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Pattern reversal visual evoked potentials in adults: variability with age

Abstract

Purpose: Pattern reversal visual evoked potential (PRVEP) is an electrophysiological test for evaluating the visual pathway. This study measured the changes in the latencies and amplitudes of the PRVEP with age and gender in normal subjects.

Methods: Healthy participants (n=81; 162 total eyes), between the ages of 20 and 92 years were recruited for the study. Stimulation was performed monocularly with a high-contrast (>50%) black-white checkerboard pattern with a check size of 30° at a reversal rate of 2 Hz, a band-pass of 1-100 Hz, a sweep of 250 msec and an average of 150 stimulations in a dark room. Mean and standard deviations for three latencies (N75, P100 and N145) and the amplitude (N75-P100) for each decade were measured.

Results: There was a linear trend by age for all three latencies, indicating that the higher age groups had longer latencies. The latencies decreased in the 5th decade before increasing in the higher age groups. The amplitude of N75-P100 decreased with age. The P100 latencies were longer in males than females in all age groups and the difference increased with increasing age.

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Pattern reversal visual evoked potential (PRVEP) is an electrophysiological test aimed at evaluating the visual pathway. It is based upon stimulating the retina with pattern-reversal stimulation and recording from the occipital cortex using surface electrodes. The cortical wave is averaged against the cortical EEG activity, producing a reproducible wave with a certain latency and amplitude. This test was established in the 1930s [1]. Several attempts through the years have been made to standardize the criteria of the wave in relation to the stimulus size, contrast, pattern angle, light intensity and frequency of pattern reversal, as well as the subjects' age and gender [1,2].

In healthy subjects, PRVEP latencies appear to be significantly influenced by stimulus-related variables such as luminance, spatial frequency and contrast and subject distance from the light screen [2]. Some investigators have reported shorter PRVEP latencies in females than in males, attributing these differences to hormonal factors [3]. Other studies reported that the major determinants of difference in VEP latencies among gender was not gender but head size [4].

The normal values published in the literature do not cover all age categories. The purpose of this study is to standardize the results of the PRVEP with the new systems and software created for electrophysiological studies, as well as to produce reliable data (means and standard deviations) for each wave across the different age categories and to define the changes in latencies and amplitudes with age and gender.

Materials and Methods

The PRVEP was measured on 81 healthy participants, a total of 162 eyes, between the ages of 20 and 92 years. The average number of subjects per decade was limited to 23 participants. The study was performed on NICOLET Viking IV, as described in the literature and according to the recommendations of the International Society for Clinical Electrophysiology of Vision on performing visual evoked potentials [5]. The study has been approved by, and carried out according to the instructions of, the authors' institutional ethics committee. Stimulation was performed monocularly, with the second eye covered, with a 1° visual angle, black-to-white checkerboard pattern from a 15 inch CRT screen, with a luminance of 50 cd/m² at a rate of 2 reversals per second, a bandpass of 1-100 Hz and a sweep of 250 msec. The response was the average of 150 consecutive stimulations. The final response was analyzed with measurement of the positive and negative peaks N75, P100, N145 (in milliseconds) and the N75-P100 amplitude (in microvolts).

The stimulation was performed in a dark and quiet room, with the patient seated 100 centimeters from the screen. To correct for refractive error, the patient was examined with his eyeglasses on. Each eye was stimulated separately with the other eye covered. The recording electrode was placed at Oz, the reference electrode at Fz, and the ground electrode at the forehead according to the International 10/20 system [2]. The patients were allowed to rest for few minutes before the second eye was studied [1,5]. The test was performed by the same trained technicians and interpreted by a trained clinical neurophysiologist (Dr. R. Sawaya).

The participants were recruited from the Society of Members of the American University of Beirut Medical Center in Lebanon where the study was performed. The participants were healthy students, teachers, staff and retired personnel or their parents. All the participants were Caucasian. They were informed of the procedure and provided consent. They were screened by history and physical examination and recruited only if healthy with normal visual acuity and no medical illnesses and not on medications that may interfere with vision or the central nervous system, such as diabetes mellitus, hypertension, coronary artery disease, malignancies or macular degeneration. The elderly participants did not have cataracts as assessed by ophthalmoscopy, or had undergone lens replacement surgery. Smoking and alcohol habits were not corrected for.

Statistical analysis

Means and standard deviations for the N75, P100, N145 latencies and N75-P100 amplitude were calculated for each age group. To compare the mean latency and amplitude across age groups, a one-way ANOVA was conducted for each of the latencies and amplitude measurements. Welch's F test was used to make adjustments for the differences in group variances due to the heterogeneity of variances across groups. Robust *post hoc* tests were then carried out to determine which pairs of age groups differed significantly in latency and amplitude. Four *post hoc* tests were carried out, one for each of the variables (N75, P100, N145 latencies and N75-P100 amplitude), corrected for multiple comparisons.

A trend analysis was also conducted to determine whether there was a general trend in latency and amplitude with age. Linear and polynomial trends were investigated to determine whether the relationship between age and latencies and amplitude were linear or curvilinear. Linear models were built to investigate whether gender is a predictor of N75, P100 and N145 latencies and N75-P100 amplitude. The main effects of gender and age group by gender interaction were investigated.

TABLE 1. Patient demographics

Study group age	Mean age (SD)	Median age (years)	Age range (years)	M, F	L, R
20s	25.21 (2.86)	25.5	20 – 29	8, 20	14, 14
30s	34.5 (3.05)	34.5	30 – 39	16, 8	12, 12
40s	44.09 (3.1)	44.00	40 – 49	10, 12	11, 11
50s	55.00 (2.8)	55.00	50 – 59	14, 12	13, 13
60s	64.5 (2.95)	64.5	60 – 69	14, 6	10, 10
70s	74.5 (2.95)	74.5	70 – 79	14, 6	10, 10
≥ 80	85.55 (3.84)	85.00	80 – 92	14, 8	11, 11

Abbreviations: M, F: number of eyes in males and female in study population; L, R: number of left and right eyes in study population; SD: standard deviation

TABLE 2. Means and standard deviations of latencies and amplitude for each gender and the total number of subjects in the different age groups.

	N 75 latency (ms)			P100 latency (ms)			N145 latency (ms)			Amplitude (μv)		
	Mean (SD)			Mean (SD)			Mean (SD)			Mean (SD)		
Age groups	M	F	All	M	F	All	M	F	All	M	F	All
20s	78	73.8	74.9 (3.96)	108.6	104.6	105.8 (3.57)	149	146.2	146.9 (12.13)	9.55	13.2	11.3 (5.79)
30s	78	76.4	77.4 (4.64)	108.1	106.5	107.5 (3.56)	152.9	141.3	149.1 (9.98)	8.4	8.1	8.4 (3.86)
40s	71.4	73.7	72.7 (3.46)	105.7	101.2	103.3 (4.5)	139	134.3	136.4 (9.76)	6.6	8.3	7.6 (3.67)
50s	72.4	75.9	74.1 (5.39)	104.4	107.3	105.7 (6.67)	141.5	145.8	143.5 (12.87)	6.4	7.3	6.7 (3.18)
60s	77.5	76.7	77.3 (9.73)	114	108.8	112.4 (6.23)	153	140.2	149.2 (9.78)	5.9	5	5.6 (2.38)
70s	78.6	79.8	78.9 (7.09)	116.5	111.8	115.1 (5.53)	153.6	141.7	150.1 (14.57)	5.5	5.7	5.5 (2.41)
≥ 80s	88.4	75.5	83.6 (9.74)	121.5	112	118.1 (7.79)	156.9	148.8	153.9 (11.23)	4.6	8.2	5.9 (3.52)

Abbreviations: M, male; F, female.

TABLE 3. Comparison of mean P100 latencies in males and females

Age (years)	P100 latency in males	P100 latency in females	Reference
17-35	95.3	91	Tandon et al., 1989
20-55	108	105.6	Gregori et al., 2006
17-21	93.4	88.78	Sharma et al., 2015
20-75		94.7	Kenneth et al., 1964
20-92	108.9	106.5	data from this study

FIGURE 1. : P100 latency in males (triangles; grey line) and females (circles; full line) with age

Various techniques were used to ensure our dependent variables approximated a normal distribution. Normality was assessed visually (histograms and QQ plots) and statistically (Shapiro-Wilk normality test). The distributions of N75, P100 and N145 latencies approximated the Gaussian distribution. The distribution of N75-P100 latency was slightly positively skewed, but this seem to be driven by outliers (amplitudes >16). When the outliers were removed, the distribution appeared closer to normality. The Shapiro-Wilk tests supported normality ($P>0.05$) for the N75 and N145 latencies (after removal of an outlier for the latter) and a significant, but minor, deviation from normality for the P100 latency ($P<0.05$). Due to our large sample ($n=162$), we relied more strongly on the visual representations of normality, as statistical tests become sensitive to even minute deviations from normality in large samples.

FIGURE 2. N75 (lower curve), P100 (middle curve) and N145 (upper curve) latencies across age revealing the “dip” in the 4th decade

Results

The number of patients studied, distribution over decades, mean and standard deviations, gender differences and other demographics are presented in Table 1. The mean and standard deviation of each latency and the N75-P100 amplitude were calculated for each age category (Table 2).

ANOVA tests revealed significant differences across the groups in N75 latency ($P<0.0001$), P100 latency ($P<0.0001$) and N145 latency ($P<0.0001$). Results also revealed a significant group difference in N75-P100 amplitude ($P=0.0002$).

Post hoc tests revealed significant differences in the N75 latency between the following age groups: 20s vs ≥ 80 s ($P=0.0004$); 40s vs 70s ($P<0.0001$); 40s vs ≥ 80 s ($P<0.0001$); 50s vs ≥ 80 s ($P<0.0001$); and 30s vs 40s ($P=0.0004$). For all the above comparisons, except 30s vs 40s, the higher age groups had a significantly longer N75 latency.

For the P100 latency, significant differences were found between the following age groups: 20s vs 60s ($P=0.0008$); 20s vs 70s ($P<0.0001$); 20s vs ≥ 80 s ($P<0.0001$); 30s vs 70s ($P<0.0001$); 30s vs 80s ($P<0.0001$); 40s vs 60s ($P<0.0001$); 40s

FIGURE 3. P-100 latency in msec vs age in three previous studies. All studies reveal a “dip” in the latency around the 4th decade. Adapted from a) Celestia *et al.*, 1987 (ref 6); b) De Graff *et al.*, 1985 (ref 7); c) Chiappa 1989 (ref 2) and d) our data in this study.

vs 70s ($P<0.0001$); 40s vs ≥ 80 ($P<0.0001$); 50s vs 60s ($P=0.0008$); 50s vs 70s ($P<0.0001$); 50s vs 80s ($P<0.0001$); and 30s vs 40s ($P=0.0004$). In all of the above comparisons except 30s vs 40s, the higher age groups had longer P100 latencies.

For the N145 latency, significant differences were found between the following groups: 40s vs 60s ($P<0.0001$); 40s vs 70s ($P<0.0001$); 40s vs ≥ 80 s ($P<0.0001$); 20s vs 40s ($P<0.0001$); and 30s vs 40s ($P<0.0001$). In all of the above comparisons, except 20s vs 40s and 30s vs 40s, the higher age groups had longer N145 latencies.

Trend analyses showed that there was a significant linear trend by age for the N75 ($B=6.36$, $SE = 1.35$, $P<0.0001$), P100 ($B=11.56$, $SE=1.14$, $P<0.0001$) and N145 ($B=6.75$, $SE=2.39$, $P=0.005$) latencies, indicating that the higher age groups had longer latencies. Due to the unexpected finding of shorter latencies in the 40s age group compared with the 30s

across all latencies studied, polynomial trends were also investigated to determine whether the 40s age group deviated from the upward linear trend. Results showed a significant quadratic trend for the N75 ($B=5.1$, $SE = 1.34$, $P=0.0002$), P100 ($B=5.36$, $SE=1.13$, $P<0.0001$) and N145 ($B=8.07$, $SE=2.37$, $P=0.0008$) latencies where the straight line shows a “dip” in this age group. Trend analyses for the N75-P100 amplitude showed a significant downward linear ($B=-4.48$, $SE=.79$, $P<0.0001$) and quadratic trend ($B=2.1$, $SE=.78$, $P=0.008$), with shorter amplitudes at higher age groups. The trends suggest a downward inclination that tended to plateau after the age of 60. This suggests that although generally older age groups have longer latencies compared with their younger counterparts, hence the upward linear trend, the 40s age group exhibited a shorter latency compared with the younger age groups (20s and 30s). There is also a quadratic trend by age for the amplitude: generally the older age groups had lower

amplitudes, but the downward slope was steeper among the younger groups (i.e., the decrease in amplitude among younger groups was greater). The amplitudes were similar among groups aged 60 years and older.

The secondary aim of the study was to determine whether the differences in N75, P100 and N145 latencies across age groups also differ between males and females. Results showed a marginally significant group by gender interaction ($P=0.08$) for the N75 latency. For the P100, results show a significant group by gender interaction ($P=0.01$), with the older males having higher P100 latencies compared with the older females. No significant main effect of gender or interaction with group was found for the N145 latency ($P>0.05$). No effect of gender or interaction with group was found for the N75-P100 amplitude ($P>0.05$).

Discussion

The studies of normative data for PRVEP in the literature are few and quite deficient in methodology [7-13]. The values for P100 latency in the literature range between 90 and 108 msec, but these numbers were not correlated with age categories, included a small number of participants and most of the studies were performed only on subjects in the second and third decades (Table 2). Our study defines the mean latencies and standard deviations of N75, P100, and N145, and the amplitude of N75-P100, for each decade allowing standardization of the numbers for clinical purposes (Table 1). We found the latencies to be consistent between participants in each decade, with a relatively small standard deviation.

Our data reveals progressive increase in the latencies of all three waves with increasing age. The N75 increased from 74.93 msec in the second decade to 83.61 in the 8th decade. The P100 increased from 105.75 to 118.05 msec and the N145 increased from 146.96 msec to 153.95 msec. Furthermore, there was a progressive decrease in the N75-P100 amplitude with age from 11.26 microvolts in the second decade to 5.9 microvolts in the 8th decade.

Age-related increase in latency and decrease in amplitude of the VEP components has been reported previously in the literature and may be related to senescence and cell-death of neurons of the visual system [7, 14-16]. Evidence is accumulating that cell-death occurs throughout the visual system, and that cell axon loss in the optic nerve is prominent after the seventh decade [15]. Other age-associated changes occurring in the optic nerve are corpora amylacea, deposit of lipofuscin in the astrocytes and axonal swelling [15]. A statistically significant decrease in nerve fiber population of the optic nerve has been demonstrated in aged individuals [16]

and this age effect may account for a loss of approximately 400,000 fibers during a 70 year life span [16]. Devaney and Johnson reported a 50% reduction in neuronal density from age 20 to age 80 years in the visual cortex [17].

Variability of the latencies with gender, age and ethnicity has been reported in the literature, with contradictory results. A PRVEP study performed in 1985 on 276 normal adult subjects aged 15 – 73 years revealed shorter latency of the P100 in Black subjects compared with Caucasian [8]. It has been confirmed that PRVEP have a shorter latency in females than in males, but this difference was attributed to head size rather than gender; males and females with similar head circumference had similar P100 latencies [9]. Head size has a weak, though significant, influence on the latency of PRVEP with 30% variability in head size yielding 3% variability in VEP latencies [9]. This gender difference might also be due to shorter axial eye length in females than in males [11]. A study comparing PRVEP latencies in males and females reported in 2015 revealed longer P100 latencies in males than females, with higher amplitudes in females compared with males, and no significant correlation with head circumference [10]. The weakness of this study is that the population was limited to subject between the ages of 17 and 20 years [10]. Another study found shorter P100 latencies in pregnant woman, raising the possibility of effect of hormonal changes on the VEP latencies [18].

Our study showed a clear gender difference with age, with males having longer P100 latency than females. Moreover, the difference increases with age, becoming more pronounced between the ages of 50 and 80 years (Figure 1).

The most astonishing finding in this study is that we found a clear drop in the latency of each wave in the fifth decade. The N75 latency drops from 74.93 msec in the third decade to 72.67 in the 5th decade. The P100 latency drops from 105.75 msec to 103.25 msec and the N145 latency drops from 146.96 to 136.41 msec. This “dip” in latency is revealed in the mean and standard deviation of each of the waves. This drop in latency reverted by around 50% in the sixth decade, and then progressively followed an ascending trend to the eighth decade (Table 1 and Figure 2).

This characteristic dip has been observed, but not commented upon, in previous studies in the literature (Figure 3) [2,7, 8]. Chiappa reported age-related changes in latency of the P100 component of PRVEP, revealing a dip in the latency in the fifth decade without presenting an explanation [2]. A study on 276 normal adults, aged 15 to 73 years, revealed a non-linear relationship of the major positive components with age, with the shortest latency being at about 35 years of age [8].

A similar study done in 1987 on 112 normal individuals aged between 20 and 75 years revealed that the slope for the aging effect on N70 was almost linear, while for the P100 was U shaped [7]. Shaw also noted a different effect of aging on the N70 and P100 components of the VEPs [12].

The exact explanation of this dip in latencies of the PRVEP waves in the 5th decade has not yet been determined. It cannot be attributed to maturity of myelination of the optic tracts as myelination of the optic nerves starts *in utero*, extends from the brain to the globe, is observed first at 32 weeks gestation and is completed by the second year of life [6,19]. Shaw discusses the possibility that changes in P100 are related to extra-striate areas [12]. It may be postulated that the decrease in latency in males and females in the 5th decade could be secondary to initiation of cortical shrinkage and decrease in cerebral volume. It has been reported that 50% of neurons degenerate between the third and eighth decades [17]. The eventual increase in latency after the 6th decade and progressive increase in the 7th and 8th decade, and the commensurate decrease in the amplitude of the N75 – P100 wave, could be secondary to slow neuronal death of the retinal and cortical neurons and axonal degeneration of the optic tracts [15]. Presbyopia and changes in lens characteristics may alter the VEP latencies in the 5th decade, but this should, in principle, cause a delay, rather than a decrease, in the latencies and these changes should revert as age progresses. Furthermore, patients were examined with their eyeglasses on to counteract the possibility of decreased vision.

In summary, we defined specific latencies and amplitudes for the N75, P100 and N145 cortical wave of the PRVEP for the different age categories in our population (Table 2). We found a definite increase in the latencies of all components and a decrease in the major wave amplitude with increasing age, which we attribute to neuronal death and optic tract degeneration. We found a definite gender difference, with longer latencies in the PRVEP in males compared with females in all age groups, with the difference becoming more pronounced with age. We defined a dip in the latencies of all the components of the PRVEP wave at the 4th decade and suggest that this could be secondary to the first stages of cerebral volume loss before the development of neuronal death and axonal loss, which is reflected in the loss of amplitude and delayed latencies of the PRVEP with increasing age.

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References

1. Vernon Odom J, Bach M, Brigell M, Holder G, McCulloch D, Tormene AP. ISCEV standard for clinical visual evoked potentials (2009 update). *Doc Ophthalmol*.2010;120:111-9.
2. Chiappa KH. Pattern-shift visual evoked potentials: methodology. In Chiappa KH. *Evoked potentials in clinical medicine*, 2nd ed, Raven press, New York 1989; Chapter 2, 37-109.
3. Emmerson-Hanover R, Shearer DE, Creel DJ, Dustman RE. Pattern reversal evoked potentials: gender differences and age-related changes in amplitude and latency. *Electroencephalogr Clin Neurophysiol*. 1994; 92: 93-101
4. Guthkelch AN, Bursick D, Sclabassi RJ. The relationship of the latency of the visual P100 wave to gender and head size. *Electroencephalogr Clin Neurophysiol*. 1987; 68:219-22.
5. Vernon Odom j, Bach M, Brigell M, Holder G, McCulloch D, Mizota A, Tormene AP. ISCEV standard for clinical visual evoked potentials: (2016 update). *Doc Ophthalmol* 2016;133:1-9.
6. Drislane FW. Visual Evoked Potentials. In Blum AS, Rutkove SB, editors. *The Clinical Neurophysiology Primer*. Humana Press Inc;2007;25:461-73.
7. Celesia G, Kaufman D, Cone S. Effects of age and gender on pattern electroretinograms and visual evoked potentials. *Electroencephal and Clinical Neurophys* 1987;68:161-71
8. de Graaf AS, Lombard CJ, Hart GL. Standards for the assessment of the visual evoked potentials in an ethnically heterogeneous adult population. *S Afr Med J* 1985;16:243-5
9. Gregori B, Pro S, Bombelli F, La Riccia M, Accornero N. VEP latency: sex and head size. *Clinical Neurophys*. 2006;117:1154-7
10. Sharma R, Joshi S, Singh KD, Kumar A. Visual evoked potentials: normative values and gender differences. *Journal of clinical and diagnostic research*. 2015;9:12-5.
11. Tandon OP, Sharma KN. Visual evoked potential in young adults: a normative study. *Indian J Physiol Pharmacol*.1989;33:247-9.
12. Shaw N A. Changes in the cortical components of the visual evoked potential with age in man. *Aust J Exp Biol Med Sci*.1984; 62:771-8
13. Kooi KA, Bagchi BK. Visual evoked responses in man: normative data. *Ann NY Acad Sci*.1964;112: 254-69.

14. Weale RA. Senile ocular changes, cell death, and vision. In R. Sekuler, D. Kline, and K. Dismukes (Eds.) *Aging and Human visual Function* New York: Liss. 1982; 161-171.
15. Dolman C L, McCornnck A Q, Drance S M. Aging of the optic nerve. *Arch Ophthal* 1980; 98:2053-2058.
16. Balazsl AG, Rootman J, Drance S M, Schulzer M, Douglas G R. The effect of age on the nerve fiber population of the human optic nerve. *Amer J Ophthal*. 1984; 97:760-6.
17. Devaney KO, Johnson HA. Neuron loss in the aging visual cortex of man. *J Gerontol*. 1980;35: 836-41.
18. Marsh MS, Smith S. Differences in the pattern visual evoked potential between pregnant and non-pregnant women. *Electroencephalogr Clin Neurophysiol*. 1994;92:102-6.
19. Magoon EH, Robb RM. Development of myelin in human optic nerve and tract, a light and electron microscopic study. *Arch ophthalmol*. 1981;99:655-9.