AMERICAN ACADEMY™ OF OPHTHALMOLOGY

Segmentation Errors in Macular Ganglion Cell Analysis as Determined by Optical Coherence Tomography

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Purpose: To investigate the prevalence, features, associated factors, and reproducibility of segmentation errors in macular ganglion cell inner plexiform layer (GCIPL) thickness measurement as determined by optical coherence tomography (OCT).

Design: Cross-sectional study.

Participants: Five hundred thirty-eight glaucomatous and healthy eyes from 290 subjects with OCT-measured macular GCIPL thickness were enrolled. Eyes with macular disorders, including epiretinal membrane, macular degeneration, macular hole, and myopic maculopathy, were excluded.

Methods: By inspecting 128 cross-sectional OCT B-scan images per eye, the presence (yes vs. no), layer (anterior vs. posterior border), location (quadrants), and area (diffuse vs. focal) of macular GCIPL segmentation error were investigated. The effects of age, refractive error, mean deviation of visual field test, circumpapillary retinal nerve fiber layer thickness obtained by OCT, and signal strength of OCT scan on the presence of macular GCIPL segmentation errors were evaluated. In eyes with segmentation errors, repeated OCT examinations were performed to investigate the reproducibility of the segmentation errors.

Main Outcome Measures: The prevalence, features, associated factors, and reproducibility of macular GCIPL segmentation errors were assessed.

Results: Among the 538 eyes, 52 eyes (9.7%) showed segmentation errors in macular GCIPL thickness measurement. The most common features of segmentation errors were that they affected both the anterior and posterior borders, were located at the nasal quadrant (centered to the fovea), and were diffuse. In univariate analysis, the presence of segmentation error was associated significantly with younger age ($P < 0.001$), higher degree of myopia ($P < 0.001$), and lower signal strength of OCT scan ($P = 0.038$). In multivariate analysis, only higher degree of myopia was associated significantly with the presence of segmentation error ($P < 0.001$). In repeated examinations, segmentation errors were reproducible in 24 eyes (46.2%). In other cases, the features of segmentation errors changed or disappeared.

Conclusions: Although the OCT segmentation algorithm accurately detected macular GCIPL thickness in most eyes without macular disorders, in some cases, segmentation errors were found, especially in myopic eyes. In repeated examinations, approximately half of the errors were nonreproducible. These findings should be considered when assessing macular GCIPL thickness using OCT. *Ophthalmology 2016;123:950-958 © 2016 by the American Academy of Ophthalmology.*

Glaucoma is a progressive optic neuropathy that is characterized by progressive changes in the circumpapillary retinal nerve fiber layer (RNFL), optic nerve head (ONH), and macular retinal ganglion cells (RGCs). Optical coherence tomography (OCT) is an imaging technique that can measure RNFL thickness, ONH parameters, and macular RGC thickness. Recent studies have reported that OCT-derived parameters show excellent glaucoma diagnostic ability[1–7]; however, there remains the possibility of segmentation errors in RNFL thickness, ONH parameters, and macular RGC thickness, as determined by an automated OCT segmentation algorithm. Investigation of the prevalence, as well as associated factors, of segmentation errors in OCT parameters would be helpful to prevent misinterpretation of glaucoma evaluation using OCT. Several studies reported the prevalence and associated factors of segmentation errors in RNFL thickness and ONH parameters[8–11]; however, to date, little is known about segmentation errors in macular RGC thickness measurement.

Previous studies reported that segmentation errors in total retinal thickness measurement, as determined by OCT, were found in eyes with various macular disorders, such as epiretinal membrane, age-related macular degeneration, diabetic retinopathy, retinal vein occlusion, and macular hole.[12–15] Given that macular disorders can cause a structural distortion in retinal layers, this finding is predictable. Interestingly, in our clinical practice, we have found that even in eyes without macular disorders, segmentation errors in macular ganglion cell inner plexiform layer (GCIPL) thickness measurement were found. Thus, we focused on

http://dx.doi.org/10.1016/j.ophtha.2015.12.032
ISSN 0161-6420/16

the prevalence, features, and associated factors of segmentation errors in macular GCIPL thickness measurement in eyes without macular disorders. Furthermore, to investigate whether the development of macular GCIPL segmentation error is reproducible, repeated OCT examinations were performed on eyes with macular GCIPL segmentation errors.

Methods

Participants

The study protocol was approved by the Institutional Review Board at Kim's Eye Hospital, Seoul, Korea. All procedures conformed to the guidelines of the Declaration of Helsinki. Participants who visited a glaucoma specialist (Y.H.H.) between March 2015 and April 2015 were recruited consecutively at the glaucoma clinic of Kim's Eye Hospital.

Each subject underwent a complete ophthalmic examination that included visual acuity and refractive error assessments using a model TX-20P autorefractor keratometer (Canon, Tokyo, Japan), intraocular pressure measurements using a Goldmann applanation tonometer, anterior segment examination using slit-lamp biomicroscopy, ONH evaluation and fundus examination using a 90-diopter (D) lens, the 24-2 Swedish interactive threshold algorithm standard automated visual field test using a Humphrey Visual Field Analyzer (Carl Zeiss Meditec, Dublin, CA), red-free fundus photography using a Kowa Nonmyd7 fundus camera (Kowa, Tokyo, Japan), and circumpapillary RNFL and macular GCIPL thickness measurements using the Cirrus high-definition spectral-domain OCT device (Cirrus HD-OCT; Carl Zeiss Meditec).[7] A glaucomatous visual field defect was defined as (1) a cluster of 3 points with probabilities of less than 5% on the pattern deviation map in at least 1 hemifield, including at least 1 point with a probability of less than 1% or a cluster of 2 points with a probability of less than 1%; (2) glaucomatous hemifield test results outside of normal limits; or (3) a pattern standard deviation beyond 95% of normal limits as confirmed by at least 2 reliable examinations (false-positive or false-negative results, <15%; fixation losses, <15%).[7]

The inclusion criteria for healthy eyes were best-corrected visual acuity of 20/30 or better, normal anterior segment results on slit-lamp examination, no RNFL defects on red-free fundus photographs, no visual field defects, and an intraocular pressure of 21 mmHg or less. Inclusion criteria for glaucomatous eyes were best-corrected visual acuity of 20/30 or better, a normal anterior segment on slit-lamp examination, ONH with glaucomatous changes (i.e., an increased cup-to-disc ratio and neuroretinal rim narrowing), and an RNFL defect on red-free fundus photography.[7] Given that early-stage glaucomatous visual field defects may not be evident in standard automated perimetry, in the current study, the presence of glaucoma was defined based on ONH and RNFL results; visual field test results were not considered in our definition of glaucoma. Eyes with concurrent macular disorders, including a vascular disorder, diabetic retinopathy, epiretinal membrane, age-related macular degeneration, macular hole or edema, retinoschisis, retinal detachment, or myopic maculopathy (atrophic change, lacquer cracks, or choroidal neovascularization),[16] were excluded.

Optical Coherence Tomography Measurement

A macular cube scan was obtained using the Cirrus HD-OCT as described previously.[7] The subjects were seated and properly positioned, and scanning laser images were focused for image acquisition. As soon as the macula was centered on the live scanning laser image, a 6×6-mm square of data was captured with the use of a retinal tracking system. The pupils were not dilated during the OCT examinations. The RGC analysis algorithm of the Cirrus HD-OCT identifies the outer boundary of the macular RNFL and the outer boundary of the inner plexiform layer (IPL). The difference between the RNFL and the IPL outer boundary segmentation yields the macular GCIPL thickness. The sectoral (superotemporal, superior, superonasal, inferonasal, inferior, and inferotemporal) macular GCIPL thicknesses are measured in an elliptical annulus with a vertical outer radius of 2.0 mm and a horizontal radius of 2.4 mm (Figs 1–4).[7] The macular cube scan of the Cirrus HD-OCT includes 128 horizontal scan lines (B-scans), each consisting of 512 A-scans per line. To investigate the segmentation errors in the macular GCIPL thickness measurement, 128 cross-sectional OCT B-scan images per eye were inspected. In B-scan images, the outer border of the RNFL is presented as a solid purple line and the outer border of the IPL is presented as a solid yellow line (Figs 1–4). The presence of a macular GCIPL segmentation error was defined as when these 2 lines were not located in the proper positions between the retinal layers in at least 1 cross-sectional image.

A segmentation error was classified by its layer, location, and size. When the outer border of the RNFL was not located between the RNFL and the RGC layer, it was defined as an anterior border segmentation error (Fig 2); when the outer border of the IPL was not located between the IPL and the inner nuclear layer, it was defined as a posterior border segmentation error (Fig 3). The location of the segmentation error was classified into superior, nasal, inferior, and temporal quadrants centered to the fovea. Furthermore, the segmentation error was classified as diffuse or localized. We arbitrarily defined a localized segmentation error when the longest length of an area of segmentation error was less than 1.0 mm (half of the vertical outer radius of the macular GCIPL scan; Fig 2).

To identify whether segmentation error development was consistent in subsequent examinations, repeated OCT examinations were performed within 1 month in eyes with GCIPL segmentation errors. Based on the reproducibility of the segmentation error, segmentation errors were classified into reproducible (presence of segmentation error in the same layer, location, and area), partially reproducible (changes in layer, location, or area of segmentation error), and nonreproducible (disappearance of segmentation error in the repeated examination). The OCT machine was calibrated regularly by technicians from the manufacturer. According to the manufacturer's guidelines, only images with signal strengths of 6 or more (range, 0–10) were included.

The OCT images were assessed by 2 investigators (Y.H.H. and M.K.K.), each of whom was blinded to the other's judgment. Any disagreements between the 2 investigators were resolved by a third adjudicator (D.W.K.). The same investigators were involved in reviewing repeated OCT scans. When evaluating the OCT images, only macular cross-sectional images were assessed. Other factors, including age, refractive error, ONH and RNFL results, and visual field test results, were masked to the investigators.

Statistical Analyses

Eyes were classified into 2 groups based on the presence of GCIPL segmentation error. To identify associated factors for the development and reproducibility of segmentation errors, age, refractive error, mean deviation (MD) and pattern standard deviation of visual field test, circumpapillary RNFL thickness measured by OCT, and the signal strength of the OCT scan were compared between the 2 groups using the independent t test. In addition, univariate and multivariate logistic regression analyses were performed. The dependent variable was the presence and reproducibility of macular