evoked by various stimuli, such as loud air-conduction sounds (Colebatch et al., 1994), bone-conduction vibrations applied to the skull (Todd et al., 2007), and galvanic stimulation (Watson et al., 1998). The vestibular system is typically unresponsive to acoustic stimulation; however, it can be stimulated by high-intensity acoustic sound.

Originally, VEMP was recorded from the sternocleidomastoid muscle, known as the Cervical vestibular evoked myogenic potential (cVEMP) (Colebatch & Halmagyi, 1992; Colebatch et al., 1994). cVEMP is a short-latency inhibitory electromyographic response that is recorded from the tonically activated sternocleidomastoid muscle in response to loud acoustic stimulation. The saccule is stimulated by loud acoustic stimulation. From the saccule, information is carried along the inferior vestibular branch of the vestibulocochlear nerve, then relayed to the vestibular nucleus, and finally descends through the vestibular spinal tract to the sternocleidomastoid muscle. Thus, cVEMP basically assesses the functional integrity of the saccule, the inferior vestibular nerve or the sacculo-ocular reflex.

Later, another myogenic response potential, the ocular vestibular evoked myogenic potential (oVEMP), was recorded by placing the electrode over the inferior oblique muscle (Rosengren et al., 2005). It assesses the functional integrity of the utricle and the superior vestibular nerve by checking the integrity of the utriculo-ocular reflex. oVEMP is an excitatory contralateral electromyogenic potential. cVEMP and oVEMP have been widely used to assess and evaluate the integrity of sacculo-collic and utriculo-ocular pathways, respectively.

In recent years, many researchers have successfully recorded vestibular-evoked myogenic potentials from the masseter muscle, which have been referred to as masseter vestibular-evoked myogenic potentials (Deriu et al., 2005). The masseter vestibular evoked myogenic potential test evaluates the vestibulomasseteric reflex neural pathway between the sound-stimulated hair cells and the trigeminal nerve motor neurons. Masseter vestibular evoked myogenic potentials are short-latency bilateral inhibitory electromyograms elicited by galvanic or acoustic stimulation (Deriu et al., 2003).

Masseter vestibular-evoked myogenic potentials are vestibular-dependent inhibitory reflexes (Thirusangu & Sinha, 2023). It is a bilaterally generated, electromyographically (EMG) mediated response innervated by the Trigeminal nerve. It was first recorded using transmastoid electrical stimulation from tonically activated masseter muscles in healthy humans (Deriu et al., 2003), followed by acoustic stimulation (Deriu et al., 2005). Although masseter vestibular-evoked myogenic potential recording was initiated a decade ago, there has recently been renewed interest in using this, especially in patients with brainstem dysfunction (Thirusangu & Sinha, 2023).

The vestibulomasseteric reflex pathway originates from the saccule located in the inner ear. When the sensory hair cells of the saccule macula get stimulated, the information is passed on to Scarpa's ganglion, which is then carried along the inferior vestibular branch of the vestibulocochlear nerve to the ipsilateral vestibular nucleus in the brainstem. From there, the nerve undergoes a decussation and ends in the ipsilateral and contralateral trigeminal motor nuclei via the vestibulo-trigeminal monosynaptic pathway (VTMP). From the Trigeminal nucleus, signals are sent to the masseter muscles via the mandibular branch of the trigeminal motor nerve. It is said that the lower third of the masseter muscle receives comparatively heavy innervation; therefore, electrodes placed on this site will generate responses of higher amplitude

(Giacconi et al., 2006). Various animal studies have confirmed the existence of the vestibulomasseteric reflex pathway (Tolu & Pugliatti, 1993; Giacconi et al., 2006).

Initially, various stimuli, such as clicks, chirps, and tone bursts, were used to elicit mVEMP (De Natale et al., 2019; Romero et al., 2022; Sangu Srinivasan et al., 2023; Neupane, et al. (2023). Vignesh et al. (2021) compared VEMP responses elicited by tone bursts with those elicited by clicks in a study by de Natale et al. (2015b). Tone burst stimuli produced more robust p11/n21 peaks than click stimuli; the latencies of the peaks were more prolonged, and the amplitude was larger when tone burst was used to elicit mVEMP.

mVEMP has shown potential clinical relevance in neurological conditions such as Parkinsonism, multiple sclerosis and idiopathic REM sleep disorders (De Natale et al., 2015a; Magnano et al., 2014; De Natale et al., 2018). It was reported that the rate of abnormality was higher in mVEMP (66.7%) than in cVEMP (41.7%) and oVEMP (45.8%), thus providing evidence of the efficiency and sensitivity of masseteric VEMP in identifying brainstem pathological conditions such as Parkinson's disease (De Natale et al., 2015a). An increase in the abnormality of mVEMP was observed with increasing severity of Parkinson's disease (De Natale et al., 2015b). The abnormality rate is higher for mVEMP than for cVEMP and oVEMP in both the early and advanced stages of Parkinson's disease (De Natale et al., 2015b). In multiple sclerosis, greater abnormality was observed in mVEMP than in cVEMP and oVEMP (Sangu Srinivasan et al., 2023). The frequency of abnormal masseter vestibular-evoked myogenic potential has been reported to be 62.1 percent in patients with multiple sclerosis (Magnano et al., 2014). In case of idiopathic REM sleep disorders, the rate of abnormality of cVEMP was 45% for cVEMP, 50% for oVEMP, and 65% for masseter VEMP (De Natale et al., 2018). It is also evident from various studies that mVEMP is superior to cVEMP and oVEMP in identifying neural lesions in numerous Pathologies.

Three distinct electrode montages can be used to record mVEMP. The first montage (Mandibular montage) involves placing the reference/inverting electrode on the mandibular angle, termed as mandibular reference. The active /non-inverting electrode is placed on the lower third of the masseter muscle, and the ground electrode is placed on the lower forehead. In the second method (Zygomatic montage), the inverting/reference electrode is placed on the medial point of the zygomatic arch. Non-inverting on the lower third of the masseter muscle, ground on the lower forehead (Deriu et al., 2005). The third type of montage included reference electrodes at Fz; the active electrode was placed on the lower third of the masseter, and the ground electrode was placed at Fpz.

NEED FOR THE STUDY

mVEMP has become a promising neurophysiological tool to assess the vestibulo-masseteric reflex pathway. Few published studies have explored the clinical application of mVEMP in a few neural disorders. However, the wide clinical application of mVEMP warrants thorough exploration. As it has become a promising tool, there should be a standardised, best-practice protocol for recording mVEMP. Various studies have used different electrode montages; the influence of electrode montage on response parameters has not yet been investigated. Most existing studies have compared only two montages, predominantly the zygomatic and mandibular configurations, or zygomatic versus forehead montages, with findings suggesting differences in amplitude, response rate, and test–retest reliability (De Natale et al., 2019; Loi et al., 2020). Few studies have evaluated the effects of electrode montages on mVEMP responses. Very few studies have compared the effects of forehead and zygomatic electrode montages on mVEMP responses. However, there is a lack of systematic investigations that have simultaneously evaluated commonly employed montages (mandibular, zygomatic, and forehead/Fz-based). Furthermore, while some studies report adequate reproducibility of mVEMP responses within a given montage, comprehensive test–retest

reliability analyses across different montages are limited. Establishing such reliability is essential to ensure that the technique can be applied consistently in clinical and research contexts, particularly for diagnostic purposes in conditions affecting the vestibulo-trigeminal Pathway.

Therefore, there is a clear need to investigate comparative response characteristics and reliability across different electrode montages to identify the most robust and clinically applicable configuration. Addressing this gap will not only enhance the standardisation of mVEMP protocols but also improve their diagnostic utility in both otological and neurological disorders.

Aim of the study

The study aims to compare mVEMP response characteristics across two different electrode montages and to evaluate their test–retest reliability.

Objectives of the study

- To compare the response rates of 500Hz tone burst-evoked mVEMP recorded using two different electrode montages (zygomatic, and forehead/Fz).
- To compare the individual peak latencies (p11 and n21) of mVEMP recorded using the two electrode montages.
- To compare the peak-to-peak amplitude of mVEMP recorded between two electrode montages.
- To compare the inter-aural amplitude ratio between the two electrode montages for the 500Hz tone burst-evoked mVEMP.
- To compare the test–retest reliability of mVEMP responses between the two electrode Montages.

CHAPTER II

REVIEW OF LITERATURE

Vestibular evoked myogenic potential, a promising neurophysiological tool which is used to assess the functional integrity of the otolith organs and its connecting vestibular nerve. VEMPs are obtained from the otolith organs, saccule and utricle and their respective pathways. It is a short-latency electromyographic potential that can be recorded using air-conduction sounds (Colebatch et al., 1994), bone-conduction sounds applied to the skull (Todd et al., 2007), and galvanic stimulation (Watson et al., 1998).

The vestibular system is typically insensitive to acoustic stimuli. However, the vestibular response can be recorded using a high-intensity acoustic stimulus. Bickford et al.(1964) identified a response recorded at theinion in response to loud clicks, which was obtained only during the contraction of the sternocleidomastoid muscle. Various researchers have obtained the vestibular evoked myogenic responses from various muscles such as neck extensor, masseter muscle, soleus muscle, gastrocnemius muscle, triceps muscle and inferior oblique muscle (Wu et al., 1999; Deriu et al.,2007; Todd et al., 2007; Rudisill & Hain, 2008; Cherchi et al.,2009; Cunha et al., 2014).

2.1 Masseter Vestibular Evoked Myogenic Potentials

The masseter vestibular evoked myogenic potential (mVEMP) assesses the integrity of the vestibulotrigeminal pathway (Deriu et al., 2005). Previous studies have demonstrated that the tonically contracted masseter muscle produces bilateral short-latency inhibitory EMG responses to galvanic and acoustic stimulation in healthy individuals (Meier-Ewert et al., 1974; Deriu et al., 2003, 2005).

These short-latency inhibitory EMG responses are known as the masseter vestibular evoked myogenic potential. The vestibulo-masseteric reflex pathway helps stabilise the jaw during head movement. The innervation of trigeminal motor neurons is also influenced by

vestibular, auditory, visual and somatosensory inputs (Deriu et al., 2003). Deriu et al (2003) did a study in which they obtained short-latency myogenic responses from the masseter muscle in response to unilateral and bilateral transmastoid electrical stimulation in healthy individuals. Also, no response was recorded in 5 subjects to electrical stimulation of cutaneous receptors over the mastoid and external ear. The results of this study confirmed that the observed responses were not mediated by cutaneous receptors but by otolith afferents, known as the vestibulo-masseteric reflex.

A study done by Meier-Ewert et al. (1974) revealed that the acoustic jaw reflex was present only in individuals with normal hearing and intact auditory nerves. It was found that hearing-impaired individuals with normal vestibular function did not exhibit any jaw reflex in response to high-intensity acoustic stimulation. Based on these findings, the researchers suggested that the acoustic reflex's origin might be cochlear receptors rather than the vestibular end organs. They also said that this response could be related to a protective mechanism or a startle reflex triggered by loud sounds (Meier-Ewert et al., 1974).

In contrast, a study done by Deriu et al., (2003) demonstrated the presence of inhibitory reflex in response to both unilateral and bilateral galvanic stimulation. The response obtained was a biphasic waveform with two peaks having latencies p11 and n15. They further tried to determine its origin; for that, they used a head-tilt method, tilting participants about 30 degrees to either side. It was found that the p11/n15 responses were asymmetrically modulated, indicating that they originate in the vestibular system rather than the auditory system.

Deriu et al. (2005) reported that during loud click presentations, a p11 peak was observed, and at lower stimulus levels, a p16 peak was observed. Similarly, De Natale et al. (2018) found that at lower intensities (around 98-113dB SPL), p16 is typically observed, whereas at higher intensities, an earlier p11 response was observed. The p11 component was found to be asymmetrically modulated during head tilt, indicating a vestibular origin. P16

response did not show modulation with head movement, indicating that it primarily originates in the cochlea. Hence, the authors concluded that, upon presentation of a loud acoustic stimulus such as 100 dBnHL, two partially overlapping reflexes were elicited, originating from both the vestibular and hearing end organs. The visible peaks which were observed during such loud stimulation are p11 and n21 which is just the overlap of p16/n21 (cochlear origin) responses and p11/n15 (vestibular origin) responses. Only p16/n21 response is observed at the threshold level of stimulus, which is predominantly cochlear in origin (Deriu et al., 2005).

2.2 Vestibulo masseteric Reflex Pathway

The vestibulo-massteric reflex pathway originates from the saccule. When the sensory hair cells of the saccule are stimulated, the information is carried along the inferior vestibular branch of the vestibulocochlear nerve to the brainstem. From the vestibular nerve, the information is carried out to the ipsilateral medial vestibular nucleus. From there, the nerve takes a decussation path and terminates at the ipsilateral and contralateral trigeminal motor nuclei via the vestibulo-trigeminal monosynaptic pathway (VTMP). The decussated VTMP fibres are more numerous than the ipsilateral ones (Tolu & Pugliatti, 1993; Giaconi et al., 2006). From the trigeminal nucleus, the signals are carried to the masseter muscles via the mandibular branch of the trigeminal motor nerve. The lower third of the masseter muscle is said to receive comparatively heavy innervation, and thus electrodes placed at that site yield responses of larger magnitude (Giaconi et al., 2006).

Various studies have shown that masseter muscle activity can also be influenced by stimulation of the auditory pathway, producing inhibitory reflex responses (Meier-Ewert et al., 1974; Deriu et al., 2005). When the cochlea is stimulated by the sound, the auditory nerve fibres transmit the information to the dorsal cochlear nucleus, from there the signal is transferred bilaterally to the trigeminal motor nucleus. This bilateral projection can be explained as a reason for the fact that masseter responses can be recorded bilaterally even when

a stimulus is given to only one ear. The information is further conveyed to the masseter muscle through the mandibular branch of trigeminal nerve.

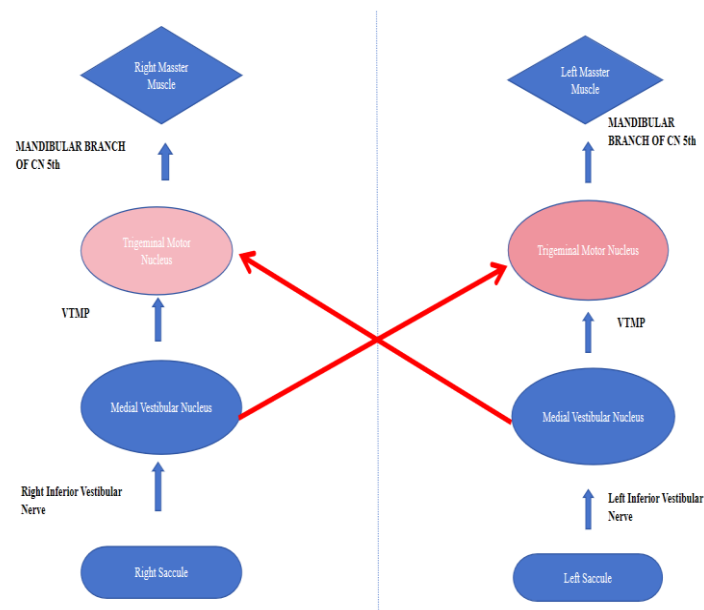


Figure 2.1 - Vestibulo masseteric reflex Pathway. VTMP - Vestibulo Trigeminal Monosynaptic Pathway.

It has also been said that this pathway exhibits few similarities with the tensor tympani reflex, as both are mediated by the trigeminal motor system and can also be triggered by high level acoustic stimulation and also from non-auditory stimuli, such as swallowing and chewing (Mukerji et al., 2010; Edmonson et al., 2022).

2.3 Effect of stimulus parameters

Various studies have shown that mVEMP responses can be evoked using both electrical and acoustic stimulation. (Deriu et al., 2003, 2005). Electrical stimulation of the vestibular system elicits bilateral responses in the masseter muscle, whether unilateral or bilateral, because the underlying pathways are bilateral. Across electrical and acoustic stimulation, no significant difference was found in the latency or amplitude of the P11 component, but a difference was observed in the N15 component, which was more clearly observed with electrical stimulation, possibly due to better neural synchrony (Deriu et al., 2005).

2.3.1 Stimulus Type and Frequency

Various acoustic stimuli like tone bursts, clicks and chirps have been used to elicit mVEMP responses (Deriu et al., 2005, 2007; Vignesh et al., 2021; Romero et al., 2022; Sangu Srinivasan et al., 2022; Thirusangu & Sinha, 2022; Neupane et al., 2023; Vinayagar & Sinha, 2023)

Nagarajan et al. (2024) investigated the effects of three different types of stimuli (click, 500Hz Tone burst, 500Hz narrow-band chirp) on EMG rectified amplitude, absolute latencies, interpeak interval, and interaural asymmetry of mVEMP. They recorded mVEMP at an intensity of 125 dB peSPL at a repetition rate of 5.1/s in 25 young healthy adults aged 18-27 years and found significantly earlier latencies for chirps, followed by clicks, and then tone bursts. Larger rectified amplitudes were obtained for chirps than tone bursts, followed by clicks, wider interpeak intervals for chirps in comparison to the other two stimuli and higher interaural asymmetry ratios for chirps. The authors concluded that 500Hz narrow-band chirps tend to produce higher amplitudes and earlier latencies and are therefore considered better stimuli. Neupane et al., (2023) studied the effects of three stimulus types on mVEMP in 21 normal-hearing individuals. Results showed that Chirp stimuli elicit earlier latencies and larger amplitudes, and are thus better stimuli.

Vignesh et al. (2021) established the normative values and determined the test-retest reliability of mVEMP evoked using a tone burst. He recorded mVEMP in 44 healthy individuals with normal vestibular function and normal hearing aged 18-50 years, and results showed no significant ear or sex differences, although males had higher normalised peak-to-peak EMG amplitude than females. Results showed excellent test-retest reliability for ipsilateral and contralateral p11 latencies, the amplitude asymmetry ratio, and the ipsilateral EMG-normalised p11-n21 peak-to-peak amplitude. Fair and good test-retest reliability was

found for the normalised peak-to-peak amplitude of contralateral EMG and the ipsilateral and contralateral n21 latencies.

Vinayagar and Sinha (2023) compared click stimulus and 500Hz tone burst for mVEMP responses across ipsilateral, contralateral and bilateral stimulation conditions. mVEMP was recorded in 20 healthy individuals aged 19-28years. Results showed a significant difference between the ipsilateral and contralateral conditions for both the click and the 500Hz tone burst stimuli. As compared to click stimuli, the amplitude of the p11-n21 complex was higher for 500Hz tone burst stimuli across all three stimulations-ipsilateral, bilateral and contralateral. Hence, suggesting tone burst as a better stimulus.

Arkadi and Neupane (2023) compared mVEMP responses across multiple tone burst frequencies(250-2000Hz) in a zygomatic electrode montage. They recorded mVEMP in 20 healthy individuals. Results showed no ear or gender differences at any frequency. Shortening of peaks p1 and n1 latencies was observed with an increase in the frequency of tone burst stimulation. Peak-to-peak amplitude was found to be maximum at 500Hz and minimum at 2000Hz for tone bursts under both unscaled and scaled conditions.

2.3.2 Number of Stimuli Averages

Deriu et al. (2005) used 300-500 stimuli to average acoustically evoked mVEMP and 800 averages for electrically evoked mVEMP. Many studies have used almost the same number of stimuli for both tone burst-evoked mVEMP and click-evoked mVEMP(de Natale et al., 2015a, 2019; Magnano et al., 2014; Deriu et al., 2007; Vignesh et al., 2021). Deriu et al. (2007) have recommended a high number of averages for the mVEMP. Although no studies have been done to compare the effect of no of averages on mVEMP response.

2.3.3 Intensity

Usually, a higher stimulus is required to evoke an mVEMP response. Deriu et al. (2005) used different intensity levels ranging from 80 to 100dB nHL to evoke mVEMP response. He found 80 dB HL to be a threshold for recording mVEMP. The amplitude of mVEMP varies

with intensity. The mVEMP response at 90 dB nHL was 40% lower than the 100 dB-evoked mVEMP response (Deriu et al., 2005). De Natale et al. (2019) reported that the mVEMP response can be obtained at 128-138dB SPL. Vignesh et al. (2021) recorded mVEMP responses with clear, replicable waveforms in 44 healthy subjects at 125 dB peSPL. At various standard institutes, such as Aiiish, the mVEMP is usually recorded at 125 dBSPL.

2.3.4 Stimulus Duration

Various studies on cVEMP and oVEMP have found that increasing the tone-burst duration increases response amplitude. Lim et al. (2013) revealed that in cVEMP, an increase in duration of 6-8ms or more tends to reverse the increase in amplitude. The amplitude of oVEMP decreased with increasing stimulus duration. The maximum amplitude was observed with a 2-3 ms stimulus duration, and increasing stimulus duration from 1 to 8 ms resulted in latency prolongation (Singh & Apeksha, 2014). Considering this relationship between intensity and duration, the intensity needs to be decreased to keep the stimulus duration within acceptable limits. Studies in masseter VEMP have used a 500Hz Blackman-gated tone burst with a duration of 4msec (2-0-2) cycles (Romero et al., 2022; Vignesh et al., 2021). A 500Hz tone burst with a rise/fall time of 2-1-2 cycles has been used as a stimulus in a few studies (Ravichandran, 2020; Thirusangu & Sinha, 2022).

2.3.5 Stimulus Rate

A study evaluated the effect of stimulus repetition rate on cervical vestibular evoked myogenic potentials and found that amplitude was higher at 1Hz and 5Hz stimulus rates (Wu & Murofushi, 1999). The investigator suggested a higher signal-to-noise ratio and 5Hz stimulus rate for a short examination time. To avoid synchronisation with the 50Hz frequency of the Indian electrical power supply, stimulation rates are generally set to non-integer values. Because of this rationale, studies of masseter VEMP have also used 5.1 Hz as a stimulation rate, as there are very few studies directly examining mVEMP (Kilinc et al., 2023;

Ravichandran, 2020; Vignesh et al., 2021). A stimulation rate of 5.4/second has also been used to record mVEMP in one study (Romero et al., 2022).

2.3.6 Amplifier gain

Many studies have used a 5000 amplification gain to record mVEMP (de Natale et al., 2015b, 2019; Magnano et al., 2014; Deriu et al., 2005; Loi et al., 2020; Vignesh et al., 2021). de Natale et al. (2018) used an amplification gain of 3000 to record all three VEMPs without any apparent reason. To date, no studies have compared the effects of different amounts of gain/amplification on recorded mVEMP.

2.3.7 Bandpass Filter

Various studies have used a 0.3-2000 Hz bandpass filter to evoke masseter vestibular evoked myogenic potential (Deriu et al., 2003, 2005, 2007; Vignesh et al., 2021). A few other studies have used a 5-5000 Hz bandpass filter to elicit mVEMP responses (De Natale et al., 2019; Loi et al., 2020; Ravichandran, 2020). A bandpass filter from 0.1 to 3000 Hz was used in one study (Thirusangu & Sinha, 2022).

2.3.8 Electrode Montage

To record the vestibular evoked myogenic potential, disc or cup electrodes are usually used. Different electrode montages are used to evoke the masseter Vestibular Evoked myogenic potential in various studies. Specifically, two electrode montages -the Zygomatic and Mandibular electrode montages are used in studies to record mVEMP responses. In mandibular montage, the reference electrode is placed on the mandibular angle (Deriu et al., 2003, 2005). In the Zygomatic montage, the reference electrode is placed midway along the zygomatic arch. The placement of active and ground electrodes is the same in both montages, active electrode at the lower third of the masseter muscle and the ground electrode at the forehead (de Natale et al., 2019; Loi et al., 2020; Thirusangu & Sinha, 2022; Vinayagar & Sinha, 2023).

According to a study by De Natale et al (2019), larger amplitudes and more consistent responses are observed with the zygomatic Montage. De Natale et al. (2019) compared the two montages in their study and found that a higher elicitation rate was observed with the zygomatic montage, and they cited a larger inter-electrode distance as a reason. Better test-retest reliability and larger amplitude were observed for the zygomatic montage (Loi et al., 2020). Thirasangu and Sinha et al. (2022) found no significant difference between the two montages when using a 500Hz tone burst as a stimulus to record mVEMP. Nowadays, a more recent electrode montage, known as the forehead montage, has come into use, in which the reference electrode is placed at the upper forehead (fz) position, and the placement of the active and ground electrodes is the same as in the zygomatic and mandibular montage. A study was done to compare the latency and amplitude characteristics of mVEMP response between the zygomatic and forehead montage and results of the study revealed significant increase in P1 latency, decrease in N1 latency, reduction of P1-N1 interpeak interval, an enlargement of both rectified and unrectified amplitude, and lower interaural amplitude asymmetry ratio for the forehead montage as compared to zygomatic montage (Nagarajan et al., 2024).

2.4 Characteristics of mVEMP in Healthy Individuals

2.4.1 Latency

Studies have demonstrated mVEMP response as a biphasic deflection with a positive peak with a latency of around 11-12msec, followed by a negative peak with a latency of around 21msec. mVEMP obtained with electrical stimulation had shown a p11/n15 response (Deriu et al., 2003). Masseter response to click stimulation showed a bilateral p11 peak with an onset latency of 8.4 ± 0.7 ms, a peak latency of 11.9 ± 0.9 ms, and a poorly defined n15 peak with a poor response rate. The p16 peak was also observed symmetrically, with an onset latency of 12.0 ± 1.3 ms and a peak latency of 16.6 ± 1.1 ms. In the same study, it was observed that p11/n15 peaks appeared only at higher stimulation intensities, even though p15 was not clear. Hence, it

suggests that the saccule is the activating receptor for p11/n15, and the cochlea is the receptor for p16/n21 (Deriu et al., 2007).

De Natale et al. (2019) sought to determine the latency values of mVEMP response peaks for both the mandibular and zygomatic montages. He found the mean ipsilateral p11 latency to be 11.17 ± 0.98 , contralateral p11 latency to be 11.38 ± 0.9 and ipsilateral n21 latency to be 19.68 ± 1.81 and contralateral n21 latency to be 19.53 ± 1.9 for the zygomatic montage. And for the mandibular montage, the value of ipsilateral p11 latency is 11.37 ± 0.91 , while that of contralateral p11 latency is 11.5 ± 0.87 , and the value of ipsilateral n21 latency is 19.75 ± 1.84 and contralateral n21 latency is 19.5 ± 1.84 . Values of mean interpeak latency were also mentioned in the study. For the zygomatic montage, the mean ipsilateral stimulation value was 8.52 ± 1.78 , and the contralateral value was 8.15 ± 1.87 . And for mandibular montage, the mean interpeak latency for ipsilateral stimulation was 8.38 ± 1.78 , and that for contralateral stimulation was 7.99 ± 1.73 .

Ravichandran (2020) gave values of latency and amplitude characteristics of mVEMP response evoked by click and tone burst stimuli. They observed a click-evoked p11 latency of 11.45 ± 0.87 , n21 latency of 21.85 ± 1.65 and interpeak latency of 10.4 ± 0.78 , and a tone burst-evoked p11 latency of 12.13 ± 0.81 , n21 latency of 22.54 ± 1.30 and interpeak latency of 10.42 ± 0.49 . Vignesh et al. (2021) reported that when tone burst stimuli were used, the ipsilateral p11 mean latency was found to be 13.20 ± 1.25 msec and the contralateral latency of 13.48 msec, the ipsilateral n21 latency of 21.40 ± 1.27 , and the contralateral n21 latency of 21.55 ± 1.32 were observed. The study also reported that tone burst stimuli tend to prolong the latencies of mVEMP response. A study used a stimulus of 100 dBnHL and found that mVEMP had a mean P1 latency of 15.90 ± 1.68 ms, a N1 latency of 25.86 ± 1.48 ms, and an interpeak latency of 9.96 ± 1.50 ms. It was also reported that, as stimulus intensity decreased, P1 and N1 latencies prolonged (Kilinc et al., 2023).

2.5.2 Amplitude

The integrity of the vestibular reflex pathway is related to the response and amplitude of the VEMP response. De Natale et al. (2019) reported amplitude measures for both the zygomatic and mandibular montages. They found that the peak-to-peak ipsilateral amplitude was 0.72 ± 0.31 (microvolts) and the contralateral amplitude was 0.74 ± 0.31 (microvolts), and the amplitude asymmetry ratio was 14.56 ± 11.8 for the zygomatic montage. For mandibular montage, the ipsilateral peak-to-peak rectified amplitude was 0.68 ± 0.33 (microvolts), and the contralateral amplitude was 0.68 ± 0.35 (microvolts). Vignesh et al. (2021) in their study found an ipsilateral peak-to-peak amplitude of 0.86 ± 0.38 , an amplitude asymmetry ratio of 15.07 ± 11.40 msec, and a contralateral peak-to-peak amplitude of 0.83 ± 0.31 , with an asymmetry ratio of 15.48 ± 9.83 . A study done by Kilinc et al. (2023) used 100dBnHL as a stimulus and found p11-n21 amplitude to be 97.89 ± 37.34 (microvolt) and an asymmetry ratio of 0.13 ± 0.07 (microvolt).

Romero et al. (2022) found that the effect of EMG on the VEMP response was reduced when rectified amplitudes were used. They also reported in their study that the average rectified amplitude asymmetry ratios were small and did not depend on the EMG target level.

2.5.3 Test Retest Reliability

Loi et al. (2020) reported excellent test-retest reliability of the mVEMP response for both montages. It was also reported that the mVEMP response was more reliable with the zygomatic montage than with the mandibular montage. Similarly, Vignesh et al. reported good test-retest reliability of mVEMP in healthy individuals and found no significant effects of ear or gender on mVEMP latency or amplitude. A study by Vignesh et al. (2021) revealed excellent test-retest reliability for p11 latency and fair test-retest reliability for n21 latency of tone-burst-evoked mVEMP responses, although differences across participants were observed, hypothesised to be related to differences in the degree of masseter muscle activation.

CHAPTER -III

METHOD

The aim of the present study was to compare the mVEMP response characteristics between two electrode montages and to evaluate their test-retest reliability. To achieve the objectives of this study, 30 healthy adults (15 males and 15 females) aged 18–40 years were recruited. Participants initially underwent testing to compare various electrode montages for recording masseter vestibular evoked myogenic potentials (mVEMPs). The same cohort was retested 1 week later to assess the test–retest reliability of responses across electrode configurations. Informed consent was obtained from all participants prior to enrollment.

Participant Selection Criteria

All participants in this group had hearing thresholds not exceeding 15dB HL at audiometric frequencies from 250 Hz to 8000 Hz in response to air-conduction tones and from 250 Hz to 4000 Hz in response to bone-conduction pure tones. All participants had normal middle ear function, as identified by a type A/As tympanogram and the presence of ipsilateral and contralateral acoustic reflexes at 100 dB HL to reflex-eliciting tones of 500 Hz, 1000 Hz, 2000 Hz, and 4000 Hz. Loudness discomfort level was greater than 100 dB HL for the speech stimulus. No retrocochlear lesion was confirmed by conventional click-evoked auditory brainstem response elicited at two repetition rates (11.1/s & 90.1/s). No presence of history of vestibular symptoms. Participants with a history of neurological, vestibular, otological disorders, temporomandibular joint dysfunction, cervical or masticatory muscle pathology, prior ear surgery, or exposure to ototoxic medication were excluded.

Test environment and instrumentation

The experiments were conducted inside well-illuminated sound-treated rooms, with ambient noise levels within the permissible limits (ANSI S3.1, 1991). The Inventis Piano audiometer with TDH-49 headphones and a B-71 bone vibrator was used to estimate the hearing

threshold. GSI TYMPSTAR was used to record tympanometry and reflexometry. A calibrated two-channel Neuro Audio auditory evoked potential system (Neuro-Audio Version, 2010; Neurosoft, Ivanovo, Russia) with impedance-matched transducers was used to record ABR and mVEMP responses.

Procedure

Case History

A detailed case history was obtained from all the participants. The case history included questions like: any conductive hearing loss symptoms and a history of middle ear pathology. Questions regarding any vestibular symptoms, such as vertigo, imbalance, nausea, vomiting, and blurred vision, were included.

Pure Tone Audiometry

Pure tone audiometry was carried out and hearing thresholds were obtained for all the participants using a modified version of Hughson and Westlake at all the octave frequencies between 250-8000Hz for Air conduction and between 250-4000Hz for bone conduction.

Immitance

Tympanogram was acquired in both ears using 226 Hz probe tone and acoustic thresholds were measured were estimated for both contralateral and ipsilateral recording using 500, 1000, 2000 and 4000Hz stimulus.

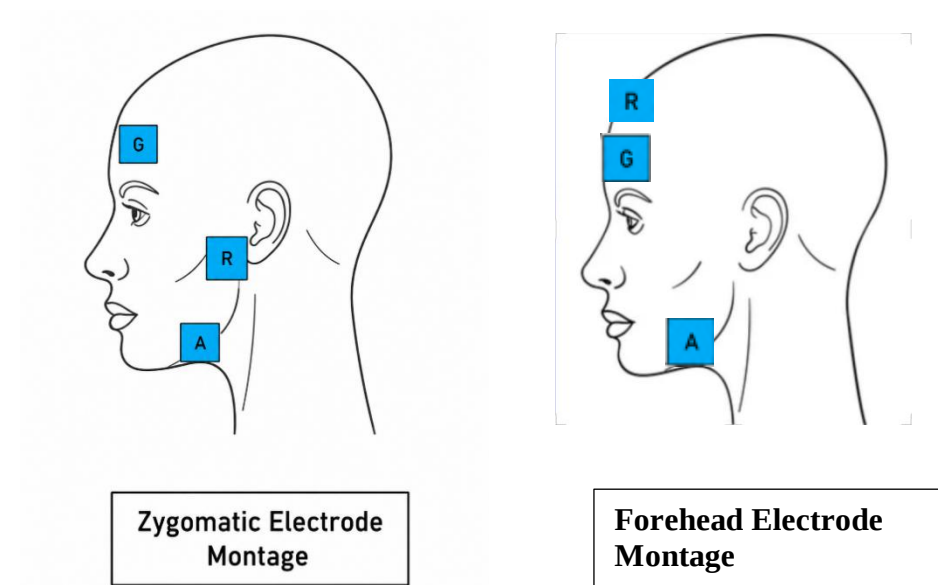
Mvemp recording Procedure

For each participant, mVEMP was recorded for 2different electrode montages 1) Zygomatic electrode montage:- inverting electrode(reference) at the midpoint of the zygomatic arch; non-inverting (active) electrode on the lower third(belly) of the masseter muscle ; ground on the forehead(Fpz).

2) Forehead Electrode montage: This montage includes an inverting electrode(reference) on Fz, the active electrode on the lower third of the masseter

muscle and a ground electrode on Fpz.

Figure 3.1 Two different electrode montages



G - Ground electrode, R-Reference electrode, A-Active electrode

Participants were seated on a non-inclined chair in a relaxed, upright position. With the help of abrasive gel (Nuprep), the skin over the electrode placement sites (lower third of the masseter muscle, lower forehead, midpoint of the zygomatic arch, mandibular angle) was cleaned and prepped for electrode placement. After electrode placement, absolute and inter-electrode impedances were measured. The absolute and inter-electrode impedances were maintained within 5 k Ω and 2 k Ω , respectively. Participants were instructed to clench their teeth to contract the masseter muscle throughout the recording. Subjects received muscle-tension feedback throughout the recording. The baseline for muscle contraction was set at 30%–50% of maximum voluntary contraction to reduce amplitude variation caused by differences in muscle tension.

For each type of electrode montage, stimuli were presented monaurally via calibrated insert earphones (ER-3A or equivalent). Responses were elicited by using a stimulus with the following stimulus parameters and acquisition parameters.

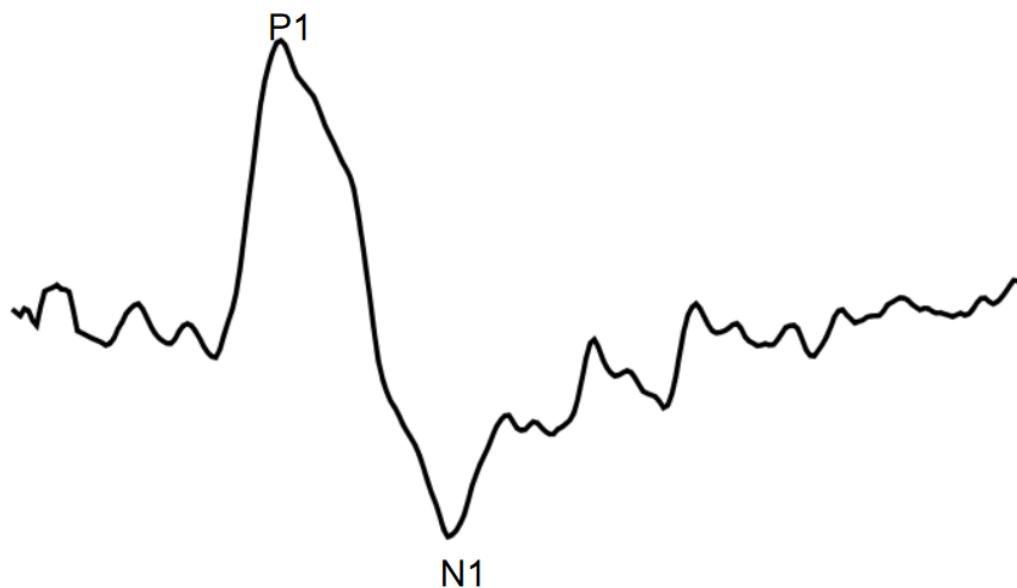
Table 3.1

Stimulus and Acquisition Parameters used for recording masseter VEMP

Stimulus Parameters		Acquisition parameters	
Stimulus Type	Tone Bursts	Analysis time	100 msec (20msec pre stimulus)
Frequency	500Hz	Filter settings	0.1-2000 Hz
Gating	Blackman	No of sweeps	200
Stimulus Duration	2-1-2	Amplifier Gain	3000
Polarity	Alternating	No of channels	1
Repetition rate	5.1/s	Electrode Montage	Zygomatic Inverting -mid point of the zygomatic arch Non-inverting - lower third of masseter muscle Ground - forehead Forehead Inverting -Fz, Non inverting-lower third of masseter muscle
Stimulation Mode	Monoaural		
Transducer	Insert earphones		

Data Analysis

Figure 3.2 Waveform of mVEMP



- Latency of p11 was analysed.
- Latency of n21 was analysed.
- Peak-to-peak amplitude of p11-n21 was analysed.
- Interaural amplitude asymmetry ratio for each electrode montage was analysed.

Test-retest procedure: mVEMP was recorded for the same participants a second time, after a fixed interval of 1 week, using 2 different electrode montages. The parameters of stimuli were kept same as those of the previous session. Recording Parameters were also the same.

Statistical Analysis

The statistical package for Social Sciences (SPSS) software Version 20 was used for the statistical analyses. The following statistical analyses were done.

- Descriptive statistics were done to find out the mean and standard deviation for the absolute latency of P1 and N1 peaks and the peak-to-peak amplitude of the P1-N1 complex.
- The Shapiro-Wilk test of Normality was used for all parameters to determine whether the data had a normal distribution.

- A paired-samples t-test was performed to determine whether there were individual-ear differences.
- Descriptive statistics were done to obtain the mean and standard deviation for the combined ear data.
- A paired-samples t-test was performed to compare the P1 latency, N1 latency, and peak-to-peak amplitude between the two montages.
- A paired-samples t-test was performed again on another set of data to examine ear differences.
- Then again, descriptive statistics were performed on another set of data to obtain the mean and standard deviation for the combined ear data.
- Again, a paired-samples t-test was performed to compare the P1 latency, N1 latency, and rectified amplitude between the two montages for another set of data.
- An intraclass correlation coefficient test was performed to check the reliability of the data.

CHAPTER IV

RESULT

The present study was conducted to compare the latency and amplitude of masseter VEMP across two electrode montages in young healthy individuals. The study also aimed to assess the test-retest reliability of mVEMP responses for the two electrode montages. A total of 30 healthy individuals (60 ears) with normal hearing and normal vestibular function were included in the study. The same cohort was retested one week later to assess the test-retest reliability of mVEMP responses between two electrode configurations.

4.1 Response Rate of Masseter VEMP

The response rate of the 500 Hz tone burst-evoked mVEMP response was analysed for both the Zygomatic and forehead electrode montages across the two sessions. In the two-electrode montages, masseter VEMP responses were present in all participants across the two trials. The individual and grand-averaged mVEMP waveforms for trials 1 and 2 in both montages are shown in Figures 1 and 2, respectively.

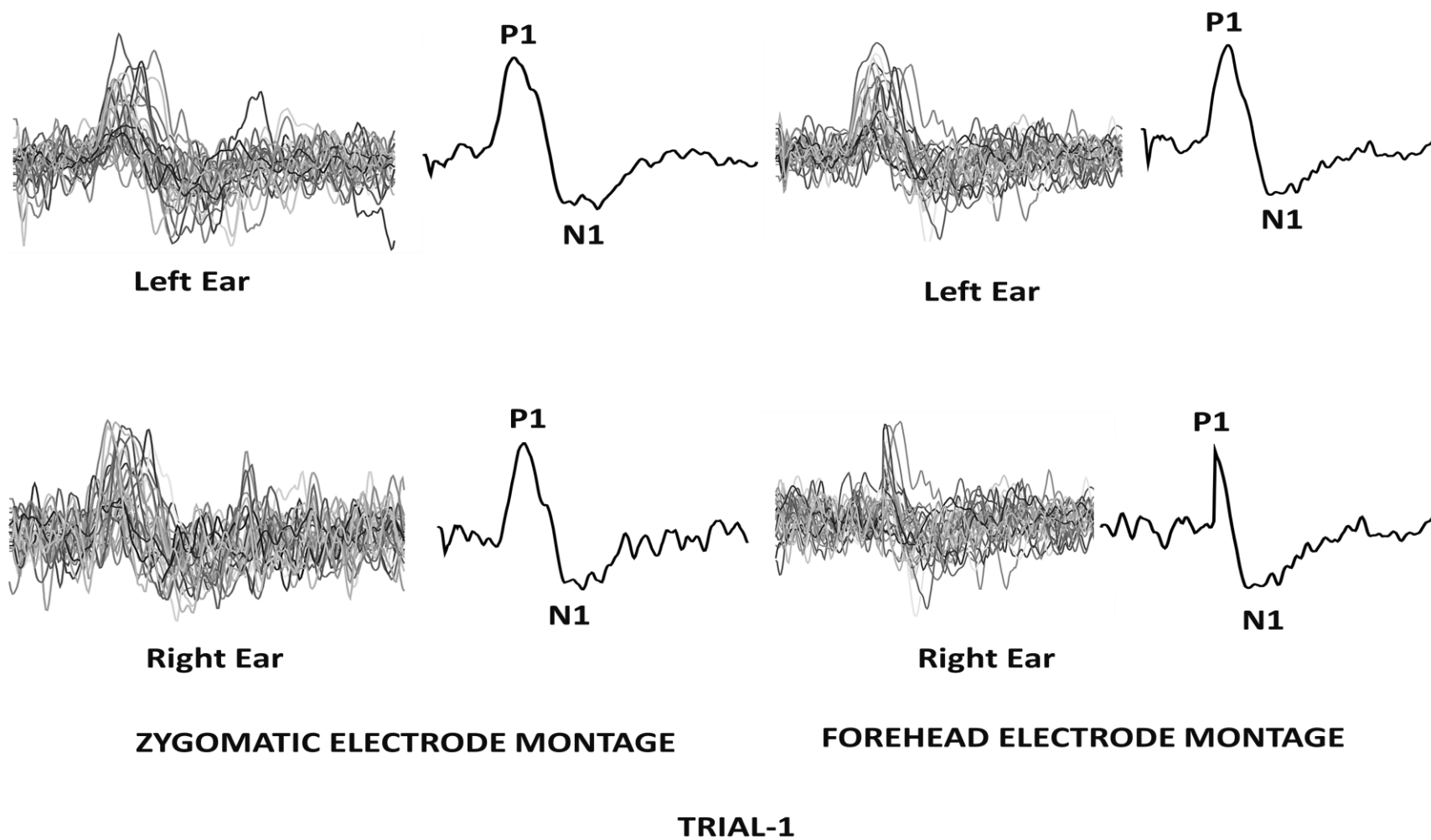


Figure-4.1. Individual and grand-averaged waveform for Zygomatic and forehead electrode montage for Trial-1

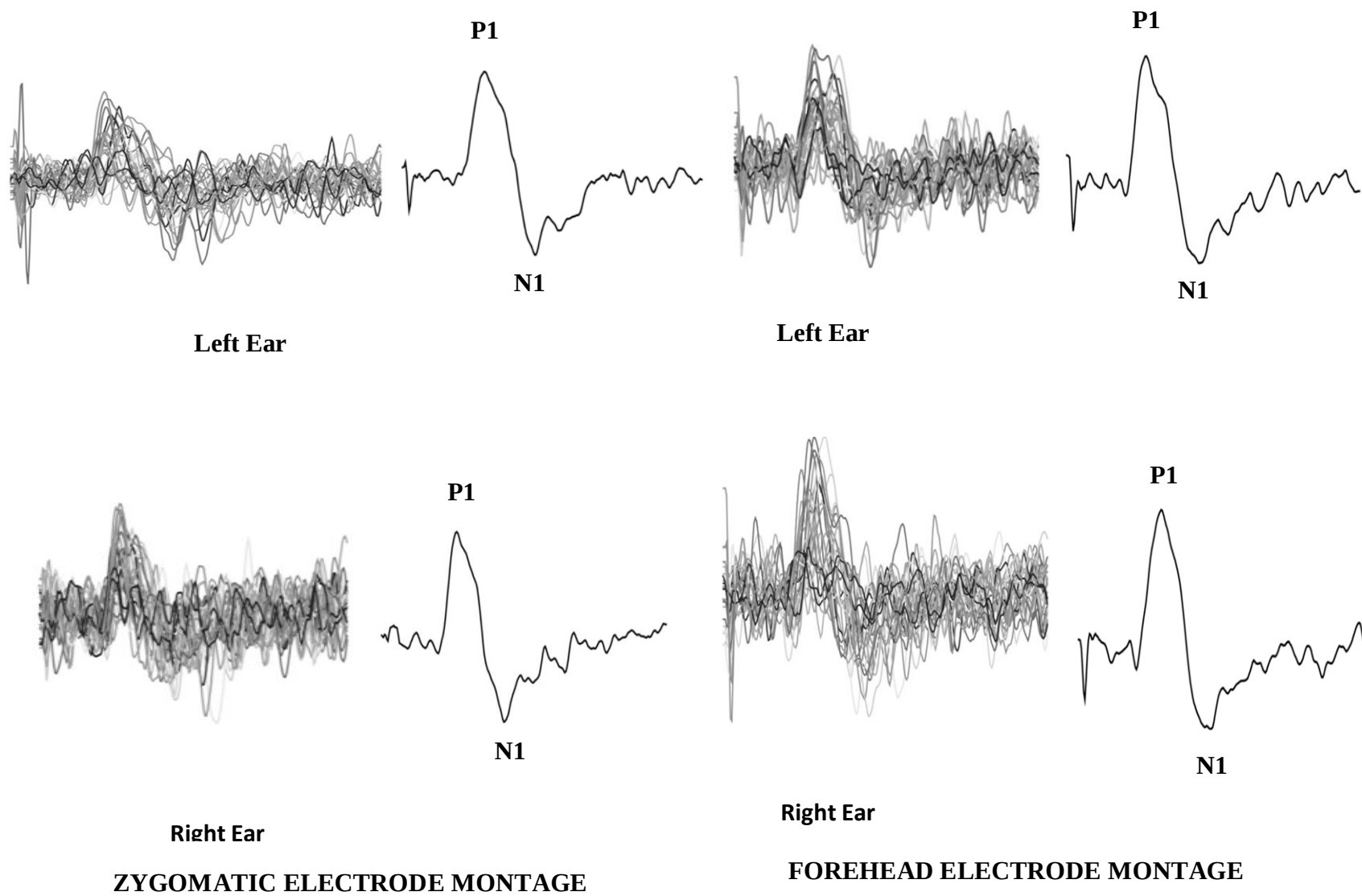


Figure-4.2. Individual and grand-averaged waveform for Zygomatic and forehead electrode montage for Trial-2

4.1 Latency and amplitude measures of mVEMP in two electrode montages - Zygomatic and Forehead for Trial 1

The mean and standard deviation of the P1, N1 latency and amplitude measures of the P1-N1 complex of mVEMP elicited by a 500Hz tone burst for two montages - Zygomatic and forehead for 'Trial 1' are presented in Table 4.1

Table 4.1

Mean and standard deviation of P1, N1 latency measures and amplitude measures of the P1-N1 complex of mVEMP elicited for two different montages -zygomatic and forehead in trial 1

Electrode Montage	Ear	P1 peak(msec)		N1 peak		Amplitude of p1-n1 complex	
		Mean	SD	Mean	SD	Mean	SD
Zygomatic Montage	Right	13.58	1.74	21.27	2.08	0.76	0.31
	Left	13.58	1.45	22.04	1.65	0.79	0.38
Forehead Montage	Right	14.09	1.73	22.10	2.29	0.82	0.35
	Left	14.08	1.47	22.79	2.08	0.81	0.37

Note: The mean latency values suggest that P1 and N1 latencies are earlier with the masseter VEMP recorded from the zygomatic than with the forehead electrode montage. However, the mean amplitude appears to be larger for the forehead electrode montage than the zygomatic electrode montage.

The Shapiro-Wilk test was performed to check the normality of the data. Results of the Shapiro-Wilk test revealed that the data were normally distributed for both electrode montages. Parametric tests were used because the Shapiro-Wilk test indicated the data were normally distributed. Paired sample t-test for 500Hz tone burst evoked mVEMP response in zygomatic montage showed no significant difference between the two ears for P1 latency ($t=0.012$, $p=0.99$), N1 latency ($t=1.826$, $p=0.078$), and P1-N1 amplitude ($t=0.414$, $p=0.68$). For the

forehead electrode montage, Paired Sample t-test showed no significant difference between the left and right ears for P1 latency ($t=0.06$, $p=0.95$), N1 latency ($t=-1.91$, $p=0.066$), and P1-N1 amplitude ($t=0.109$, $p=0.914$). Hence, the data were combined for both electrode montages. Descriptive statistics were performed on the combined data to obtain the mean and standard deviation. The combined ear data of P1 and N1 latency for both montages are given below in Table 4.2

Table: 4.2

Mean and standard deviation (SD) of combined data for P1 latency, N1 latency and P1-N1 complex amplitude measures in both the montages

Electrode Montage	P1 peak		N1 peak		P1-N1 Complex amplitude	
	Mean	SD	Mean	SD	Mean	SD
Zygomatic electrode Montage	13.58	1.59	21.56	1.90	0.77	0.34
Forehead electrode Montage	14.05	1.59	22.44	2.20	0.81	0.36

Note: Table 4.2 shows that the mean latencies of P1 and N1 are earlier with the zygomatic than with the forehead electrode montage. However, the amplitude of the P1-N1 complex is higher for the forehead than for the zygomatic electrode montage.

4.1.2 Comparison of latency and amplitude parameters of masseter VEMP between Zygomatic and Forehead Electrode Montage for ‘Trial 1’

Results of the Paired sample t-test revealed a significant early latency of P1 ($t= 5.789$, $p= 0.000$) and N1 ($t= 5.332$, $p= 0.000$) for zygomatic electrode montage compared to forehead electrode montage. However, statistically no significant difference in P1-N1 amplitude measures ($t= 1.313$, $p=0.194$) was observed between the two electrode montages.

4.2 Latency and amplitude measures of mVEMP in two electrode montages - Zygomatic and Forehead for Trial-2

The mean and standard deviation of the P1, N1 latency and amplitude measures of the P1-N1 complex of mVEMP elicited by a 500Hz tone burst for two montages - Zygomatic and forehead for 'Trial 2' are presented in Table 4.3

Table 4.3

The mean and standard deviation of the P1, N1 latency and amplitude measures of the P1-N1 complex of mVEMP for two montages - Zygomatic and forehead for 'Trial-2'

Electrode Montage	Ear	P1 latency		N1 Latency		P1-N1 Complex amplitude	
		Mean	SD	Mean	SD	Mean	SD
Zygomatic Montage	Right	13.76	1.59	21.39	1.85	0.72	0.28
	Left	13.65	1.38	22.36	2.12	0.73	0.32
Forehead Montage	Right	14.17	1.71	21.78	1.84	0.82	0.35
	Left	13.92	1.49	22.80	2.33	0.75	0.30

Note: The mean latency values suggest that P1 and N1 latencies are earlier with the masseter VEMP recorded from the zygomatic than with the forehead electrode montage. However, the mean amplitude appears to be larger for the forehead electrode montage than the zygomatic electrode montage

Paired sample t-test for 500Hz tone burst evoked mVEMP response in zygomatic montage showed no significant difference between the two ears for P1 latency ($t=0.42$, $p=0.676$), N1 latency ($t= 1.827$, $p=0.078$), and P1-N1 amplitude ($t=0.131$, $p=0.896$). For Forehead montage, Paired Sample t test showed no significant difference between left and right ear for P1 latency ($t=0.88$, $p=0.382$), N1 latency ($t=1.837$, $p=0.77$) and P1-N1 amplitude ($t=0.14$, $p=0.165$). Hence, the data were combined for both electrode montages. Descriptive statistics were performed on

the combined data to obtain the mean and standard deviation. The combined ear data of P1 and N1 latency for both montages are given below in Table 4.4

Table 4.4

The combined data and standard deviation of latency and amplitude measures of mVEMP in two different montages for 'trial 2'

Parameters	P1 latency		N1 latency		P1-N1 Complex amplitude	
	Mean	SD	Mean	SD	Mean	SD
Zygomatic	13.70	1.47	21.87	2.04	0.73	0.30
Forehead	14.05	1.59	22.29	2.14	0.78	0.33

Note: Table 4.4 shows that the mean latencies of P1 and N1 are earlier with the zygomatic than with the forehead electrode montage. However, the amplitude of the P1-N1 complex is higher for the forehead than for the zygomatic electrode montage.

4.2.1 COMPARISON OF ZYGOMATIC AND FOREHEAD ELECTRODE MONTAGE FOR 'TRIAL 2'

Results of the paired-samples t-test revealed statistically significant differences in P1 latency ($t = 3.501$, $p = 0.003$) and N1 latency ($t = 2.274$, $p = 0.027$), and no significant difference for P1-N1 amplitude ($t = 1.780$, $p = 0.078$) between the two montages.

4.3 Test-Retest reliability measures of mVEMP across two montages

Intra-class coefficient (ICC) was used to assess the test-retest reliability of mVEMP measures between two trials.

4.3.1 Test-retest reliability of P1 latency, N1 latency and P1-N1 amplitude measures between the two trials in zygomatic montage

An intraclass correlation coefficient (ICC) analysis was performed to assess the test-retest reliability of p11 latency in Montage 1 across the two recording sessions. The analysis

demonstrated good reliability, with an average ICC of 0.882 (95% CI: 0.803-0.929). Intraclass correlation coefficient analysis revealed moderate test–retest reliability for n21 latency between the two recording sessions, with an average ICC value of 0.618 (CI: 0.360-0.772). Intraclass correlation coefficient analysis demonstrated good test–retest reliability for the amplitude measure between the two recording sessions, with an average ICC value of 0.874 (CI:0.789-0.925).

4.3.2 Test-retest reliability of P1 latency, N1 latency and P1-N1 amplitude measures between the two trials in forehead montage

Intraclass correlation coefficient analysis demonstrated good test–retest reliability for p11 latency in the second electrode montage between the two recording sessions, with an average ICC value of 0.890 (CI:0.816-0.934). Intraclass correlation coefficient analysis revealed moderate test–retest reliability for n21 latency in the second electrode montage between the two recording sessions, with an average ICC value of 0.630. (CI:0.378 -0.779). Intraclass correlation coefficient analysis demonstrated good test–retest reliability for the amplitude measure in the second electrode montage between the two recording sessions, with an average ICC value of 0.832 (CI:0.719-0.899).

CHAPTER -V

DISCUSSION

The aim of the present study was to compare the response rate, amplitude, and latency characteristics of mVEMP across two electrode montages and to assess the test-retest reliability of mVEMP across these montages.

5.1 Response Rate

In the present study, all 30 participants showed a mVEMP response in both the electrode montages during both trials.

This finding is consistent with earlier reports demonstrating 100% response rates of masseter VEMPs in healthy individuals using zygomatic, forehead, and mandibular electrode montages (De Natale et al., 2019; Loi et al., 2020; Vignesh et al., 2021; Nagarajan & Sinha, 2025). The robust response rate across all electrode montages suggests that the 500 Hz tone-burst stimulus at 125 dBpeSPL is sufficient to elicit masseter VEMPs. The presence of mVEMP responses in all healthy participants for both electrode montages suggests that both can be used reliably to elicit masseter VEMPs.

5.2 Latency and amplitude characteristics of Masseter VEMP across two electrode montages

The results of the study suggest that the P1 and N1 latencies for masseter VEMPs recorded with the zygomatic electrode montage were shorter than those recorded with the forehead electrode montage in both trials. However, there was no significant difference in the amplitude of the P1-N1 complex between the two electrode montages.

Earlier studies on masseter VEMPs have utilised either zygomatic or mandibular electrode montages to record masseter VEMPs in healthy and clinical populations (de Natale et al., 2019; Loi et al., 2020; Thirusangu & Sinha, 2022). There are equivocal findings regarding the effect of these two electrode placements on response rate and amplitude of masseter VEMPs in healthy individuals. De Natale et al. (2019) and Loi et al. (2020) reported

that the EMG rectified amplitude, response rate, and reliability are superior with the zygomatic electrode montage compared to the mandibular electrode montage. The test-retest reliability of masseter VEMP has also been reported to be superior when recorded with the zygomatic electrode montage compared to the mandibular electrode montage (Loi et al., 2020). The better test-retest reliability of the zygomatic electrode montage could be due to the reduction in reference contamination in the zygomatic electrode montage, as the active and reference electrodes are placed at a greater distance in the zygomatic electrode montage than in the mandibular electrode montage (Nagarajan & Sinha, 2024).

However, Thirusangu & Sinha (2022) reported no significant differences in latency or amplitude parameters of mVEMP between the zygomatic and mandibular electrode montages. The authors suggested that either of the two electrode montages can be used for recording masseter VEMPs. Loi et al. (2020) suggested using both montage configurations to obtain the highest detection rates in mVEMP. Behmen et al. (2024) and Tas Elibol et al. (2024) first reported a different electrode placement. In this electrode placement, the reference electrode was used on the upper forehead, the ground electrode on the lower forehead and the active electrode on one-third of the masseter muscle.

The forehead electrode montage is expected to provide higher amplitudes than the mandibular or zygomatic electrode montage. This is because, as the distance between the reference and the active electrode increases, noise contamination decreases (Nagarajan & Sinha, 2025; Makowiec et al., 2017). Nagarajan & Sinha (2025) reported a greater amplitude of masseter VEMP with the forehead electrode montage than with the zygomatic electrode montage. The authors stated that the greater amplitude of masseter VEMP in the forehead electrode montage may be due to the greater distance between the reference and active electrode montage. However, in the present study, there was no significant difference in the amplitude of masseter VEMPs recorded with either electrode montage. In the previous study

by Nagarajan et al. (2025), the number of healthy participants was the same. Based on the results of the present study, it is recommended to use either of the electrode placements for recording masseter VEMPs.

The current study's results demonstrated that zygomatic montage consistently yielded shorter P1 and N1 latencies compared to forehead montage across both recording sessions.

This finding is consistent with those of Nagarajan et al. (2024), who also reported a significant increase in P1 latency for the forehead electrode montage compared with the zygomatic montage. The prolongation in P1 latency observed with the forehead reference electrode placement may be attributable to the inter-electrode distance and the electrical field geometry created by the reference electrode. The forehead (Fz) placement is away from the masseter muscle relative to the zygomatic reference, which may alter the differential recording and introduce a slight phase shift, resulting in an apparent latency prolongation. Some earlier studies on oVEMP also reported better latency when the active and reference electrodes were placed together (Makowiec et al., 2017; Raveendran & Singh, 2024). The oVEMP responses recorded using the belly tendon montage showed significantly shorter latencies when compared to other electrode configurations, including the chin-referenced and sternum-referenced montages that had a larger interelectrode distance (Raveendran & Singh, 2024). In a similar investigation of cVEMP, Wei et al. (2013) examined different electrode placements on the sternocleidomastoid muscle and observed that absolute latencies increased significantly as the distance between the active and reference electrodes increased.

5.3 Test–Retest Reliability of mVEMP across two electrode montages

In the present study, intraclass correlation coefficients (ICCs) were calculated for P1 latency, N1 latency, and P1–N1 amplitude in both the zygomatic and forehead montages. For the zygomatic montage, good test–retest reliability was observed for P1 latency ($ICC \approx 0.88–0.89$), moderate reliability for N1 latency, and good reliability for P1–N1 amplitude ($ICC \approx$

0.83–0.87). For the forehead montage, a comparable pattern was observed: good reliability for P1 latency and P1–N1 amplitude, and moderate reliability for N1 latency.

The finding of greater reliability for P1 latency compared to N1 latency is consistent with previous reports. Vignesh et al. (2021) observed excellent test–retest reliability for P1 latency and fair-to-good reliability for N1 latency in tone burst-evoked mVEMP recordings. They suggested that the relatively lower reliability of N1 latency may be related to the composite and partially cochlear origin of the N21 peak, which could introduce greater variability across recording sessions.

The moderate reliability of N1 latency observed across both montages in the present study is therefore noteworthy. As reported by Deriu et al. (2005) and De Natale et al. (2019), the n21 component reflects overlapping vestibular and cochlear contributions, making it inherently more variable than the p11 component, which is predominantly vestibular in origin and influenced by head position. In addition, minor inter-session differences in the level of masseter muscle activation, despite the use of a standardised biofeedback protocol, may have contributed to variability in N1 latency, as the temporal characteristics of the myogenic response can be affected by baseline EMG activity. Thus, either of the electrode montages can be used for recording the masseter VEMPs.

CHAPTER -VI

SUMMARY AND CONCLUSION

The masseter vestibular evoked myogenic potential is a short-latency inhibitory electromyographic response that is recorded from the tonically activated masseter muscle in response to acoustic or galvanic stimulation. It evaluates the functional and structural integrity of the vestibulo-masseteric reflex pathway, which originates from the saccule, goes through the inferior vestibular nerve and vestibular nuclei, then crosses to the trigeminal motor nuclei and finally reaches the masseter muscle via the mandibular branch of the trigeminal nerve. Because this pathway includes the brainstem, various studies on mVEMP have provided strong evidence for the high sensitivity of mVEMP in identifying brainstem dysfunction in conditions such as multiple sclerosis, Parkinson's disease, and REM sleep behaviour disorder, compared with cVEMP and oVEMP.

Although mVEMP is emerging as a clinically promising neurophysiological tool, its recording protocol and stimulus parameters are not yet standardised, particularly regarding electrode montage and filter settings. Several studies have used different electrode montages, such as mandibular, zygomatic, and forehead/Fz. Most of the earlier studies have compared response rate, latency and amplitude characteristics of mVEMP between the two montages - zygomatic and mandibular only. However, there are very few studies that have included forehead montage for comparison with mandibular and zygomatic montage. And there is limited data on the test-retest reliability of mVEMP responses across different electrode montages.

Therefore, the present study was done to compare the response rate, latency, and amplitude characteristics of 500Hz evoked mVEMP response between two widely used electrode montages- zygomatic and forehead and also to assess the test-retest reliability of mVEMP across those two electrode montages.

To achieve the aim of this study , 30 healthy individuals aged 18-40 years with normal hearing sensitivity and normal vestibular function were included in this study. Normal middle ear function was identified by a type A/A tympanogram with ipsilateral and contralateral reflexes. Retrocochlear pathology was ruled out using conventional click-evoked auditory brainstem responses. All the participants underwent mVEMP recording for two electrode montages- zygomatic and forehead montage using 500Hz as a tone burst stimulus. For the zygomatic montage, the reference electrode was placed at the midpoint of the zygomatic arch; for the forehead montage, it was placed at the Fz position. The positions of the active electrode at the lower third of the masseter muscle and the ground electrode on the forehead (Fpz) were identical in both montages. For each type of electrode montage, a 500Hz tone burst was presented monaurally via calibrated insert earphones (ER-3A). The same cohort group was taken once again to record mVEMP to assess the test-retest reliability of latency and amplitude parameters of mVEMP response across two electrode montages.

The individual waveforms were then analysed for absolute latencies of P1 and N1, and peak-to-peak rectified amplitude for the P1-N1 complex was measured for both electrode montages. Test retest reliability was then determined. Appropriate statistical analysis was carried out using the Social Sciences (SPSS) software Version 20. The following analysis was done

- Descriptive statistics were performed to determine the mean and standard deviation of the absolute latencies of the P1 and N1 peaks and the peak-to-peak amplitude of the P1-N1 complex.
- The Shapiro-Wilk test of Normality was used for all parameters to determine whether the data had a normal distribution.
- A paired-samples t-test was performed to assess differences between ears for mVEMP latency and amplitude parameters across both electrode montages and trials.

- As the ear differences were not seen the data from the two ears were combined and Descriptive statistics were done to obtain the mean and standard deviation for the combined ear data.
- A paired sample t test was performed to compare the P1 latency, N1 latency and peak to peak amplitude between the two montages.
- Intraclass correlation coefficient test was performed to check the reliability of data.

The results of the study are as follows:

- For both set of data, mVEMP response was present in all the 30 individuals for both the electrode montages- zygomatic and forehead
- For both the trials the latency of P1 and N1 peak was shorter for zygomatic electrode montages compared to the forehead electrode montages.
- For both the trials, the P1-N1 amplitude was significantly not different between the zygomatic and forehead electrode montages.
- Considering the test reliability measures, for zygomatic montage, good test-retest reliability was found for P1 latency, moderate test-retest reliability for N1 latency and good test-retest reliability for P1-N1 amplitude measures.
- For forehead montage, good test-retest reliability for P1 latency and P1-N1 amplitude measures and moderate test-retest reliability for N1 latency was observed.

Conclusion

The results of the study suggest that P1 and N1 latency of masseter VEMPs for the zygomatic electrode montages are shorter compared to the forehead electrode montage. However, there is no difference in amplitude of masseter VEMPs between the two electrode montages. Also, the test-retest reliability of the two electrode montages are moderate to good. It can be concluded that either of the electrode montage can be used to record masseter VEMPs.

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INSTITUTE REVIEW BOARD (IRB) APPROVAL FOR BIO-BEHAVIOURAL RESEARCH PROPOSAL INVOLVING HUMAN SUBJECTS AT AIISH

AIISH Institute Review Board (IRB)

Ref: SH/IRB/M.4/AUD.03/2025-26

Date: 06.01.2026

Title of the Dissertation

: Test -Retest Reliability of Masster
VEMP between 2 electrodes
Montages

Name of the Investigator

: Ankita

Guide

: Dr Sujeet Kumar Sinha

Co-Guide (if any)

: Nil

Proposed Duration of the Project

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Date on which the IRB meeting was held

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A clear statement of the decision reached IRB meeting

(In the vent of the proposal is not approved, a statement

of reasons for the same must be indicated)

: APPROVED

Advice & suggestions (if any)

: NIL

Signature & Name of the Member Secretary-IRB

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