A Combined Index of Structure and Function for Staging Glaucomatous Damage

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Objectives: To present and evaluate a new combined index of structure and function (CSFI) for staging and detecting glaucomatous damage.

Methods: Observational study including 333 glaucomatous eyes (295 with perimetric glaucoma and 38 with preperimetric glaucoma) and 330 eyes of healthy subjects. All the eyes were tested with standard automated perimetry and spectral domain optical coherence tomography within 6 months. Estimates of the number of retinal ganglion cells (RGCs) were obtained from standard automated perimetry and spectral domain optical coherence tomography and a weighted averaging scheme was used to obtain a final estimate of the number of RGCs for each eye. The CSFI was calculated as the percent loss of RGCs obtained by subtracting estimated from expected RGC numbers. The performance of the CSFI for discriminating glaucoma from normal eyes and the different stages of disease was evaluated by receiver operating characteristic curves.

Results: The mean CSFI, representing the mean estimated percent loss of RGCs, was 41% and 17% in the perimetric and preperimetric groups, respectively ($P < .001$). They were both significantly higher than the mean CSFI in the healthy group ($P < .001$). The CSFI had larger receiver operating characteristic curve areas than isolated indexes of structure and function for detecting perimetric and preperimetric glaucoma and differentiating among early, moderate, and advanced stages of visual field loss.

Conclusion: An index combining structure and function performed better than isolated structural and functional measures for detection of perimetric and preperimetric glaucoma as well as for discriminating different stages of the disease.

patients who manifest severe glaucomatous damage. In this situation, SAP losses are still the best method to quantify the effect of the disease and monitor its progression.

The apparent disagreement between structural and functional measurements of the disease seem to be largely derived from the different algorithms and measurement scales as well as the different variability characteristics of the tests commonly used to assess structural and functional losses. In fact, Harwerth et al demonstrated that structural and functional tests agree as long as one uses appropriate measurement scales for neural and sensitivity losses and considers factors, such as the effect of aging and eccentricity, on the estimates of neural losses. In a series of investigations, they demonstrated that estimates of RGC losses obtained from clinical SAP agreed closely with estimates of RGC losses obtained from RNFL thickness assessment by optical coherence tomography (OCT). The results of their model provided a common domain for expressing results of structural and functional tests, that is, the estimates of RGC losses, opening the possibility of combining these different tests to improve the reliability and accuracy of estimates of the amount of neural losses and develop a combined staging system for glaucoma severity.

In the current study, we propose a new index to estimate glaucoma severity based on a combination of functional measurements obtained by SAP and structural assessment by spectral domain optical coherence tomography (SDOCT). We show that the index performed well in discriminating diseased from nondiseased patients and provided a better estimate of the stage of glaucoma severity compared with the isolated use of functional or structural measures.

METHODS

This was an observational study. Participants from this study were included in 2 prospective longitudinal studies designed to evaluate optic nerve structure and visual function in glaucoma (the African Descent and Glaucoma Evaluation Study [ADAGES] and the Diagnostic Innovations in Glaucoma Study [DIGS]). The 3-site ADAGES collaboration includes the Hamilton Glaucoma Center at the Department of Ophthalmology, University of California, San Diego ([UCSD] data coordinating center), the Department of Ophthalmology, University of Alabama, Birmingham, and the New York Eye and Ear Infirmary, New York. Although DIGS includes only patients recruited at UCSD, the protocols of the 2 studies are identical. The institutional review boards at all 3 sites included if they were seen with a best-corrected visual acuity less than 20/40, spherical refraction outside ±5.0 diopters (D) and/or cylinder correction outside 3.0 D, or any other ocular or systemic disease that could affect the optic nerve or the visual field.

The study included 333 eyes of 246 patients with glaucoma diagnosed based on evidence of the presence of repeatable glaucomatous visual field defects or documented history of progressive glaucomatous optic neuropathy. From the 333 eyes, 295 had evidence of glaucomatous visual field defects based on repeatable abnormal visual field test results defined as a pattern standard deviation outside of the normal 95% CIs or a Glaucoma Hemifield Test result outside normal limits. An additional group of 38 eyes had evidence of progressive glaucomatous change in the appearance of the optic disc as assessed by masked grading of simultaneous stereoscopic optic disc photographs (Topcon Instrument Corporation of America), despite absence of statistically significant visual field losses. The evidence of progressive glaucomatous damage had to be present before the imaging test date, and the details of the methodology used to grade optic disc photographs at the UCSD Optic Disc Reading Center have been provided elsewhere. This latter group was used to assess the ability of the proposed staging system to quantify damage in patients with confirmed peripapillary glaucoma.

The control group consisted of 330 eyes from 171 healthy participants. These subjects were recruited from the general population and were required to have a normal ophthalmologic examination and intraocular pressure below 22 mm Hg OU, but results of visual field tests were not used as inclusion or exclusion criteria.

VISUAL FIELD TESTING

All the patients underwent SAP testing using SITA Standard 24-2 strategy less than 6 months apart from imaging. All the visual fields were evaluated by the UCSD Visual Field Assessment Center. Visual fields with more than 33% fixation losses or false-negative errors or more than 13% false-positive errors were excluded. The only exception was the inclusion of visual fields with false-negative errors of more than 33% when the field showed advanced disease (mean deviation [MD] < −12 dB). Visual fields exhibiting a learning effect (ie, initial tests showing consistent improvement on visual field indexes) were also excluded. Visual fields were further reviewed for the following artifacts: eyelid and rim artifacts, fatigue effects, inappropriate fixation, evidence that the visual field results were caused by a disease other than glaucoma (eg, homonymous hemianopia), and inattention. The UCSD Visual Field Assessment Center requested repeats of unreliable visual field test results, and these were obtained whenever possible.

SPECTRAL DOMAIN OCT

The Cirrus HDOCT (software version 5.2; Carl Zeiss Meditec, Inc) was used to acquire RNFL measurements in the study. It uses a superluminescent diode scan with a center wavelength of 840 nm and an acquisition rate of 27,000 A-scans per second at an axial resolution of 5 µm. The protocol used for RNFL thickness evaluation was the optic disc cube. This protocol is based on a 3-dimensional scan of a 6 × 6-mm² area centered on the optic disc in which information from a 1024 (depth) × 200 × 200-point parallelepiped is collected. Then, a 3.46-mm diameter circular scan (10.87-mm length) is automatically placed around the optic disc, and the information about parapapillary RNFL thickness is obtained. Because information from the whole region is obtained, it is possible to modify the position of the scan after the examination is conducted. To be included, all the images were reviewed for noncentered scans and had to have a signal strength greater than 6, the absence of movement artifacts, and good centering around the optic disc.
COMBINED INDEX OF STRUCTURE AND FUNCTION

The development of the combined index of structure and function (CSFI) to measure disease severity was based on previous work by Harwerth et al. on the development and validation of a model linking structure and function in glaucoma. Based on experimental studies in monkeys, the authors first derived an empirical model relating sensitivity measurements in SAP to histological RGC counts as a function of retinal eccentricities. The experimental results were then translated to clinical SAP in humans. The following formulas were proposed to estimate the number of RGC somas in an area of the retina corresponding to a specific SAP test field location at eccentricity ec with sensitivity s in decibels:

\[ m = 0.054 \times (ec \times 1.32) + 0.9 \]

\[ b = 1.5 \times (ec \times 1.32) - 14.8 \]

\[ gc = 1 + s - b/m \]

\[ SAP_{rgc} = \sum 10^{g+c} \times g \times 0.1 \]

Where \( m \) and \( b \) represent the slope and intercept, respectively, of the linear function relating ganglion cell quantity \( g \) in decibels to the visual field sensitivity \( s \) in decibels at a given eccentricity \( ec \). By applying the formulas, one can obtain a SAP-derived estimate of the total number of RGCs (\( SAP_{rgc} \)) by adding the estimates from all the locations in the visual field.

The structural part of the model consisted in estimating the number of RGC axons from RNFL thickness measurements obtained by OCT. The model considered the effect of aging in the axonal density and the effect of disease severity on the relationship between the neuronal and nonneuronal components of the RNFL thickness estimates obtained by OCT. To derive the total number of RGC axons from the global RNFL thickness measurement obtained by OCT (\( OCT_{rgc} \)), one can apply the following formulas:

\[ d = (-0.007 \times age) + 1.4 \]

\[ c = (-0.26 \times MD) + 0.12 \]

\[ OCT_{rgc} = 10^d (\log(average \ RNFL \ thickness \times 10^{870 \times d})) \times 10^{-c} \times 0.1 \]

where \( d \) corresponds to the axonal density (axons per micrometers squared) and \( c \) is a correction factor for the severity of disease to consider remodeling of the RNFL axonal and nonaxonal composition. The above calculations allow one to estimate the number of RGCs from 2 sources, 1 functional and 1 structural, and a strong relationship was demonstrated between the 2 estimates in external validation cohorts. However, although Harwerth et al. proposed a model linking structure and function, no attempt was made to develop an index combining structural and functional estimates that could be clinically used to stage glaucoma severity. We propose the following calculations to develop such an index. To derive a combined index, we simply averaged the estimates of RGC numbers obtained from SAP and OCT but weighting according to severity of disease. As clinical SAP and OCT test accuracies have been proposed to be inversely related to disease severity, we propose a weighted scale combining the estimates of RGC numbers (\( wrgc \)) from both tests:

\[ wrgc = \frac{(1 + MD/30) \times OCT_{rgc} + (-MD/30) \times SAP_{rgc}}{1} \]

The weights were chosen to reflect the inverse relationship with disease severity of SAP and OCT estimates, along the scale of MD values ranging from 0 to −30 dB. After estimates of \( wrgc \) were obtained, a linear regression model was run to relate \( wrgc \) estimates to age and optic disc area in the normal control population. The purpose was to develop a model to predict expected RGC numbers according to age and optic disc area. To avoid model overfitting, the regression parameters were obtained using only half of the normal eyes (development sample). After the expected number of RGCs was calculated for each eye, an estimate of the percent RGC loss for each eye was obtained by subtracting measured from estimated RGC numbers. The percent estimate of RGC loss by the CSFI should reflect an estimate of glaucomatous damage obtained by combining data from the structural and functional measurements, as calculated below:

\[ CSFI = \frac{|(\text{expected RGC number} - wrgc)|}{(\text{expected RGC number})} \times 100 \]

STATISTICAL ANALYSIS

The performance of the CSFI for discriminating glaucomatous eyes from normal eyes and the different stages of disease was compared with those of other indexes previously used to stage disease severity, such as MD and the visual field index, as well as with the SD OCT parameter average RNFL thickness. Receiver operating characteristic (ROC) curves were built, and the area under the ROC curves (AUC) was used to summarize the diagnostic accuracy for each parameter. Perimetric and preperimetric glaucomatous eyes were compared with normal eyes in the validation sample, that is, excluding the eyes previously used to obtain the regression parameters described earlier. An AUC equal to 1 represents perfect discrimination, whereas an AUC of 0.5 represents chance discrimination. The AUCs and 95% CIs were obtained for each parameter after adjusting for age. A bootstrap resampling procedure (n = 1000 samples) was used to derive CIs. Age adjustment was performed using an ROC regression model, as previously described. The model is able to adjust for the differences in variables between control and cases by fitting a linear regression of the marker distribution on the adjustment variables among controls. Standardized residuals based on this fitted linear model are used in place of the marker values for cases and controls. To account for the potential correlation between eyes, the cluster of data for the study subject was considered as the unit of resampling when calculating standard errors. This procedure has been previously used to adjust for the presence of multiple correlated measurements from the same unit.\(^{13}\)

All statistical analyses were performed with commercially available software (STATA, version 12; StataCorp LP). The \( \alpha \) level (type I error) was set at .05.

RESULTS

From the 333 glaucomatous eyes, 295 (89%) had perimetric glaucoma and 38 (11%) had preperimetric glaucoma. The eyes were compared with 165 eyes from 85 healthy subjects included in the validation sample. The mean (SD) ages of perimetric glaucoma and preperimetric glaucoma participants were 69 (11) and 66 (10), respectively. They were both significantly higher than that of control subjects (60 [11]; \( P < .01 \) for both comparisons). Age differences were adjusted for in the ROC analyses.

Table 1 lists estimates of the different parameters obtained in the study. There was a strong correlation between RGC estimates obtained from SAP and SD OCT data in the eyes included in the study (\( r = 0.89; \ P < .001 \) (Figure 1). Figure 2 shows histograms of calculated weighted estimates of RGC numbers combining structural and functional tests (\( wrgc \)), according to the diagnostic categories. The mean estimated number of RGCs...
Table 1. Mean Values of the Different Parameters Calculated in the Study in Perimetric Glaucoma, Preperimetric Glaucoma, and Healthy Eyes

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Perimetric Glaucoma (n = 295)</th>
<th>Preperimetric Glaucoma (n = 38)</th>
<th>Healthy Eyes (n = 165)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>MD, dB</td>
<td>-4.01 (-1.79 to -9.40)</td>
<td>-0.32 (-1.33 to 0.47)</td>
<td>0.17 (-0.85 to 1.05)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>PSD, dB</td>
<td>4.80 (2.59 to 9.77)</td>
<td>1.52 (1.41 to 1.76)</td>
<td>1.60 (1.34 to 1.85)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>VFI, %b</td>
<td>92 (77 to 97)</td>
<td>99 (99 to 100)</td>
<td>99 (99 to 100)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>Average RNFL thickness, µm</td>
<td>69 (13)</td>
<td>78 (10)</td>
<td>94 (9)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>SAPrgc, ×1000 cells</td>
<td>660 (277)</td>
<td>944 (148)</td>
<td>1075 (208)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>OCTrgc, ×1000 cells</td>
<td>502 (221)</td>
<td>749 (107)</td>
<td>977 (156)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>wrgc, ×1000 cells</td>
<td>525 (210)</td>
<td>749 (105)</td>
<td>973 (154)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>CSFI, %</td>
<td>41 (22)</td>
<td>17 (10)</td>
<td>4 (7)</td>
<td>&lt; .001</td>
</tr>
</tbody>
</table>

Abbreviations: CSFI, combined index of structure and function; MD, mean deviation; OCT, optical coherence tomography; PSD, pattern standard deviation; rgc, retinal ganglion cell; RNFL, retinal nerve fiber layer; SAPrgc, number of rgcs estimated from standard automated perimetry (SAP) sensitivity values; VFI, visual field index; wrgc, weighted estimate of the number of rgcs.

Table 2. Results of the Linear Regression Model Evaluating the Association Between the Weighted Number of Retinal Ganglion Cells and Age and Optic Disc Area in Healthy Eyes

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Coefficient (95% CI)</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, per year older</td>
<td>-9249 (-10 613 to -7885)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>Optic disc area, per 0.1 mm² larger</td>
<td>11 607 (6077 to 17 138)</td>
<td>&lt; .001</td>
</tr>
<tr>
<td>Constant</td>
<td>1 301 098 (1 163 399 to 1 438 796)</td>
<td>&lt; .001</td>
</tr>
</tbody>
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<tr>
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<td>Constant</td>
<td>1 301 098 (1 163 399 to 1 438 796)</td>
<td>&lt; .001</td>
</tr>
</tbody>
</table>

a Values are given as mean (SD) unless otherwise indicated.
b Median (first quartile, third quartile).

Figure 1. Scatterplot illustrating the relationship between the number of retinal ganglion cells (RGCs) derived from standard automated perimetry (SAP) sensitivity data and the number of RGCs estimated from analysis of the retinal nerve fiber layer by spectral domain optical coherence tomography (SDOCT).

Figure 2. Histograms illustrating the distribution of the number of estimated retinal ganglion cells (RGCs) according to the different diagnostic categories.

Figure 3. Boxplot graph of the CSFI values according to diagnostic category.

Table 3 summarizes the AUCs parameters investigated in the study. The CSFI had an ROC curve area of 0.94 to discriminate glaucomatous from normal eyes. The performance of the CSFI was superior to that of SDOCT parameter average RNFL thickness (AUC=0.92; P=.008) and the global visual field index MD (AUC=0.88; P < .001) and visual field index (AUC=0.89; P < .001). Analyses were also performed by subgroups of perimetric and preperimetric glaucoma. For detection of perimetric glaucoma, the CSFI also performed significantly better than average RNFL thickness and MD (P < .001 for both comparisons) but not sig-

in the group with perimetric glaucoma was 524 545 compared with 748 731 in the preperimetric group and 973 120 in normal eyes. The results of the linear regression model relating estimated RGC numbers to age and optic disc area in the normal eyes from the development sample are given in Table 2. A significant relationship was noted between RGC number and age, with an estimated loss of 9249 RGCs per year of older age in normal subjects (Figure 3). Also, each 0.1-mm² larger optic disc area corresponded to an increase in 11 607 RGCs.

The mean CSFI, representing the mean estimated percent loss of RGCs, was 41% and 17% in the perimetric and preperimetric groups, respectively (P < .001). They were also both significantly higher than the mean CSFI in the normal group (P < .001) (Table 1). Figure 4 shows a boxplot graph of the CSFI values according to diagnostic category. Table 3 summarizes the AUCs parameters investigated in the study. The CSFI had an ROC curve area of 0.94 to discriminate glaucomatous from normal eyes. The performance of the CSFI was superior to that of SDOCT parameter average RNFL thickness (AUC=0.92; P=.008) and the global visual field index MD (AUC=0.88; P < .001) and visual field index (AUC=0.89; P < .001). Analyses were also performed by subgroups of perimetric and preperimetric glaucoma. For detection of perimetric glaucoma, the CSFI also performed significantly better than average RNFL thickness and MD (P < .001 for both comparisons) but not sig-

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significantly different from the visual field index (P = .16) (Table 4). For detecting preperimetric glaucoma, the CSFI had an ROC curve area of 0.85, which was superior to that of the visual field index (AUC = 0.51; P < .001) and MD (AUC = 0.63; P < .001). The ability to detect preperimetric glaucoma with the CSFI was similar to that of the SDOCT parameter average RNFL thickness (AUC = 0.88; P = .32). Figure 5 shows ROC curves for the different parameters for detection of perimetric and preperimetric glaucoma.

We also evaluated the ability of the CSFI in discriminating eyes with different stages of glaucomatous visual field loss as determined by the Hodapp-Anderson-Parrish classification system. According to the Hodapp-Anderson-Parrish system, from the 295 eyes with glaucomatous visual field loss, 189 had early damage, 49 had moderate damage, and 57 had advanced damage. Table 4 lists the values of the parameters calculated in the study for these different severity groups. The AUC for the CSFI for separating early from moderate visual field loss was significantly better than that for average RNFL thickness (AUC = 0.70 [SD, 0.05]; P < .001). The CSFI also performed better than average RNFL thickness to discriminate eyes with preperimetric glaucoma from those with early visual field loss (AUC = 0.70 [SD, 0.05]; P < .001) and RNFL thinning, with average thickness of 68 µm. The CSFI for the eye was 39%, indicating a loss of 39% of the estimated number of RGCs compared with the age-expected number. Figure 8 shows 2 eyes with advanced glaucoma, 1 with MD of −15.12 dB and another with MD of −23.61 dB. Despite the important differences in visual field damage between the 2 patients, SDOCT results were similar in the 2 eyes, with the same value of average thickness of 50 µm. The CSFI clearly distinguished between the eyes with values of 74% for the former and 85% for the latter.

Table 3. Areas Under the Receiver Operating Characteristic (ROC) Curves and Standard Errors for the Parameters Evaluated in the Study

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Glaucoma vs Healthy Eyes</th>
<th>Perimetric Glaucoma vs Healthy Eyes</th>
<th>Preperimetric Glaucoma vs Healthy Eyes</th>
</tr>
</thead>
<tbody>
<tr>
<td>MD</td>
<td>0.88 (0.01)</td>
<td>0.92 (0.01)</td>
<td>0.63 (0.05)</td>
</tr>
<tr>
<td>PSD</td>
<td>0.88 (0.01)</td>
<td>0.94 (0.01)</td>
<td>0.46 (0.05)</td>
</tr>
<tr>
<td>VFI</td>
<td>0.89 (0.01)</td>
<td>0.94 (0.01)</td>
<td>0.51 (0.04)</td>
</tr>
<tr>
<td>Average RNFL thickness</td>
<td>0.92 (0.01)</td>
<td>0.93 (0.01)</td>
<td>0.88 (0.04)</td>
</tr>
<tr>
<td>wrgc, ×1000 cells</td>
<td>0.95 (0.01)</td>
<td>0.96 (0.01)</td>
<td>0.88 (0.03)</td>
</tr>
<tr>
<td>CSFI, %</td>
<td>0.94 (0.01)</td>
<td>0.96 (0.01)</td>
<td>0.85 (0.04)</td>
</tr>
</tbody>
</table>

Abbreviations: CSFI, combined index of structure and function; MD, mean deviation; PSD, pattern standard deviation; RNFL, retinal nerve fiber layer; VFI, visual field index; wrgc, weighted estimate of the number of retinal ganglion cells.

aValues are given as mean (SD).

In the current study, we propose a new index combining information from structural and functional damage in glaucoma that can be used to stage and provide diagnostic information on the disease. The index performed significantly better than isolated measures of structure and function for diagnosing preperimetric and perimetric glaucoma. In addition, the index also performed better in discriminating different stages of the disease.
gesting that it might also be helpful for staging and monitoring patients over time.

Several staging systems for glaucoma have been proposed in the literature. Most of them have been based solely on information extracted from visual fields. Visual field-based staging systems assume that all the patients with statistically normal fields should be grouped at a single stage and, therefore, they do not differentiate whether the patient is actually a healthy subject, has suspicious findings for the disease, or has evidence of glaucomatous neuropathy despite absence of detectable field losses. Experimental and clinical research findings, however, have shown that a substantial number of RGCs may

Table 4. Values of the Parameters Obtained in the Study for the Different Stages of Glaucoma Severity Based on the Hodapp-Anderson-Parrish Classification

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Early Glaucoma (n = 189)</th>
<th>Moderate Glaucoma (n = 49)</th>
<th>Advanced Glaucoma (n = 57)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MD, dB^b</td>
<td>−2.3 (−3.7 to −1.0)</td>
<td>−8.2 (−9.7 to −7.0)</td>
<td>−17.4 (−23.3 to −14.7)</td>
</tr>
<tr>
<td>PSD, dB^b</td>
<td>3.0 (2.1 to 4.6)</td>
<td>9.9 (7.2 to 11.6)</td>
<td>11.6 (9.4 to 13.6)</td>
</tr>
<tr>
<td>VFI, %^b</td>
<td>96 (93 to 98)</td>
<td>80 (75 to 84)</td>
<td>51 (32 to 58)</td>
</tr>
<tr>
<td>Average RNFL thickness, µm</td>
<td>74 (12)</td>
<td>65 (10)</td>
<td>57 (9)</td>
</tr>
<tr>
<td>SAPrgc, ×1000 cells</td>
<td>812 (180)</td>
<td>540 (157)</td>
<td>260 (138)</td>
</tr>
<tr>
<td>OCTrgc, ×1000 cells</td>
<td>628 (156)</td>
<td>376 (82)</td>
<td>193 (70)</td>
</tr>
<tr>
<td>wrgc, ×1000 cells</td>
<td>641 (147)</td>
<td>422 (82)</td>
<td>227 (100)</td>
</tr>
<tr>
<td>CSFI, %</td>
<td>28 (13)</td>
<td>52 (8)</td>
<td>75 (11)</td>
</tr>
</tbody>
</table>

Abbreviations: CSFI, combined index of structure and function; MD, mean deviation; OCT, optical coherence tomography retinal ganglion cells (rgcs); PSD, pattern standard deviation; RNFL, retinal nerve fiber layer; SAPrgc, number of rgcs estimate from standard automated perimetry (SAP) sensitivity values; VFI, visual field index; wrgc, weighted estimated of the number of rgcs.

^a Values are given as mean (SD).

^b Median (first quartile, third quartile).

Figure 5. Receiver operating characteristic (ROC) curves for discriminating between perimetric glaucoma and healthy eyes (A) and between preperimetric glaucoma and healthy eyes (B). ROC curves are shown for the parameters of combined index of structure and function (CSFI), average retinal nerve fiber layer (RNFL) thickness, and visual field index (VFI).

Figure 6. Scatterplots showing the relationship between mean deviation (MD) and the combined index of structure and function (CSFI) and the average retinal nerve fiber layer (RNFL) thickness. A, Scatterplot showing the relationship between MD and the CSFI with superimposed locally weighted scatterplot smoothing (lowess). B, Scatterplot illustrating the relationship between MD and average RNFL thickness with superimposed lowess. There is much more scatter around the lowess curve for the average RNFL thickness compared with the CSFI.
need to be lost before detectable changes are observed in the visual field.\(^3\) Evidence of structural damage to the optic disc and RNFL has been demonstrated in patients with statistically normal visual fields using different imaging technologies and conventional stereophotographs.\(^5,8,20,21\) More important, these structural changes have been shown to carry prognostic information, being strongly associated with risk of development of future functional losses in the disease.\(^3\) In our study, patients with preperimetric glaucoma had a mean number of RGCs of 748 731 that was approximately 23% lower than the mean number of 973 720 cells measured in the healthy eyes included in the validation sample. Differences in the number of cells could be partially explained by age differences in the 2 groups. Therefore, we calculated the CSFI that corresponds to a percent estimate of loss compared with the age-expected number of RGCs. Patients with preperimetric glaucoma had a mean CSFI of 17% that was still significantly higher than that of healthy subjects. The diagnosis of preperimetric glaucoma in our study was based on documented evidence of progressive optic disc change in stereophotographs. Because of the wide variability of the optic nerve appearance, a single optic disc examination is frequently not diagnostic in the early stages of glaucoma.\(^5,21\) In the absence of visual field loss, a diagnosis of certainty of glaucoma can only be given by demonstrating a previous history of progressive glaucomatous changes to the optic nerve. We demonstrated that the CSFI performed well in differentiating eyes with preperimetric glaucoma from healthy subjects, with an ROC curve area of 0.85, similar to what can be obtained from analysis using SDOCT average thickness.

Staging systems based on optic disc appearance or quantitative assessment of the optic disc and RNFL have also been proposed.\(^27,30\) These classification systems are limited by the decreasing performance of imaging instruments to discriminate among the different stages of disease with increasing severity of damage. Sihota et al\(^31\) reported an AUC of only 0.705 for discriminating early to moderate visual field losses with the SDOCT parameter average thickness. A weak performance was also reported in separating moderate from advanced cases, with an ROC curve area of only 0.737. These values are similar to those found in our study for the SDOCT parameter average thickness, with corresponding AUCs of 0.77 and 0.70, respectively. Longitudinal studies have also shown an inverse relationship between disease severity and ability to detect change with imaging devices.\(^5,17,32\)

These findings collectively suggest that the use of a struc-
ture-only staging system is likely to be inadequate once the patient has been diagnosed with visual field loss. In contrast, the use of a CSFI allowed excellent separation between the different stages of the disease. The CSFI had AUCs of 0.94 to separate early from moderate loss and 0.96 for discriminating moderate from advanced loss. Although these results may seem obvious as the CSFI actually incorporates visual function information used to define severity or to classify the groups, they need to be seen in the context of the overall performance of the CSFI. The CSFI performed well not only to differentiate the different stages of glaucomatous visual field loss but also in detecting preperimetric glaucoma. Therefore, using a single CSFI, we were able to detect the earliest stages of different stages of glaucomatous visual field loss but also in detecting preperimetric glaucoma. Therefore, using a single CSFI, we were able to detect the earliest stages of the disease in more advanced cases, a task...
that was poorly performed when visual field data or SDOCT data were used in isolation.

Some overlap in CSFI values was seen among the different studied groups as shown in Figure 4. However, this is a limitation inherent to any parameter assessing biologic variables and could also be related to the variability of the tests used to obtain estimates of RGC numbers. Both SAP and SDOCT have test-retest variability and this will translate into CSFI variability. This should not have affected the comparisons performed in our study; however, it indicates the need for clinicians to obtain multiple tests to improve reproducibility, as currently performed in clinical practice.

We based our estimates of SAP- and SDOCT-derived RGC numbers on previously published work by Harwerth et al. Using normal monkeys and monkeys with laser-induced experimental glaucoma, they showed that SAP sensitivity values can provide good estimates of the amount of histologically measured RGC counts in the retina. These estimates agreed closely with those obtained from OCT RNFL thickness data. They showed a strong linear relationship between the number of RGC somas and axons obtained from functional and structural measures, respectively, when retinal eccentricity and appropriate measurement scales for neural and sensitivity losses were used. The linear relationship suggests that the lack of sensitivity of SAP for detection of early glaucomatous damage is most likely not the result of true structural changes occurring in the absence of functional losses but is rather related to the logarithmic scale used for SAP sensitivity measurements, as well as the magnitude of change required to reach statistically significant levels of abnormality. The logarithmic scale compresses the range of losses in early stages of the disease while expanding the range in later stages. These findings could suggest that a simple linearization of SAP data could improve detection of early damage. However, this is usually not the case. In fact, the ROC curve for detecting peripapillary glaucoma using estimates of RGC numbers from SAP (SAPrgc) in our study was still only 0.69, much inferior to that of RGC estimates from SDOCT data (0.88). As SAP data are originally acquired using staircase procedures based on a logarithmic scale (decibels), SAP is not good at estimating small amounts of ganglion cell losses at early stages of the disease. In contrast, by expanding the range of the scale at later stages, SAP might be more sensitive to small changes in the number of RGCs that do not seem to produce detectable changes in RNFL thickness. Despite these observations, the ability to express results of functional and structural tests in the same domain opens the possibility of combining the information from the 2 tests to increase the precision of RGC estimates, as performed in our study. By combining the estimates, one increases the precision of the final estimate of neuronal losses to better stage glaucomatous damage. However, instead of simply averaging the 2 estimates, we used a weighing scheme based on MD values. This was done to consider differences in performance of SAP and SDOCT tests at different stages of the disease for the reasons described above.

Our study has limitations. We used empirically derived formulas to estimate the number of RGCs from SAP and SDOCT data. Although estimates obtained from these formulas have been validated in multiple external cohorts, the original formula for estimating RGCs from OCT data was based on an older version of the technology, time-domain OCT. In our study, we used the same previously derived formulas, but data were obtained by SDOCT, and it is possible that modifications would be necessary to compensate for the change in technologies. However, the agreement between SAP and SDOCT data found in our study was similar to that reported by Harwerth et al, suggesting that major modifications are probably unnecessary. Another potential limitation of our study is that we used only global measures of visual function and structural damage. A sectoral analysis may provide a better representation of localized damage and improved detection of glaucoma. However, the use of sectoral information may be difficult to interpret in the context of a staging system. Additionally, sectoral information will be more variable and not necessarily better for monitoring changes over time. Further studies should evaluate whether a combination of sectoral structure and function data could improve detection and staging of glaucomatous damage. Another limitation of our study is that the presence of media opacities could potentially affect SAP-derived estimates of RGCs and, therefore, calculations of the CSFI. This is a potential limitation of most visual field-based staging systems, as they usually base their classifications at least in part on values of the MD index. However, by combining functional and structural measurements, our approach potentially reduces the effect of media opacities by relatively decreasing the influence of SAP-derived data on the final estimates of neuronal losses. Nevertheless, clinicians should be aware of the effect of media opacities when evaluating functional changes and quality of imaging test results in patients with glaucoma.

The CSFI has several desirable properties for use as a staging index. It discriminates well among the different stages of the disease and has an intuitive interpretation as the overall percent loss of neuronal tissue. In addition, it is provided on a continuous scale, avoiding the artificial categorization of the disease continuum. However, an ideal staging system for glaucoma would be highly predictive of the degree of disability from the disease. Although SAP measurements have been related to measures of quality of vision in patients with glaucoma, such a relationship is usually weak. Recent studies have proposed different methods to evaluate the degree of functional impairment caused by the disease. Future studies should be performed to attempt to correlate proposed staging systems to results of these tests or to develop staging systems based on results of tests directly measuring functional impairment in glaucoma. The methods described in our study to estimate RGC counts from a combination of structure and function could also be used to provide a useful parameter for longitudinal monitoring of glaucomatous changes. We are conducting additional studies to investigate this possibility.

In conclusion, an index combining structure and function performed better than isolated structural and functional measures for detection of perimetric and preperimetric glaucoma as well as for discriminating different
stages of the disease. Further studies should evaluate the ability of the proposed CSFI to monitor glaucomatous changes over time.

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REFERENCES


34. Medeiros FA, Weinreb RN, Boer ER, Rosen PN. Driving simulation as a performance-based test of visual impairment in glaucoma [published online April 1, 2011]. J Glaucoma. doi:10.1097/JJG.0b013e3181e7f7ed.