A Comparison between Multifocal and Conventional VEP Latency Changes Secondary to Glaucomatous Damage

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PURPOSE. To compare latencies of conventional visual evoked potentials (cVEPs) and multifocal VEPs (mfVEPs) in the same patients. Previous reports of prolonged cVEP latency suggest a vehicle for detecting abnormal ganglion cells and for monitoring neuroprotection.

METHODS. Seventy-five glaucomatous eyes (47 patients), 75 eyes with suspected glaucoma (46 patients), and 41 control eyes (22 subjects) underwent achromat standardized automated perimetry and mfVEP and cVEP testing. The mfVEP stimulus was a scaled dart board with 60 sectors; each sector was a pattern-reversing checkerboard. The cVEP stimulus was a reversing checkerboard with checks of either 15 minutes or 60 minutes in width.

RESULTS. Relatively few glaucomatous eyes had latencies that fell outside the range of control eyes, and there was little difference between the cVEP and mfVEP results. In the glaucoma group, 12.3% (15 minutes cVEP), 8% (60 minutes cVEP), and 17.3% (mfVEP) of the eyes and 5.3% (15 minutes cVEP), 6.7% (60 minutes cVEP), and 5.3% (mfVEP) of the suspect eyes exceeded the normal range. The glaucomatous eyes had, on average, relatively small increases in latency, compared with the control or suspect groups. Further, the latency of both the mfVEP and cVEP bore no obvious relationship to the mean deviation of the visual field.

CONCLUSIONS. Contrary to previous reports, prolonged VEP delays were present in a minority of patients with glaucoma. Either a delayed VEP is not a good indicator of damaged, as opposed to dead, retinal ganglion cells, or there are relatively few patients who exhibit evidence of damaged ganglion cells.

Neuroprotection is currently an avidly sought goal for the treatment of patients with glaucoma. If affected retinal ganglion cells (RGCs) are alive, but functioning abnormally, then neuroprotection becomes a viable therapy. The latency of the conventional pattern-reversal visual evoked potential (cVEP) has been reported to increase markedly with glaucomatous damage,1–4 and there is evidence that citicoline treatment can improve the latency of the cVEP in these patients.5–8 Given the potential usefulness of cVEP latency as a measure of RGC health, it is important to understand the prevalence of abnormal cVEP latency among patients with glaucoma.

Although earlier work1–3 is consistent with delayed responses due to glaucoma, the recent work by Parisi et al.4 provides the clearest evidence. The mean latency of their open angle glaucoma (OAG) group was 27.8 ms longer than that of their control group. Further, all 84 of their patients with OAG showed cVEP latencies that were longer than that of any of the 80 control subjects. In other words, using the upper limit of normal latencies as the criterion measure yielded a sensitivity and specificity of 100%.

These results appear to conflict with measures of local VEP activity provided by the multifocal visual evoked potential (mfVEP). The mfVEP shows relatively modest increases in latency in patients with glaucoma (Klistorner A, et al. IOVS 2002;43:ARVO E-Abstract 2165).9,10 Recently, Rodarte et al.14 measured the latency of the mfVEP in 50 patients with glaucoma and found that the delays were modest (<5.7 ms) and involved fewer than one third of the patients. Further, the sensitivity (<38%) was poor. In other words, there was considerable overlap between the latencies of the glaucoma and control groups. Given the recent results of Parisi et al.,4 this overlap is surprising, as one would expect that the mfVEP, being a measure of local response, would be more likely to detect latency changes.

Because of the potential importance of delayed VEPs, both in detecting glaucomatous damage and in monitoring neuroprotective therapies, this apparent discrepancy must be resolved. In the present study, cVEPs and mfVEPs were recorded in the same patients, and the effects of glaucoma on latency were compared.

METHODS

Subjects

Subjects underwent a full ophthalmic examination including visual acuity, slit lamp biomicroscopy, achromatic automated perimetry, stereoscopic optic nerve head photography, and mfVEP and cVEP testing. Standard, full-threshold or SITA-standard 24-2 automated perimetry was performed (Humphrey Field Analyzer II; Carl Zeiss Meditec, Inc., Dublin, CA). All subjects had reliable visual fields with fewer than 33% fixation losses, false positives, and false negatives. None of the subjects enrolled in this study had other known abnormalities of the visual system besides the ones studied. Eyes were excluded if visual acuity was worse than 20/30, the pupil diameter was smaller than 2 mm, or...
refractive error exceeded ±6 D. The eyes of 115 subjects—22 control subjects (41 eyes) and 93 patients (150 eyes)—were placed into three groups as will be described later. There was virtually no overlap with the data in our recently published study,10 which examined the latency of the mfVEP in 47 normal control subjects and 50 patients with glaucoma. These two studies had only one mfVEP recording from a single control subject in common. All the other data were obtained after the data collection by Rodarte et al.10 was completed. However, in addition to the single control, nine patients, approximately 10% of the patients in the present study, were retested on all tests for the present study.

**Glaucoma (GL).** Seventy-five eyes of 47 patients comprised the glaucoma group. This group consisted of patients with a clinical diagnosis of primary open-angle glaucoma (n = 17), normal-tension glaucoma (n = 19), chronic angle-closure glaucoma (n = 4), pigmentary glaucoma (n = 3), exfoliative glaucoma (n = 3), and juvenile open-angle glaucoma (n = 1). Patients ranged in age from 29 to 78 years (average age 61 ± 12 years). Eyes were classified as glaucomatous if there was both an abnormal-appearing disc (including cup-disc [C/D] asymmetry between fellow eyes of greater than 0.2, rim thinning, notching, C/D ratio >0.6, or retinal nerve fiber defects) and an abnormal 24-2 visual field (VF). The 24-2 visual field was classified as abnormal if the glaucoma hemifield test (GHT) result was abnormal and/or if there was a significant cluster of abnormal points in the total deviation plot in either hemifield. A significant cluster was defined as the presence of two or more contiguous points at P < 0.01, or three or more contiguous points at P < 0.05 with at least one point at P < 0.01. Only one point could be on the rim. Ninety-three percent of the eyes were abnormal based on the GHT, whereas 7% showed a GHT within normal limits or borderline and a significant cluster. Fellow eyes that did not meet these criteria were suspected of having glaucoma. The average MD of the 24-2 VF for the glaucomatous eyes was −5.9 ± 4.4 dB. Sixty-nine percent (52/75) of the eyes had abnormal mfVEP amplitudes. An abnormal amplitude was defined, as in previous work,11 as the presence of two or more contiguous locations within a hemifield with a probability (P) < 5%, one of which had P < 1%, or two locations with P < 1%.

**Suspected Glaucoma.** This group (GLS) included 75 eyes of 46 patients. Forty-one of these eyes (27 patients) had a normal 24-2 VF (normal GHT and no significant clusters), but suspicious or abnormal optic disc appearance (e.g., large cup [i.e., C/D > 0.6], nerve fiber layer thinning, notching, or hemorrhage) and/or asymmetric discs (i.e., C/D ratio difference > 0.2). Thirty-four eyes (19 patients) had a normal VF result and optic disc appearance (as defined above), but one or more of the following exfoliation syndrome (XFS), pigment dispersion syndrome (PDS), ocular hypertension (intraocular pressures >21 mm Hg) or positive family history for glaucoma. Patients ranged in age from 14 to 78 years (average age, 60 ± 16). The average mean deviation [MD] of the 24-2 VF was −1.5 ± 1.7 dB.

**Control Group.** Forty-one eyes of 22 healthy subjects (C) with normal ophthalmic examination and normal VF, as described earlier, were included. Three eyes were excluded from the study because one eye had a refractive error of −7 D, another eye had a macular scar and abnormal VF results, and the third eye had a GHT result outside normal limits. The subjects ranged in age from 18 to 71 years (average age, 43 ± 20). The average MD of the 24-2 VF was −0.88 ± 1.7 dB. Informed consent was obtained from all subjects before their participation. Procedures adhered to the tenets of the Declaration of Helsinki, and the protocol was approved by the committee of the Institutional Board of Research Associated of Columbia University.

**Stimuli and Recording**

**mfVEP.** Figure 1 is a schematic of the stimulus array produced by the VEP software (VERIS Dart Board 60 with Pattern; Electro-Diagnostic Imaging, Inc., EDI, San Mateo, CA). The stimulus display, viewed through natural pupils with the appropriate refractive correction, consisted of 60 sectors, each with 16 checks: 8 white (200 cd/m²) and 8 black (<1 cd/m²). The sectors were scaled for cortical magnification with the central 12 sectors falling within the central 5.2° (diameter). The entire dart board display subtended 44.4° in diameter at the viewing distance. The stimulus array was displayed on a black and white monitor driven at a frame rate of 75 Hz. On each frame change, each of the 16-element sectors had a 0.5 probability of reversing in contrast or staying the same. The mean luminance was 100 cd/m² with a contrast close to 100%. Stimulation was monocular after occlusion of the other eye. For more details about the mfVEP see Baseler et al.12 and Hood and Greenstein.13

The recording procedures have been described in detail in previous publications (e.g., Hood et al.14,15; Hood and Greenstein13). Briefly, three channels of continuous VEP (EEG) records were obtained by using gold cup electrodes. For the midline channel, electrodes were placed 4 cm above the inion (active), at the inion (reference), and on the forehead (ground). For the other two channels, the same ground and reference electrodes were used, but the active electrodes were placed 1 cm above and 4 cm lateral to the inion on either side. By taking the difference between pairs of channels, three additional “derived” channels were obtained, resulting in effectively six channels of recording. The records were amplified with the high- and low-frequency cutoffs set at 3 and 100 Hz (one-half amplitude; Grass Instruments preamplifier PS11J, Quincy, MA), and were sampled at 1200 Hz (every 0.83 ms). The impedance was < 5 k for all subjects. In a single session, two 7-minute recordings were obtained from monocular stimulation of each eye (ABBA order). Second-order response components were extracted (VERIS 4.3 software; EDI).

**cVEP.** The cVEP test was run after completion of the mfVEP. The conditions of stimulation and recording followed the ISCEV guidelines.16 The display, a reversing checkerboard, was 48° in diameter and had a mean luminance of 70 cd/m² and a contrast close to 100% (produced by Espion System Software V4.0.12, 2004; Diagnosys, Boston, MA). Two checkerboard stimuli with check sizes of 15 and 60 minutes were used, each reversed at two reversals per second. Subjects were refracted for the viewing distance and wore the appropriate refractive correction. The stimuli were viewed through natural pupils. Recordings were obtained for each eye separately, the nontested eye was occluded. A small red dot was placed at the center of the stimulus to aid in fixation. The cVEPs were recorded with the system (Espion; Diagnosys), with cutoff frequencies of 3 and 100 Hz. A reference electrode, Fz was added. Impedance was kept below 5 k. For each eye and each check size, two recordings were obtained between the inion + 4-cm electrode and Fz with a forehead electrode serving as the ground.
Analysis of Latency

mfVEP. The mfVEP responses from each channel were exported from the system (VERIS; EDI) and the two recordings from each eye were averaged. This averaging, as well as all other analyses, was computed with programs written in commercial software (MatLab; The MathWorks Inc., Natick, MA). Analyses were performed on the “best” responses—that is, those with the largest signal-to-noise ratio (SNR), from the six “channels” as previously described (see Refs. 13,15). Monocular latencies were measured and analyzed as previously described.\(^{17}\) Briefly, to obtain a measure of the monocular latency of responses, a cross-correlation was calculated between the patient’s response and a template. A template was created for each location, eye, and channel, and derived from averaging the responses of 100 normal subjects.\(^{17,18}\) The relative mfVEP latency shown in Figures 3 and 5 is the shift in time that maximized the cross-correlation with the template, with amplitude scaling of the template as is typically done. Records with small signal-to-noise ratios (<0.23 log unit) or with cross-correlation values of less than 0 were excluded as previously described.\(^{17}\)

cVEP. Typically in the literature, the method for obtaining the latency of the cVEP’s P100 is not specified beyond a general statement that the peak was identified and the latency measured. Parisi et al.,\(^4\) for example, averaged the waves from separate sessions after checking for “repeatability” and then determined the amplitude and latency of P100 “directly on the display records by means of a pair of cursors.” These methods work, in general, because the location of P100 is usually, but not always, easy to determine.

To obtain the latency of the peak near 100 ms (P100), we exported the responses for both eyes and both check sizes (60 and 15 minutes) to a graphics program for analysis. The two responses for each condition were averaged after visual inspection to assure that they were reasonably similar. In one pair of responses (15 minutes cVEP for one of the GL patients), there was no sign of a P100, and this pair was omitted. In one of the remaining 580 pairs of cVEP responses, one of the responses was discarded because it was aberrant (i.e., there was no clear cVEP with a definable peak), and in a second pair only one response was recorded because of an equipment malfunction. Figure 2 shows examples of averaged cVEP responses from 7 controls and 7 patients from the GL group. The latency of the four averaged responses (two eyes \(\times\) two check sizes) for each individual was measured by using the following technique. In nearly all cases, a P100 peak was obvious in a time window between 75 and 150 ms, and its latency measured with a cursor. In a few cases when it was not obvious, we required the peak to be in a similar place in both runs. In the relatively rare cases in which the peak was still ambiguous, the other channels of recording were examined to see whether the peak was in a similar location. In all cases, when the peak of P100 was not localizable to a single time, two lines were drawn, each line representing an estimated best fit to the rising or declining phases of the wave. The point of intersection of these lines provided the latency measure.

RESULTS

mfVEP Latency

Each data point in Figure 3 represents the relative mfVEP latency in a single eye plotted against the MD of the 24-2 HVF for that eye. To obtain the relative mfVEP latency, each of the 60 mfVEP responses was compared to a template from a normative group (see the Methods section), and the average difference in latency (shift of the scaled template) was calculated for that eye and plotted as one of the points in Figure 3. First, there was no clear relationship between the latency of the mfVEP and the MD of the VF. Second, although the GL group had a mean relative latency (4.1 ms) that was longer than that (1.7 ms) of the control group the difference was relatively small, consistent with past work (Klistermar A, et al. \(\text{IOVS}\) 2002;43:ARVO E-Abstract 2165).\(^9,10\) The GLS group showed a mean relative latency of 2.7 ms (Table 1).

Recall that a recent cVEP study\(^5\) found no overlap between GL and control groups. In contrast, there was considerable overlap in our data (Fig. 3) with only 17.3% of the GL eyes falling outside the limits (dashed line) of the control values.

cVEP Latency

Each point in Figure 4 represents the cVEP latency for a single eye plotted against the MD of the 24-2 for that eye. For each eye, the latency of the cVEP response (P100) was measured.
(see the Methods section) and plotted as one of the points in Figure 4. First, as with the mfVEP, for both the 60 minutes (Fig. 4A) and 15 minutes (Fig. 4B) check sizes, there was no clear relationship between the latency of the cVEP and the MD of the HVF. Second, there was a relatively small difference, less than 7 ms, between the mean latency of the GL group, 107.7 ms (60 minutes) and 120.2 ms (15 minutes), and that of the control group, 101.9 ms (60 minutes) and 113.9 ms (15 minutes). The latencies in the GLS group fell between these values: 105.1 ms (60 minutes) and 120.1 ms (15 minutes; Table 1).

More to the point, there was considerable overlap of the latencies for the GL and control groups. Only 8% (60 minutes) and 12.3% (15 minutes) of the GL eyes fell above the range of the control subjects (Fig. 4; dashed line). This was slightly less than the 17.3% falling above the control values for the mfVEP (Fig. 3) and substantially below the 100% value reported recently for the cVEP.4 This discrepancy will be discussed in the following sections.

A Comparison of mfVEP and cVEP Latencies

Figure 5 provides a comparison between the relative mfVEP latency and the latency of the cVEP for the 60 minutes (Fig. 5A) and 15 minutes (Fig. 5B) stimuli. The range of 60 minutes cVEP latencies tended to be greater than the range of changes in mfVEP latency (44 vs. 24.6 ms in the GL group). The cVEP and mfVEP latencies showed modest correlations in all groups for the 60 minutes stimulus (Spearman’s rank correlation coefficient $r_s = 0.58$ [GL], $0.57$ [GLS] and $0.83$ [C]) and the 15 minutes stimulus ($r_s = 0.56$ [GL], $0.53$ [GLS] and $0.39$ [C]). Similar values were obtained when the latencies of the two cVEP stimuli were compared ($r_s = 0.56$ [GL], $0.42$ [GLS], and $0.40$ [C]). That is, the correlation between the cVEP and the mfVEP was about as good as between the cVEP for the two check sizes.

DISCUSSION

This study was motivated by previous studies, some reporting that glaucoma had a relatively small effect on the latency of the mfVEP (Klistorner A, et al. IOVS 2002;43:ARVO E-Abstract 2165),9,10 while another suggested that glaucoma could have a major effect on the latency of the conventional pattern-reversal visual evoked potential (cVEP).4 The present study examined this apparent discrepancy by comparing cVEP and mfVEP latencies in the same group of patients. In general, our cVEPs

![Figure 4](image-url)

**Figure 4.** The cVEP latency versus MD of the 24-2 VF for individual eyes of the control (+), GL, and GLS groups. Dashed line: the upper limit for the control subjects; (A) 60 minutes check size; (B) 15 minutes check size.

![Figure 5](image-url)

**Figure 5.** (A) Relative mfVEP latency versus cVEP (60 minutes checks) latency for each eye of the three subject groups. (B) Same as (A), but for the 15 minutes checks.
and mVEPs agreed and showed only modest delays in the VEP response of eyes with glaucomatous damage.

Qualitatively, our results agree with previous studies that found that patients with glaucoma had significantly delayed cVEP responses.1–4 The cVEP and mfVEP latencies for our patients with glaucoma were longer than those of our control subjects. Quantitatively, however, there is a major discrepancy, especially with the recent findings of Parisi et al.4 In the latter, by far the largest of the previous studies, stimulus conditions were used that were similar to our 15 minutes cVEP stimulus. In particular, for 15 minutes checks, Parisi et al. reported a sensitivity and specificity of 100% for a cVEP latency criterion. That is, the cVEP latencies of all their 84 patients fell outside the 95% CI based on a normative group, whereas none of the controls fell outside these limits. In other words, there was no overlap in groups; 100% of the patients had cVEP latencies greater than the control subjects. In our study, fewer than 12.3% of the patients had latencies falling outside the range of the controls for the 15 minutes stimulus. In addition, we found a difference in mean 15 minutes cVEP latency between the glaucoma and control groups of less than 7 ms, compared with a difference of 27.8 ms in Parisi et al.

Although our stimuli (e.g., check size, reversal rate, and mean luminance) were essentially the same as that in Parisi et al.,4 there were two differences in procedure. First, our field of view was 48° in diameter, so as to match the mfVEP, whereas they used an 18° wide field. Given that the cVEP is generated largely from the foveal region, this is unlikely to be the explanation for the discrepant results. Second, our “active” electrode was placed 4 cm above the inion, as in our previous mfVEP work, whereas they placed the electrode at Oz, which is 10% of the distance between the inion and nasion. On average, Oz is approximately 3.5 cm above the inion. This difference between our placement and theirs is small compared with the large variation in the location of the striate cortex in relation to the external markers (e.g., the inion).15 Given that the testing conditions were similar, this raises the question about the possible difference in the patient populations.

All of the patients with glaucoma in Parisi et al.4 were classified as having OAG. They did not specify whether patients with exfoliative glaucoma or pigmentary glaucoma were included in their group or whether all their patients had POAG. In our study, we included patients with a variety of types of glaucoma. To address the possible differences in our patient populations, we analyzed the results for our patients with recorded IOPs of 23 mm Hg or greater on at least two different occasions (26 eyes). These inclusion criteria were similar to the ones used by Parisi et al. Only 8% of these 26 eyes had a cVEP (15 minutes) latency that fell outside the range of the control. Parisi et al. also included a group of patients with ocular hypertension (OHT). Eighty-five percent of these patients fell outside the normal range. We analyzed a subgroup of our suspect eyes that approximately fit their OHT inclusion criteria, but again only 7.2% (1/14) fell outside the range for the control subjects.

Finally, we identified two other differences between their study and ours, but these differences would lead to our overestimating, not underestimating, the effects of glaucoma. Our control group was, on average, younger than the patient groups. In addition, we included patients with pupil diameters as small as 2 mm, whereas the smallest pupil diameter in the Parisi et al.4 was 3 mm. A smaller pupil diameter20 and an older control group21–25 would tend to increase, not decrease, the latency of the cVEP. Dropping the 10 youngest individuals from our control group left 10 individuals with about the same ages (60 ± 10 years) as those in our patient groups. This subset of 10 control subjects had an increase in mean latency of 0.7 (60 minutes) and 5.3 (15 minutes) ms for the cVEP and 1.7 ms for the mfVEP, values consistent with published results for the cVEP2, 21–25 and mfVEP.17 Thus, these two factors (age and pupil size), taken together, suggest that the relatively small latency increases we find for the patients would be even smaller if age and pupil size were taken into consideration.

Although we cannot account for the discrepancy with the Parisi et al. study, our patients represent a fairly typical sample of patients seen in a glaucoma service. In this group, the effects on latency are relatively small, on average a few milliseconds. Further, only 12.3% of our patients with glaucoma fell outside the normal limits for the cVEP (15 minutes). For the mfVEP, 17.3% of the patients fell outside the normal limits. Thus, only a small subset of our patients with glaucoma had prolonged latencies, and these delays were not large. Either a delayed VEP is not a good indicator of damaged, as opposed to dead, RGCs or there are relatively few patients who exhibit evidence of damaged RGCs.

### References


