Normal values of pattern reversal visual evoked potentials (PRVEP) and pattern electroretinography (PERG) in healthy adults in Sri Lanka

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Abstract

Background: *Visual electrophysiological tests can be used effectively to evaluate the functional integrity of retina and post retinal pathways. The main aim of this study was to determine the normal values for pattern reversal visual evoked potentials (PRVEP) and pattern electroretinography (PERG) in a group of healthy adults in Sri Lanka.*

Methods: *Fifty healthy adults were recruited for the study. PRVEP and PERG tests were performed using surface skin electrodes. Amplitude (μV) and peak latency (msec) values of N75, P100 and N145 components in PRVEP and N35, P50 and N95 components in PERG were measured. Mean values of each PRVEP and PERG measures in both sides were compared between males and females using independent sample t-test to determine the effect of gender on those measures.*

Results: *There were 11 males and 39 females (age range: 20-62, mean (SD): 37.2 (10.94) years). There was no significant difference in PRVEP or PERG measures with regard to gender.*

Conclusions: *Only few studies are reported on normative data of PRVEP and PERG in the world. Ideally, each laboratory has to establish normal values for its own equipment and patient population. However, the normal values of PRVEP and PERG in normal healthy adults obtained in this study would be of immense value to evaluate the patients with several post retinal and retinal disorders in the future visual electrophysiological studies done in Sri Lanka using the similar technical parameters*

Key words: visual evoked potentials, pattern electroretinography, normal values, surface skin electrodes

Introduction

Electrophysiological testing of the visual system consists of a range of tests based upon the recording of electrical potentials evoked by visual stimuli. These tests can be used for an evaluation of function of the visual pathway including different locations and cell types within the visual system[1]. Visual evoked potentials (VEP) and pattern electroretinography (PERG) are widely used electrophysiological tests to measure the functional integrity of the post retinal visual pathways and macula region. Though there are many types of VEP, the pattern reversal VEP (PRVEP) is the preferred stimulus for most purposes, because it has relatively low variability of waveforms and latency values both within a subject and over the normal population[2]. The PRVEP indicates the function of the post retinal pathway mainly reflecting the central retinal projection to the occipital poles[1].

PERG is derived largely from the macular photo-receptors and ganglion cells. It complements the full-field ERG, in differentiating between maculopathy and generalized retinopathy. It is also used to exclude a macular cause of VEP abnormality. It is increasingly used as a tool to assess the ganglion cells of the macula[1]. Though the international society for clinical electro-physiology of vision (ISCEV) recommends the use of corneal electrodes in PERG measurement, many studies have shown that there is no significant difference in PERG measures obtained by using corneal electrodes when compared with the surface skin electrodes[3]. Increasingly, surface skin electrodes are used as they are easier to place and obviate the risk of corneal abrasion and infections[4].

According to the ISCEV guidelines, it is recommended that each laboratory establish normal values for its own equipment and patient population[5]. There are only few studies done in the world regarding the normative values of PRVEP and PERG[6,3]. Studies done in this regard in South Asia are even less[7]. Since South Asian population has a different genetic background from the Western population, it is essential to study the normative values of visual electrophysiological data in the South Asian population. Also, it is essential

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to ascertain normative data to the population of a particular country under similar laboratory facilities to compare the results obtained from the patients in different studies rather than referring only to the international normative data[7]. To the best of our knowledge, there are no reported studies on normative data of visual electrophysiological tests in Sri Lanka up to now.

Given the importance of establishing the normative values of PRVEP and PERG in each laboratory, the aim of this study was to establish normal values for visual electrophysiological measures in a group of healthy adults in Sri Lanka using PRVEP and PERG using surface skin electrodes.

Methods

This is a cross-sectional study in which we tested a healthy adult group. The study was carried out in Sri Lanka from March 2017 to September 2019. The study design and protocols complied with the code of ethics of the World Medical Association Declaration of Helsinki[8]. The procedures were explained and informed written consent was obtained from all participants.

The study group comprised 50 healthy subjects of age ≥18 years with normal or corrected to normal vision in neuro-ophthalmological examination done by a specialist ophthalmologist (SS) who is a co-author. Subjects having a history of any disorder which could influence the results, like central nervous system diseases (multiple sclerosis, stroke, meningitis, parkinsonism, etc), diabetes mellitus, hypertension, ophthalmological conditions (cataract, glaucoma, retinopathy, optic atrophy) and subjects taking drugs which may affect normal functioning of the central nervous system (antidepressants, antipsychotics, sedatives, opioids etc) were excluded from the study. All participants underwent a neuro-ophthalmological assessment which included monocular measurements of distant visual acuity with Snellen's chart, colour vision with Ishihara chart, pupillary reflexes, fundoscopy and visual fields tested with 30-2 Humphrey automated perimetry. They did not have any visual complaint or fundoscopic changes suggestive of any ophthalmological disorder. All the subjects were instructed to take a sound sleep in the previous night and to avoid using any eye drops, at least 48 hours before the test.

Pattern reversal visual evoked potentials

A Natus EMG/NCV/EP machine (Natus Neurology Inc. USA) was used to present visual stimuli, to record and average the PRVEP waveforms. The recording technique conformed to the International Society of Clinical Electrophysiology of Vision (ISCEV) guidelines[9]. PRVEPs were measured in response to 2 Hz pattern-reversal checkerboard stimuli presented on a cathode ray tube monitor with a black and white alternating checkerboard pattern with check sizes of 1° and 0.25°. The field size was 15° of visual angle at the smallest point. The contrast between black and white squares was ≥80% as defined by Michelson contrast. Silver-silver chloride recording electrodes were used. An active electrode was fixed at the Oz position, reference electrode was fixed at Fz position and ground electrode was placed at Cz position[10]. The electrode impedances were maintained below 5 kΩ. Mean photopic luminance was 50 cdm-^{2}. The mean luminance of the stimulus screen was constant during checkerboard reversals and varied less than 30% between the centre and periphery of the visual field. Each eye was tested separately. Each subject was asked to focus the centre of the pattern field (at a corner of the four checks located at the center of the field). Background illumination was kept darkened. A band pass filter with low-and high-cut off frequencies of 1 Hz and 100 Hz was used. One hundred sweeps were averaged, and two averaged waveforms were recorded for reproducibility. The N75, P100 and N145 component peaks that represent successive stages of processing of afferent visual pathways up to the visual cortex were marked on the averaged PRVEP wave forms and the absolute peak latency and amplitude values of each component were measured. Among the three components, P100 is the most robust component as per the ISCEV guidelines.

Pattern electroretinography

A Nicolet Viking Quest machine (Natus Neurology, USA) was used to record the PERG waveforms. Active electrode was placed on the lower eyelids. These were preferred to corneal electrodes because of the complications associated with the latter[11]. Silver- silver chloride recording electrodes were used. The reference electrode was placed near the ipsilateral, outer canthus of each eye. The ground electrode was located on Cz position. The stimuli were a black and white reversing checkerboard pattern with check size of 0.8° (± 0.2°). The stimulus field size was 15°. Transient PERG stimuli had a reversal rate of 4 per second (2 Hz). The band pass filters were set to 1 Hz and 100 Hz. A photopic luminance of the white areas was greater than 80 cdm-^{2}. The mean luminance of the stimulus screen was constant during checkerboard reversals. The sweep time was 250 ms. A minimum of

100 artifact-free sweeps were averaged. Background illumination was kept darkened. At least two trials for each stimulus condition confirmed reproducibility. Binocular recording was done. The amplitudes and peak latencies of the N35, P50 and N95 peaks were measured[12]. Functional integrity of photoreceptor cells and ganglion cells of the macular region could be assessed by P50 and N95 components of PERG, respectively[13].

Data analysis

Main outcome measures were the peak latency and amplitude values of P100 component in PRVEP which assess the functional integrity of post retinal pathways; peak latencies and amplitudes of P50 and N95 components of PERG which assess the functional integrity of the photoreceptor cells and ganglion cells of the macular region. The other outcome measures were peak latencies and amplitudes of N75, N145 components in PRVEP, N35 in PERG as per the ISCEV guidelines. Continuous outcomes measures, i.e. peak latencies and amplitudes of PRVEP and PERG, are reported as means and standard deviations (SD) and were compared between groups using independent sample t-test. The comparisons were interpreted as significant at a cut-off p value of 0.05. IBM SPSS Statistics for Windows, version 22.0 was used to analyze the data.

Results

This healthy adult group had a mean age of 37.22 (SD:10.94) with a range of 20-62 years. There were 11 males and 39 females. Fundoscopic examination was normal as was color vision in all of them. There were no visual field defects in any of them. All had best-corrected Snellen visual acuity of 6/6.

PRVEP measures

Peak latency and amplitude values of PRVEP waveforms were determined in each side in each subject. Then the mean values for each PRVEP measure in each side and inter-ocular peak latency and amplitude differences were calculated based on the individual data.

Mean inter-ocular peak latency and amplitude differences were calculated for the entire group.

Table 1. Mean peak latency and amplitude values of PRVEP in right and left sides (n=50)

PRVEP parameter Peak latency (msec) and amplitude (μV)	*Right side Mean (SD)*	*Left side Mean (SD)*	*Inter-ocular difference Mean (SD)*
N75 peak latency	74.4 (5.86)	75.92 (7.73)	7.04 (5.49)
P100 peak latency	102.38 (5.86)	101.61 (6.30)	4.12 (3.41)
N145 peak latency	139.02 (7.79)	140.17 (7.45)	6.36 (4.86)
N75 amplitude	1.08 (0.82)	0.92 (0.85)	0.64 (0.58)
P100 amplitude	5.01 (3.39)	4.29 (3.32)	2.07 (1.95)
N145 amplitude	5.14 (3.43)	5.20 (3.51)	4.98 (4.55)

Then independent sample t-test was used to determine whether there were differences in peak latencies and amplitudes between males and females. No difference was found.

Table 2. Comparison of mean peak latency and amplitude values of PRVEP between males and females

PRVEP parameter Peak latency (msec) and amplitude(μV)	*Males (n=11) Mean (SD)*	*Females (n=39) Mean (SD)*	*P value*
Right N75 peak latency	73.09 (4.65)	74.77 (6.13)	0.40
Left N75 peak latency	74.64 (7.11)	76.28 (7.95)	0.54
Right P100 peak latency	104.04 (5.17)	101.91 (6.02)	0.29
Left P100 peak latency	103.54 (6.35)	101.07 (6.27)	0.25
Right N145 peak latency	141.18 (5.25)	138.41 (8.33)	0.30
Left N145 peak latency	142.86 (5.75)	139.41 (7.76)	0.18
Right N75 amplitude	1.42 (1.12)	0.98 (0.71)	0.24
Left N75 amplitude	0.86 (0.66)	0.94 (0.90)	0.79
Right P100 amplitude	5.76 (3.50)	4.80 (3.38)	0.41
Left P100 amplitude	4.54 (2.56)	4.23 (3.53)	0.78
Right N145 amplitude	4.63 (3.62)	5.28 (3.41)	0.58
Left N145 amplitude	5.92 (2.75)	4.99 (3.70)	0.45

PERG measures

Peak latency and amplitude values of PERG waveforms were determined in each side in each subject. Then the mean values for each PERG measure in each side and mean inter-ocular differences for each measure were calculated based on the individual data (Table 3). Mean inter-ocular peak latency and amplitude differences were calculated for the entire group.

Table 3. Mean peak latency and amplitude values of PERG in right and left sides (n=50)

PRVEP parameter Peak latency (msec) and amplitude(μV)	*Right side Mean (SD)*	*Left side Mean (SD)*	*Inter-ocular difference Mean (SD)*
N35 peak latency	36.09 (5.61)	36.05 (4.82)	5.96 (4.75)
P50 peak latency	51.93 (5.64)	52.57 (6.11)	7.14 (5.27)
N95 peak latency	92.53 (4.18)	91.63 (4.18)	4.89 (4.24)
N35 amplitude	0.47 (0.56)	0.46 (0.39)	0.35 (0.50)
P50 amplitude	1.66 (1.42)	2.00 (1.45)	1.07 (1.28)
N95 amplitude	1.91 (1.56)	1.84 (1.05)	0.99 (1.29)

Then the independent sample t-test was used to determine whether there were differences in peak latencies and amplitudes between males and females. No difference was found.

Table 4. Comparison of mean peak latency and amplitude values of PERG between males and females

PRVEP parameter Peak latency (msec) and amplitude(μV)	*Males (n=11) Mean (SD)*	*Females (n=39) Mean (SD)*	*P value*
Right N35 peak latency	36.73 (4.02)	35.92 (6.02)	0.68
Left N35 peak latency	34.09 (5.72)	36.60 (4.47)	0.13
Right P50 peak latency	55.32 (5.00)	50.97 (5.49)	0.05
Left P50 peak latency	51.09 (6.99)	52.99 (5.87)	0.37
Right N95 peak latency	92.77 (4.25)	92.46 (4.21)	0.39
Left N95 peak latency	92.13 (5.19)	91.49 (3.92)	0.65
Right N35 amplitude	0.79 (0.92)	0.38 (0.38)	0.18
Left N35 amplitude	0.71 (0.51)	0.39 (0.31)	0.07
Right P50 amplitude	2.02 (2.18)	1.55 (1.14)	0.50
Left P50 amplitude	3.08 (2.29)	1.70 (0.96)	0.08
Right N95 amplitude	1.67 (1.33)	1.98 (1.63)	0.56
Left N95 amplitude	2.03 (1.61)	1.79 (0.85)	0.65

Discussion

In our study cohort of normal healthy adults in Sri Lanka, there were no significant differences in PRVEP or PERG measures with regard to gender. These normal values can be used for comparison of corresponding visual electrophysiological data obtained using similar technical parameters in patients with various visual disorders in Sri Lankan popu-lation in the future. Most laboratories consider PRVEP latencies are abnormal, when the peak latencies and inter-ocular latency differences of PRVEP exceed the mean and standard deviation values as in an age-matched control sample from the normal population, subjected to same stimulating and recording parameters[14]. However, all the other retinal and ocular disorders have to be excluded by appropriate ophthal-mological examination before establishing a diagnosis of optic nerve dysfunction based on the PRVEP findings. Several studies have determined the nor-mative values of PRVEP measures at regional level and have evaluated factors affecting PRVEP waveforms. It was shown that P100 latency values in PRVEP were longer in males compared to the females[15,7]. However some studies have shown that there is no significant difference in P100 latency values with regard to gender[6,16]. With regard to amplitude values, it was shown that P100 amplitude was higher in females in some studies[6,7], while in another study done in England it was found that there is no effect of gender on the P100 amplitude values[16] (Table 5).

Table 5. Normal values of PRVEP in the present study and studies done in other countries

Study, Country	*Number of patients (males)*	*Mean age (SD) (range) in years*	*Overall P100 peak latency (msec) Mean (SD)*	*P100 peak latency (msec) Mean (SD)*		*Overall P100 amplitude (μV) Mean (SD)*	*P100 amplitude (μV) Mean (SD)*		*Remarks*
				Males	*Females*		*Males*	*Females*	
Gregori et al, 2006[15] Italy	54 (17)	30.15 (9.2) (20-55)	106.6 (4.94)	108.65 (5.0)	105.68 (4.6)	NR	NR	NR	P100 latency slightly shorter in females No difference with regard to head size in both sexes
Sharma R et al, 2015[7] India	100 (50)	(17-20)	NR	LE:93.21 (10.65) RE:93.41 (10.62)	LE:88.31 (8.79) RE:88.78 (8.9)	NR	RE:5.70 (0.48) LE: 5.68 (0.49)	RE:6.37 (0.66) LE:6.39 (0.66)	P100 latency significantly longer in males P100 amplitude is higher in females No difference with regard to head circumference in both sexes
Mahjoob M et al, 2018[6] Iran	59 (27)	22.5 (3.79)	RE: 102. 42 (5.37) LE: 100. 26 (3.58)	NR	NR	N75-P100 A RE:15.04 (6.26) LE:14.26 (6.90)	NR	NR	Amplitude of N75-P100 is larger in females compared to males.
Mitchell KW et al, 1987[16] England	68 (31)	61 (8.6)	NR	40-61 years: 111.1 (3.7) 62-80 years: 119.6 (4.7)	40-61 years: 109.2 (3.6) 62-80 years: 115.9 (5.7)	NR	40-61 years: 10.6 (4.0) 62-80 years: 15.5 (5.3)	40-61 years: 11.1 (5.1) 62-80 years: 15.5 (6.9)	No effect of gender and age on the parameters
Present study, Sri Lanka	50 (11)	37.2 (10.9) 20-62 years	RE:102. 38 (5.86) LE: 101. 61 (6.30)	RE:104. 04 (5.17) LE: 103. 54 (6.35)	RE:101. 91 (6.02) LE: 101. 07 (6.27)	RE: 5.01 (3.39) LE: 4.29 (3.32)	RE: 5.75 (3.50) LE:4.54 (2.56)	RE: 4.80 (3.38) LE: 4.23 (3.53)	No effect of gender on the parameters

NR: Not reported, A: amplitude, RE: right eye, LE: left eye

According to the above studies, it is obvious that the effect of gender on PRVEP measures is not conclusive and there are different findings which vary from country to country. This variation in the values of VEP in different studies may be due to the difference in recording instruments and ethnicity[6].

Different laboratories use different techniques to perform PERG measurements adhering to their own protocols based on the available resources. Though the standard guideline recommends the use of corneal electrodes for PERG, there are practical issues when using them. The use of these corneal electrodes requires better compliance from patients for stable recordings. In addition, prolonged recording sessions may cause corneal damage, optical degradation of the pattern stimulus and electrode instability. Use of skin electrodes taped on lower eye lids in PERG eliminates the above mentioned problems of corneal electrodes [3,17,18]. The disadvantages of skin electrodes are amplitude reduction and reduced signal to noise ratio when compared to that of corneal electrodes[18]. Interestingly, PERG measurements obtained from surface skin electrodes were comparable to those obtained from corneal electrodes[19]. Based on all these facts, we have used surface skin electrodes to record PERG in our study. There are very few normative studies done using PERG in the world. In those studies, it was found that there was no significant difference in latency and amplitude values of PERG with regard to gender as in our study[20, 21] (Table 6).

Table 6. Normal values of PERG in the present study and studies done in other countries

Study, year, Country	*Number of patients (males)*	*Mean age (SD) (range) in years*	*Type of electrodes*	*Overall P50 peak latency (msec)*	*P50 peak latency (msec) Mean (SD)*		*Overall P50 amplitude (μV) Mean (SD)*	*P50 amplitude (μV) Mean (SD)*		*Remarks*
					Males	*Females*		*Males*	*Females*	
Porciatti et al, 2004[3], USA	93 (56)	43.5 (18) (22-85)	Surface skin electrodes	NR	NR	NR	NR	NR	NR	Responses are compatible with the standard protocols done with corneal electrodes
Alves LD et al, 2010[21] Brazil	30 (15)	30.8 (8.7) (18-61)	Disposable fiber electrodes	60'-64.4 15'-65.6	NR	NR	P50-N95 60':3.8 15':2.8			No effect of gender on parameters at different stimulus sizes
Korth M. et al, 1989[21] Germany	147	(14-79)	DTL electrodes	<30y: 54.7 (48.5-60.6) 30-50y: 54 (47.6 -60.3) >50y:56.5 (50-65)	55 (48.5-61)	54 (49.4-60)	<30y: 4.9 (2.2-7.5) 30-50y: 4.9 (1.8-8.1) >50y:3.85 (1.8-5.95)	4.45 (1.7-7.1)	4.75 (2.2-7.3)	Increase in latency and reduction in amplitude with increasing age
Present study, Sri Lanka	50 (11)	37.2 (10.9) 20-62 years	Surface skin electrodes	RE: 51.93 (5.64) LE:52.57 (6.11)	RE: 55.32 (5.00) LE: 51.09 (6.9)	RE: 50.97 (5.49) LE: 52.99 (5.87)	RE: 1.65 (1.42) LE: 2.00 (1.45)	RE: 2.02 (2.18) LE: 3.08 (2.29)	RE: 1.55 (1.14) LE: 1.70 (0.96)	No effect of gender on parameters

NR: Not reported, A: amplitude, RE: right eye, LE: left eye

Considering all the above facts, it is evident that the normal values of PRVEP and PERG in a group of normal healthy adults in Sri Lanka would be of immense value as they can be used to compare the data of visual electrophysiological tests obtained under similar technical conditions in patients with different visual disorders in Sri Lanka in future studies.

The main limitation of our study is the small sample size. Since this is a part of an ongoing study, further studies using larger sample size are warranted to assess the influence of various other factors on PRVEP and PERG measures.

Conclusions

The normal values of PRVEP and PERG for normal healthy Sri Lankan adults of this study would be valuable to compare the visual electrophysiological data of Sri Lankan adult patients with different visual disorders in the future ophthalmological studies.

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