Analysis of Peripapillary Choroidal Thickness in Unilateral Amblyopia

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Abstract

Purpose: To evaluate the peripapillary choroidal thickness (CT) in children with unilateral amblyopia using spectral-domain optical coherence tomography (SD-OCT).

Methods: One hundred and six eyes of 53 children with unilateral amblyopia and 20 eyes of 20 children with normal vision were involved in this study. Of the 53 children with unilateral amblyopia, 29 (54.7%) had hyperopic anisometropic amblyopia and 24 (45.3%) had strabismic amblyopia. Peripapillary CT was measured from 6 mm length radial B-scans at the optic nerve head using the enhanced depth imaging program of an SD-OCT (Heidelberg Engineering, Germany). Age, sex, refractive error, and best-corrected visual acuity were also recorded.

Results: The average peripapillary CT was greater in amblyopic eyes than in the fellow eyes of the children with amblyopia (P = 0.002), and control eyes (P < 0.001). There was no significant difference between the fellow eyes of children with amblyopia and the control eyes (P = 0.158). The average peripapillary CT was negatively correlated with axial length (AL) in amblyopic eyes (r = -0.381; P = 0.005) and fellow eyes (r = -0.392; P = 0.004) but not in control eyes (r = -0.232; P = 0.325). After adjustment for the possible effects of AL, the average peripapillary CT in amblyopic eyes was still greater than in fellow eyes (P = 0.014) and control eyes (P = 0.022).

Conclusion: The peripapillary choroid of eyes with amblyopia was thicker than that of the fellow eyes and control eyes. No significant difference was observed between fellow eyes and control eyes.

Keywords: Amblyopia; Anisometropia; Optical Coherence Tomography; Peripapillary Choroidal Thickness; Strabismus

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INTRODUCTION

During the critical visual development period in childhood, reduced best-corrected visual acuity (BCVA) is called as amblyopia, which is the most common cause...
of monocular visual loss in children. Amblyopia can be bilateral, but it most commonly affects one eye of children with anisometropia, strabismus or visual deprivation (e.g., congenital cataract, ptosis). A large number of studies have been recently performed to assess the structural variations in retinal nerve fiber layer (RNFL) in eyes with amblyopia. Contrary to the general belief that the amblyopic eyes are structurally normal, significant alterations have been found in the RNFL and foveal thickness in patients with amblyopia. However, a consensus on retinal abnormalities has not been reached.

Another region under investigation in amblyopic eyes is the choroid. Spaide et al defined enhanced depth imaging (EDI) method using conventional spectral-domain optical coherence tomography (SD-OCT). This new procedure enables measurement of the choroidal thickness (CT) at different locations within the macular and peripapillary regions. Recent studies have reported an increase in macular CT in amblyopic eyes. However, peripapillary CT has not been well investigated in children with amblyopia. The aim of this study was to measure the peripapillary CT in children with unilateral amblyopia and to compare the results among amblyopic and fellow eyes of children with unilateral amblyopia, as well as the eyes of healthy children.

METHODS

This single-center, cross-sectional comparative study was approved by the institutional medical ethics committee and followed the tenets of the Declaration of Helsinki for research involving human subjects. All participating children and their parents received information about the study and signed an informed consent document.

The inclusion criteria were as follows: a diagnosis of unilateral strabismic or hyperopic anisometropic amblyopia, a BCVA of 20/32 (0.22 LogMAR) or worse in the amblyopic eye or at least 0.2 LogMAR values difference between the two eyes, and a BCVA of 20/25 (0.1 LogMAR) or better in the fellow eye. Anisometropia was defined as 1.5 diopters (D) or more difference in cycloplegic spherical equivalent (SE) refraction between the two eyes. Strabismus was defined as any manifest heterotropia. SE refraction was calculated as the sum of the spherical plus one-half of the cylindrical error. Patients with any other type of amblyopia, a history of previous ocular surgery or trauma, history of any systemic disease with ocular findings or any ocular disease were excluded from the study. Subjects without sufficient cooperation for visual acuity assessments or OCT examinations were also excluded.

Twenty healthy children less than 13 years old, with an uncorrected visual acuity equal to or better than 20/20 (0.0 LogMAR) in each eye, were enrolled as control subjects. One eye was randomly chosen for analysis in the control group.

All participants underwent full ophthalmic evaluation, including cycloplegic refraction, visual acuity testing, cover and cover-uncover test, slit-lamp biomicroscopy, dilated fundus examination, and axial length (AL) measurement (IOLMaster, Carl Zeiss Meditec, Jena, Germany). A standard Snellen chart was used to measure the visual acuity, and the results were converted to LogMAR units for further statistical analyses.

Peripapillary CT measurements were obtained using the EDI system of an SD-OCT (Spectralis OCT; Heidelberg Engineering, Heidelberg, Germany). All images were captured by a single experienced technician who was masked to the identity of the participants and was not involved in the data analysis. All images were acquired with 6 radial B-scans centered on the optic nerve head (ONH) [Figure 1]. Using the electronic caliper within the OCT, the distance between the outer portion of the hyperreflective line corresponding to the Bruch’s membrane beneath the retinal pigment epithelium (RPE) and the inner portion of the sclerochoroidal interface was measured. The method used to quantify the peripapillary CT has been previously described in detail by Park et al. Two examiners (G.B. and E.M.) who were masked as to whether the eye was amblyopic, independently measured the choroidal thicknesses. The measurements of one examiner were used for overall statistical analyses and the measurements of the second examiner were used for calculating the inter-observer agreement. The measurements were taken at 0.3-mm intervals from the ending point of the Bruch’s membrane of the ONH at 6 points in each sector [Figure 2]. The measured CTs from 6 B-scans were divided into 12 sectors and the averaged values of every 3 sectors in each quadrant were obtained at the optic nerve head. The averaged choroidal thickness values of three sectors in each quadrants were defined as the superior, inferior, nasal, and temporal thicknesses.
defined as the superior, inferior, nasal, and temporal thicknesses. The average peripapillary CT was calculated by averaging the thicknesses at these 4 quadrants. Thus, to obtain the average peripapillary CT of one eye, CT was measured at 72 different points. To reduce the potential confounding influence of diurnal variations in CT, all EDI-OCT measurements were performed between 9 and 12 AM.

Statistical analysis was performed using SPSS ver. 17.0 (Chicago, IL, USA) software. Basic descriptive statistics were calculated on all the gathered data and are reported as the mean ± SD. The Pearson χ² test was used to compare the categorical parameters. Normal distribution of continuous variables was confirmed with the Kolmogorov-Smirnov test. Analysis of variance (One-Way ANOVA) test followed by Tukey multiple comparison test was used to compare the SE, AL, and peripapillary CT measurements among the amblyopic, fellow, and control eyes. Multivariate regression analysis was performed to adjust for the possible effects of AL on the peripapillary CT. Pearson correlation coefficient was used to determine the correlation between peripapillary CT and other continuous variables. Inter-observer agreement for peripapillary CT measurements was calculated using the intraclass correlation coefficient (ICC). Agreement was defined according to the guidelines proposed by Landis and Koch[17] as follows: 0, chance agreement; 0.01‑0.19, poor agreement; 0.20‑0.39, fair agreement; 0.40‑0.59, moderate agreement; 0.60‑0.79, substantial agreement; and 0.80‑1.00, almost perfect agreement. For all evaluations, a P value of less than 0.05 was considered statistically significant.

RESULTS
Fifty-seven children with unilateral amblyopia and 20 age-matched healthy controls were enrolled in the study. Due to poor image quality, 4 subjects with amblyopia were excluded. Among 53 children with unilateral amblyopia, 29 (54.7%) had hyperopic anisometropic amblyopia and 24 (45.3%) had strabismic amblyopia. Of the subjects with strabismic amblyopia, 21 (87.5%) had esotropia and 3 (12.5%) had exotropia. Slit-lamp biomicroscopic evaluation of the anterior segment and dilated fundus examination were normal in all subjects. Clinical and demographic characteristics of the participants are given in Table 1. The mean age of the subjects with amblyopia was 8.94 ± 1.98 years (range 6-12 years) and the mean age in the control group was 8.50 ± 2.37 years (range 5-12 years). No statistically significant differences were observed between amblyopia and control groups with respect to age (P = 0.423) and gender (P = 0.798).

BCVA ranged between 20/32 and 20/400 in the amblyopic eyes and was significantly lower compared with the fellow eyes and control eyes (P < 0.001 for both). SE of the amblyopic eyes (+4.36 ± 2.10) was significantly higher when compared with fellow eyes (+2.57 ± 1.83)

Table 1. Demographic and clinical characteristics of the participants. Note that anisometropic group and strabismic group are further breakdown of patients with unilateral amblyopia.

| Patients with unilateral amblyopia (n=53) | Anisometropic group (n=29) | Strabismic group (n=24) | Control group (n=20) |
|----------------------------------------|---------------------------|------------------------|----------------------|
| Age (year)                             | 8.94±1.98                 | 8.83±2.05              | 9.08±2.05            | 8.50±2.37 |
| Sex (M, F)                             | 27, 26                    | 13, 16                 | 14, 10               | 11, 9     |
| Spherical error, D                     |                           |                        |                      |
| AE                                     | +4.36±2.10                | +5.38±1.69             | +3.11±1.88           | +1.52±0.58 |
| FE                                     | +2.57±1.83                | +2.71±1.95             | +2.39±1.69           |           |
| BCVA, logMAR                            |                           |                        |                      |
| AE                                     | 0.38±0.25                 | 0.44±0.29              | 0.31±0.15            | 0.02±0.04 |
| FE                                     | 0.01±0.05                 | 0.02±0.05              | -0.01±0.05           |           |
| Axial length, mm                       |                           |                        |                      |
| AE                                     | 21.65±1.09                | 21.35±0.99             | 22.01±1.12           | 23.01±0.75 |
| FE                                     | 22.17±1.11                | 22.25±1.12             | 22.07±1.11           |           |

Data present the mean±SD. AE, amblyopic eye; FE, fellow eye; BCVA, best corrected visual acuity; M, male; F, female
and control eyes (+1.52 ± 0.58) (P < 0.001 for both). AL was significantly lower in amblyopic eyes (21.65 ± 1.09) than in fellow eyes (22.17 ± 1.11) and control eyes (23.01 ± 0.75) (P = 0.031 and P < 0.001, respectively). The differences in BCVA and SE between amblyopic eyes and fellow eyes were statistically significant in both the anisometropic and strabismic groups (P < 0.001 for all). AL of the amblyopic eyes was significantly lower compared with fellow eyes in the anisometropic group (P < 0.001), but did not differ significantly in the strabismic group (P = 0.222). Control eyes had significantly higher AL compared with the fellow eyes of the patients with unilateral amblyopia (P = 0.009). No significant differences were observed between control eyes and fellow eyes in terms of BCVA and SE (P = 0.782 and P = 0.077, respectively).

The peripapillary CT measurements showed excellent inter-observer agreement, with an ICC of 0.971. The average peripapillary CT was 191.42 ± 47.41 µm in amblyopic eyes and 162.37 ± 42.43 µm in fellow eyes of patients with unilateral amblyopia (P = 0.002). Peripapillary CT in the temporal, nasal, superior, and inferior quadrants were also significantly higher in amblyopic eyes than in fellow eyes. After controlling for the possible effects of AL, the differences in all quadrants, except the nasal peripapillary CT remained significant [Table 2]. In the anisometropic subgroup, peripapillary CT in all quadrants were significantly higher in amblyopic eyes than in fellow eyes (P < 0.001 for all). When controlling for AL, the difference in superior peripapillary CT was no longer significant but the difference in peripapillary CT in other quadrants were statistically significant [Table 3]. In the strabismic subgroup, there was no significant difference in nasal peripapillary CT between amblyopic eyes and fellow eyes (P = 0.928), and the difference in superior and inferior peripapillary CT only slightly reached the threshold of significance (P = 0.048 and P = 0.049, respectively). After adjustment for AL, no statistically significant difference was observed in any of the quadrants [Table 4]. When comparing the anisometropic and strabismic amblyopia subgroups, the average peripapillary CT did not show any significant difference in the amblyopic eyes (P = 0.092) and fellow eyes (P = 0.226) of the two groups.

The average peripapillary CT was 141.60 ± 27.59 µm in control eyes. Amblyopic eyes had significantly higher CT in all quadrants compared with control eyes. After adjusting for the effects of AL, the differences in average, temporal and inferior peripapillary CT remained significant between amblyopic eyes and control eyes. There was no statistically significant difference between fellow eyes and control eyes in any of the quadrants [Table 2].

There was a significant negative correlation between the average peripapillary CT and AL in both amblyopic eyes (r = −0.381; P = 0.005) and fellow eyes (r = −0.392; P = 0.004), but not in control eyes (r = −0.232; P = 0.325). There was no significant correlation between the average peripapillary CT and age in any of the groups (P > 0.05 for all).

### DISCUSSION

In the present study, we used the EDI program of an SD-OCT to evaluate the peripapillary CT in patients with amblyopia and healthy control subjects. The average peripapillary CT was greater in amblyopic eyes than in fellow eyes and control eyes. In subgroup assessments, both the strabismic and anisometropic amblyopia subgroups had greater average peripapillary CT in amblyopic eyes compared to the fellow eyes. When possible effects of AL on peripapillary CT were controlled using multivariate regression analysis, the average peripapillary CT was still greater in amblyopic eyes than in fellow eyes in the anisometropic amblyopia subgroup but not in the strabismic amblyopia subgroup.

Visual signals on the retina provide feedback to regulate ocular emmetropization after birth. Morphological aberration and behaviors that result in inadequate use of one eye in early childhood might cause asymmetric visual feedback which promotes amblyopia formation. Different levels of the visual pathway may be affected during the amblyopic process.

### Table 2. Peripapillary choroidal thickness measurements of children with unilateral amblyopia and children with normal vision

| Amblyopic eyes, µm | Fellow eyes, µm | Control eyes, µm | Amblyopic eyes vs. Fellow eyes | Amblyopic eyes vs. Control eyes | Fellow eyes vs. Control eyes |
|-------------------|----------------|-----------------|-------------------------------|-------------------------------|-----------------------------|
| Average           | 191.42±47.41   | 162.37±42.43    | 141.60±27.59                  | 0.002                         | <0.001                      |
| Temporal          | 218.85±56.72   | 179.87±47.59    | 150.03±32.01                  | <0.001                        | <0.001                      |
| Nasal             | 178.42±47.23   | 155.87±43.45    | 143.57±29.99                  | 0.023                         | 0.008                       |
| Superior          | 197.06±54.31   | 169.34±47.58    | 149.01±27.43                  | 0.010                         | 0.001                       |
| Inferior          | 171.34±45.64   | 144.43±39.56    | 123.78±27.69                  | 0.003                         | <0.001                      |

Data present the mean±SD. *One-Way ANOVA test followed by Tukey multiple comparison test. **Adjusted for axial length using multivariate regression analysis.

### Table 3. Peripapillary choroidal thickness measurements of children with unilateral anisometropic amblyopia and children with normal vision

| Amblyopic eyes, µm | Fellow eyes, µm | Control eyes, µm | Amblyopic eyes vs. Fellow eyes | Amblyopic eyes vs. Control eyes | Fellow eyes vs. Control eyes |
|-------------------|----------------|-----------------|-------------------------------|-------------------------------|-----------------------------|
| Average           | 191.42±47.41   | 162.37±42.43    | 141.60±27.59                  | 0.002                         | <0.001                      |
| Temporal          | 218.85±56.72   | 179.87±47.59    | 150.03±32.01                  | <0.001                        | <0.001                      |
| Nasal             | 178.42±47.23   | 155.87±43.45    | 143.57±29.99                  | 0.023                         | 0.008                       |
| Superior          | 197.06±54.31   | 169.34±47.58    | 149.01±27.43                  | 0.010                         | 0.001                       |
| Inferior          | 171.34±45.64   | 144.43±39.56    | 123.78±27.69                  | 0.003                         | <0.001                      |

Data present the mean±SD. *One-Way ANOVA test followed by Tukey multiple comparison test. **Adjusted for axial length using multivariate regression analysis.
Histopathological evaluation of the lateral geniculate nucleus of monkeys with amblyopia has revealed notable shrinkage in the cells receiving input from the amblyopic eye. Similar to their findings, Hess et al. reported that pattern electroretinograms (PERG) were significantly reduced in humans with different types of amblyopia. In contrast to their findings, Hess et al. did not observe a PERG deficit in amblyopic eyes with individually optimized optical focus, fixation alignment and fixation stability.

With the advent of OCT in ophthalmology, a large number of studies have been performed to evaluate the thickness of the RNFL, foveal volume and macular thickness in eyes with amblyopia, but the relationship between choroidal structure and amblyopia has not yet been adequately studied. Current choroidal investigations have concentrated upon macular CT instead of peripapillary CT in amblyopic eyes. The literature lacks evidence regarding the evaluation of peripapillary CT in amblyopic children.

The choroid is a highly vascular tissue which provides blood supply to the outer retina, and also participates in various physiological mechanisms including ocular temperature and intraocular pressure regulation, and the absorption of stray light. Even though the regulatory function of the choroid in ocular physiology is well understood, the clinical aspects of the association between CT and amblyopia remains unclear. It has been recently reported that subfoveal CT was increased in amblyopic eyes compared with fellow eyes and control eyes in children with hyperopic anisometropic amblyopia. Although we focused on peripapillary CT instead of macular CT, the results of our study are consistent with these reports. Two other studies including children with either anisometropic or strabismic amblyopia, have also reported that subfoveal CT was thicker in amblyopic eyes. In our study, after controlling for the effect of AL on peripapillary CT, patients with anisometropic amblyopia showed increased CT measurements in the amblyopic eyes compared with the fellow eyes but there were no significant differences in the strabismic amblyopia subgroup. Previous studies have reported that there was a negative correlation between the thickness of choroid and the axial length in non-amblyopic healthy subjects. Now we have shown the persistence of the difference in peripapillary CT despite correction for the effects of AL, suggesting that there would be a separate operational mechanism for amblyopia itself. On the other hand, the reason for the dissimilarity between anisometropic and strabismic amblyopic patients is unclear. Yen et al have reported an increase in RNFL thickness in the amblyopic eyes of patients with anisometropic amblyopia while no difference was observed in patients with strabismic amblyopia. They attributed this finding to the difference in neural mechanisms associated with different amblyopia subtypes. Further histopathological studies might be useful to enlighten the reason of this dissimilarity in anisometropic and strabismic amblyopia.

Xu et al. reported that both amblyopic and fellow eyes had thicker subfoveal CT compared to control eyes. However, other investigators found no significant difference between fellow eyes of subjects with amblyopia and control eyes. Nishi et al. hypothesized that CT reduces in response to hyperopic defocus in fellow eyes and control eyes; however, in amblyopic eyes, this choroidal compensation is not observed. Our results also indicate that fellow eyes of children with amblyopia and control eyes were similar in terms of peripapillary CT measurements.

This study has some potential limitations. Automatic segmentation of the choroid is not currently supported by the software of the Heidelberg Spectralis OCT, and manual segmentation might have produced inaccuracy in the measurement. However, the excellent level of agreement seen between the observers may confirm the accuracy of the study. A further limitation is the relatively small number of subjects enrolled. Therefore, future work including a larger number of subjects will be necessary to verify our findings.
In conclusion, this study presents new data regarding the thickness of the peripapillary choroid in children with unilateral amblyopia. The current knowledge in the literature reveals that macular choroid is thicker in amblyopic eyes. Based on the results of our study, the choroid in the peripapillary region also appears to be effected in anisometropic amblyopic eyes.

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Conflicts of Interest
There are no conflicts of interest.

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