Original Article

Peripapillary Retinal Nerve Fiber Layer and Perifoveal Macula Thickness: Which One is More Helpful in the Early Diagnosis of Primary Open Angle Glaucoma Using Optical Coherence Tomography Angiography?

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Abstract

Background: Macula involvement in early glaucoma changes of the retina is still inconclusive. The objective of the study was to compare the precision of parameters of peripapillary retinal nerve fiber layer (pRNFL) thickness and the macula in the diagnosis of early primary open angle glaucoma (POAG). **Purpose:** To evaluate the pRNFL and perifoveal inner macula thicknesses in the early diagnosis of POAG using optical coherence tomography angiography (Angio-OCT). **Materials and Methods:** Fifty-five subjects were included in a prospective, cross-sectional study divided into three groups: early glaucoma (EG) group (46 eyes of 25 patients with early POAG), glaucoma suspects (GS) group (34 eyes of 20 subjects), and control group (20 eyes of 10 healthy subjects). The mean age of the respective groups was 65.47 ± 9.59, 56.53 ± 9.31, and 51.65 ± 4.16. All subjects underwent Angio-OCT scanning using RTVue-100 {Optovue.} The optic nerve head scan was used for the pRNFL and Retina Thickness Map 5 × 5 mm scan for perifoveal inner macula region. Parameters analyzed were total average, superior, inferior, temporal, and nasal thicknesses of both regions. **Results:** There was thinning in both pRNFL and perifoveal inner macula thicknesses in the EG group compared to the N group. Mann–Whitney intergroup analysis revealed statistically significant differences between the EG and the N groups in all parameters of the perifoveal inner macula thickness, while for the pRNFL thicknesses, there were differences only in total average, superior, and inferior thicknesses. The temporal and nasal perifoveal inner macula thicknesses were parameters with highest areas under the receiver operating characteristic curve (0.907 and 0.900, respectively). **Conclusion:** In early detection of glaucomatous optic neuropathy in POAG, parameters of perifoveal inner macula thickness are diagnostically more significant compared to pRNFL thickness using the Angio-OCT.

Keywords: Angio-OCT, early diagnosis, primary open angle glaucoma

Introduction

Glaucoma is a chronic progressive optic neuropathy, in which structural damages of the retina precede functional changes.[1-3] Retinal ganglion cells (RGC) damage with a subsequent retinal nerve fiber loss is considered an important step in the pathogenesis of glaucomatous optic neuropathy (GON).[4,5] The final common pathway is usually cupping of optic nerve head (ONH) and irreversible vision loss. The RGC are present in three layers of the retina: (1) the retinal nerve fiber layer (RNFL) as the axons of the RGC; (2) the ganglion cell layer (GCL) containing the cell bodies (neurons) of the RGC; and (3) the inner plexiform layer containing primarily the ganglion cell dendrites.[6] The optical coherence tomography (OCT) has remained the main noninvasive imaging device in the evaluation of structural damages of the retina in glaucoma.[7] With the development of new high-resolution spectral domain OCT (SD-OCT), equipped with improved software that can segment the retina into distinct layers including the RGC

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layers,[8] the understanding of the pattern of nerve fiber and neuronal loss in glaucoma, particularly in the macula region has improved.[7,9,10] One of the SD-OCTs with such an ability is the optical coherence tomography angiography (Angio-OCT). It is one of the currently available high-speed, high-resolution optovue SD-OCTs with a better demonstrable intraretinal layer separation capability.[11,12] It can evaluate both structural and microvascular changes of the retina due to the incorporated angiography software, with both information having quite often been presented on the same scanned images or printout.

As the ideal method of early diagnosis of glaucoma disease is still unknown, the retinal structural evaluation aspect of the optovue Angio-OCT was explored in the current study to compare the rate of affectation of the two aspects of the RGC: the axons (in the peripapillary zone) and the neurons (mainly in the macula) and their diagnostic significances in the early stage of GON.

Materials and Methods

Subjects

This was an observational, cross-sectional, hospital-based study that included 55 participants categorized into three groups: The first group [early glaucoma (EG)] comprised patients with early stage primary open angle glaucoma (POAG), consisted of 25 patients (46 eyes); all of whom had early GON. The second group comprised glaucoma suspects (GS) totaling 20 persons (34 eyes), and lastly a normal (N) control group of 10 healthy individuals (20 eyes).

Participants, mainly Caucasians (94.5%), were evaluated at the 3rd Urban Hospital, Minsk, republic of Belarus between the period of December 2015 and June 2016. These were patients referred from various clinics and polyclinics in the city. All participants were given detailed explanations about the test and informed consent was obtained from everyone. The Belarusian State Medical University review board approved the research protocol and the moral, ethical, and scientific principles of clinical trials on human beings, as reflected in the Declaration of Helsinki of 1975 (as revised in 2000), were observed.

All individuals included in the study were subjected to a comprehensive ophthalmic evaluation which included initial medical, family, and ocular histories, best-corrected distance visual acuity (BCDVA) at 6 m using the Golovin–Sivtsev table (decimal fraction), dilated fundoscopy using the Heine Beta 200 LED ophthalmoscope, slit lamp biomicroscopy, gonioscopy using the Three Mirror Universal Diagnostic lens – 18 mm, Ocular Instr., USA), intraocular pressure (IOP) measurement using the Goldman tonometry, pachymetry, and central visual field (CVF) testing on the standard automated perimetry using Humphrey Field Analyzer model 745, employing the Swedish Interactive Threshold Algorithm 30-2 (Carl Zeiss Meditec Inc., Dubling, California, USA).

IOP was recorded after a wash off period of glaucoma drugs wherever indicated, and analyzed after adjusting for central corneal thickness wherever required.

CVF second reliable testing results were used for analysis.

Recruited for the EG group were eyes that fulfilled more than one of the following criteria: (1) early signs of glaucomatous ONH changes on dilated fundoscopic examination (vertical cup/disc ratio of 0.6–0.7 or greater than the fellow eye by >0.2; a localized notch in the rim; presence of minimal neuroretinal rim narrowing or defect). (2) Glaucomatous CVF loss [repeatable mean deviation (MD) of −2 to −6; a glaucoma hemifield test (GHT) outside normal limits at 95% normal confidence limits confirmed on at least two visual field examinations]. A persistent repeated difference of >2 mmHg in the IOP of any figure between the pair of eyes on follow-up and presence of disc hemorrhage were added criteria.

Eyes of individuals with a normal CVF findings (MD within 95% limits of the normal reference and GHT within normal limits), but with history of occasional rise in IOP >21 mmHg, a suspicious asymmetry in the color saturation of the compared discs, neuroretinal rim findings including the presence of a minimal violation of the inferior superior nasal temporal (ISNT) rule and/or nerve fiber layer saturation with at least one of the above being suspicious,[13] were used to constitute the GS group. These are individuals with neither clear-cut evidence of glaucoma nor ocular hypertension, but they are also not considered healthy. Their inclusion was aimed at getting any morphological changes of the retina at the earliest process of the GON.

The N group participants had normal CVF findings (MD within 95% limits of the normal reference and GHT within normal limits), no ONH and neuroretinal rim abnormalities, normal anterior segment findings, IOP of <21 mmHg in both eyes on the day of investigation and with no history of raised IOP >20 mmHg, chronic ocular or systemic corticosteroid use in the past, confirmed from their past medical history records.

Inclusion criteria for all participants were: (1) age 40 years and above; (2) BCDVA of 0.6 and better; (3) spherical and cylindrical correction not higher than ±3.0 diopters; (4) a fully opened anterior chamber angle in its entire circumferences on gonioscopy; and (5) absence of pigmentations and/or exfoliative materials. Any participant not meeting above criteria was excluded.

Presented in Table 1 are the main clinic-demographic characteristics of the study groups. The mean age ($M \pm m$) was 65.47 ± 9.59 years in the EG group, 56.53 ± 9.31 years in the GS group and 51.65 ± 4.16 years in N group.

Angio-OCT measurement

All subjects who met the inclusion criteria underwent posterior retinal segment scans using Angio-OCT, RTVue-

100, Optovue, (Optovue, Inc. Fremont, CA, USA), with tracking. This was performed by a single, well-trained examiner. The *}*ONH 3.45 mm*}* and the *}*Retina Thickness Map 5 × 5 mm*}* scans were used to, respectively, obtain the parameters of the peripapillary retinal nerve fiber layer (pRNFL) and the perifoveal inner macula thicknesses. The total average, superior, inferior, temporal and nasal thicknesses of both regions were the parameters analyzed. Only scans with signal strength index of ≥50 were included for the analyses.

Statistical analysis

The results were collected on an excel sheet (Microsoft Office 2013) and analyzed using STATISTICA (version 12) software (Stat Soft Inc., USA). The normality of distribution was verified by inspection of the histogram. Intergroup comparative analysis using Mann–Whitney test (*U*-value) was conducted taking a *P*-value <0.05 as statistically significant.

Area under the receiver operating characteristic curves (AUCs), sensitivity, and specificity tests were calculated for all available parameters using receiver operation characteristic curves (ROC) analysis (Atte Stat package).

Results

The mean values of both the pRNFL and perifoveal inner macula thicknesses are, respectively, presented on Tables 2 and 3. The EG were noticed to have thinner pRNFL and perifoveal inner macula thickness as compared to the GS and N groups in all the analyzed parameters.

The Mann–Whitney intergroup analysis of the pRNFL thickness revealed statistically significant differences between the EG and N groups only in the total average, superior, and inferior pRNFL thicknesses. All other parameters showed no statistically significant differences between the groups [Table 4].

Table 1: The main clinic-demographic characteristics of study groups

Parameters	Early glaucoma group	Glaucoma suspect group	Control group
	N = 46	*N* = 34	*N* = 20
Sex (male/female)	10/13	7/10	3/7
Age (years) (mean ± SD)	65.47 ± 9.59	56.53 ± 9.31	51.65 ± 4.16
IOP (mmHg) (mean ± SD)	22.43 ± 43	21.31 ± 2.91	16.45 ± 1.93
Visual acuity [Me (Q25; Q75)]	1.0 (0.9; 1.0)	1.0 (1.0; 1.0)	1.0 (1.0; 1.0)

N = number of eyes, SD = standard deviation, Me = median, IOP = intraocular pressure.

Table 2: The means and standard deviations of the peripapillary 3.45 RNFL thickness by Angio-OCT among the study groups

Parameters	Early glaucoma group	Glaucoma suspect group	Control group
	Mean ± SD	Mean ± SD	Mean ± SD
	N = 46	*N* = 34	*N* = 20
Total average (μm)	106.97 ± 14.35	113.34 ± 11.97	117.65 ±14.49
Average superior (μm)	35.69 ± 25.37	147.69 ± 22.53	158.20 ± 20.80
Average inferior (μm)	125.89 ± 14.57	130.43 ± 18.85	136.10 ± 14.57
Average nasal (μm)	83.67 ± 21.69	81.26 ± 11.61	91.62 ± 19.62
Average temporal (μm)	82.67 ± 20.05	90.04 ± 19.68	85.75 ± 23.04

N = number of eyes.

Table 3: The means and standard deviations of the perifoveal inner macula thickness by Angio-OCT among the study groups

Parameters	Early glaucoma group	Glaucoma suspect group	Control group
	N = 46	*N* = 34	*N* = 20
	Mean ± SD	Mean ± SD	Mean ± SD
Total average (μm)	104.48 ± 7.83	108.26 ± 5.27	119.20 ± 9.28
Average superior (μm)	102.39 ± 15.20	111.62 ± 10.01	123.40 ± 19.15
Average inferior (μm)	103.11 ± 9.21	105.32 ± 8.62	108.95 ± 8.41
Average nasal (μm)	111.13 ± 5.90	114.56 ± 6.84	124.45 ± 8.23
Average temporal (μm)	100.48 ± 6.44	104.65 ± 4.39	120.45 ± 18.48

N = number of eyes.

For the perifoveal inner macula thickness, Mann–Whitney intergroup analysis revealed statistically significant differences between the EG and N groups in all parameters. Also, between the EG and the GS groups, statistically significant differences were noticed in the total average, superior and temporal perifoveal inner macula thickness [Table 5].

AUCs, sensitivity/specificity test of the parameters of the pRNFL and perifoveal inner macula thicknesses are presented on Tables 6 and 7, respectively. Lower values of the AUCs were recorded in the pRNFL thickness as compared to the perifoveal inner macula thickness. Nasal and temporal perifoveal inner macula thicknesses were the parameters with the highest AUCs among all the parameters of the two regions of the retina.

Among the parameters of the pRNFL, the highest sensitivity was noticed in the temporal pRNFL thickness while a better specificity was recorded in the total average pRNFL thickness [Table 6].

For the perifoveal inner macula thickness, the nasal perifoveal inner macula thickness was the parameter with the highest sensitivity, while the temporal perifoveal inner macula thickness had the highest specificity [Table 7].

Discussions

The peripapillary RNFL analysis is the most frequently performed method of investigation in the evaluation of the retinal morphological changes in glaucoma using the OCT.[7,14,15] These measurements provide good structural and functional correlation.[16] However, few data have revealed the involvement of the macula in early GON[9,10,17-21] and macula imaging can be used to improve the diagnosis of glaucoma.[21-23] Macula imaging is alleged to overcome some shortcomings associated with ONH assessment and pRNFL measurements[24] and has the tendency of becoming a very important imaging procedure in the evaluation of glaucoma patients.

Tan *et al.*[21] using the Stratus OCT identified macula parameters, especially its inner retinal layer, as parameters with higher discriminating power than the pRNFL, but Leung

Table 4: Mann–Whitney intergroup analysis of the peripapillary RNFL thickness

Parameters	*U* value	*P* value
Between early glaucoma and control groups		
Total average	257.00	0.033749
Superior	187.50	0.001215
Inferior	223.50	0.007831
Nasal	295.00	0.129997
Temporal	381.50	0.898050
Between early glaucoma and glaucoma suspect group		
Total average	313.50	0.050001
Superior	332.50	0.092359
Inferior	377.50	0.304262
Nasal	433.50	0.832656
Temporal	357.00	0.184818
Between control and glaucoma suspect group		
Total average	210.50	0.643625
Superior	157.50	0.079578
Inferior	177.00	0.201132
Nasal	165.50	0.119151
Temporal	203.00	0.518762

Table 5: Mann–Whitney intergroup analysis of the perifoveal inner macula thickness

Parameters	*U* value	*P* value
Between early glaucoma and the control groups		
Total average	97.50	0.000000
Superior	112.00	0.000001
Inferior	276.0	0.010458
Nasal	92.0	0.000000
Temporal	85.00	0.00000
Between early glaucoma and glaucoma suspect group		
Total average	477.00	0.003041
Superior	407.00	0.000268
Inferior	631.00	0.142988
Nasal	530.50	0.014570
Temporal	427.00	0.000560
Between control and glaucoma suspect group		
Total average	109.00	0.000036
Superior	184.50	0.005496
Inferior	254.00	0.127870
Nasal	123.00	0.000105
Temporal	121.00	0.000091

Table 6: AUCs/sensitivity/specificity test for the peripapillary RNFL thickness

Parameters	AUCs	Sensitivity	Specificity	*P* value
Total average	0.670	33.330	95.102	0.056075
Superior	0.759	69.230	75.001	0.005241
Inferior	0.713	76.923	70.000	0.020745
Nasal	0.621	82.051	50.000	0.133622
Temporal	0.510	97.430	15.202	0.461614

Table 7: AUCs/sensitivity/specificity test for the perifoveal inner macula thickness

Parameters	AUCs	Sensitivity	Specificity	*P* value
Total average	0.894	84.782	85.690	0.029180
Superior	0.878	89.130	85.190	0.098529
Inferior	0.700	91.304	45.190	0.025223
Nasal	0.900	95.652	75.420	0.004368
Temporal	0.907	78.260	90.420	0.005470

et al.[14] also using Stratus OCT in their work concluded that, the parameters of the pRNFL were the best in diagnosing early glaucomatous structural changes compared to the macula nerve fiber layer (NFL) and the total macula thicknesses. The inconsistency in the findings of the comparisons between the various segments of the macula and the pRNFL in some previous studies may not be unrelated to the software and the technology used by different authors as the OCT has continued to evolve since its first clinical application in 1991.[25] Very importantly to note, however, is that glaucomatous damage to the macula is common and can occur early in the disease.[21,22,26] Notable in that direction is a study conducted by Nakano *et al*.,[10] where they noted a severely thinned macula GCL in eyes with preperimetric glaucoma, while using speckle noise-reduced SD-OCT.

In the current study, we are more specific in the comparisons between the parameters of the pRNFL and the macula thicknesses using the optovue Angio-OCT, RTVue-100.

The Angio-OCT, being the newest generation of the spectral OCT with incorporated motion correction technology and split-spectrum amplitude-decorrelation angiography algorithms, seems to have an edge over the older versions of the OCT device in terms of scanning time (70,000 A scans per second), resolution, and intraretinal layer separation.[12] The retina thickness map of the OCT optovue allows more sampling points of the macula and can provide values of three different layers of the macular region: full, inner and outer macula thicknesses of the fovea, parafovea and the perifovea (with the inner plexiform layer as the reference point for the division between the inner and outer segments). Parameters of each of these sections can then be analyzed independently. The patients' eye tracking possibility of the device is also of great practical importance.[11] There is also an improvement in its signal-to-noise ratio, therefore, significantly reducing possible measurement variability.[6]

The choice of the perifoveal inner macula area in the study was to compare a segment of the macula that is relatively close to the peripapillary zone in term of thickness (as observed in the Angio-OCT scans) and at the same time not missing most area of the macula zone.

In our study, we noticed thinning in both pRNFL and perifoveal inner macula thicknesses in the EG group as compared to the GS and N groups. Thinning in the inner macula thickness in patients with the early stage of POAG in addition to these of pRNFL as observed in our study testify damage to the macula region in the initial stage of the glaucoma. In some previous studies, observations of reduction in macula volume[27] and macula thickness[28] as evidences of macula structural damage in early stage of glaucoma were made. In our study we were specific to the perifoveal inner macula thickness. Besides, measuring the whole macula thickness especially at the center on OCT test for diagnosis of glaucoma may have limited role.[20] Seong *et al*.,[19] in their study, also compared the inner macula thickness to the RNFL but only in patients with normal tension glaucoma.

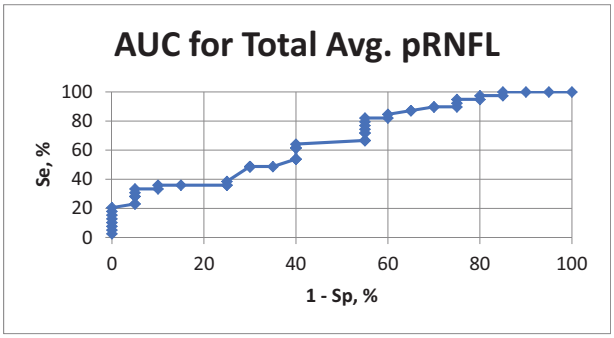

Figure 1: Area under receiver operating characteristic curve for Total Average pRNFL. Se = sensitivity, Sp = specificity

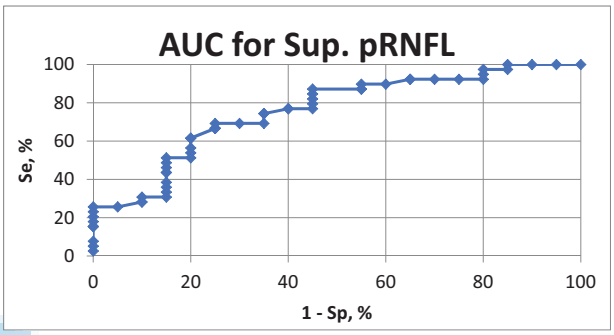

Figure 2: Area under receiver operating characteristic curve for Superior pRNFL. Se = sensitivity, Sp = specificity

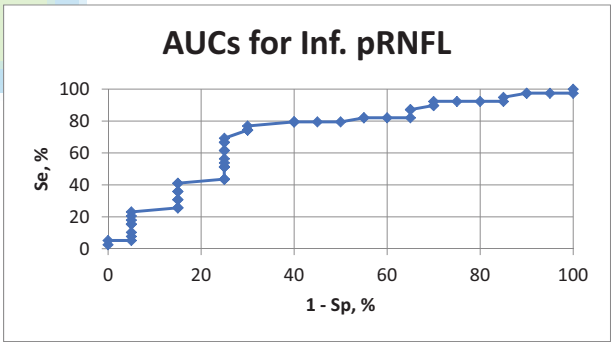

Figure 3: Area under receiver operating characteristic curve for Inferior pRNFL. Se = sensitivity, Sp = specificity

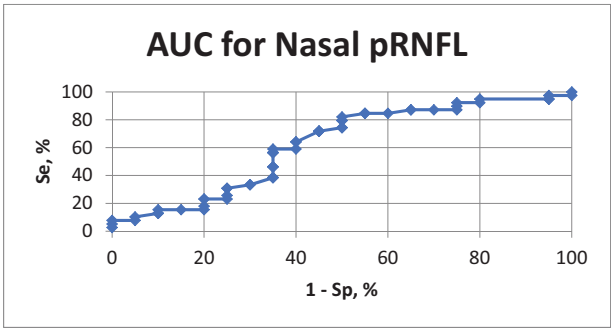

Figure 4: Area under receiver operating characteristic curve for Nasal pRNFL. Se = sensitivity, Sp = specificity

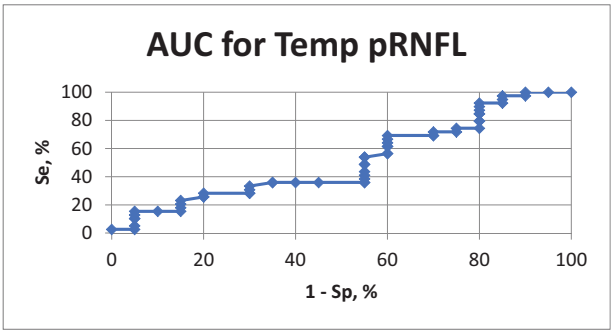

Figure 5: Area under receiver operating characteristic curve for Temporal pRNFL. Se = sensitivity, Sp = specificity

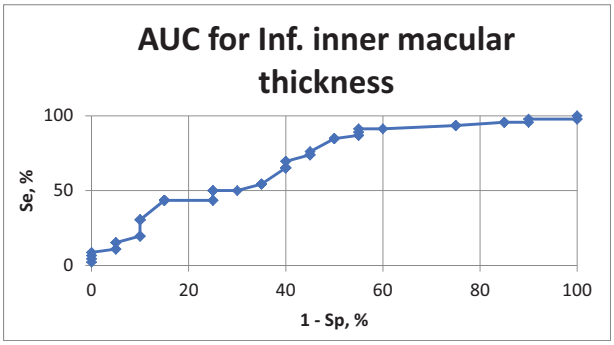

Figure 8: Area under receiver operating characteristic curve for Inferior Perifoveal inner macula thickness. Se = sensitivity, Sp = specificity.

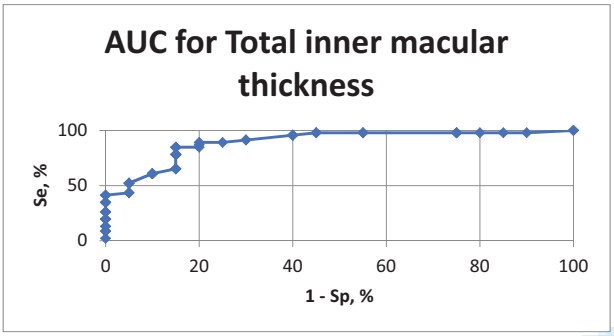

Figure 6: Area under receiver operating characteristic curve for Total average perifoveal inner macula thickness. Se = sensitivity, Sp = specificity

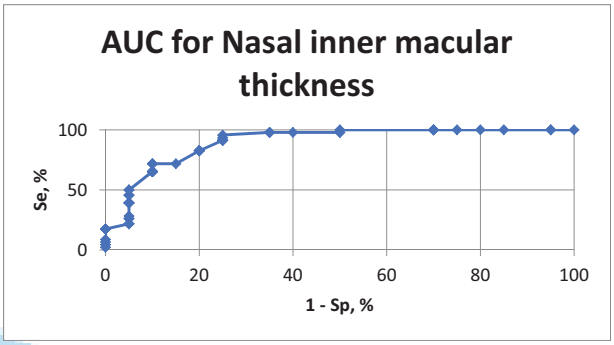

Figure 9: Area under receiver operating characteristic curve for Nasal Perifoveal inner macula thickness. Se = sensitivity, Sp = specificity

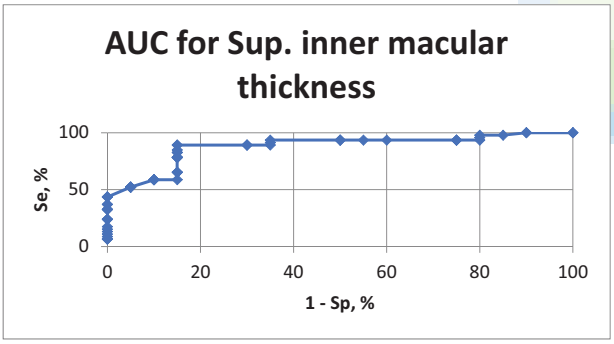

Figure 7: Area under receiver operating characteristic curve for Superior Perifoveal inner macula thickness. Se = sensitivity, Sp = specificity

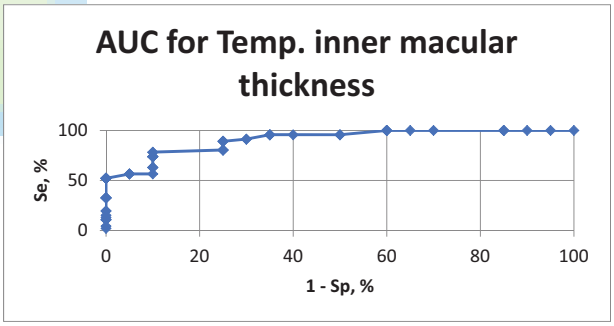

Figure 10: Area under receiver operating characteristic curve for Temporal Perifoveal inner macula thickness. Se = sensitivity, Sp = specificity

Furthermore, the Mann–Whitney intergroup analysis revealed higher statistically significant differences between groups in parameters of the perifoveal inner macula thickness. We also observed higher values of the AUCs in parameters of the inner macula thickness with the temporal and nasal perifoveal inner macula thicknesses having the highest figures (0.907 and 0.900, respectively) [Table 6, Figures 1–5 and Table 7, Figures 6–10]. This indicates higher diagnostic significance of these parameters. A better demonstrable involvement of the perifoveal inner macula thickness in early stage of glaucoma, in comparison to the pRNFL thickness, was, therefore, demonstrated in our study using the Angio-OCT. The same opinion is shared by Nakatani *et al.*[29] while using the SD-OCT, in which they concluded that the macula parameters have high discriminating power with a better reproducibility than the peripapillary RNFL parameters. However, this ratio changes with the increase in the stage of glaucoma.[19]

It may be postulated, from our study that the optovue Angio-OCT is able to detect the death of RGC and the loss of their cellular bodies at an earlier stage of glaucoma than the previous OCT versions and, therefore, probably have more ability to identify reliable early changes in the macula in the patients with POAG, which can improve the chances of early diagnoses of the disease. Further research in this area is a multiethnic approach and a larger size of participants.

Limitations of Our Study

Relatively small number of subjects itself is one concern. Secondly the study was ethnically homogeneous (94.5% Caucasians) that may limit its' universal applicability. A larger sample size and a multiethnicity approach are suggested for further studies.

Conclusion

There is thinning in both the pRNFL and perifoveal inner macula thicknesses in early stage of POAG using Angio-OCT. Parameters of the perifoveal inner macula thickness are diagnostically more significant in early detection of GON than these of pRNFL thickness using the Angio-OCT, although the sensitivity and specificity patterns of parameters of the two regions in it wholeness are similar. Inclusion of macula scans in OCT imaging can improve the early diagnosis of glaucoma.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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