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ORIGINAL RESEARCH

Evaluation of Central Macular Thickness and Retinal Nerve Fiber Layer Thickness using Spectral Domain Optical Coherence Tomography in a Tertiary Care Hospital

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ABSTRACT

Purpose: To evaluate the normative data of macular thickness and retinal nerve fiber layer thickness (RNFL) among normal subjects using spectral domain optical coherence tomography (OCT).

Materials and methods: Normal subjects presenting to a tertiary medical hospital were included in the study. All patient underwent clinical examination followed by study of macular thickness and RNFL thickness by spectral domain Topcon OCT. The data was collected and analyzed for variations in gender and age. The data was also compared with available literature.

Results: Total numbers of patients enrolled in the study were 154 (308 eyes). Numbers of males were 79 (158 eyes) and numbers of females were 75 (150 eyes). The mean age among males was 42.67 ± 12.15 years and mean age among females was 42.88 ± 11.73 years.

Overall the mean macular thickness (central 1 mm zone) with SD-OCT was 241.75 ± 17.3 microns. The mean macular volume was 7.6 cu. mm ± 0.33. On analysis of the RNFL thickness, we observed that the RNFL was thickest in the inferior quadrant (138.58) followed by superior (122.30) nasal (116.32) and temporal quadrant (73.04).

Gender-wise comparison of the data revealed no statistically significant difference for age, macular thickness parameters, volume and RFNL values except outer temporal thickness among males and females. No age-related difference was noted in the above parameters. On comparison with available normative data from India and elsewhere, we found significant variations with different machines.

Conclusion: The study is the first to provide normative data using SD-OCT from central India. The data from spectral domain OCT correlated well with the values obtained from similar studies with SD-OCT. Values obtained from time domain OCT machines are different and are not comparable.

Keywords: Spectral Domain OCT, Normative data, Central India.

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INTRODUCTION

Optical coherence tomography is a standard noninvasive diagnostic test today to visualize the morphology of retina. It provides high-resolution, cross-sectional, quantitative image of the retina and helps us measure the thickness of retina at various points. Central macular thickness can be measured with the OCT and correlated with clinical examination and visual function.[1,2] Similarly, retinal nerve fiber layer thickness (RFNL) around the disc (peripapillary RFNL) can be measured with the OCT and correlated with the health of neural retinal rim of the optic nerve head and visual fields of the patient.[3,4] With evolution and refinement of technology now we have moved from time domain to spectral domain OCT. This has lesser image acquisition time and provides high resolution images which help us delineate pathology from normal tissues.[5-8] There are very few large studies on the normative data for macular thickness using the spectral OCT. The macular thickness measurement for diagnostic function may differ with the population used as a database. There are differences in normative data with respect to age, sex, gender and race.[9,10] Such differences need to be taken into account while interpreting raw data. Most of the newer generation machines have inbuilt normative data and hence, are able to differentiate normal values from abnormal and represent it in a color coded manner. However, apart from color coding representation of data; knowledge of normal absolute values is also essential which may vary between different machines. The absolute cut-off values of central macular thickness may be a deciding factor to treat the macular edema, which may vary according to the machine being used. Similarly, in certain scenarios absolute values of RFNL may be deciding factor in diagnosis of glaucoma.[11,13] Thus, it is essential for the operator and ophthalmologist to have complete knowledge of normative data of the machine being used to examine the respective patient. Most of the studies of normative data of macular thickness and retinal nerve fiber thickness (RFNL) were done in northern[14,15]

or southern[16,17] India using time domain OCT. This study was done to establish the normal macular thickness and RFNL parameters using spectral domain OCT (3D OCT 2000, Topcon corporation, Tokyo, Japan) in central India at a tertiary medical college.

AIM

To evaluate the central macular thickness (CMT) and retinal nerve fiber layer (RFNL) thickness in normal subjects presenting at tertiary care hospital using spectral domain optical coherence tomography (OCT).

MATERIALS AND METHODS

Materials

Our study was conducted at ophthalmology department and healthy volunteers presenting to eye out patient department were included in this cross-sectional study. This study was approved from the research and ethics committee of the institute. Informed consent was obtained. All subjects underwent vision, refraction, examination of eye with slit-lamp, Goldmann applanation tonometry, gonioscopy and fundus examination with plus 90D lens.

Inclusion criteria were age > 18 years, healthy volunteers constituting attendants of patients, hospital staff, contralateral normal eye of patients were included in this cross-sectional study. Exclusion criteria were family history of glaucoma, history of prior photocoagulation, history of prior ocular disease, history of intraocular surgery, previous ocular trauma, vertical asymmetry of cup: disk (C:D) ratio (>0.2) between the two eyes, high C: D ratio (>0.6), disk hemorrhages, disk pallor, and localized RNFL defects, refractive error of > ±4 diopter, intraocular pressure >22 mm Hg. Optical coherence tomography was performed using 3D OCT 2000 (Topcon corporation, Tokyo, Japan), with software version 3.

Methods of Evaluation

Eyes that fulfilled both exclusion and inclusion criteria were selected for analysis, if both eyes fulfilled the criteria, both the eyes were included. After complete clinical examination, each eye was dilated with tropicamide 1% before recording the images, and scans were performed with a minimum pupillary diameter of 5 mm. After entry of details of patient including age, sex, race (Asian) specific examination modes were selected.

Central Macular Thickness

The macular evaluation mode was selected from the computer console. The scan was performed with 3D 6.0 × 6.0 protocol. The image was taken with green cross as the internal fixation target. After saving the computer image the analyzed data values using inbuilt protocol was noted. The report generated by the machine gives the color image of central macular with image centered at the fovea. The macular thickness is depicted as concentric circles of 1, 3, and 6 mm from the center of fovea. All the values of macular thickness and macular volume were noted, tabulated and analyzed.

For RFNL Analysis

The glaucoma evaluation mode was selected from the computer console. The scan was performed with 3D 6.0 × 6.0 protocol. The image was taken with green cross as the internal fixation target. After saving the computer image the analyzed data values using inbuilt protocol was noted. The report generated by the machine gives the color image of optic nerve head surrounded by 3.4 mm green centration ring. It gives the peripapillary RFNL thickness of superior, inferior, nasal and temporal quadrants along with total average RFNL thickness. All these values were noted and analyzed.

RESULTS

Total numbers of patients enrolled in the study were 154 (308 eyes). Numbers of males were 79 (158 eyes) and numbers of females were 75 (150 eyes). The mean age among males was 42.67 ± 12.15 years and mean age among females was 42.88 ± 11.73 years.

Overall, the mean macular thickness (central 1 mm zone) with SD-OCT was 241.75 ± 17.3 microns. The mean macular volume was 7.6 ± 0.33 cu. mm. On analysis of the RNFL thickness, we observed that the RNFL was thickest in the inferior quadrant (138.58) followed by superior (122.30) nasal (116.32) and temporal quadrant (73.04) (Table 1).

Gender-wise comparison of the data revealed no statistically significant difference age, macular thickness parameters, volume and RFNL values except outer temporal thickness (OTT) among males and females (Table 2). The standard deviation (SD) of OTT among males is 5.35 and among females is 10.21. This difference of SD is responsible for statistical significance among the two groups.

To study age-related change in macular thickness and RNFL values, the data was divided into two groups with age < 40 years (152 eyes) and second group age ≥ 40 years (156 eyes). No statistically significant difference was noted in macular thickness parameters, volume, and RFNL values (Table 3).

Table 1: Normative data of OCT parameters of 154 subjects (308 eyes)

	N	*Minimum*	*Maximum*	*Mean*	*Std. deviation*
Average macular thickness	154 (308 eyes)	269.00	295.00	279.4277	7.17867
Outer nasal thickness	154 (308 eyes)	261.00	308.00	283.7143	12.94570
Outer temporal thickness	154 (308 eyes)	244.00	286.00	253.6039	8.34659
Outer superior thickness	154 (308 eyes)	245.00	284.00	264.0195	12.15557
Outer inferior thickness	154 (308 eyes)	248.00	285.00	266.0455	11.37842
Inner nasal thickness	154 (308 eyes)	290.00	333.00	306.7078	10.46059
Inner temporal thickness	154 (308 eyes)	285.00	308.00	298.7597	5.64304
Inner superior thickness	154 (308 eyes)	284.00	333.00	303.3442	12.14906
Inner inferior thickness	154 (308 eyes)	278.00	320.00	296.9026	10.79323
Central macular thickness	154 (308 eyes)	212.00	296.00	241.7532	17.30553
Total volume	154 (308 eyes)	7.13	8.20	7.6056	0.33822
Average RNFL	154 (308 eyes)	102.75	125.50	112.5601	5.19985
Superior RNFL	154 (308 eyes)	101.00	133.00	122.2987	9.48968
Inferior RNFL	154 (308 eyes)	125.00	153.00	138.5844	7.52226
Nasal RNFL	154 (308 eyes)	90.00	137.00	116.3117	13.48614
Temporal RNFL	154 (308 eyes)	58.00	98.00	73.0455	8.95036

Table 2: Difference of parameters according to gender

	Gender	*N*	*Mean*	*Std. deviation*	*Significance*
Average macular thickness	Male	79 (158 eyes)	279.3082	7.33188	0.833
	Female	75 (150 eyes)	279.5536	7.06081	
Outer nasal thickness	Male	79 (158 eyes)	283.3165	13.37386	0.697
	Female	75 (150 eyes)	284.1333	12.55510	
Outer temporal thickness	Male	79 (158 eyes)	251.5316	5.35852	0.001
	Female	75 (150 eyes)	255.7867	10.21564	
Outer superior thickness	Male	79 (158 eyes)	264.4684	12.71365	0.640
	Female	75 (150 eyes)	263.5467	11.60533	
Outer inferior thickness	Male	79 (158 eyes)	267.0127	12.02934	0.280
	Female	75 (150 eyes)	265.0267	10.63520	
Inner nasal thickness	Male	79 (158 eyes)	306.9114	10.18318	0.805
	Female	75 (150 eyes)	306.4933	10.80962	
Inner temporal thickness	Male	79 (158 eyes)	298.3797	5.79191	0.393
	Female	75 (150 eyes)	299.1600	5.49211	
Inner superior thickness	Male	79 (158 eyes)	303.7848	12.29621	0.646
	Female	75 (150 eyes)	302.8800	12.05725	
Inner Inferior Thickness	Male	79 (158 eyes)	296.2911	10.69286	0.472
	Female	75 (150 eyes)	297.5467	10.93259	
Central macular thickness	Male	79 (158 eyes)	242.0759	16.49302	0.813
	Female	75 (150 eyes)	241.4133	18.22761	
Total volume	Male	79 (158 eyes)	7.6095	0.34302	0.884
	Female	75 (150 eyes)	7.6015	0.33534	
Average RNFL	Male	79 (158 eyes)	112.2722	4.81138	0.482
	Female	75 (150 eyes)	112.8633	5.59656	
Superior RNFL	Male	79 (158 eyes)	122.3544	9.75079	0.941
	Female	75 (150 eyes)	122.2400	9.27193	
Inferior RNFL	Male	79 (158 eyes)	138.4177	7.46892	0.779
	Female	75 (150 eyes)	138.7600	7.62436	
Nasal RNFL	Male	79 (158 eyes)	115.6329	14.21644	0.523
	Female	75 (150 eyes)	117.0267	12.72789	
Temporal RNFL	Male	79 (158 eyes)	72.6835	7.74521	0.608
	Female	75 (150 eyes)	73.4267	10.10509	

DISCUSSION

Retinal thickness or macular thickness is important for diagnosis of early diabetic macular edema, cystoid macular edema, age-related macular degeneration and choosing appropriate management strategies in other cases of retinal diseases.[18] Likewise, RNFL thickness assessment is important for detection of preperimetric glaucoma and damage to ganglion cell layer.[19-21]

Table 3: Difference of parameters according to age

	Age	*N*	*Mean*	*Std. deviation*	*Significance*
Average macular thickness	Less than 40 years	76 (152 eyes)	280.2600	7.18721	0.156
	More than equal 40 years	78 (156 eyes)	278.6168	7.12273	
Outer nasal thickness	Less than 40 years	76 (152 eyes)	284.5921	12.69664	0.408
	More than equal 40 years	78 (156 eyes)	282.8590	13.20933	
Outer temporal thickness	Less than 40 years	76 (152 eyes)	254.9211	9.03292	0.053
	More than equal 40 years	78 (156 eyes)	252.3205	7.45461	
Outer superior thickness	Less than 40 years	76 (152 eyes)	264.7500	12.76153	0.463
	More than equal 40 years	78 (156 eyes)	263.3077	11.57282	
Outer inferior thickness	Less than 40 years	76 (152 eyes)	266.3684	11.08253	0.729
	More than equal 40 years	78 (156 eyes)	265.7308	11.72263	
Inner nasal thickness	Less than 40 years	76 (152 eyes)	308.0526	11.13301	0.116
	More than equal 40 years	78 (156 eyes)	305.3974	9.65286	
Inner temporal thickness	Less than 40 years	76 (152 eyes)	298.9605	5.82510	0.664
	More than equal 40 years	78 (156 eyes)	298.5641	5.49046	
Inner superior thickness	Less than 40 years	76 (152 eyes)	304.7632	12.61837	0.153
	More than equal 40 years	78 (156 eyes)	301.9615	11.58755	
Inner inferior thickness	Less than 40 years	76 (152 eyes)	297.9474	11.41682	0.237
	More than equal 40 years	78 (156 eyes)	295.8846	10.11875	
Central macular thickness	Less than 40 years	76 (152 eyes)	241.9868	16.05033	0.869
	More than equal 40 years	78 (156 eyes)	241.5256	18.54844	
Total volume	Less than 40 years	76 (152 eyes)	7.6428	0.33884	0.179
	More than equal 40 years	78 (156 eyes)	7.5694	0.33582	
Average RNFL	Less than 40 years	76 (152 eyes)	112.7599	5.23425	0.639
	More than equal 40 years	78 (156 eyes)	112.3654	5.19251	
Superior RNFL	Less than 40 years	76 (152 eyes)	122.9079	9.38535	0.433
	More than equal 40 years	78 (156 eyes)	121.7051	9.61345	
Inferior RNFL	Less than 40 years	76 (152 eyes)	138.6053	7.92478	0.973
	More than equal 40 years	78 (156 eyes)	138.5641	7.15975	
Nasal RNFL	Less than 40 years	76 (152 eyes)	116.7632	13.23618	0.683
	More than equal 40 years	78 (156 eyes)	115.8718	13.79663	
Temporal RNFL	Less than 40 years	76 (152 eyes)	72.7632	8.94408	0.701
	More than equal 40 years	78 (156 eyes)	73.3205	9.00576	

Methods to assess macular thickness are slit-lamp biomicroscopy, fundus photography, fundus fluorescein angiography and OCT. Among these, OCT alone provides quantitative assessment of macular thickness.[22-24] OCT provides for accurate assessment of details of retina and nerve fiber layer with high reproducibility and can be correlated well with clinical disease state.[25-29] All the information thus collected needs to be analyzed and interpreted considering age, gender and racial differences.[9-11,13-17]

With various OCT machines available, we need to understand the normative data generated by both TD and SD-OCT machines before we can conclude about abnormalities and decide on management strategies. The color coding system of the analyzed report provides reasonable discrimination between normal and abnormal values.[12] Our study done in central India provides for normative data of population visiting a tertiary care hospital and the data was collected using spectral domain OCT machine, which is a standard tool today.

In our study, the mean macular thickness (central 1 mm zone) with SD-OCT was 241.75 ± 17.3 microns. Compared with this, various studies done with time domain OCT reported macular thickness as 150 microns approximately.[2,30-33]

Massin P et al[34] and Muscat S et al[28] reported mean central macular thickness as 175 approximately, while Guedes V et al[33] reported 210 microns as mean central macular thickness.

Ibrahim MA et al[35] reported, the mean thickness was 188 mm (SD ± 20 mm) in normal eyes with TD-OCT and 266 mm (SD ± 21 mm) on SD-OCT. The mean thickness in the subfields N, S, T and I was: 266, 268, 255 and 267 mm, respectively, when measured by TD-OCT and 340, 340, 327 and 336 mm, respectively, when measured by SD-OCT. The difference in average thickness as measured by both OCT technologies was statistically significant in all subfields ($p < 0.01$). This difference in measurements could be attributed to the difference in measurement protocols used by various machines. Time domain OCT machines measure

retinal thickness from IS/OS to ILM. The Topcon SD-OCT used in our study measures retinal thickness between the ILM and the posterior border of RPE. Factors other than segmentation algorithm (for example, density of sections, acquisition method, and acquisition speed) may contribute to differences in thickness measurements among devices.

Carineto P et al[36] reported significant difference in macular thickness measured by SD-OCT (approximately 227 microns) *vs* TD-OCT (approximately 144 microns) in 40 healthy subjects. Grover S et al[37] reported a difference of approximately 70 microns in the value of that mean central macular thickness between TD-OCT and SD-OCT. This increased measurement corresponds to the inclusion of the outer segment-RPE-Bruch's membrane complex by SD-OCT, which is relevant to studies using the newer SD-OCT for assessment of retinal thickness.

From above studies, it is evident that values of macular differ when measured using TD-OCT and SD-OCT. Thus, we conclude that while reviewing patients and retinal thickness, OCT machine, their protocols should be taken into account and values from different machines cannot be used for comparison or follow-up.

RNFL

In our study, the mean macular volume was 7.6 ± 0.33 cu mm. On analysis of the RNFL thickness, we observed that the RNFL was thickest in the inferior quadrant (138.58) followed by superior (122.30) nasal (116.32) and temporal quadrant (73.04) (Table 1). The mean RNFL from our study was similar to the data available from other studies (Table 4).

Sony P et al[15] in a cross-sectional study of 146 patients of OCT analysis on quadrant-wise analysis of the RNFL thickness, they observed that the RNFL was thickest in the inferior (132.34 ± 14.70 μ) and superior (131.09 ± 14.13 μ) quadrants. The thickness was lesser in nasal (85.93 ± 17.89 μ) and temporal (67.1 ± 12.77 μ) quadrants according to them, the difference between inferior and superior quadrants was not statistically significant suggesting that the ISNT rule does not apply to Indian eyes.

Kanamori et al[38] in their study of 160 normal eyes showed slightly higher values than ours. They found that superior thickness (145.5 ± 19.6 μ), was maximum followed by inferior RNFL thickness (143.1 ± 19.5 μ), temporal (98.7 ± 20.8 μ), and last in nasal quadrant (92.6 ± 20.4 μ). Their observation also did not follow the previously described ISNT rule.

Ramakrishnan R et al[16] in their study (Stratus OCT 3000; Carl Zeiss Ophthalmic Systems-Humphrey Division, Dublin, CA, USA) found that RNFL thickness for superior, inferior, nasal, and temporal quadrants were 138.2 ± 21.74 (95% CI: 134.3-142.1), 129.1 ± 25.67 (95% CI: 124.5-133.7), 85.71 ± 21 (95% CI: 81.9-89.5), and 66.38 ± 17.37 (95% CI: 63.3-69.5) μm, respectively. The mean RNFL thickness was highest in the superior quadrant followed by inferior, nasal, and temporal quadrants (ISNT rule not followed).

Table 4 gives summary of RNFL values and their comparison using various machines. It is clear from the data (Table 4) that RNFL obtained from various machines cannot be used interchangeably.

Seibold LK et al[44] in their study of RNFL thickness from 40 normal subjects using 3 SD-OCT machines and one TD-OCT machine. The mean RNFL thickness was 106.6 ± 12.8 μm for Spectralis, 98.7 ± 10.9 μm for Cirrus, 112.8 ± 13.2 μm for RTVue and 110.1 ± 12.8 μm for Stratus. Despite high correlations, RNFL values are significantly different between instruments and should not be used interchangeably.

It is evident from review of literature that RNFL values obtained using TD- and SD-OCT show correlation but are different. They may not be comparable and should not be used for follow-up and comparison.[45-47]

Johnson DE et al[48] studied RNFL thickness among 20 healthy volunteers using TD-OCT (Stratus) and SD-OCT (RTvue) and found that RNFL measurement with RTvue were thicker by approximately 20 microns as compared to values obtained with Stratus (TD-OCT), thus the technological difference does not allow direct comparison of data.

Lee ES et al[49] in their study compared RNFL values of 108 open angle glaucoma patients and 46 controls using TD-OCT (Stratus) and SD-OCT (RTvue and Cirrus OCT).

Table 4: Comparative data of our study with RNFL values from other studies

	Machine	*N (no. of subjects)*	*Mean*	*Inferior*	*Superior*	*Nasal*	*Temporal*
Current study	Topcon SD-OCT 3000	154	112.5 ± 5.1	138.5 ± 7.5	122.2 ± 9.4	116.3 ± 13.4	73.0 ± 8.9
George Kampougeris et al[39]	SD-OCT + SLO (Optos, UK)	278	114.8 ± 13.3	134.5 ± 18.1	136.7 ± 18	107.2 ± 17.8	79.5 ± 15.3
YM Tariq et al[40]	Cirrus SD-OCT	1521	99.4 ± 9.6	128.8 ± 17.1	124.7 ± 15.7	74.3 ± 12.8	69.9 ± 11.2
Hirasawa et al[41]	Topcon SD-OCT	251	101.9 ± 8.4	125.5 ± 13.1	123.9 ± 13.6	79.6 ± 13.6	78.6 ± 13.3
Bendschneider et al[42]	Spectralis SD	170	97.2 ± 9.7	123.7 ± 16.4	118.0 ± 14.5	76.4 ± 15	68.8 ± 11.1
Huynh et al[43]	Stratus TD-OCT	2132	103.6 ± 10.6	128.3 ± 18.6	129.7 ± 17.5	82.0 ± 16.7	74.6 ± 12.8

RNFL measurements were more with the RTvue, followed by the Stratus, and finally by the Cirrus OCT ($p < 0.05$). However, the tendency was reversed or no longer present in severe glaucomatous eyes and nasal quadrant maps. Thus, the study concluded that direct comparisons of RNFL thickness measurements among OCT instruments should not be done.

In our study, no significant variation was noted in mean central macular thickness and RNFL with age, gender and refractive error. Subjects with high refractive errors were excluded from the study as per protocol. Similar results were reported by Gobel et al[50] and Sony P et al.[15]

The limitation of our study was relatively smaller sample size. Long-term studies with larger population base may be required to validate the results.

Thus, we highlight the fact that macular thickness values are different from TD-OCT and SD-OCT and are not comparable. However, RNFL values do not show such variation. To conclude our study gives data of macular thickness and RNFL in normal subjects using SD-OCT from central India which should form the basis for further studies.

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