Choroidal Haller’s and Sattler’s Layers Thickness in Normal Indian Eyes

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Abstract:
AIM: This study aims to study normative choroidal thickness (CT) and Haller’s and Sattler’s layers thickness in normal Indian eyes.

MATERIALS AND METHODS: The choroidal imaging of 73 eyes of 43 healthy Indian individuals was done using enhanced depth imaging feature of spectralis optical coherence tomography. R raster scan protocol centered at fovea was used for imaging separately by two observers. CT was defined as the length of the perpendicular line drawn from the outer border of hyporeflective RPE-Bruch’s complex to inner margin of choroidoscleral junction. Choroidal vessel layer thickness was measured after defining a largest choroidal vessel lumen within 750 µm on either side of the subfoveal CT vector. A perpendicular line was drawn to the innermost border of this lumen, and the distance between the perpendicular line and innermost border of choroidoscleral junction gave large choroidal vessel layer thickness (LCVLT, Haller’s layer). Medium choroidal vessel layer thickness (MCVLT, Sattler’s layer) was measured as the distance between same perpendicular line and outer border of hyperreflective RPE-Bruch’s complex.

RESULTS: The mean age of individuals was 28.23 ± 15.29 years (range 14–59 years). Overall, the mean subfoveal CT was 331.6 ± 63.9 µm. Mean LCVLT was 227.08 ± 51.24 µm and the mean MCVLT was 95.65 ± 23.62 µm. CT was maximum subfoveally with gradual reduction in the thickness as the distance from the fovea increased.

CONCLUSION: This is the first study describing the choroidal sublayer thickness, i.e., Haller’s and Sattler’s layer thickness along with CT in healthy Indian population.

Keywords: Hallers and Sattler layer, choroidal thickness, enhanced depth imaging optical coherence tomography

Introduction

Choroid is the vascular layer of the ocular coat extending from ora serrata to optic nerve head.[1] It is the most vascular tissue of the human body. Choroid functions mainly by providing oxygenation and taking care of the nutritional and metabolic needs of the retinal photoreceptor layer.[2] Histologically, it consists of a choriocapillaris layer and choroidal stroma. The vascular component consists of the outer Haller’s layer of large blood vessels and the inner Sattler’s layer of small and medium vessels along with extravascular tissues containing the collagen and elastin fibers form the choroidal stroma.[3]

Various retinal diseases have been attributed to abnormalities of the choroid. Central serous chorioretinopathy (CSCR) has been linked with choroidal hyperpermeability.[3] Polypoidal choroidal vasculopathy has been associated with thickened choroid.[4] Vogt Koyanagi Harada syndrome has been reported to result in loss of small choroidal vessels.[5] Changes in overall choroidal thickness (CT) in various retinal conditions are known; however, our understanding...
regarding the behavior of the individual choroidal vascular layers and its response to these disease conditions is limited.

Assessment of choroidal structure and function is an important to understand the pathophysiology of various retinochoroidal diseases. Enhanced depth imaging (EDI) with spectral domain optical coherence tomography (SDOCT) is increasingly being used to study the choroidal anatomy in both healthy and diseased eyes. Branchini et al. and Chhablani et al. have studied the choroidal structure using Cirrus high definition SDOCT (Carl Zeiss Meditec, Inc., Dublin, CA, USA). Former have studied the thickness of both large and medium choroidal vessel layer along with CT. Chhablani et al. from India have correlated the CT with age and report a reduction in the CT with aging. Apart from the studies from Japan and China in Asia, most of the reports on choroidal morphology studied with SDOCT are from the Western population.

The aim of our study was to report the CT and choroidal vascular layer thickness using EDI on SDOCT in healthy Indian eyes.

Materials and Methods

The study was performed in a tertiary care center in Eastern India between June 2014 and December 2014. It was a cross-sectional observational study. Healthy adult volunteers were explained about the study, and their informed consent was obtained before recruitment. The study was approved by the Institutional Review Board and was conducted as per the tenets of Declaration of Helsinki. Eyes with myopia more than 6 D or hyperopia more than 4 D, haze due to cataract, or vitreous opacities and with history of prior ocular surgery were excluded from the study. All the individuals included in the study had 20/20 best-corrected visual acuity with no color vision abnormality, no systemic disorder, and no family history of any retinal dystrophy. Poor quality of SDOCT images due to unstable fixation and retinal or retinal pigment epithelial (RPE) abnormality were also excluded from the study. All eyes underwent comprehensive ophthalmic examination including measurement of best-corrected visual acuity with Snellen’s visual acuity chart, slit lamp examination, intraocular pressure measurement with applanation tonometer, and fundus evaluation with indirect ophthalmoscope and slit lamp biomicroscopy.

Choroidal imaging

Choroid was imaged using EDI feature of spectralis spectral-domain optical coherence tomography (SDOCT) (Heidelberg Engineering, Heidelberg, Germany). Raster scan protocol centered at fovea with 31 high-resolution B scan lines, measuring 9.3 mm in length, and spaced 240 µ apart was used for imaging. CT was defined as the length of the perpendicular line drawn from the outer border of hyperreflective RPE-Bruch’s complex to inner margin of the choroidoscleral junction. CT was measured at subfoveal location first, the subfoveal CT (SFCT). This was followed by the measurement of CT in nasal, temporal, superior, and inferior directions from fovea. In each direction, three measurements were taken at 500, 1000, and 1500 µ from the center of fovea [Figure 1]. These three readings were termed as N500, N1000, and N1500 for nasal CT measurements and accordingly the readings in other three directions were termed (T500, T1000, T1500; S500, S1000, S1500; and I500, I1000, and I1500). All measurements were taken using inbuilt calliper in the SDOCT system.

Choroidal vascular layers were measured manually. The resolution of the imaging techniques commercially available currently is unable to image the choriocapillaris separately and thus choriocapillaris and the Sattler’s layer are imaged as a single complex. Hence, two layers were imaged, large choroidal vessel layer corresponding to Hallers layer and Medium choroidal vessel layer corresponding to Sattler’s layer and choriocapillaris layer.

Measurement of large choroidal vessel layer thickness (LCVLT) and medium choroidal vessel layer/choriocapillaris thickness (MCSVLT) at subfoveal location was undertaken using the method suggested by Branchini et al. A large choroidal vessel was defined as a lumen of at least 100 µ in diameter within the choroid. The largest choroidal vessel lumen within 750 µ of either side of the fovea was used for the measurement. A perpendicular line was drawn from the SFCT vector to the innermost margin of this choroidal vessel lumen. LCVLT or Haller’s layer thickness was measured as the distance between this perpendicular line and innermost border of choroidoscleral junction. Thereafter, MCSVLT (Sattler’s layer choriocapillaris complex) was measured as the distance between same perpendicular line and outer border of hyperreflective RPE-Bruch’s complex [Figure 2]. For the central foveal thickness, automated values generated by retinal thickness analysis protocol provided with the machine software were taken.

All the SDOCT measurements were independently taken by two authors (KD-observer 1 and CV-observer 2).

All measurement values were expressed as mean ± standard deviation (SD). Intraclass correlation coefficient (ICC) was used to sought out the correlation between the CT measurements by two observers.
To describe the correlation between two variables, Pearson correlation coefficient was calculated. Statistical analysis was done using SPSS Version 15 (SPSS Inc. Chicago, IL). All values are presented as the means ± SDs. In this study, \( P < 0.05 \) was considered statistically significant.

**Results**

A total of 73 phakic eyes of 43 healthy individuals were included in this study. Twenty-seven were male, and sixteen were female. Mean age of individuals was 28.23 ± 15.29 years (range 14–59 years). Thirty-three subjects were adults (18 years or above) 10 individuals were below 18 years of age (median 16 years). The mean CFT was 261.9 ± 27.7 \( \mu \)m. The mean (SFCT) and choroidal vessel layer thickness by both observers are described in Table 1. Overall, the mean subfoveal CT was 331.6 ± 63.9 \( \mu \)m. Among 33 adult individuals, the mean subfoveal CT was 318.2 ± 56.56 \( \mu \)m. Overall mean LCVL (Haller’s layer) thickness was 227.08 ± 51.24 \( \mu \)m while mean MCVL (Sattler’s layer) thickness was 95.65 ± 23.62 \( \mu \)m. The mean ratio between the LCVLT to the mean CT subfoveally was 0.68 ± 0.80.

Table 2 represents the trend of CT change from fovea to the various locations. Choroidal was found to be thickest at the fovea with gradual reduction of CT as the distance from the fovea increases circumferentially.

Table 1 describes the intraclass correlation coefficient for CT measured over various locations. ICC for the (SFCT) was 0.857 with upper limit at 0.774 and lower limit at 0.911 with a 95% confidence interval. ICC for the LCVLT was 0.791 with upper limit at 0.676 and lower bound at 0.868 and for the small choroidal vessel layer thickness was 0.613 with upper limit 0.432 and lower bound at 0.746 with 95% confidence interval. The morphology of the sclerochoroidal junction was bowl shaped in 60 (82.25%) of eyes. The large choroidal vessels were evenly distributed in the nasal and temporal planes in 64 (87.69%) of eyes with no eyes showing focal choroidal thinning. The thickest point of choroid being subfoveally was found in 70 (96.24%). Mean (SFCT) in males was 335.96 ± 61.53 \( \mu \)m and in females was 324.24 ± 66.88 \( \mu \)m \( (P = 0.32) \). Mean LCVL thickness in males was 233.11 ± 43.36 \( \mu \)m, and females were 216.81 ± 56.02 \( \mu \)m \( (P = 0.45) \). Mean MCVL thickness in males was 95.68 ± 23.29 \( \mu \)m and in females was 95.59 ± 24.06 \( \mu \)m \( (P = 0.36) \).

Pearson correlation coefficient between age and (SFCT) was found to be −0.45 with a \( P = 0.001 \) and between age and LCVLT and MCVLT was found to be −0.34 with \( P = 0.03 \) [Table 3]. There was no significant correlation between the CT and choroidal vessel layer thickness with spherical equivalent \( (P = 0.35) \).
**Table 2:** Interobserver correlations in spectral domain optical coherence tomography-measured choroidal thickness across various locations

| Region   | ICC  | 95% CI Lower bound | 95% CI Upper bound |
|----------|------|---------------------|--------------------|
| N1500    | 0.979| 0.966               | 0.987              |
| N1000    | 0.977| 0.963               | 0.986              |
| N500     | 0.950| 0.918               | 0.969              |
| Subfoveal| 0.857| 0.774               | 0.911              |
| T500     | 0.835| 0.741               | 0.897              |
| T1000    | 0.920| 0.871               | 0.951              |
| T1500    | 0.932| 0.889               | 0.958              |
| S500     | 0.936| 0.896               | 0.961              |
| S1000    | 0.984| 0.973               | 0.990              |
| S1500    | 0.966| 0.944               | 0.979              |
| I500     | 0.992| 0.987               | 0.995              |
| I1000    | 0.992| 0.987               | 0.995              |
| I1500    | 0.098| −0.152              | 0.336              |
| LCVLT    | 0.791| 0.676               | 0.868              |
| MCVLT    | 0.613| 0.432               | 0.746              |

**Table 3:** Pearson correlation coefficient between age and subfoveal choroidal and choroidal vessel layer thickness

|           | Pearson correlation coefficient | P   |
|-----------|--------------------------------|-----|
| SFCT      | −0.45                          | 0.001|
| LCVLT     | −0.34                          | 0.03 |
| MCVLT     | −0.34                          | 0.03 |

**Discussion**

Understanding choroidal morphology is vital for diagnosis and treatment of various vision-threatening retinal diseases. Although indocyanine green angiography (ICGA) provides functional details, it does not provide three-dimensional structure of choroid. ICGA being an invasive procedure is also fraught with risk of adverse reaction to dye. SDOCT systems using EDI are increasingly being used to study choroidal morphology in different eye diseases as well as in healthy eyes. In myopic eyes, choroid has been noted to be thickest at temporal end of macula contrary to emmetropic eyes in which subfoveal choroid is thickest. The CT decreases with increase in the axial length in myopic eyes; which can be used as predictive factor visual acuity. Similarly, in eyes with retinitis pigmentosa, thinning of choroid has been associated with poor visual acuity. In eyes with CSCR thicker, choroid has been found in both involved and uninvolved fellow eyes of patients. Eyes with exudative age-related macular degeneration (AMD) have thinner choroid than those with nonexudative variety. On the contrary, eyes with polypoidal choroidal vasculopathy have thicker choroid than neovascular AMD. The knowledge of three-dimensional structure of choroid has been changing the understanding of other retinal diseases as well. Studies have reported thinning choroid with increase in the severity of diabetic retinopathy. On the other hand, diabetic retinopathy has also been associated with thickened choroid by different group of researchers.

Most studies have focused on overall CT profile in diseases. Since choroid is a dynamic structure, concentrating only on the overall CT and overlooking individual choroidal vascular layers may not fill the gaps in knowledge about pathogenesis various retinal and choroidal disease. It has been found that choriocapillaries are selectively affected in early AMD and the large vessel layer mainly thickened in CSCR. Hence, there is a need to study in detail the choroidal sublayers.
known to vary with different axial lengths and the axial length in turn varies among different ethnicities.\cite{24}

Ours was the first study from India to provide choroidal vascular layer thickness along with CT values.

We have noted that choroid was thickest at subfoveal location. This finding was in keeping with Branchini et al.\cite{25} However, mean subfoveal CT (331.6 µ) noted in our study was higher than that noted by Branchini et al. (256.8 µ) and Chhablani et al. (216.4 µ).\cite{25,26}

There has been variation in the mean subfoveal CT across the studies. Ikuno et al. from Japan and Rahaman et al. from the United Kingdom have reported mean subfoveal CT as 354 ± 111 µ and 332 ± 90 µ, respectively.\cite{27,28} On the other hand, Hirata et al. from Japan have measured this value as low as 191.5 ± 74.2 µ.\cite{29}

On the other hand, Hirata et al. from Japan have measured this value as low as 191.5 ± 74.2 µ.\cite{29} However, the ratio of LCVL to CT was identical (0.68 and 0.70, respectively). Table 4 summarizes the CT measurements in various studies reported in literature.

Measurement of CT with different SDOCT machines have shown comparable results.\cite{30}

Difference in the ethnicity of study individuals may be one of the factors responsible for the variation in the mean subfoveal CT across different studies. Moreover, CT is influenced by number of factors such as axial length, refractive error, age, and time of the day.\cite{26,31}

Different CT values across different studies dealing with different study populations may also mean that methods of measurement of CT need to be standardized further so that CT can be used as a diagnostic and prognostic marker. Understandably, choroid being a vascular layer of the eye must be influenced by various other systemic changes in the body which also need to be studied in further studies. One such study by Wong et al. had noted that CT was significantly higher in individuals with hypercholesterolemia.\cite{32}

There can be other factors which may be influencing CT and would be responsible for different values across different studies. 3D scan can also have potential use in the analysis of the choroidal vasculature. Zhang et al. have utilized 3D scan SDOCT to describe the choroidal vascular layers thickness. En face OCT to analyze choroidal vasculature has been described by Sohrab et al. However, both studies needed an intricate array of postacquisition processing, thereby their limiting their usefulness in the present clinic scenario.\cite{33,34}

Our study has few drawbacks. It was a single-center study which did not undertake axial length of the eyes into consideration while measuring CT. We had excluded eyes with high myopia (more than 6 D) and high hyperopia (more than 4 D) which would exclude the eyes with extreme axial lengths.

Despite several attempts of image the choroidal morphology, no standard protocol is available due to the highly variable nature of the choroidal sublayers.

Role of choroid and the changes in the CT in AMD and CSCR has been described in the past.\cite{35,36,37} However, the knowledge about the role of choroidal vascular layer in the pathogenesis of these diseases still remain elusive. Qualitative and quantitative analysis of the Haller’s and Sattler’s layer thickness may provide better understanding of the diseases and may help in formulation of better therapeutic strategies.

**Conclusion**

The present study provides first spectralis SDOCT-based data on CT and vascular layers of choroid in Indian participants. Our findings can be used to study and compare CT and CT vascular layer measurements in different disease conditions in different populations. Variation in the CT values across different studies also emphasizes on the need for standardization of the methods of measurement of CT.

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**Table 4: Comparison of the present study to the previously published literature on choroidal thickness in healthy adults**

| Studies                          | Place of study | Number of subjects (eyes) | Mean age (years) | Ethnicity       | Machine              | Mean CT±SD (µ) |
|----------------------------------|----------------|---------------------------|------------------|-----------------|----------------------|----------------|
| Margolis and Spaide\cite{38}      | USA            | 30 (54)                   | 50.4             | Not mentioned   | Spectralis SD-OCT    | 287±76         |
| Ikuno et al (2010)\cite{39}       | Japan          | 43 (43)                   | 39.4             | Japan           | HP-OCT prototype     | 354±111        |
| Hirata et al\cite{29}             | Japan          | 31 (31)                   | 64.6             | Japan           | Topcon SS-OCT        | 191.5±74.2     |
| Manjunath et al\cite{40}          | USA            | 34 (34)                   | 51.1             | Not mentioned   | Cirrus SD-OCT        | 272±81         |
| Ding et al\cite{41}               | China          | 210 (420)                 | 49.7             | China           | Spectralis SD-OCT    | 261.93±88.4    |
| Rahman et al\cite{42}             | UK             | 50 (100)                  | 38               | Mixed           | Spectralis SD-OCT    | 332±90         |
| Chhablani et al\cite{43}          | India          | 71 (124)                  | 42.8             | India           | Cirrus SD-OCT        | 280.1±46.5     |
| Present study (2015)              | India          | 43 (73)                   | 28.23            | India           | Spectralis SD-OCT    | 331.6±63.9     |

SD: Standard deviation, CT: Choroidal thickness, SD-OCT: Spectral-domain optical coherence tomography, HP-OCT: High penetration optical coherence tomography, SS-OCT: Swept-source optical coherence tomography.
Conflicts of interest
There are no conflicts of interest.

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