Correlation between Optical Coherence Tomography, Pattern Electroretinogram, and Visual Evoked Potentials in Open-angle Glaucoma Patients

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Objective: To correlate the nerve fiber layer (NFL) thickness and the visual function evaluated by electrophysiologic retinal and cortical responses assessed in open-angle glaucoma (OAG) eyes.

Design: Prospective case-control study.

Participants: Thirty glaucoma patients (mean age, 47.1 ± 7.15 years; refractive error range, ± 2 spherical equivalent) with a mean deviation of computerized static perimetry (24/2 Humphrey, Dublin, CA) from −5 to −28 dB and intraocular pressure less than 21 mmHg on pharmacologic treatment and 14 age-matched control participants.

Methods: Nerve fiber layer thickness was measured by optical coherence tomography. Retinal and visual pathway function was assessed by simultaneously recording pattern electroretinograms (PERGs) and visual evoked potentials (VEPs) using high-contrast (80%) checkerboard stimuli (the single check edges subtend 15 minutes of the visual arc) reversed at the rate of two reversals per second. Linear regression analyses were adopted to establish the correlation between NFL thickness and PERG and VEP parameters.

Main Outcome Measures: Nerve fiber layer thickness measurements in each quadrant (superior, inferior, nasal, and temporal) were taken and then averaged (12 values averaged) and identified as NFL overall, whereas the data obtained in the temporal quadrant only (three values averaged) were identified as NFL temporal. PERG P50 implicit time and P50-N95 amplitude and VEP P100 implicit time and N75-P100 amplitude were also measured.

Results: In OAG eyes, we found a significant (P < 0.01) reduction in NFL thickness in both NFL overall and NFL temporal evaluations with respect to the values observed in control eyes. PERG and VEP parameters showed a significant (P < 0.01) delay in implicit time and a reduction in peak-to-peak amplitude. In OAG eyes, the NFL overall and NFL temporal values were significantly correlated (P < 0.01) with the PERG P50 implicit time and P50-95 peak-to-peak amplitude. No correlations (P > 0.01) between NFL values and VEP parameters were found.

Conclusions: There is a correlation between PERG changes and NFL thickness, but there is no correlation between VEP changes and NFL thickness in patients affected by OAG. Ophthalmology 2001;108:905–912 © 2001 by the American Academy of Ophthalmology.
Recently, optical coherence tomography (OCT), a new noninvasive technology allowing cross-sectional imaging of the eye, has been described as offering the possibility of assessing the retinal nerve fiber layer (NFL) thickness with good reproducibility.29–34 Interestingly, a high degree of correlation between OCT-estimated values of NFL thickness29–33 and the stage of diseases affecting primarily the ganglion cells (for example, of glaucoma) has been reported in humans.34 In addition, in vivo measurements of the inner retina thickness obtained by OCT have been significantly correlated with PERG responses in living human eyes that show some degree of ocular hypertension.15

In the present study, a widespread selection of glaucoma patients, with eyes affected by different degrees of glaucomatous optic neuropathy, were tested for PERG, VEP, and OCT. Our aim was to correlate the inner retinal thickness with the visual function evaluated by electrophysiologic retinal and cortical responses assessed in eyes with open-angle glaucoma (OAG).

Materials and Methods

Patients

Thirty eyes of 30 consecutive patients (mean age, 47.1 ± 7.15 years) affected by OAG were recruited. Because it is known that the PERG responses could be modified by the reduction of the ganglion cells (for example, of glaucoma) has been reported in humans. In addition, in vivo measurements of the inner retina thickness obtained by OCT have been significantly correlated with PERG responses in living human eyes that show some degree of ocular hypertension. In the present study, a widespread selection of glaucoma patients, with eyes affected by different degrees of glaucomatous optic neuropathy, were tested for PERG, VEP, and OCT. Our aim was to correlate the inner retinal thickness with the visual function evaluated by electrophysiologic retinal and cortical responses assessed in eyes with open-angle glaucoma (OAG).

Optical Coherence Tomography Examination

Optical coherence tomography (Humphrey Instruments, Dublin, CA), including the fiber optic delivery system coupled with slit-biomicroscope, was used. This system provides the operator with a video-camera view of the scanning probe beam on the fundus and OCT imaging acquired in real time on a computer monitor. After dilatation with 1% tropicamide, each eye was scanned three times using a circle size of 3.4 mm (1.7-mm radius). Near-infrared light (840 nm wavelength) was used. Throughout scanning, the subject kept his or her eyes constantly fixed on an internal target provided by the equipment. The measurements were obtained from three nonconsecutive scans (i.e., the subject was allowed to rest for a few seconds before being repositioned to proceed to the following scan). As previously reported, the OCT software provides an automated computer algorithm that identifies the anterior and posterior borders of the retina. This has been claimed to offer the possibility of calculating both NFL and total retinal thicknesses. The software allows the mapping of the thickness data according to both quadrant-by-quadrant and clock-hour analyses. Retinal thickness was determined by computer as the distance between the first reflection at the vitreoretinal interface and the anterior boundary of the second reflective layer, corresponding to the retinal pigment epithelium and the choriocapillaris. As discussed elsewhere, NFL thickness was automatically assessed by the computer assuming the correlation with the red, highly reflective layer at the vitreoretinal interface.

We considered the average values of three different measurements per quadrant (superior, inferior, nasal, and temporal); the overall data obtained in all quadrants (12 values averaged) were identified as NFL overall, whereas the data obtained in the temporal quadrant only (three values averaged) were identified as NFL temporal. Nerve fiber layer temporal was recorded to evaluate the temporal fiber in which the papillomacular bundle fibers are included.

Electrophysiologic Examination

According to previously published studies, simultaneous PERG and VEP recordings were performed using the following methods.

The subjects under examination were seated in a semidark, acoustically isolated room in front of the display surrounded by a uniform field of luminance of 5 cd/m². Before the experiment, each participant was adapted to the ambient room light for 10 minutes, and the pupil diameter was approximately 5 mm. Mydriatic or miotic drugs were never used. Stimulation was monocular after occlusion of the other eye. Visual stimuli were checkerboard patterns (contrast 80%, mean luminance 110 cd/m²) generated on a television monitor and reversed in contrast at the rate of two reversals per second; at the viewing distance of 114 cm, the check edges subtended 15 minutes of visual angle. The screen of the monitor subtended 18°, and a small red target (0.5°) was placed in the center of the stimulus field to maintain stable fixation. The refraction of all participants was corrected for viewing distance.
Pattern Electroretinogram Recordings

The bioelectrical signal was recorded by a small Ag/AgCl skin electrode placed over the lower eyelid. Pattern electroretinograms were derived tipically between the stimulated (active) electrode and the patched (reference) electrode using the method previously described. The ground electrode was in Fpz. The inter-electrode resistance was less than 3 KOhms. The signal was amplified (gain, 50,000), filtered (band pass, 1–30 Hz), and averaged with automatic rejection of artifacts (200 events from artifacts were averaged for every trial) by BM 6000 (Biomedica Mangoni, Pisa, Italy). The analysis time was 250 milliseconds. The transient PERG response is characterized by a number of waves with three subsequent peaks, of negative, positive, and negative po-
Statistics

The data are reported as mean values ± 1 standard deviation. The differences between control and OAG eyes were statistically evaluated with a one-way analysis of variance without post hoc Bonferroni analysis for repeated measures. To assess whether a correlation exists between OCT and electrophysiologic parameters, linear regression analysis (Pearson’s test) was adopted. In both statistical analyses, a P value less than 0.01 was considered significant.

Optical coherence tomography scans were performed by one observer (GM). Test–retest variability was maintained within the 5% limit. In case of a variability of more than 5%, a new set of three independent scans was repeated until proper values were recorded.

In the recording session, simultaneous PERG and VEP values were recorded at least twice, and the resulting waveforms were superimposed to confirm the repeatability of the results. The test–retest variability of both PERG and VEP parameters proved to be less than 5%.

Results

The main clinical, morphologic, and electrophysiologic data pertaining to control participants and OAG patients are reported in Table 1. The statistical results are shown in Tables 2 and 3.

Visual Evoked Potential Recordings

Cup-shaped electrodes of Ag/AgCl were fixed with collodion in the following positions: active electrode in Oz, reference electrode in Fpz; ground in the left arm. The interelectrode resistance was kept at less than 3 KOhms. The bioelectric signal was amplified (gain, 20,000), filtered (band-pass, 1–100 Hz), and averaged (200 events) free from artifacts were averaged for every trial by BM 6000. The analysis time was 250 milliseconds. The transient VEP response is characterized by a number of waves with three subsequent peaks, of negative, positive, and negative polarity, respectively. In normal subjects, these peaks have the following implicit times: 75, 100, and 145 milliseconds (N75, P100, N145).

We accepted PERG and VEP signals with a signal-to-noise ratio of more than 2. The noise was measured by recording the bioelectrical signals (200 averaged events), while the monitor was screened by a cardboard, and a retinal noise (peak-to-peak measure) less than 0.1 microvolt (mean 0.086 microvolt) was observed in all subjects tested. For all PERG and VEP values, the implicit time and the peak-to-peak amplitude of each of the averaged waves were measured directly on the displayed records by a pair of cursors.

Table 2. Mean Values and One (±) Standard Deviation of Electrophysiologic Parameters in Control Subjects and in Patients Affected by Open-angle Glaucoma (One-way Analysis of Variance with Respect to Control Subjects)

<table>
<thead>
<tr>
<th>Group</th>
<th>Pattern Electroretinogram</th>
<th>Pattern Electroretinogram</th>
<th>Visual Evoked Potential</th>
<th>Visual Evoked Potential</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>P50 Implicit Time (msec)</td>
<td>P50-N95 Amplitude (µV)</td>
<td>P100 Implicit Time (msec)</td>
<td>N75-P100 Amplitude (µV)</td>
</tr>
<tr>
<td>Control (n = 14)</td>
<td>56.92 ± 4.04</td>
<td>1.71 ± 0.17</td>
<td>106.35 ± 3.47</td>
<td>6.70 ± 0.91</td>
</tr>
<tr>
<td>Open-angle glaucoma (n = 30)</td>
<td>F(1,42) = 28.62</td>
<td>F(1,42) = 281.8</td>
<td>F(1,42) = 142.1</td>
<td>F(1,42) = 18.95</td>
</tr>
<tr>
<td></td>
<td>P &lt; 0.001</td>
<td>P &lt; 0.001</td>
<td>P &lt; 0.001</td>
<td>P &lt; 0.001</td>
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</tbody>
</table>

Table 3. Linear Regression and Correlation between Electrophysiologic and Perimeter Parameters and Nerve Fiber Layer Overall or Temporal Evaluated in Open-angle Glaucoma Eyes

<table>
<thead>
<tr>
<th>Age (yrs)</th>
<th>Intraocular Pressure</th>
<th>Corrected Pattern Standard Deviation (Humphrey 24-2)</th>
<th>Mean Deviation (Humphrey 24-2)</th>
<th>Pattern Electroretinogram</th>
<th>Visual Evoked Potential</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<tr>
<td>Nerve fiber layer overall</td>
<td>r = 0.172</td>
<td>r = 0.071</td>
<td>r = 0.663</td>
<td>r = 0.393</td>
<td>r = 0.847</td>
</tr>
<tr>
<td>t = 0.361</td>
<td>r = 0.707</td>
<td>P &lt; 0.001</td>
<td>P = 0.031</td>
<td></td>
<td></td>
</tr>
<tr>
<td>t = 0.009</td>
<td>r = 0.041</td>
<td>r = 0.688</td>
<td>r = 0.400</td>
<td></td>
<td></td>
</tr>
<tr>
<td>t = 0.048</td>
<td>t = 0.022</td>
<td>t = 2.311</td>
<td>t = 0.028</td>
<td></td>
<td></td>
</tr>
<tr>
<td>P = 0.961</td>
<td>r = 0.827</td>
<td>P &lt; 0.001</td>
<td>P = 0.001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nerve fiber layer temporal</td>
<td>r = 0.283</td>
<td>r = 0.632</td>
<td>r = 0.198</td>
<td>r = 0.115</td>
<td>r = 0.891</td>
</tr>
<tr>
<td>t = 0.009</td>
<td>r = 0.041</td>
<td>r = 0.688</td>
<td>r = 0.400</td>
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Pattern Electroretinogram. Open-angle glaucoma eyes showed P50 implicit time significantly delayed and P50-N95 amplitudes significantly reduced with respect to the control eyes.

Visual Evoked Potential. In OAG eyes, it was observed that P100 implicit time was significantly delayed and N75-P100 amplitude was significantly reduced with respect to those of controls.

Optical Coherence Tomography versus Pattern Electroretinogram and Visual Evoked Potential

The correlation between NFL thickness and PERG and VEP parameters is shown in Figures 2 and 3 and in Table 3.

In OAG eyes, the NFL overall and NFL temporal values were significantly correlated (P < 0.01) with the PERG parameters (P50 implicit time and P50-N95 amplitude). No correlations (P > 0.01) between NFL values and VEP parameters (P100 implicit time and N75-P100 amplitudes) were observed. Open-angle glaucoma eyes showed a weak correlation between NFL (overall and temporal) and mean deviation values. Interestingly, corrected pattern standard deviation values correlated well with both NFL overall and NFL temporal values. In control eyes, no significant correlation between electrophysiologic (PERG and VEP) parameters and NFL thickness was observed.

Discussion

In our OAG patients, we observed a significant reduction in NFL thickness that correlated significantly with PERG responses, whereas no correlations were found between NFL values and VEP responses.

The OCT readings are comparable to those previously observed in normal and glaucomatous eyes by several authors. Recently, Chauhan and Marshall raised some criticism regarding the accuracy of OCT in NFL thickness measurements. However, in the same report, they showed a good correlation between excimer laser-induced ablation of the inner retina and the signal recorded by OCT, stating that “...the thickness of the inner band was reduced by the same amount as the ablation step height.” Therefore, although the accuracy of OCT in quantifying the NFL thickness is still a matter of debate, we can assume that progressive changes in the OCT signal coming from the inner retina (including NFL, inner plexiform layer, and ganglion cell layer) are paralleled by similar changes occurring in the tissue. Actually, the NFL thickness showed an
excellent correlation with the corrected pattern standard deviation of the Humphrey Field Analyzer, whereas the mean deviation showed a poor correlation with NFL. In fact, the corrected pattern standard deviation is considered a more accurate index of localized defects in the visual field and is known to reproduce considerably well the areas of thinner papillary rim in glaucoma.

On OCT scan, the mean IOP between the study groups was superimposable (see Table 1). Therefore, we can rule out any possible IOP-induced bias in the OCT reading procedure between normal and glaucomatous eyes. In the glaucomatous eyes, we did not find a correlation between NFL thickness and age, and this may result from the NFL reduction induced by both disease and age.

The wide range of NFL readings observed in glaucomatous eyes provided enough heterogeneity to perform correlations with electrophysiologic analysis. The scatterplots shown in Figure 2 indicate a high degree of correlation between the NFL thickness and the PERG response. A similar correlation was observed in humans with multiple sclerosis.44 In this study, a significant correlation between NFL thickness and PERG P50 implicit time or the P50-N95 amplitude was observed only when the PERG was recorded using 15-minute checks. By contrast, when PERG was recorded using high-contrast checks, subtending 60 minutes of visual arc, NFL thickness did not correlate with PERG P50 implicit time or P50-N95 amplitude. Furthermore, because NFL thickness did not correlate with PERG N35-P50 amplitudes, this suggests that not every P50 component arises from the innermost retinal layers.44 These results may have a possible explanation if we consider that the transient PERG to checkerboard stimuli is a complex response, with the contribution of both contrast- and luminance-sensitive retinal generators (ganglion and preganglionic cells).44

Following this line of reasoning, we chose to adopt the response to 15-minute checks as the most sensitive index of inner retinal dysfunction. The P50 implicit time and the P50-N95 amplitude were considered as those PERG parameters that could be more accurately related to the NFL thickness.

The relationship observed between PERG responses and NFL thickness found in our glaucoma patients confirms previous results obtained for multiple sclerosis patients,44 thus indicating that the integrity of ganglion cell fibers is
essential for the generation of a normal PERG response. This evidence agrees with available studies carried out by Maffei and Fiorentini,24 who showed that PERG originates from the innermost retinal layers (ganglion cells and their fibers).

In OAG eyes, we observed delayed VEP P100 implicit times and reduced VEP N75-P100 amplitudes, and this is in agreement with previous studies.5–7,16,19–23 Nevertheless, the correlation between VEP and NFL thickness did not reach statistical significance.

The lack of correlation between NFL thickness and VEP responses could also be explained by considering that VEP responses depend on the magnitude and timing of afferent inputs to the visual cortex and result from both retinal activity and neural conduction along the postretinal visual pathways.7 Previous electrophysiologic evidence6,7,23 indicated that the impaired VEP responses observed in glaucoma patients could be ascribed to impaired neural conduction in the optic nerve and in the whole postretinal visual pathways as a consequence of the dysfunction of the innermost retinal layers. Additional postretinal factors could then contribute to the observed reduced magnitude and delayed timing of the input to the visual cortex. Interestingly, structural and functional damage in the dorsal lateral geniculate nucleus of persons or animals affected by well-documented glaucomatous optic neuropathy has been recently reported.45–47 An impairment at the dorsal lateral geniculate nucleus level could cause functional changes in those cells that produce the visual cortical evoked responses; this is likely to be related to the delay and to the reduction of the VEP responses observed in our glaucoma patients.

In conclusion, our results indicate that there is a correlation between PERG changes and NFL thickness, but there is no detectable correlation between VEP changes and NFL thickness in patients affected by OAG.

References

26. Maffei L, Fiorentini A, Bisti S, Holländer H. Pattern ERG in...


