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Influence of scanning density on macular choroidal volume measurement using spectral-domain optical coherence tomography

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Abstract

BACKGROUND—To evaluate the impact of scanning density on macular choroidal volume measurement using spectral-domain optical coherence tomography (SD-OCT).

METHODS—Thirty eyes of normal subjects underwent consecutive raster choroidal scanning protocols using SD-OCT in enhanced-depth imaging mode. Manual choroidal segmentation was performed using the built-in automated retinal segmentation software to obtain five analyses with different inter-scan distances including inter-scan distances of 30 microns, 60 microns, 120 microns, 240 microns, and 480 microns. The built-in software of the device automatically generated the choroidal thickness and volume map in the similar manner as for retinal volume map, using the standardized Early Treatment Diabetic Retinopathy Study (ETDRS) grid. For each raster scan, mean absolute difference and relative difference of mean foveal choroidal thickness (FCT), foveal choroidal volume (FCV) and total macular choroidal volume (TCV) in comparison to “true value” (i.e., 30-micron inter-scan distance) were calculated.

RESULTS—The maximum relative differences were 10% and 16% for TCV and FCV, respectively. For mean FCT, the maximum absolute difference was 31 microns, and maximum relative difference was 12.7%. No statistically significant differences were found in measurements of mean foveal choroidal thickness ($p=0.912$) and volume ($p=0.944$), as well as macular choroidal volume ($p=0.912$), with varying inter-scan distance.

CONCLUSIONS—Our study shows that approximately 16 scans over the macula with a inter-scans distance of 480 microns is sufficient to provide clinically relevant and reliable choroidal thickness/volume map. This information could be useful in design of choroidal scanning protocols for future clinical trials.

Keywords

Scanning density; Choroidal volume; Choroidal thickness; raster protocol

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Introduction

The enhanced depth imaging (EDI) system on spectral-domain optical coherence tomography (SD-OCT) has improved the visualization of the structure of the choroid.^[1] Recent studies have shown changes in choroidal thickness in various chorioretinal disorders such as decrease in age-related macular degeneration (AMD),^[2, 3] and high myopia^[4]; increase in central serous chorioretinopathy,^[5] Vogt-Koyanagi-Harada syndrome,^[6] and other uveitic syndrome.^[7] Quantitative analysis of choroidal volume/thickness would be important for evaluation and understanding the pathophysiology of these diseases. Recently, measurement of choroidal thickness and volume by manual segmentation has been reported using swept-source OCT and high-resolution OCT devices, however, these methods are not yet approved by the Food and Drug Administration.^[8, 9] We reported a novel technique of manual choroidal segmentation to measure the choroidal thickness and volume using a commercial Spectralis HRA2 (Heidelberg Engineering, Heidelberg, Vista, CA) in enhance depth imaging (EDI) mode.^[10] The built-in retinal software of the device automatically calculated choroidal volume map of 6 mm macula scans. The built-in EDI software of the Spectralis HRA2 reverts the inverted image automatically.

Achieving reliable choroidal thickness and volume measurements depends on the number of scans and interpolation between the scans. Obviously, denser scans (i.e., less distance between the scans) provide the more reliable measurements. Sparse scans (i.e., larger distance between the scans) may provide false results or artifactually extensive interpolation of data between the scans. Performing a raster scan requires longer test duration, thus eye movements may thus also affect the measurements. Eye-tracking during acquisition of volume scans is one of the key advantages of the Spectralis HRA2, and although it requires longer exam time, eye-tracking assures foveal centration.^[11] At present, no automated software is available for choroidal segmentation, and it is laborious to perform manual choroidal segmentation.

With the increasing importance of choroidal analysis in the evaluation of various chorioretinal pathologies, it is critical to understand the adequate number of B-scans to obtain reliable and consistent measurements of the posterior choroidal volume with good compliance of the patient. If reliable choroidal thickness and volume measurements were possible with fewer scans, the time taken for performing a raster scan and the effect of eye movements would be reduced, resulting in an easier manual segmentation process. In the present study, we evaluate the impact of reducing the density of B-scan choroidal volume maps in normal eyes using an SD-OCT device in EDI mode.

Methods

For this prospective observational study, 30 eyes of 15 consecutive healthy volunteers were included. Written informed consent for diagnostic procedure was obtained from each subject before examination. The study was approved by the Institutional Review Board at Shiley Eye Center at the University of California, San Diego (UCSD) and was conducted in adherence with the tenets of the Declaration of Helsinki. Time period for this study was from August 2011 to October 2011. A single trained retina specialist (JC) performed SD-OCT raster scans in EDI mode without pupillary dilation using a Spectralis HRA2 device (Heidelberg Engineering, Heidelberg, Vista, CA). Raster scans with any single poor choroidal scan were excluded.

Choroidal volume imaging protocol

Three consecutive raster choroidal scanning protocols were performed on each eye and were acquired in a continuous, automated sequence, covering a $30^\circ \times 25^\circ$ area centered on the

fovea. Inter-scan distances in the three protocols were 30 microns, 120 microns, and 240 microns, respectively. The scans obtained after 25 frames were averaged using built-in automatic averaging (TruTrack) to obtain a good-quality choroidal image.

Choroidal image analysis

Two experienced retina specialists (GB & JC) analyzed all scans to assure the foveal centration. Choroidal segmentation was performed manually after disabling the automated retinal layer segmentation software. Reference lines of the built-in automated retinal segmentation were moved from the retinal boundaries to the choroidal boundaries. The internal limiting membrane (ILM) line was moved to the outer part of the hyper-reflective line corresponding to the base of the RPE. The basement membrane line, reference line for the posterior edge of the retina, was moved to the posterior edge of the choroid, as demarcated by hyper-reflective margin line corresponding to the choriocleral interface (Figure 1). The built-in software of the device automatically generated the choroidal thickness and volume map in the similar manner as for retinal volume map, using the standardized Early Treatment Diabetic Retinopathy Study (ETDRS) grid. The ETDRS grid was automatically positioned in the macula by the software of the device and was visualized by the software designed to map macular thickness (Figure 1). The grid divides the macula into three rings with a diameter of 1 mm (fovea), 3 mm (inner) and 6 mm (outer). Intra and inter-observer concordance correlation was calculated for manual choroidal segmentation. The two observers checked the automatic foveal centration of the grid and, if necessary, moved the grid to assure exact foveal centering.

Scanning density modulation

After collecting results of the choroidal maps of the three scanning protocols, every other line was disabled by the two observers in the first protocol (30-microns inter-scan distance) and the third protocol (240-microns inter-scan distance). As a result, five analyses with different inter-scan distances were obtained, including inter-scan distances of 30 microns (97 scans), 60 microns (49 scans), 120 microns (37 scans), 240 microns (31 scans), and 480 microns (16 scans). Values of the average choroidal volume and choroidal thickness for the fovea, as well as total choroidal volume of macular grid, were recorded for all the analyses.

Statistical analysis

Choroidal thickness and volume measurements computed using the closest raster scanning protocol (i.e., 30-micron inter-scan distance) were considered the “true value” to which measurements from all other sampling densities were compared. For each raster scan, mean absolute difference and relative difference of mean foveal choroidal thickness, foveal choroidal volume and total macular choroidal volume were calculated. Absolute values instead of non-absolute values were used to avoid masking of differences. Relative (percentage) difference was calculated by dividing the value of the difference between the two measurements by the mean of the two and multiplying by 100. Generalized Estimating Equations (GEE) analysis was used to assess differences in the results of the measurement between each protocol and the “true value” protocol (30-micron inter-scan distance). Bland-Altman plots were used to assess comparisons between values for each inter-scan distance group and “true value” measurements.^[12] All statistical analysis was performed using SAS statistical software.

Results

This prospective study included 30 eyes of 15 subjects. The mean age of the 15 subjects was 35.1 years (range, 22-51 years) with 7 men and 8 women. The racial distribution included

Caucasians (4 subjects), Asians (8 subjects) and Hispanics (3 subjects). The mean spherical equivalent was -1.86 D (Standard deviation = 0.7D).

In our previous study the inter-observer reproducibility and intra-observer repeatability for manual choroidal segmentation in eyes with or without chorioretinal diseases was very high with concordance correlation coefficient (CCC) 0.9956 and 0.98 to 0.99 respectively. The inter-observer coefficient of reproducibility was approximately 0.42 (95% CI, 0.34–0.5 mm³) for the average CV.[10] Table 1-3 shows absolute and relative differences for all inter-scan distance protocols: no significant differences between the five protocols were noted regarding mean foveal choroidal thickness ($p=0.912$), foveal choroidal volume ($p=0.944$), and total choroidal volume of the 3mm-diameter macula ($p=0.912$). Bland-Altman plots of the mean difference between the “true-value” and each sampling inter-scan distance for mean foveal choroidal thickness, foveal subfield choroidal volume, and total choroidal volume measurements are shown in Figures 2-5. The effect of scanning density on measurements of total choroidal volume, foveal choroidal volume and mean foveal choroidal thickness are shown in Figure 6.

Discussion

Imaging of the choroid has significantly improved since the advent of the enhanced depth imaging on SD-OCT devices. Recent literature shows choroidal volume measurements using swept-source and high-resolution OCT devices.^[8, 9] Previously we showed that manual choroidal segmentation and choroidal volume measurement resulted in highly reproducible measurements.[10] The reproducibility would not change with the number of scans because the reproducibility assesses only the accuracy of manual segmentation. Reducing the number of scans would assess the acceptable interpolation and reliability of choroidal volume measurements. However, adequate number of B-scans in the choroidal raster scanning protocol to obtain reliable and clinically relevant choroidal volume measurement remains unknown.

Denser scans are obviously closer to the true measurements; however, longer scan duration and strenuous manual choroidal segmentation are the drawbacks. The need for fewer scans to obtain a clinically reliable measurement is helpful for the operator as well as for compliance of the patient. Patients with poor fixation can be quickly scanned to achieve the reliable volume measurements. Devices with eye tracking software also prolong the image acquisition time, and thus a smaller number of scans would be preferable. Choroidal imaging quality can be improved by averaging for better visualization of fine structures; however, the scanning period increases markedly for a raster scan.^[13] Fewer scans would allow for increasing averaging and further improving choroidal visualization.

In the present study, we found that there was a gradual increase in relative percentage differences with increasing distance between the scans (Tables 1-3). However, the maximum relative differences were 10% and 16% for total choroidal volume and foveal choroidal volume, respectively. For mean foveal choroidal thickness, the maximum absolute difference was 31 microns, and maximum relative difference was 12.7%. No statistically significant differences were found in measurements of mean foveal choroidal thickness ($p=0.912$) and volume ($p=0.944$), as well as macular choroidal volume ($p=0.912$), with varying inter-scan distance. An inter-scan distance of 480 microns is sufficient to obtain a reliable good-quality macular choroidal volume, allowing for good compliance of the patient and a short scanning duration.

Our study provides important information for future trials in relation to choroidal diseases. At present, no automated software is available for choroidal segmentation. We assume our

results would be applicable to automated segmentation software also, after assuring the accurate choroidal segmentation.

We acknowledge a number of limitations of our study. Our study results may not be applicable to eyes with localized choroidal involvement, such as macular degeneration or choroidal nevi where choroidal assessment may need denser scans. Only one SD-OCT device was used in our study. We expect that our results would be applicable to other SD-OCT devices also, however, additional studies are required to confirm this. In our study, it was necessary to perform three scanning protocols, as the built-in software of the Spectralis HRA2 does not provide choroidal volume measurements when disabling two consecutive scans. As a result, three different raster scans were used to obtain five groups of different inter-scan distances; thus, this may have influenced the measurements. However, two retina specialists checked the foveal centration and performed the manual choroidal segmentation in all the scans. Our study results are limited to quantitative assessment of the choroid. For qualitative assessment, denser scans may be required.

In conclusion, we report a critical aspect about adequate number of scans for clinically relevant and practically feasible choroid raster scanning protocol. We report that approximately 16 scans over the macula with a inter-scans distance of 480 microns is sufficient to provide clinically relevant and reliable choroidal thickness/volume map. Our findings may be helpful for establishing a choroidal volume protocol for future studies.

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REFERENCES

1. Spaide RF, Koizumi H, Pozzoni MC. Enhanced depth imaging spectral-domain optical coherence tomography. *Am J Ophthalmol.* 2008; 146:496–500. [PubMed: 18639219]
2. Chung SE, Kang SW, Lee JH, Kim YT. Choroidal thickness in polypoidal choroidal vasculopathy and exudative age-related macular degeneration. *Ophthalmology.* 2011; 118:840–845. [PubMed: 21211846]
3. Koizumi H, Yamagishi T, Yamazaki T, Kawasaki R, Kinoshita S. Subfoveal choroidal thickness in typical age-related macular degeneration and polypoidal choroidal vasculopathy. *Graefes Arch Clin Exp Ophthalmol.* 2011; 249:1123–1128. [PubMed: 21274555]
4. Fujiwara T, Imamura Y, Margolis R, Slakter JS, Spaide RF. Enhanced depth imaging optical coherence tomography of the choroid in highly myopic eyes. *Am J Ophthalmol.* 2009; 148:445–450. [PubMed: 19541286]
5. Imamura Y, Fujiwara T, Margolis R, Spaide RF. Enhanced depth imaging optical coherence tomography of the choroid in central serous chorioretinopathy. *Retina.* 2009; 29:1469–1473. [PubMed: 19898183]
6. Fong AH, Li KK, Wong D. Choroidal evaluation using enhanced depth imaging spectral-domain optical coherence tomography in Vogt-Koyanagi-Harada disease. *Retina.* 2011; 31:502–509. [PubMed: 21336069]
7. Aoyagi R, Hayashi T, Masai A, Mitooka K, Gekka T, Kozaki K, Tsuneoka H. Subfoveal choroidal thickness in multiple evanescent white dot syndrome. *Clin Exp Optom.* 2011
8. Hirata M, Tsujikawa A, Matsumoto A, Hangai M, Ooto S, Yamashiro K, Akiba M, Yoshimura N. Macular choroidal thickness and volume in normal subjects measured by swept-source optical coherence tomography. *Invest Ophthalmol Vis Sci.* 2011; 52:4971–4978. [PubMed: 21622704]

9. Ouyang Y, Heussen FM, Mokwa N, Walsh AC, Durbin MK, Keane PA, Sanchez PJ, Ruiz-Garcia H, Sadda SR. Spatial distribution of posterior pole choroidal thickness by spectral domain optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2011; 52:7019–7026. [PubMed: 21810980]
10. Chhablani J, Barteselli G, Wang H, El-Emam S, Kozak I, Doede AL, Bartsch DU, Cheng L, Freeman WR. Repeatability and reproducibility of manual choroidal volume measurements using enhanced depth imaging optical coherence tomography. *Invest Ophthalmol Vis Sci*. 2012; 53:2274–2280. [PubMed: 22427584]
11. Menke MN, Dabov S, Knecht P, Sturm V. Reproducibility of retinal thickness measurements in healthy subjects using spectralis optical coherence tomography. *Am J Ophthalmol*. 2009; 147:467–472. [PubMed: 19026403]
12. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet*. 1986; 1:307–310. [PubMed: 2868172]
13. Sakamoto A, Hangai M, Yoshimura N. Spectral-domain optical coherence tomography with multiple B-scan averaging for enhanced imaging of retinal diseases. *Ophthalmology*. 2008; 115:1071–1078. e1077. [PubMed: 18061270]

Figure 1.

Spectral domain optical coherence tomography scan using enhanced depth imaging showing automated retinal segmentation (left-top), manual choroidal segmentation (left-bottom) and standardized grid (right) including three concentric rings with total nine subfields centered on the fovea. Internal limiting membrane line (blue arrows) and basement membrane line (orange arrows) on automated retinal segmentation was moved to base of retinal pigment epithelium (blue arrows) and choriocleral interface (orange arrows) to demarcate choroidal boundaries (left-bottom).

Figure 2.

Bland-Altman plots demonstrating the 95% level of agreement between the “True value” (inter-scan distance of 30 microns) and each sampling inter-scan distance for total choroidal volume. TCV (30)= total choroidal volume with inter-scan distance of 30 microns, TCV (60)= total choroidal volume with inter-scan distance of 60 microns, TCV (120)= total choroidal volume with inter-scan distance of 120 microns, TCV (240)= total choroidal volume with inter-scan distance of 240 microns, TCV (480)= total choroidal volume with inter-scan distance of 480 microns.

Figure 3.

Bland-Altman plots demonstrating the 95% level of agreement between the “True value” (inter-scan distance of 30 microns) and each sampling inter-scan distance for foveal choroidal volume. FCV (30)= foveal choroidal volume with inter-scan distance of 30 microns, FCV (60)= foveal choroidal volume with inter-scan distance of 60 microns, FCV (120)= foveal choroidal volume with inter-scan distance of 120 microns, FCV (240)= foveal choroidal volume with inter-scan distance of 240 microns, FCV (480)= foveal choroidal volume with inter-scan distance of 480 microns.

Figure 4.

Bland-Altman plots demonstrating the 95% level of agreement between the “True value” (inter-scan distance of 30 microns) and each sampling inter-scan distance for mean foveal choroidal thickness. FCT (30)= mean foveal choroidal thickness with inter-scan distance of 30 microns, FCT (60)= mean foveal choroidal thickness with inter-scan distance of 60 microns, FCT (120)= mean foveal choroidal thickness with inter-scan distance of 120 microns, FCT (240)= mean foveal choroidal thickness with inter-scan distance of 240 microns, FCT (480)= mean foveal choroidal thickness with inter-scan distance of 480 microns.

Figure 5. Effect of reduced B-scan sampling density on measurements of total choroidal volume (TCV), foveal choroidal volume (FCV) and foveal choroidal thickness (FCT): (Left panel) mean absolute error, (Right panel) mean percentage error.

Table 1

Absolute difference (mm^3) and relative difference (%) for total choroidal volume (TCV) for various inter-scan distance groups

Absolute and relative difference of TCV			
Inter-scan Distance (Microns)	Mean	Standard Deviation	Maximum Difference
<i>Absolute difference (mm^3)</i>			
60	0.008	0.012	0.05
120	0.050	0.039	0.12
240	0.069	0.051	0.18
480	0.081	0.052	0.21
<i>Relative difference (%)</i>			
60	0.383	0.580	2.62
120	2.835	2.122	7.10
240	3.922	2.666	9.76
480	4.609	2.661	10.27

Table 2

Absolute difference (mm^3) and relative difference (%) for foveal choroidal volume (FCV) for various inter-scan distance groups

Absolute and relative difference of FCV			
Inter-scan Distance (Microns)	Mean	Standard Deviation	Maximum Difference
<i>Absolute difference (mm^3)</i>			
60	0.002	0.005	0.02
120	0.007	0.005	0.02
240	0.008	0.008	0.03
480	0.010	0.009	0.03
<i>Relative difference (%)</i>			
60	0.942	2.270	9.10
120	3.338	2.899	10.53
240	4.091	3.796	12.25
480	5.095	4.482	16.20

Table 3

Absolute difference (mm^3) and relative difference (%) for foveal choroidal thickness (FCT) for various inter-scan distance groups

Absolute and relative difference of FCT			
Inter-scan Distance (Microns)	Mean	Standard Deviation	Maximum Difference
<i>Absolute difference (mm^3)</i>			
60	1.867	2.177	11
120	9.167	4.488	18
240	11.033	8.927	36
480	13.067	9.078	31
<i>Relative difference (%)</i>			
60	0.753	0.816	3.98
120	3.672	1.815	7.87
240	4.299	3.246	11.93
480	5.209	3.493	12.71