Relationship Between Macular Inner Retinal Layer Thickness and Corresponding Retinal Sensitivity in Normal Eyes

Makoto Araie,[1,2] Hitomi Saito,[1,2] Atsuo Tomidokoro,[3] Hiroshi Murata,[2] and Aiko Iwase[4]

[1]Kanto Central Hospital of The Mutual Aid Association of Public School Teachers, Tokyo, Japan
[2]Department of Ophthalmology, University of Tokyo Graduate School of Medicine, Tokyo, Japan
[3]Nakano Tomidokoro Eye Clinic, Tokyo, Japan
[4]Tajimi Iwase Eye Clinic, Tajimi, Gifu, Japan

Correspondence: Makoto Araie, Kanto Central Hospital of The Mutual Aid Association of Public School Teachers, 6-25-1, Kamiyoga, Setagaya-ku, Tokyo, Japan 153-8531; araie-tky@umin.net.

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Purpose. The correlation between standard automated perimetry (SAP) sensitivity and macular inner retinal layer thickness in eyes with glaucoma is well known. We examined whether the corresponding correlation is also significant in normal eyes.

Methods. One eye of each of 195 normal subjects was included. The average thickness of the macular ganglion cell-inner plexiform layers (GCIPL) and the macular retinal nerve fiber layer/GCIPL (ganglion cell complex, GCC) in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement, were measured using spectral-domain optical coherence tomography (SD-OCT) and correlated to the mean SAP sensitivity (in 1/Lambert scale) at the corresponding test points with a multiple regression analysis using age, refraction, disc size, sex, and laterality of the eye as other explanatory variables.

Results. In normal eyes, GCIPL and GCC thickness (in micrometers) showed significant correlation to SAP sensitivity in corresponding areas, with partial regression coefficients of 0.0016 ($P = 0.036$) and 0.0022 ($P = 0.023$), respectively. Other significantly correlated factors were age and GCIPL (−0.18, $P = 0.000$), age and GCC (−0.20, $P = 0.000$), and refraction and GCIPL (0.92, $P = 0.012$). Similar analyses at each of the four test points yielded essentially the same results, although partial correlation coefficients were not always significant.

Conclusions. A thicker macular GCIPL or GCC was weakly but significantly associated with higher SAP sensitivity in the corresponding macular region in normal eyes.

Keywords: normal eyes, retinal sensitivity, macular ganglion cell–related layer thickness

Glaucoma is a disease primarily associated with damage to the retinal ganglion cell (RGC) bodies and axons, which causes characteristic patterns of visual field (VF) defects and changes in the appearance of the optic nerve head (ONH). The loss of RGCs, accompanied by thinning of the ONH neuroretinal rim and the RGC-related layers of the retina, is associated with a loss of sensitivity in perimetric tests of the VF. Structural and functional tests are thus both indispensable tools in assessing the extent of glaucomatous damage, especially in its early stage, and the progression of damage over time.[1] In fact, a number of studies have used optical coherence tomography (OCT) to show that there are significant, strong correlations between standard automated perimetry (SAP)-determined VF sensitivity and OCT-determined circumpapillary retinal nerve fiber layer (cpRNFL) thickness, macular ganglion cell complex (GCC) thickness, the combined thickness of the macular RNFL (mRNFL), ganglion cell layer (GCL) and inner plexiform layer (IPL), and the combined thickness of the macular GCL and IPL (GCIPL).[2–11]

It is well known that there is a relatively large normal range for cpRNFL and macular inner retinal layer thickness,[12–16] giving rise to the intuitive assumption that eyes with thicker RGC-related retinal layers have a greater functional reserve and that regions with higher thickness should therefore have a correspondingly higher original SAP sensitivity. Nevertheless, SAP sensitivity in regions where it is in the normal range is known to be relatively independent of the thickness of the RGC-related retinal layers.[2,5,7,8] A 1-dB difference in SAP sensitivity in normal eyes (e.g., 32 dB vs. 33 dB) corresponds to a much greater difference in SAP sensitivity on a linear scale (e.g., $10^{3.2}$ vs. $10^{3.3}$) than a 1-dB difference in glaucoma eyes (e.g., 21 dB vs. 20 dB corresponds to $10^{2.1}$ vs. $10^{2.0}$). As SAP sensitivity on a linear scale has been suggested to be linearly correlated to the thickness of the RGC-related layers,[2] the slope of the thickness of the RGC-related retinal layers, such as the cpRNFL, plotted against SAP sensitivity in decibels should be much flatter in normal eyes than in glaucoma eyes.[2] However, there are several drawbacks to this method of comparing regional cpRNFL thickness and SAP sensitivity. Some of the axons composing the cpRNFL originate from different retinal regions, and interindividual variation is believed to exist in the correspondence of VF regions to disc sectors.[17–20] Region- or eccentricity-dependent differences in the relationship between the density of RGC distribution and SAP sensitivity can also

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affect such an analysis.[3] However, recent studies have revealed the early involvement of the macular region in glaucoma,[21–28] and it may be possible to mitigate some of these disadvantages of cpRNFL thickness measurement by measuring local RGC-related layer thickness in the macular region, which contains most of the RGCs.

In the current study, therefore, we set out to determine whether there was a significant correlation between regional GCIPL or GCC thickness and SAP sensitivity in the macular region of normal eyes, after adjusting possible influence of external factors such as age and refraction.[12,14–16,29–32]

Materials and Methods

Subjects

Normal subjects and open-angle glaucoma (OAG) patients were recruited using identical inclusion criteria at the six institutes that participated in this study (the University of Tokyo, Gunma University, Kanazawa University, Kyoto University, Osaka University, and Tajimi Municipal Hospital). The study protocol was approved by the institutional review board of each institution and adhered to the tenets of the Declaration of Helsinki. Written informed consent was obtained from each subject after an explanation of the study protocol.

Self-reported healthy volunteers older than 20 years underwent the following ocular examinations: refraction and corneal curvature radius measurement with an automatic refractometer (ARK-900; NIDEK, Tokyo, Japan), best-corrected visual acuity measurement with the 5-meter Landolt chart, axial length measurement with the IOL Master (Carl Zeiss Meditec, Dublin, CA, USA), slit-lamp examination, intraocular pressure (IOP) measurement with a Goldmann applanation tonometer (Haag-Streit, Koeniz, Switzerland), dilated funduscopy, and VF testing with the Humphrey Field Analyzer (HFA) 24-2 Swedish Interactive Threshold Algorithm Standard (SITA-S) program (Carl Zeiss Meditec). Exclusion criteria for the normal group included presence of family history of glaucoma; contraindications to pupil dilation; IOP 22 mm Hg or higher; best-corrected visual acuity worse than 0.8, refractive error $<$ -6.0 or $>$ $+3.0$ diopters (D); unreliable HFA results (fixation loss, false-positives, or false-negatives $>$ 20%); VF defects suggestive of glaucoma according to the Anderson-Patella criteria as described below[33]; any abnormal visual field loss consistent with ocular disease; a history of intraocular or refractive surgery; and a history of ocular or systemic diseases that could affect the results of OCT examinations, including clinically significant cataracts, diabetic retinopathy, epiretinal membrane, age-related macular degeneration, and optic nerve or retinal abnormalities. Abnormal VFs were identified with the HFA 24-2 program and were defined as (1) a cluster of $\geq$3 points in the pattern deviation plot in a single hemifield (upper or lower) with $P < 5\%$ and at least one with $P < 1\%$, (2) glaucoma hemifield test results out of the normal limits, or (3) an abnormal pattern standard deviation (PSD) with $P < 5\%$.[33]

In addition, to produce a rough estimate of the dynamic range of GCIPL and GCC thickness in the area corresponding to the four central test points of the HFA 24-2 test program, we recruited OAG patients with moderate-to-advanced glaucomatous damage meeting the following criteria: (1) experience of HFA VF testing and ability to provide reproducible results; (2) visual acuity and refractive error in the selected eye better than 0.6 and between -6.0 and $+3.0$ D, respectively; (3) absence of any other ocular pathologic changes that could affect the results of the HFA or OCT measurements listed above; (4) total deviation values in the central four test points reproducibly worse than -13.0 dB.

If both eyes of a study participant fulfilled the inclusion criteria, the eye with better image quality in spectral-domain OCT (SD-OCT) scanning was used.

OCT Measurement

OCT scanning was performed using a SD-OCT 1000 device (3D-OCT 1000; Topcon, Tokyo, Japan) equipped with a nonmydriatic fundus camera function equivalent to a commercially available nonmydriatic fundus camera (TRC-NW200; Topcon).

The SD-OCT data sets were obtained with the raster-scan protocol, in which data were obtained from a 6.0×6.0-mm^2 area (512×128 pixels) centered on the fixation over a period of approximately 2.5 seconds. To obtain accurately sized fundus images, the magnification was corrected according to the manufacturer-provided formula (a modified Littman's equation), which is based on refractive error, corneal radius, and axial length. The correspondence of the fundus photographs and OCT images was automatically confirmed using an OCT projection image (generated from OCT data by summing different retinal depth levels) and by localization of the major retinal vessels.

The data obtained during apparent eye movements were discarded and the examination repeated. Images that were influenced by involuntary blinking or saccades, indicated by breaks, shifting of the vessels, or the presence of a straight line across the fundus OCT image, or those with a quality factor $<$ 60%, were also excluded. In normal subjects, OCT measurements were repeated three times with several-second intervals, and the image with the best quality factor was used. In the macular area, the fovea was automatically identified in the OCT image as the pixel with the thinnest total retinal thickness adjacent to the fixation. The mRNFL and GCIPL were segmented automatically in all B-scan images.[34] Confirmation of the segmentation of the images was performed by an experienced examiner (AT). In the OAG patients, OCT measurements were not repeated after an image was obtained that met the above criteria and in which segmentation of the mRNFL and GCIPL was confirmed. Data for the GCC were obtained with the following formula: (mRNFL + GCIPL).

Immediately after SD-OCT measurement, a color fundus photograph centered on the disc was taken with the nonmydriatic fundus camera function of the 3D-OCT 1000 device. The disc area in the fundus photographs was identified by determining the area inside a closed spline curve fitted to seven manually determined points on the disc margin. The magnification was corrected as described above.

Relationship Between Thickness of GCIPL and GCC and SAP Sensitivity in Corresponding Retinal Areas

GCIPL and GCC thickness in a circular retinal area with a diameter of 0.6 mm (corresponding to approximately 2° of visual angle) in the four central test points of the HFA 24-2 program, adjusted for RGC displacement, were assessed in SD-OCT raster scan data and averaged. The diameter of the retinal area (2°) was similar to the grid size of the HFA 10-2 test program and roughly twice as large as the size-III stimulus points projected onto the fundus in addition to the maximum range of small physiological eye movements during fixation, such as drift. Ganglion cell displacement was approximated with the following formula: $[y = 1.29 \times (x + 0.46)^{0.67}$, where y is ganglion cell eccentricity and x is cone eccentricity].[35] The decibel values for VF sensitivity in the four central test points of the HFA 24-2 test program were antilogged to obtain the sensitivity in the linear scale (1/Lambert = $10^{0.1 \times dB}$; liner

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Table 1. Characteristics of Normal Eyes

Age, y	48.5 ± 16.5
Male/female	103/92
Right/left	95/100
Spherical equivalent refraction, D	−0.31 ± 1.46
Intraocular pressure, mm Hg	14.1 ± 2.3
Mean deviation, dB	−0.26 ± 1.5
Mean sensitivity of the four central test points	
1/Lambert; dB scale	2064 ± 679; 32.9 ± 1.5
Disc area, mm²	2.06 ± 0.45
$GCIPL_{4\ test\ points}$, μm	91.3 ± 6.8
$GCC_{4\ test\ points}$, μm	123.2 ± 8.5

$GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) represents the mean thickness of the RGC-IPL (GCC) in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement.

sensitivity),[2,3,36,37] and the mean of linear sensitivity of the four test points was calculated using the above four antilogged decibel values. Age,[12,14–16,30,31] sex,[14,30,31] laterality of the eye,[29,32] refraction,[14,16,31] and disc size[12] are external factors that have been reported to influence OCT-measured thickness of RGC-related retinal layers such as the cpRNFL, GCIPL, and GCC. The possible influence of these factors was taken into account with a multiple linear regression analysis:

$$\begin{aligned} &GCIPL_{4\ test\ points}(GCC_{4\ test\ points}) \\ &= A_1 \times Age + A_2 \times Spherical\ equivalent\ refraction \\ &\quad + A_3 \times Sex + A_4 \times Laterality + A_5 \times Disc\ size \\ &\quad + A_6 \times (SAP\ sensitivity\ in\ the\ linear\ scale) \\ &\quad + Intercept \end{aligned} \tag{1}$$

where $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) indicates the mean of the measured GCIPL (GCC) thickness in the above-described retinal area. Similar calculations were performed to study the relationship between the thickness of the GCIPL (GCC) and SAP sensitivity in the linear scale at each of the four central test points. That is,

$$\begin{aligned} &GCIPL_{each\ of\ 4}(GCC_{each\ of\ 4}) \\ &= B_1 \times Age + B_2 \times Spherical\ equivalent\ refraction \\ &\quad + B_3 \times Sex + B_4 \times Laterality + B_5 \times Disc\ size + B_6 \\ &\quad \times (SAP\ sensitivity\ in\ the\ linear\ scale\ at\ each\ test\ point) \\ &\quad + Intercept \end{aligned} \tag{2}$$

where $GCIPL_{each\ of\ 4}$ ($GCC_{each\ of\ 4}$) indicates the measured GCIPL (GCC) thickness at each of the four central test points.

The data were analyzed using SPSS (21.0J for Windows; SPSS Japan, Inc., Tokyo, Japan).

Results

A total of 195 eyes of 195 normal subjects (female/male = 92/103) and a total of 10 OAG eyes with moderate to advanced glaucomatous damage of 10 OAG patients (female/male = 6/4) were included. All subjects were Japanese, and the majority of the normal subjects had previous experience of VF testing with the HFA. The characteristics of the normal and OAG eyes are summarized in Tables 1 and 2. In the normal eyes, mean retinal sensitivity in the four central test points averaged 2064 ± 679 (SD) in the linear scale (1/Lambert) and 32.9 ± 1.5 dB in the decibel scale. Mean GCIPL and GCC thickness in the 0.6-mm-diameter circular retinal area corresponding to the four central test points ($GCIPL_{4\ test\ points}$ and $GCC_{4\ test\ points}$) was 91.3 ± 6.8 μm and 123.2 ± 8.5 μm, respectively (Table 1; Fig. a, b). In the OAG group, the corresponding figures were 22.7 ± 14.2 in the linear scale (1/Lambert), 12.2 ± 4.7 dB in the decibel scale, 63.5 ± 6.6 μm and 73.9 ± 11.0 μm, respectively (Table 2; Fig. c, d).

Table 2. Characteristics of Open Angle Glaucoma Eyes With Advanced Central Visual Field Damage

Age, y	57.3 ± 11.4
Male/female	4/6
Right/left	5/5
Spherical equivalent refraction, D	−1.73 ± 2.42
Intraocular pressure, mm Hg	14.0 ± 2.7
Mean deviation, dB	−15.8 ± 6.4
Mean sensitivity of the four most central test points	
1/Lambert; dB scale	22.7 ± 14.2; 12.2 ± 4.7
Disc area, mm²	2.52 ± 0.71
$GCIPL_{4\ test\ points}$, μm	63.5 ± 6.6
$GCC_{4\ test\ points}$, μm	73.9 ± 11.0

$GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) represents the mean thickness of the RGC-IPL layer (GCC) in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement.

The results of the multiple regression analysis are summarized in Table 3. Among the explanatory variables, age and retinal sensitivity, age and refraction, and retinal sensitivity and refraction showed significant intercorrelation (Pearson's correlation coefficients: –0.526, 0.506, and –0.293, $P < 0.001$, respectively), but colinearity was judged to be absent (variance inflation factor value <2.0).

For the $GCIPL_{4\ test\ points}$ (in micrometers), SAP sensitivity in the corresponding area (1/Lambert), age (years), and spherical equivalent refraction (diopters) showed significant correlation, with partial regression coefficients of 0.0016 ± 0.0008 (SE) ($P = 0.036$), –0.18 ± 0.04 ($P = 0.000$), and 0.92 ± 0.35 ($P = 0.012$), respectively (Table 3). For the $GCC_{4\ test\ points}$ (in micrometers), SAP sensitivity (1/Lambert) in the corresponding area and age (years) showed a significant correlation, with partial regression coefficients of 0.0022 ± 0.0010 ($P = 0.023$) and –0.20 ± 0.04 ($P = 0.000$), respectively (Table 4).

Analyses at each of the four central test points yielded essentially the same results, but at one or two test points, partial correlation coefficients did not reach significance level. For the $GCIPL_{each\ of\ 4}$ (in micrometers), SAP sensitivity (1/Lambert) showed a significant correlation, with partial regression coefficients of 0.0019 ± 0.0009 and 0.0020 ± 0.0009 ($P = 0.046$ and $P = 0.031$) at two of four test points, age with partial correlation coefficients of –0.11 ± 0.04 ~ –0.25 ±

Table 3. Results of Multiple Regression Analysis for the $GCIPL_{4\ test\ points}$

Variable	Partial Regression Coefficient	*P* Value
Age, y	−0.18 (0.04)	0.000
Sensitivity, 1/Lambert	0.0016 (0.0008)	0.036
Disc area, mm²	0.39 (0.83)	0.631
Refraction, D	0.92 (0.35)	0.012
Sex	−0.32 (0.88)	0.720
Laterality	−0.11(0.88)	0.720

$GCIPL_{4\ test\ points}$ represents the mean thickness (in micrometers) of the RGC-IPL in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement. Figures in parentheses indicate SE.

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Table 4. Results of Multiple Regression Analysis for $GCC_{4\ test\ points}$

Variable	Partial Regression Coefficient	*P* Value
Age, y	−0.20 (0.04)	0.000
Sensitivity, 1/Lambert	0.0022 (0.0010)	0.023
Disc area, mm^2	0.43 (1.05)	0.679
Refraction, D	0.50 (0.45)	0.270
Sex	−0.63 (1.11)	0.571
Laterality	−0.36 (1.12)	0.749

$GCC_{4\ test\ point}$ indicates the mean thickness of the GCC in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement. Figures in parentheses indicate SE.

0.05 ($P = 0.008 \sim 0.000$) at all four test points, and refraction with partial correlation coefficients of 1.26 ± 0.45 and 1.40 ± 0.46 ($P = 0.003$ and 0.008) at two of four test points, respectively. For the $GCC_{each\ of\ 4}$ (in micrometers), SAP sensitivity (1/Lambert) showed a significant correlation with partial regression coefficients of 0.0023 ± 0.0011 and 0.0023 ± 0.0010 ($P = 0.038$ and $P = 0.023$) at two of four test points and correlation of marginal significance with partial correlation coefficients of 0.0020 ± 0.0011 and 0.21 ± 0.0011 ($P = 0.062$) at two of four test points, age with partial correlation coefficients of −0.12 ± 0.05 ∼ −0.25 ± 0.05 ($P = 0.009 \sim 0.000$) at all four test points, and refraction with partial correlation coefficients of 1.14 ± 0.50 ($P = 0.023$) at one of four test points, respectively.

Discussion

Greenfield et al.[38] reported that mean deviation values were significantly correlated with mean macular thickness measurements made with time domain-OCT (TD-OCT) in eyes with moderately advanced glaucoma. Several studies have found that GCIPL and GCC thickness are correlated to global or corresponding regional SAP sensitivity (in the decibel and linear scale) in groups consisting only of glaucoma eyes or of both glaucoma and normal eyes, and these studies reported correlation coefficients of approximately 0.6.[5,6,7,9,11,26] This is somewhat greater than the correlation between regional SAP sensitivity and thickness in corresponding sectors of the cpRNFL.[7,11,26] However, to the best of our knowledge, this is the first study of the correlation between GCIPL and GCC thickness and SAP sensitivity in corresponding macular regions in normal eyes: In the corresponding macular region of normal eyes, both age and SAP sensitivity showed a significant correlation to GCIPL and GCC thickness. In addition, we found that refraction showed a significant correlation to $GCIPL_{4\ test\ points}$. The strong correlation we observed of age to GCIPL and GCC thickness was entirely expected.[13–15] The correlation of axial length to GCIPL and GCC thickness has also been previously reported.[13,14,39] Analyses at each of the four central test points showed essentially the same results, although at some test points, correlation between $GCIPL_{each\ of\ 4}$ or $GCC_{each\ of\ 4}$ and SAP sensitivity (1/Lambert) at the corresponding test point or refraction did not reach significance level. This is probably due to the variability inherent in the SAP sensitivity and OCT-based thickness measurements, which were averaged and rounded in the original analysis, assuming that GCIPL or GCC thickness and SAP sensitivity relationship would not be different among the most central four test points, as far as the normal eye is concerned.

In the normal eyes, after adjusting for the possible influence of external factors, we found that the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) increased in thickness by approximately 1.6 (2.2) μm for every increase in SAP sensitivity of 1000 on the linear scale (1/Lambert), which corresponds to an increase in decibels of approximately 2.1 (from 32.0 to 34.1 dB). Even in cases with advanced glaucomatous damage, in which almost all the RGCs are seriously damaged, GCIPL (GCC) thickness cannot decrease to zero because these layers include retinal neuronal cells other than RGCs, such as glial cells or blood vessels.[2,40] In the eyes with glaucoma, which had TD values in all of the central four test points worse than −13 dB, the thickness of the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) averaged approximately 64 (74) μm, while SAP sensitivity in the corresponding area was approximately 23 on the linear scale, or 13 dB (representing a TD value of approximately −20 dB). On the other hand, the thickness of the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) in the normal eyes averaged 91 (123) μm, while SAP sensitivity in the corresponding area was approximately 2000 on the linear scale, or 33 dB. Thus, in this study we observed a difference in $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) thickness of approximately 27 μm (= 91 − 64; 49 = 123 − 74), between the normal and glaucoma eyes, and a corresponding difference in the SAP sensitivity of approximately 2000 (≈2000 – 23) on the linear scale. It is clear that this difference cannot be explained by our finding that in normal eyes, a difference of 1.6 (2.2) μm corresponded to a difference in SAP sensitivity of approximately 1000. Furthermore, the reported slopes of GCIPL (GCC) thickness plotted against the corresponding SAP sensitivity were much steeper in eyes affected by primary open-angle glaucoma (POAG)[5,11] than the partial regression coefficient values currently obtained in normal eyes. In the literature, a similar finding has also been reported for cpRNFL thickness: cpRNFL thickness plotted against SAP sensitivity in the corresponding area in normal eyes tended to increase as SAP sensitivity increased, although the slope did not reach significance level.[5,8] The fact that the number of normal eyes included was twice or more in the current study and that the results obtained from the four most central test points were averaged probably increased detectability of a small, but statistically significant correlation. The current findings indicated that only a small increase in the number of RGCs may result in a relatively large increase in the SAP sensitivity in normal eyes and vice versa in POAG eyes. It may be possible that the relationship between the SAP sensitivity and OCT-based GCIPL (GCC) thickness is different between normal and OAG eyes. For example, plasticity in visual cortex or normal adjustment of the brain to chronic modifications in the visual input may modify the relationship between input from RGCs and perceived sensitivity.[41]

It is well known that GCIPL and GCC thickness decreases with age.[13–15] Age-related changes in the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) in the normal eyes in this study were estimated to be approximately 1.8 (2.0) μm/decade. Published data indicates that age-related loss of the RGCs is approximately 6%/decade.[42–46] If the RGC-related thickness of the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) may be roughly assumed to be approximately 27 (49) μm, then a 6%/decade loss in the $GCIPL_{4test\ points}$ ($GCC_{4\ test\ points}$) would correspond to a loss of thickness of approximately 1.6 (2.9) μm/decade. The slightly higher value for GCC may be partly attributable to the fact that a portion of the mRNFL comprising the GCC originates from external retinal areas. These values for age-related losses are compatible with the value extrapolated with partial regression coefficients for age in our multiple linear regression analysis, that is, 1.8 (2.0) μm/decade.

Mwanza et al.[13] reported that GCIPL thickness measured with Cirrus HD-OCT (Carl Zeiss Meditec) decreased by 1.06%

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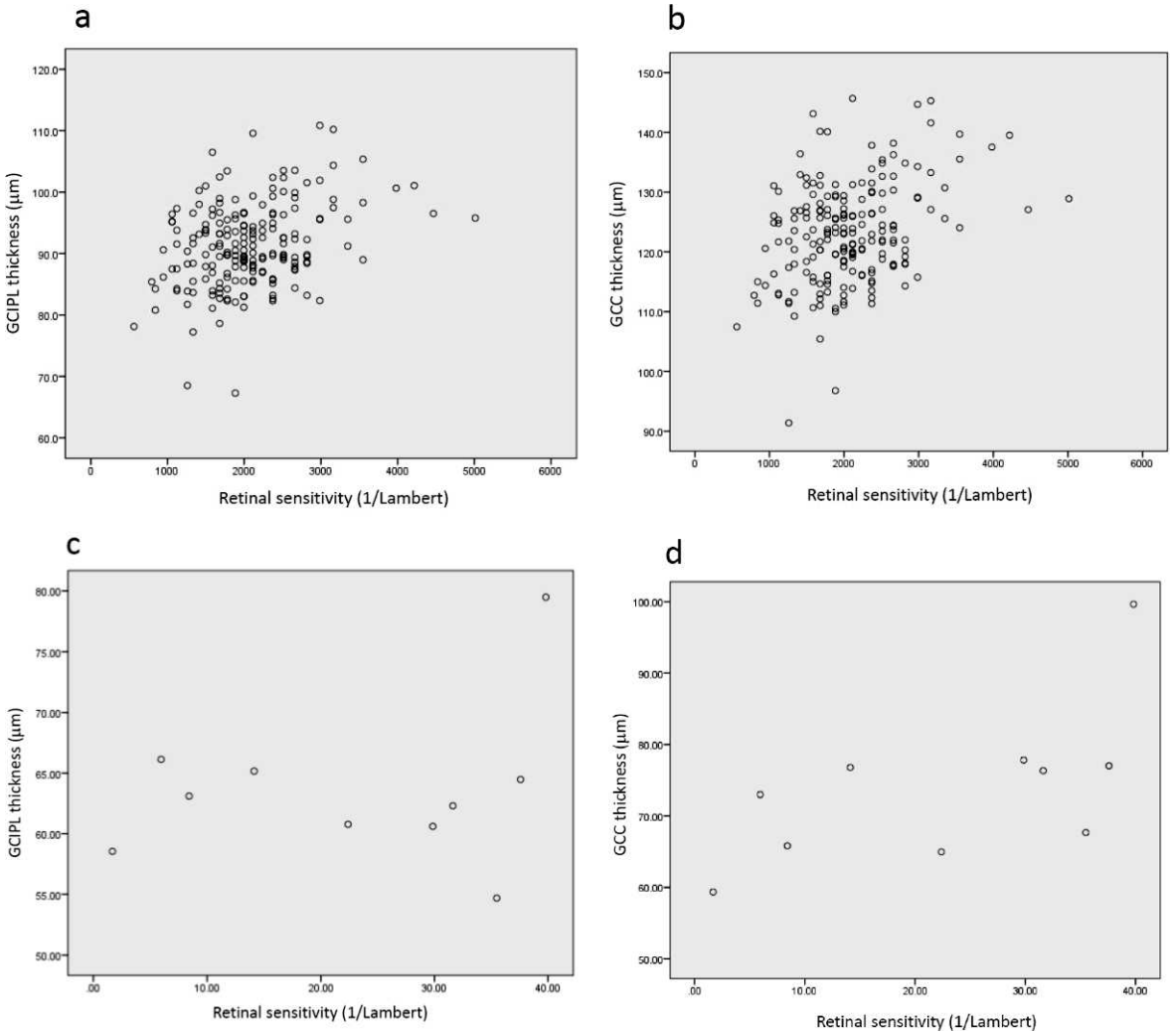

Figure. (**a**) Plots of GCIPL thickness (in micrometers) versus retinal sensitivity in the linear scale in normal subject eyes. The GCIPL thickness represents the mean thickness of the RCG-IPL in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement. (**b**) Plots of GCC thickness (in micrometers) versus retinal sensitivity in the linear scale in normal subject eyes. The GCC thickness represents the mean thickness of the RGC complex layer in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement. (**c**) Plots of GCIPL thickness (in micrometers) versus retinal sensitivity in the linear scale in subject eyes with advanced glaucomatous damage. The GCIPL thickness represents the mean thickness of the RGC-IPL layer in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement. (**d**) Plots of GCC thickness (in micrometers) versus retinal sensitivity in the linear scale in subject eyes with advanced glaucomatous damage. The GCC thickness represents the mean thickness of the RGC complex layer in four regions with 0.6-mm-diameter circular area corresponding to the four central test points of the Humphrey Field Analyzer 24-2 test program, adjusted for ganglion cell displacement. (**a**–**d**) Retinal sensitivity (1/Lambert) represents the mean of the sensitivities (1/Lambert) of the four central test points.

for every 1-mm increase in axial length. In the 195 normal eyes included in this study, we found, using a simple regression analysis, that spherical equivalent refraction decreased by approximately 2 D for every 1-mm increase in axial length (R^2 = 0.47). Extrapolating from the current multiple regression analysis of the $GCIPL_{4\ test\ points}$ suggests that there should be a decrease of approximately 1.8 µm for each 1-mm increase in axial length, while calculations based on the results of Mwanza et al.[13] suggest that the decrease should be approximately 1.0 µm (91 µm × 0.0106 ≈ 1 µm)/mm. The fact that the current subject eyes included only those with refractive error between −6 and + 3 D and that the SD-OCT instrument used was different may at least partly explain the discrepancy between 1.8 and 1.0 µm.

Standard automated perimetry sensitivity decreases with age at a rate that has been reported to be approximately 0.7 dB/decade.[47–49] It has also been reported that this decline in visual function rapidly accelerates after 50 years of age,[50,51] and according to Lachenmayr et al.,[51] the rate of age-related decrease in SAP sensitivity is approximately 2.0 dB/decade after 46 years of age. A decrease of 0.7 dB (32.9 dB − 32.2 dB)/decade corresponds to a decrease in the linear scale of 300/decade. In our multiple regression analysis, the estimated value of the partial regression coefficient for decrease per decade in $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) thickness in normal eyes was 1.8 µm (2.0)/decade, while a 1.6-µm (2.2-µm) change in $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) thickness corresponded to a change of 1000 in the linear scale, or a 2.1-dB change in SAP sensitivity. Calculating the expected decrease per decade in SAP sensitivity from these results returns a value for age-related decrease in the linear scale of approximately 900 (1000 × 1.6/1.8, or 2.0/2.2)/decade,

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which is an apparent discrepancy with a 300/decade (0.7 dB/decade) decrease. However, if we use the rate of age-related decrease in SAP sensitivity after 46 years of age reported by Lachenmayr et al.[51] (the mean age of our subjects was 48.5 years of age) as a basis for calculation, we find that the decrease in SAP sensitivity per decade in normal eyes can be expected to be 2.0 dB (= 32.9 − 30.9)/decade or 720/decade in the linear scale, a result that is in broad agreement with the result of calculations based on our findings for the estimated relationship between thickness in the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) and the corresponding SAP sensitivity in normal eyes, namely, 900/decade in the linear scale.

There were several limitations to this study. We assumed that there was a linear relationship between $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) thickness and SAP sensitivity in the linear scale in normal eyes. However, the linearity of the relationship between the thickness of the RGC-related retinal layers and SAP sensitivity in the linear scale was originally proposed in glaucoma eyes, not in normal eyes.[2] In order to estimate the thickness of the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) after all the RGCs had died out, we measured $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) thickness in OAG eyes that had undergone severe damage to the four central test points and had SAP sensitivity in the linear scale approximately 1/100 that of normal eyes. Since it was relatively difficult to find such severely damaged OAG eyes that also met the other inclusion criteria, we calculated the residual $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) thickness, attributable to glial cells, interstitial cells, and neural cells other than RGCs, from measurements of only 10 eyes of 10 patients. It is therefore possible that our estimate of residual thickness was not sufficiently accurate. In fact, our estimated value, 64 (74) μm, was somewhat greater than that reported by previous studies.[9] Thus, our calculation that the thickness of the RGC-related layers in the $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) was 27 (49) μm may have been prone to error. There is a variability in both SAP sensitivity and SD-OCT–based GCIPL (GCC) thickness measures, which should not be constant across the range of values. Furthermore, the rates of $GCIPL_{4\ test\ points}$ ($GCC_{4\ test\ points}$) change per change in the SAP sensitivity of 1000 in the linear scale currently estimated in normal eyes were approximately 2 μm, which is close to the reproducibility range of GCIPL (GCC) thickness measurements with the SD-OCT instrument currently used.[52] Presence of potential limitation of the model applied, a linear regression model, and variability of SAP- and SD-OCT–based measures must be taken into consideration in interpreting the values of partial correlation coefficients currently obtained. Another limitation of this study was that approximately one-fifth of our normal subjects were relatively inexperienced with automated perimetric testing. Confounding effects attributable to this, however, were likely to have been minor, as it has been reported that the learning effect in normal eyes on perimetric results is relatively small for test points in the central subfield.[53–55]

In summary, we found that a thicker macular GCIPL or GCC was weakly, but significantly associated with higher SAP sensitivity in the corresponding macular regions in normal human eyes after adjustment for external confounding factors such as age and that the change in layer thickness per linear unit of visual sensitivity was considerably smaller than that found in glaucomatous eyes.

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References

1. Johnson CA, Cioffi GA, Liebmann JR, Sample PA, Zangwill LM, Weinreb RN. The relationship between structural and functional alterations in glaucoma: a review. *Semin Ophthalmol*. 2000;15:221–233.
2. Hood DC, Kardon RH. A framework for comparing structural and functional measures of glaucomatous damage. *Prog Retin Eye Res*. 2007;26:688–710.
3. Harwerth RS, Wheat JL, Fredette MJ, Anerson DR. Linking structure and function in glaucoma. *Prog Retin Eye Res*. 2010; 29:249–271.
4. Hood DC, Anderson SC, Wall M, Raza AS, Kardon RH. A test of linear model of glaucomatous structure-function loss reveals sources of variability in retinal nerve fiber and visual field measurements. *Invest Ophthalmol Vis Sci*. 2009;50:4254–4266.
5. Cho JW, Sung KR, Lee S, et al. Relationship between visual field sensitivity and macular ganglion cell complex thickness as measured by spectral-domain optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2010;51:6401–6407.
6. Kim NR, Lee ES, Seong GJ, Kim JH, An HG, Kim CY. Structure-function relationship and diagnostic value of macular ganglion cell complex measurement using Fourier-domain OCT in glaucoma. *Invest Ophthalmol Vis Sci*. 2010;51:4646–4651.
7. Rao HL, Zangwill LM, Weinreb RN, Leite MT, Sample PA, Medeiros FA. Structure-function relationship in glaucoma using spectral-domain optical coherence tomography. *Arch Ophthalmol*. 2011;129:864–871.
8. Alasil T, Wang K, Yu F, et al. Correlation of retinal nerve fiber thickness and visual fields in glaucoma: a broken stick model. *Am J Ophthalmol*. 2014;157:953–959.
9. Raza AS, Cho J, de Moraes CGV, et al. Retinal ganglion cell layer thickness and local visual field sensitivity in glaucoma. *Arch Ophthalmol*. 2011;129:1529–1536.
10. Wheat JL, Rangaswamy NV, Harwerth RS. Correlating RNFL thickness by OCT with perimetric sensitivity in glaucoma patients. *J Glaucoma*. 2012;21:95–101.
11. Shin HY, Park HY, Jung KI, Park CK. Comparative study of macular ganglion cell-inner plexiform layer and peripapillary retinal nerve fiber layer measurement; structure-function analysis. *Invest Ophthalmol Vis Sci*. 2013;54:7344–7353.
12. Hirasawa H, Tomidokoro A, Araie M, et al. Peripapillary retinal nerve fiber layer thickness determined by spectral-domain optical coherence tomography in ophthalmologically normal eyes. *Arch Ophthalmol*. 2010;128:1420–1426.
13. Mwanza JC, Durbin MK, Budenz DL, et al.; for Normative Database Study Group. Profile and predictors of normal ganglion cell-inner plexiform layer thickness measured with frequency-domain optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2011;52:7872–7879.
14. Ooto S, Hangai M, Tomidokoro A, et al. Effects of age, sex, and axial length on the three-dimensional profile of normal macular layer structures. *Invest Ophthalmol Vis Sci*. 2011; 52:8769–8779.
15. Girkin CA, McGwin G Jr, Sinai MJ, et al. Variation in optic nerve and macular structure with age and race with spectral-domain optical coherence tomography. *Ophthalmology*. 2011; 118:2403–2408.
16. Knight OJ, Girkin CA, Budenz DL, Durbin MK, Feuer WJ; Cirrus OCT; for Normative Database Study Group. Effect of race, age, and axial length on optic nerve head parameters and retinal nerve fiber layer thickness measured by Cirrus HD-OCT. *Arch Ophthalmol*. 2012;130:312–318.

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17. Wirtschafter JD, Becker WL, Howe JB, Younge BR. Glaucoma visual field analysis by computed profile of nerve fiber function in optic disc sectors. *Ophthalmology*. 1982;89:255–267.
18. Weber J, Dannheim F, Dannheim D. The topographical relationship between optic disc and visual field in glaucoma. *Acta Ophthalmol*. 1990;68:568–574.
19. Garway-Heath DF, Poinoosawmy D, Fitzke FW, Hitchings RA. Mapping the visual field to the optic disc in normal tension glaucoma eyes. *Ophthalmology*. 2000;107:1809–1815.
20. Ferreras A, Pablo LE, Garway-Heath DF, Fogagnolo P, García-Feijoo J. Mapping standard automated perimetry to the peripapillary retinal nerve fiber layer in glaucoma. *Invest Ophthalmol Vis Sci*. 2008;49:3018–3025.
21. Tan O, Li G, Lu AT-H, Varma R, Huang D; for Advanced Imaging for Glaucoma Study Group. Mapping of macular substructures with optical coherence tomography for glaucoma diagnosis. *Ophthalmology*. 2008;115:949–956.
22. Tan O, Chopra V, Lu AT-H, et al. Detection of macular ganglion cell loss in glaucoma by Fourier-domain optical coherence tomography. *Ophthalmology*. 2009;116:2305–2314.
23. Seong M, Sung KR, Choi EH, et al. Macular and peripapillary retinal nerve fiber layer measurements by spectral domain optical coherence tomography in normal-tension glaucoma. *Invest. Ophthalmol Vis Sci*. 2010;51:1446–1452.
24. Inuzuka H, Kawase K, Sawada A, Aoyama Y, Yamamoto T. Macular retinal thickness in glaucoma with superior or inferior visual hemifield defects. *J Glaucoma*. 2013;22:60–64.
25. Hood DC, Raza AS, de Moraes CGV, et al. Initial arcuate defects within the central 10 degrees in glaucoma. *Invest Ophthalmol Vis Sci*. 2011;52:940–946.
26. Na JH, Kook MS, Lee Y, Yu SJ, Choi J. Detection of macular and circumpapillary structural loss in normal hemifield areas of glaucomatous eyes with localized visual field defects using spectral-domain optical coherence tomography. *Graefes Arch Clin Exp Ophthalmol*. 2012;250:595–602.
27. Mwanza JC, Durbin MK, Budenz DL, et al. Glaucoma diagnostic accuracy of ganglion cell-inner plexiform layer thickness: comparison with nerve fiber layer and optic nerve head. *Ophthalmology*. 2012;119:1151–1158.
28. Takayama K, Hangai M, Durbin M, et al. A novel method to detect local ganglion cell loss in early glaucoma using spectral-domain optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2012;53:6904–6913.
29. Budenz DL. Symmetry between the right and left eyes of the normal retinal nerve fiber layer measured with optical coherence tomography (an AOS thesis). *Trans Am Ophthalmol Soc*. 2008;106:252–275.
30. Ooto S, Hangai M, Sakamoto A, et al. Three-dimensional profile of macular retinal thickness in normal Japanese eyes. *Invest Ophthalmol Vis Sci*. 2010;51:465–473.
31. Mwanza JC, Oakley JD, Budenz DL, Chang RT, Knight OJ, Feuer WJ. Macular ganglion cell-inner plexiform layer: automated detection and thickness reproducibility with spectral domain-optical coherence tomography in glaucoma. *Invest Ophthalmol Vis Sci*. 2011;52:8323–8329.
32. Mwanza JC, Durbin MK, Budenz DL.; for Cirrus OCT Normative Database Study Group. Interocular symmetry in peripapillary retinal nerve fiber layer thickness measured with the Cirrus HD-OCT in healthy eyes. *Am J Ophthalmol*. 2011;151:514–521.
33. Anderson DR, Patella VM. *Automated Static Perimetry*. St. Louis, MO: Mosby Inc.; 1999:152–153.
34. Yang Q, Reisman CA, Wang Z, et al. Automated layer segmentation of macular OCT images using dual-scale gradient information. *Opt Express*. 2010;18:21293–21307.
35. Sjöstrand J, Popovic Z, Conradi N, Marshall J. Morphometric study of the displacement of retinal ganglion cells subserving cones within the human fovea. *Graefes Arch Clin Exp Ophthalmol*. 1999;237:1014–1023.
36. Swanson WH, Felius J, Pan F. Perimetric defects and ganglion cell damage: interpreting linear relations using a two-stage neural model. *Invest Ophthalmol Vis Sci*. 2004;45:466–472.
37. Gardiner SK, Demirel S, Johnson CA, Swanson WH. Assessment of linear-scale indices for perimetry in terms of progression in early glaucoma. *Vision Res*. 2011;51:1801–1810.
38. Greenfield DS, Bagga H, Knighton RW. Macular thickness changes in glaucomatous optic neuropathy detected using optical coherence tomography. *Arch Ophthalmol*. 2003;121:41–46.
39. Takeyama A, Kita Y, Kita R, Tomita G. Influence of axial length on ganglion cell complex (GCC) thickness and on GCC thickness to retinal thickness ratios in young adults. *Jpn J Ophthalmol*. 2014;58:86–93.
40. Sihota R, Sony P, Gupta V, Dada T, Singh R. Diagnostic capability of optical coherence tomography in evaluating the degree of glaucomatous retinal nerve fiber damage. *Invest Ophthalmol Vis Sci*. 2006;47:2006–2010.
41. Safran AB, Landis T. From cortical plasticity and unawareness of visual field defects. *J Neuroophthalmol*. 1999;19:84–88.
42. Balazsi AG, Rootman J, Drance SM, et al. The effect of age on the nerve fiber population of the human optic nerve. *Am J Ophthalmol*. 1984;97:760–766.
43. Jonas JB, Schmidt AM, Müller-Bergh JA, et al. Human optic nerve fiber count and optic disc size. *Invest Ophthalmol Vis Sci*. 1992;33:2012–2018.
44. Blanks JC, Torigoe Y, Hinton DR, et al. Retinal pathology in Alzheimer's disease. I. Ganglion cell loss in the foveal/parafoveal retina. *Aging*. 1996;17:377–384.
45. Mikelberg FS, Drance SM, Schulzer M, et al. The normal human optic nerve. Axon count and axon diameter distribution. *Ophthalmology*. 1998; 96:1325–1328.
46. Harman A, Abrahams B, Moore S, et al. Neural density in the human retinal ganglion cell layer from 16–77 years. *Anat Rec*. 2000;260:124–131.
47. Heijl A, Lindgren G, Olsson J. Normal variability of static perimetric threshold values across the central visual field. *Arch Ophthalmol*. 1987;105:1544–1549.
48. Iwase A, Kitazawa Y, Ohno Y. On age-related norms of the visual field. *Jpn J Ophthalmol*. 1988;32:429–437.
49. Johnson CA, Adams AJ, Lewis RA. Evidence for neural basis of age-related visual field loss in normal observers. *Invest Ophthalmol Vis Sci*. 1989;30:2056–2064.
50. Johnson MA, Choy D. On the definition of age-related norms for visual function testing. *Appl Opt*. 1987;26:1449–1454.
51. Lachenmayr BJ, Kojetinski S, Ostermaier N, Angstwurm K, Vivell PMO, Schaumbergar M. The different effects of aging on normal sensitivity in flicker and light-sense perimetry. *Invest Ophthalmol Vis Sci*. 1994;35:2741–2748.
52. Hirasawa H, Araie M, Tomidokoro A, et al. Reproducibility of thickness measurements of macular inner retinal layers using SD-OCT with or without correction of ocular rotation. *Invest Ophthalmol Vis Sci*. 2013;54:2562–2570.
53. Heijl A, Lindgren G, Olssen J. The effect of perimetric experience in normal subjects. *Arch. Ophthalmol*. 1989;107:81–86.
54. Werner EB, Krupin T, Adelson A, Feitl ME. Effect of patient experience on the results of automated perimetry in glaucoma suspect patients. *Ophthalmology*. 1990;97:44–48.
55. Wood JM, Wild JM, Hussey MK, Crews SJ. Serial examination of the normal visual field using Octopus automated projection perimetry. Evidence for a learning effect. *Acta Ophthalmol*. 1987;65:326–333.

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