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Macular Thickness and Volume Analysis in Primary Open Angle Glaucoma by Optical Coherence Tomography

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Short title: Macular Thickness and Volume Analysis in POAG By OCT

Abstract

Purpose: the aim of the current study was to compare the difference of retinal macular thickness and macular volume using optical coherence tomography (OCT) between normal subjects and POAG patients.

Methods: This was a case-control observational study held in Mansoura university ophthalmic center during the period from April 2021 to June 2022 on a sample size of 100 eyes. Eyes were divided into 2 groups; group A which include glaucoma group (50 eyes) and Control group (group B) of normal subjects (50eyes) . Eyes underwent O.C.T macula (macular volume and macular thickness) by using SS-OCT (3D DRI OCT Triton [plus], Topcon Corporation, Tokyo, Japan).

Results: Macular thicknesses and macular volume had significant decreased in all macular quadrants (fovea), Inner (Superior, inferior nasal, temporal) and Outer (Superior, inferior, nasal, temporal) quadrants in the glaucoma patients compared to the controls. RNFL thicknesses had significant decreased in Inferior, superior, nasal, temporal quadrants in the glaucoma patients compared to the controls.

Conclusion: This data showed a significant difference in macular volume and macular thickness between normal, compared to glaucomatous eyes in all macular quadrants including fovea, Inner (Superior, inferior, nasal, temporal) and Outer (Superior, inferior, nasal, temporal) so they could be used as reliable indicators to differentiate between cases and the normal subjects.

Keywords: Macular Thickness, Macular Volume, Primary Open Angle Glaucoma, Optical Coherence Tomograph, retinal ganglion cells.

INTRODUCTION:

Glaucoma is a progressive optic neuropathy characterized by a loss of retinal ganglion cells (RGC) which results in characteristic visual field impairment. Glaucoma is diagnosed clinically by observing optic disc changes and by measurement of visual function with perimetry[1].

Studies have shown that the visual field starts to be affected when about 40% of the axons are no longer functional. The visual field loss appears only after the retinal nerve fiber layer (RNFL) and retinal ganglion cell disturbance. In glaucoma, the RNFL defect represents one of the first signs that can be found in a patient However, in a myopic patient with tilted disc and peripapillary atrophy, it is difficult to determine accurate disc margins and the RNFL analysis might not be exact. In order to confirm RNFL analysis, studies tried to use the macular GCC scan[2].

In the macular area, ganglion cells are arranged in 4 to 6 layers making up 30 to 35% of retinal macular thickness, so that the loss of macular ganglion cells results in significant retinal or retinal nerve fiber layer thinning. Several studies indicated that in glaucomatous eyes decrease in macular thickness and volume are due to loss of RGCs and that this finding correlate with RNFL thickness and visual field defects[4].

We decided to do this study because of shortage in studies made about macular volume and macular thickness in

glaucomatous patients. In this study, we aimed to evaluate Inner macular thickness (IMT) (Central 3 mm), Outer macular thickness (OMT) (outer 6 mm zone) and Macular volume (MV) in primary open angle glaucoma (POAG) patients using optical coherence tomography (OCT) and compared it with healthy subject in a case control observational method.

At certain cut point this study could suspect primary open angle glaucoma from O.C.T macula.

PATIENTS AND METHODS

This was a case-control observational study held in Mansoura University Ophthalmic center included 100 adults eyes which were undergoing O.C.T macula from 1 April 2021 to 30 June 2022 after approval from Institutional review board (IRB) code number MS.21.03.1412. Eyes were divided into 2 groups; glaucoma group that included 50 eyes and control group with 50 normal eyes.

The included patients were diagnosed with primary open angle glaucoma with; IOP more than 21 mmHg, glaucomatous optic disc changes (notching of the neuroretinal rim(NRR),symmetrically enlarged cup-to-disc ratio greater than 0.5,increased vertical cup to disc ratio (CDR) and thinning of NRR ,asymmetry of CDR of 0.2 or more ,hemorrhage at or around the optic disc ,peripapillary atrophy, baring of circumlinear vessels ,bayonetting of vessels,Very deep (excavated) cup, laminar dot sign, nasalization of optic disc vessels and diffuse or focal (arcuate) thinning/defect of the retinal nerve fiber layer (RNFL).

Visual field changes according to Hoddap-Parish Anderson criteria. (Mild-moderate POAG was defined where Mean deviation (MD) value was (-4.00dB to−12.00 dB), and advanced POAG was defined where the(MD was −12.01 dB or worse). But excluded patients with media opacity precluding good-quality OCT scans such dense cataract or corneal opacity, patients with coexisting retinal disease as diabetic retinopathy, patients with high myopia and patients with age related macular Degeneration.

Methods:

All the included patients were submitted to complete personal history taking including age, and sex, family history and medical history regarding any intraocular surgery, neurologic, metabolic or systemic disease.

The ophthalmic examination included; best corrected visual acuity (BCVA) using landolt's broken ring chart and then converted to LogMAR, refractive errors were measured using an autorefractor (Canon RK 5 Auto Ref-Keratometer; Canon Inc., Ltd., Tochigiken, Japan), slit lamp examination of the anterior segment by slit Lamp biomicroscopy to assess; corneal clarity, depth of the anterior chamber, state of iris, pupillary reaction, and lens morphology, applanation tonometer was used to measure IOP, assessment of anterior chamber angel using gonioscopy and fundus examination was done by using direct and indirect ophthalmoscope to exclude retinal and macular diseases.

Optical Coherence Tomography Examination

Swept-source OCT (SS-OCT) was performed using (3D DRI OCT Triton [plus], Topcon Corporation, Tokyo, Japan), with a high speed of 100, 000 axial scans/second and center wavelength of 1, 050 nm (version 10.07), digital and optical axial resolution of 2.6 μm and 8 μm in tissue, respectively and transverse resolution of 20 μm.

The Steps of OCT Scanning

Mydriatic eye drops were used to achieve as much pupil dilatation as we can to assure maximal OCT signal and analysis in patients eyes prior to OCT examination. The cornea needed to be well hydrated, with lacrimal film or artificial tears. Each step was explained to the patient before doing it. The chair height, chin rest and imaging machine were adjusted to approximate position. The patient's chin was positioned in the chin rest.

The patient was asked to fixate on a target point inside the instrument. OCT scanning was performed for optic disc & macula at least 2 good quality images were taken then averaged for analysis to confirm reproducible results. Optic disc map for Peripapillary RNFL thickness used three-dimensional raster scan protocol to cover an area of 6.0× 6.0 mm centered on the optic disc with 512 A-scans × 256 B-scans [3D (6.0×6.0 mm512×256)].

Optical coherence tomography imaging of macular area {inner macular layers (inner nuclear, inner plexiform, RNFL and GCL) and outer macular layers} was performed. In both

these groups, parameters analyzed were macular thickness, Inner Macular Thicknesses (IMT) (Central 3 mm), Outer Macular Thicknesses (OMT) (outer 6 mm zone), Central Macular Thickness (CMT) and Macular Volume (MV).

Statistical Analysis of the Data

Software used:

Data entered and analyzed using IBM-SPSS software (IBM Corp. Released 2017. IBM SPSS Statistics for Windows, Version 25.0.

Armonk, NY: IBM Corp.).

Data expression:

Qualitative data was expressed as absolute frequency (N) and percentage.

Quantitative data was initially tested for normality using ShapiroWilk's test with data being normally distributed if p>0.050.

Quantitative data was expressed as mean ± standard deviation (SD) if normally distributed or median and interquartile range (IQR) if not.

Data comparison:

Qualitative data:

Chi-Square test or Fisher's exact test was used.

Quantitative data for two groups:

Independent-Samples t-Test or its non-parametric equivalent; Mann-Whitney U test was used.

Ethics Considerations

This observational study was approved by Mansoura medical research ethics committee, faculty of medicine, Mansoura University. All subjects provided written informed consent prior to study participation.

RESULTS

This study included 100 eyes. The eyes were classified into; control group that included 50 normal eyes and patients group that included 50 glaucomatous eyes. Table (1) shows no statistically significant differences between gender and laterality of eye between glaucoma patients compared to the controls.

Table 1: Comparison of demographic data in case vs. control groups:

Characteristic	Control group (50 eyes)	Case group (50 eyes)	Total	p-value
Age (years)	62.5 (49.8-69)	46 (43-52.8)	51 (46-65)	<.001
Gender				.161
Male	30 (60%)	23 (46%)	53 (53%)	
Female	20 (40%)	27 (54%)	47 (47%)	
Side				.548
Right	27 (54%)	24 (48%)	51 (51%)	
Left	23 (46%)	26 (52%)	49 (49%)	

Age data is median (Q1-Q3), and the test of significance is Mann-Whitney U-test. Sex and side data are N (%), and the test of significance is chi-square test. *: P is significant when < 0.05.

Table **(2)** shows that RNFL thicknesses had significant decreased in Inferior, superior, nasal, temporal quadrants in the glaucoma patients compared to the controls.

Table 2: Comparisons of RNFL measurements in case vs. control groups:

	Control group (50 eyes)	Case group (50 eyes)	t	p-value
RNFL [µm] (Inferior)	135.6 ± 22.8	84.8 ± 32.5	9.058	<0.001 *
RNFL [µm] (Superior)	124.6±21.6	89.7±31.4	6.47	<0.001*
RNFL [µm] (Nasal)	86.4±16.6	62.3±22.8	6.04	<0.001 *
RNFL [µm] (Temporal)	67.66±15.7	60.5±20.6	1.94	0.055 *

t: Student t-test p: p value for comparison between two studied groups. *: Statistically significant at p ≤ 0.05

RNFL thickness: Retinal Nerve Fiber layer thickness

Table **(3)** shows that Macular thicknesses had significant decreased in fovea, Inner (Superior, inferior, nasal, temporal) and Outer (Superior, inferior, nasal, temporal) quadrants in the glaucoma patients compared to the controls.

Table 3: Comparisons of Macular thickness measurements in case vs. control group

Measurement	Control group (50 eyes)	Case group (50 eyes)	t-test	p-value
Macular thickness [µm] (Foveal)	249.8±18.62	233.0±23.6	3.95	<0.001*
Macular thickness [µm] (Inner, Superior)	306.9±20.8	278.5±31.5	5.30	<0.001*
Macular thickness [µm] (Inner, inferior)	304.12±20.3	278.14±32.1	4.82	<0.001*
Macular thickness [µm] (Inner, nasal)	306.5±14.5	276.9±31.8	5.98	<0.001*
Macular thickness [µm] (Inner, temporal)	295.5±17.9	274.9±29.1	4.25	<0.001*
Macular thickness [µm] (Outer, Superior)	264.7±19.9	240.3±22.4	6.11	<0.001*
Macular thickness [µm] (Outer, inferior)	264.4±18.8	237.9±21.5	6.56	<0.001*
Macular thickness [µm] (Outer, nasal)	280.4±18.3	255.5±26.8	5.42	<0.001*
Macular thickness [µm] (Outer, temporal)	261.3±24.5	234.9±20.7	5.82	<0.001*

t: Student t-test p: p value for comparison between the two studied groups *: Statistically significant at $p \leq 0.05$

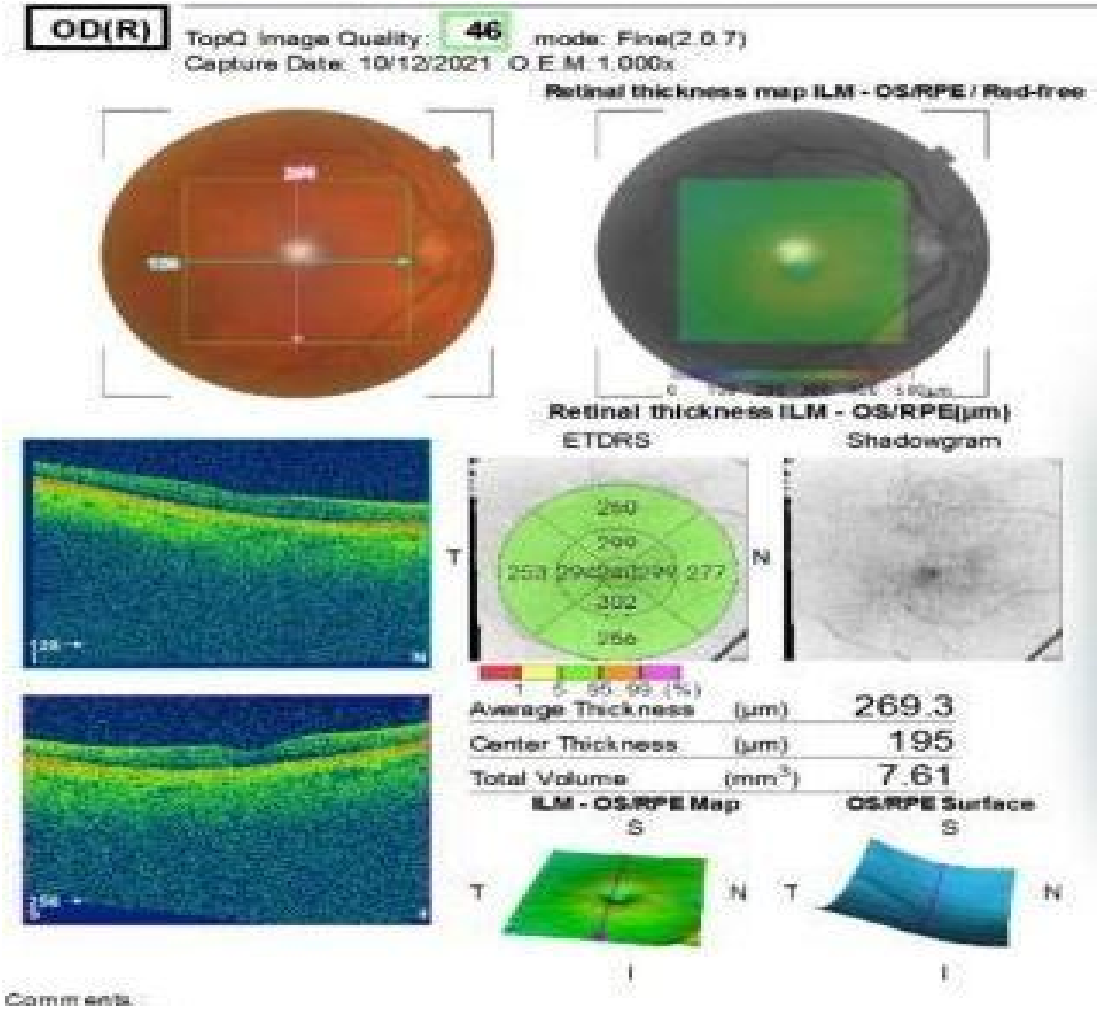

Fig. 1: Macular thickness analysis radial scan of normal subject

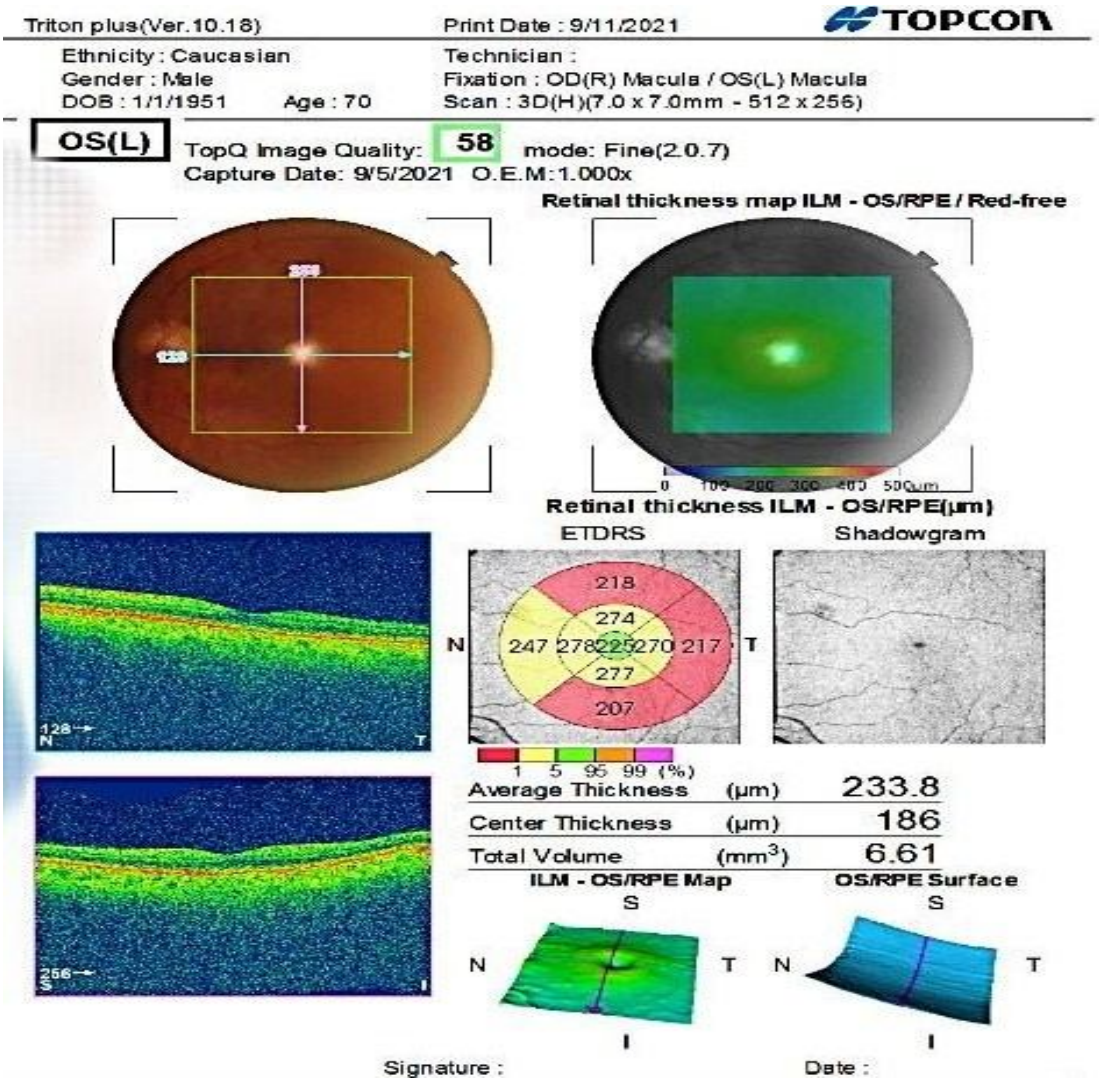

Fig. 2: Macular thichness analysis radial scan of glaucoma patient

Table **(4)** shows that Macular volume had significant decreased in fovea, inner (superior, inferior, nasal, temporal) and outer (superior, inferior, nasal, temporal) quadrants in the glaucoma patients compared to the controls .

Table 4: Comparisons of Macular Volume measurements in case vs. control group:

Measurement	**Control group (50 eyes)**	**Case group (50 eyes)**	**t**	**p-value**
Macular Volume [µm] (Foveal)	0.20±0.013	0.19±0.03	2.36	0.020*
Macular Volume [µm](Inner superior)	0.47±0.04	0.43±0.07	3.47	0.001*
Macular Volume [µm] (Inner inferior)	0.47±0.04	0.42±0.06	3.89	<0.001*
Macular Volume [µm] (Inner nasal)	0.47±0.03	0.44±0.05	3.15	0.002 *
Macular Volume [µm] (Inner temporal)	0.46±.04	0.42±0.07	2.92	0.004 *
Macular Volume [µm] (Outer superior)	1.4±0.10	1.24±0.14	6.28	<0.001*
Macular Volume [µm] (Outer inferior)	1.4±0.098	1.3±0.14	5.27	<0.001*
Macular Volume [µm] (Outer nasal)	1.5±0.096	1.4±0.13	6.87	<0.001*
Macular Volume [µm] (Outer, Temporal)	1.3±0.1	1.2±0.14	5.15	<0.001*

t: Student t-test p: p value for comparison between the two studied group *: Statistically significant at $p \leq 0.05$

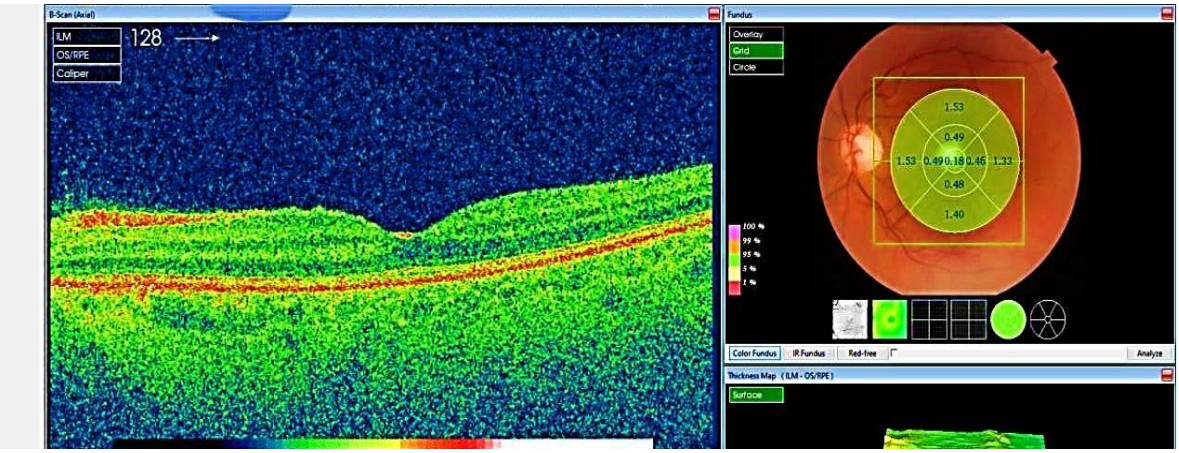

Fig. 3: Macular volume analysis radial scan of normal subject

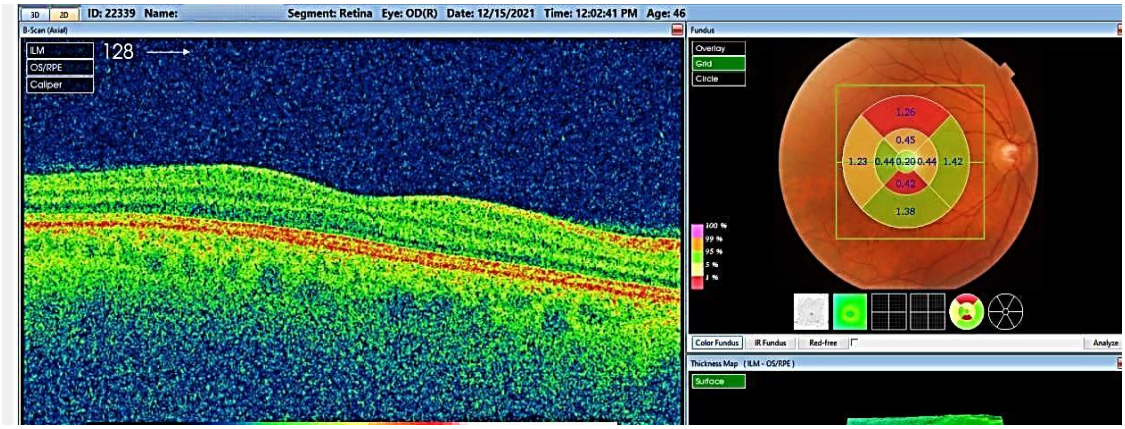

Fig. 4: Macular volume analysis radial scan of glaucoma patient.

Table (5) shows the diagnostic accuracy of RNFL, macular thickness and macular volume measurements in case group vs. Control group. RNFL measurements showed high significance regarding RNFL (inferior, superior, nasal) , macular thickness measurements showed significant macular thickness at (fovea, inner superior, inner inferior, inner nasal, inner temporal, outer superior, outer inferior, outer nasal, and outer temporal) and macular volume measurements showed significant macular volume at(fovea, inner superior, inner inferior, inner nasal, inner temporal, outer superior, outer inferior, outer nasal and outer temporal) in control group than glaucoma group

Table 5: Diagnostic accuracy of RNFL, Macular Thickness and Macular Volume measurements in case vs. control group

	Cutoff value	**AUC (Area Under Curve)**	**p-value**	**Sensitivity**	**specificity**
RNFL Measurement					
RNFL [µm] (Inferior)	≤113	0.916	<0.001	82%	88%
RNFL [µm] (Superior)	≤95	0.809	<0.001	54%	94%
RNFL [µm] (Nasal)	≤66	0.816	<0.001	64%	88%
RNFL [µm]	≤57	0.648	0.009	48%	84%

(Temporal)					
Macular Thickness Measurements					
Macular Thickness [µm]					
(Foveal)	≤235	0.731	<0.001	60%	82%
Macular Thickness [µm]					
(Inner, Superior)	≤285	0.806	<0.001	62%	90%
Macular Thickness [µm]					
(Inner, inferior)	≤289	0.764	<0.001	66%	80%
Macular Thickness [µm]					
(Inner, Nasal)	≤285	0.832	<0.001	64%	90%
Macular Thickness [µm]					
(Inner, Temporal)	≤279	0.730	<0.001	58%	84%
Macular Thickness [µm]					
(Outer, Superior)	≤252	0.822	<0.001	78%	78%
Macular Thickness [µm]					
(Outer, inferior)	≤247	0.817	<0.001	68%	82%
Macular Thickness [µm]					
(Outer, nasal)	≤275	0.793	<0.001	80%	66%
Macular Thickness [µm]					
(Outer, temporal)	≤232	0.834	<0.001	54%	98%
Macular Volume Measurements					
Macular Volume [µm]					
(Foveal)	≤0.19	0.642	0.012	60%	70%
Macular Volume [µm]					
(Inner superior)	≤0.43	0.702	<0.001	50%	90%
Macular Volume [µm]					
(Inner inferior)	≤0.45	0.721	<0.001	64%	74%
Macular Volume [µm]					
(Inner nasal)	≤0.47	0.680	0.001	76%	60%
Macular Volume [µm]					
(Inner temporal)	≤0.42	0.659	0.005	50%	84%
Macular Volume [µm]					
(Outer superior)	≤1.27	0.818	<0.001	60%	92%
Macular Volume [µm]					
(Outer inferior)	≤1.32	0.786	<0.001	74%	76%
Macular Volume [µm]					
(Outer Nasal)	≤1.49	0.838	<0.001	90%	62%
Macular Volume [µm]					
(Outer Temporal)	≤1.36	0.776	<0.001	88%	58%

DISCUSSION

Glaucoma is the leading cause of irreversible vision loss worldwide. Quigley estimated that the number of glaucoma patients would increase to 79.6 million by 2020. Glaucoma is characterized by optic atrophy, visual field defects, and eventual progression to blindness. Glaucoma can be classified as open-angle glaucoma or closed-angle glaucoma. Primary open-angle glaucoma (POAG) is the most common form, accounting for approximately 74% of all glaucoma patients [5].

The factors affecting glaucoma progression are not fully understood, but intraocular pressure (IOP) is considered the most important risk factor for glaucoma. Although reducing the IOP can slow or prevent disease progression, in some cases, glaucomatous optic nerve damage can be further aggravated even if the IOP is controlled, indicating that factors other than IOP may play important roles in glaucoma progression [6].

Glaucomatous optic neuropathy results in death of RGC which are more densely populated in the macular region. On the basis of this anatomical relationship Zeimer *et al*. [7] first observed the large losses in total macular thickness in patients with glaucomatous damage. There is a significant loss of RGCs in perifoveal region. Thus, the aim of the current study was to compare the difference of retinal macular thickness and macular volume using optical coherence tomography (OCT). This was a prospective observational comparative study held in Mansoura university ophthalmic center on a sample size of 100 eyes which were primary open angle glaucoma underwentO.C.T macula. Patients were divided into 2 groups; group A which include glaucoma group (50 eyes) and Control group (group B) of normal subjects (50 eyes).

Recent studies show that thinning of RNFL is related to the thinning of macular ganglion cell complex (GCC), which is defined as three innermost retinal layers: (1) RNFL (made of ganglion cell axons), (2) ganglion cell layer (GCL) made of ganglion cell bodies and (3) the inner plexiform layer (IPL) made out of ganglion cell dendrites. All three layers of ganglion cell complex are significantly thinner in glaucoma patients, reflecting the proportion of dead ganglion cells [8, 9].

With regard to RNFL, the present study demonstrated that there were significant reductions in Inferior, Superior and Nasal among cases compared to control group, while no significant difference was recorded as regards temporal.

In the same line, Shaheen and his colleagues have demonstrated that; there was a significant negative correlation between corrected IOP Vs. total, superior and inferior RNFL in the studied POAG patients [10]. Likewise, Patel and his colleagues conducted their study on a total of 100 eyes, in which the mean RNFL thickness was significantly less in glaucomatous eyes (83.165±15.938) than in normal's (102.42±15.2) and ocular hypertensive's (100.45±7.38). RNFL, average thicknesses in all four quadrants in POAG patients were significantly decreased compared with the OHT and the control groups [11]. Thus, they concluded that; RNFL measurement with SD-OCT could provide important information for detection of early stages of glaucoma. (pre-perimetric glaucoma) as well as help in evaluating progression of glaucoma [11].

In agreement, Bhat and his colleagues have demonstrated that; the average RNFL loss in mild, moderate, and severe POAG was 25.44%, 29.67%, and 44.15%, respectively. A statistically significant correlation ($P < 0.05$) between RNFL loss and severity of glaucoma was found in all except the superior and temporal sectors. A statistically significant ($P < 0.05$) negative correlation was noted between visual field index and RNFL loss in all sectors except the nasal-superior in moderate POAG and all sectors in severe POAG. Mean deviation and RNFL loss showed a significant positive correlation in temporal-inferior (TI) sector in mild POAG and all sectors in the severe group [12].

Also, Abd El-Naby and his colleagues, have demonstrated was that patients with glaucoma had significantly lower RNFL quadrant measurements when compared with controls in microns (65.1±4.03 vs. 77.4±6.87; P=0.0001 for temporal quadrant; 73.7±8.8 vs. 88.5±4.7; P=0.0001 for nasal quadrant; 91.06±18.86 vs. 124.55±6.95; P=0.0001 for superior quadrant; and 94.16±19.17 vs. 129.25±6.65; P=0.0001 for inferior quadrant) [13].

Guedes and his colleagues in their cross-sectional OCT study of total 534 eyes of macular and RFNL thickness in normal and glaucomatous eyes concluded that both parameters showed statistically significant correlation with glaucoma. They concluded that macular thickness may be used as an additional parameter in clinical assessment of glaucoma [14].

Kanadani and his colleagues in their study of glaucoma patients studied macular parameters using OCT and correlated the same with visual field changes and multifocal visual evoked potential (mfVEP) of macular area. They found good correlation between assessment of structural changes of macula on OCT with functional changes on Visual field and mfVEP [15].

Regarding volume, the current study demonstrated that there were highly statistically significant decreases in all macular volume parameters (Inner superior, Inner inferior, Inner nasal, Inner temporal, Outer superior, Outer inferior, Outer nasal and Outer, Temporal) compared to control group (P<0.001).

This came in the same line with Sharma and his colleagues who conducted their study on a total of 144 subjects and were divided into two groups; Group A included 76 patients of primary open angle glaucoma (POAG, n = 124 eyes) and group B included 68 normal subjects (Controls, n = 124 eyes). They have displayed that; the POAG group had significantly decreased value of total macular volume compared to control group [16].

Also, Giovannini and his colleagues conducted their study on 50 eyes of 30 patients (age: 49–68); 20 eyes normal, 15 eyes with early glaucoma and 15 eyes with advanced glaucoma who have been studied with the commercially available OCT unit. They observed significant differences between groups. Normal (7.35 ± 0.455 mm3) and early glaucoma (7.09 ± 0.475 mm3), each had significantly greater volume than subjects with advanced glaucoma (6.678 ± 0.455 mm3) [1]. Thus, they have displayed that; volumetric analysis of macular thickness with OCT tomograms may be a useful method of documenting and monitoring patients with early glaucoma and advanced glaucoma. In their analysis, and according to the observations of other authors, OCT macular volumes correlates significantly with glaucoma status [1].

Also, Lederer and his colleagues evaluated macular volume in normal, glaucoma suspect and glaucomatous subjects using a time domain OCT. Their results demonstrated a significant correlation between the macular volume and glaucoma status with decreased macular volume in patients with advanced disease as well as significant difference of macular volume between normal and glaucomatous eyes [17].

Concerning macular thickness, the current study demonstrated that there were highly statistically significant decreases in all macular thickness parameters (Foveal, (Outer, temporal), (Inner, inferior), (Inner, nasal), (Inner, temporal), (Outer, Superior), (Outer, inferior), (Outer, nasal) and (Inner, Superior) compared to control group (P<0.001).

In accordance, Sharma and his colleagues have demonstrated that; the POAG group had significantly decreased values of outer macular thicknesses (OMT) (212.61 m in cases versus 237.48m among normal subjects) and inner macular thicknesses (IMT) (243.96 m in cases versus 263.56m among normal subjects), compared to control group [16]. These findings were in correlation with previously recorded studies [18, 19].

On the contrary, Li and his colleagues have demonstrated that; the mean thicknesses of the choroid in the macular area in the POAG group and healthy group were 207.97 ± 62.83 μm and 208.24 ± 47.97 μm, and the mean volumes were 0.63 ± 0.19 μm3 and 0.64 ± 0.14 μm3. There were no significant differences in macular choroidal thickness, volumes of various macular regions, or mean choroidal thickness or volume between the POAG and healthy groups (all P>0.05). The macular choroidal thickness of various macular regions was not correlated with visual field MD in the POAG group (all P>0.05) [6].

With regard to the diagnostic accuracy of measurements in case vs. control group, the present study demonstrated that; RNFL, macular Volume and macular Thickness were associated with a high accuracy in the diffranation between the case and the control groups. In the same line, Sung *et al* [20] and Delbarre *et al* [21] in their study evaluated the diagnostic

use of macular thickness and RFNL thickness for diagnosing glaucoma. Both the studies found similar changes in macular thickness which correlated well with RFNL thickness among glaucomatous patients.

On the other hand, Li and his colleagues have demonstrated that; macular choroidal thickness is not an appropriate parameter to evaluate damage caused by POAG, and the role of the macular choroid thickness in POAG needs to be further investigated [6].

CONCLUSIONS:

Macular thickness and macular volume analysis with OCT tomograms may be a useful method of documenting patients with glaucoma. Macular parameters such as inner and outer macular thickness and macular volume can be used in addition to RNFL thickness parameters to aid glaucoma diagnosis in certain conditions as disc abnormalities and peripapillary atrophy. This data showed a significant difference in macular volume and macular thickness between normal, compared to glaucomatous eyes, so they could be used as reliable indicators to differentiate between cases and the normal subjects.

Disclosures

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No financial support was received for this submission.

Data Availability

All data are included in his article**.**

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Ethics declaration

Conflict of Interests

Marina N. George, Amgad M. Elnokrashy, Nashaat S. Zaky, Tarek A. Mohsen. All authors declare have no conflicts of interest that are directly relevant to the content of this review.

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