Reduced macular vessel density and capillary perfusion in glaucoma detected using OCT angiography

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ABSTRACT

Aims: To evaluate retinal vasculature changes in primary open angle glaucoma (POAG) and whether the functional visual loss correlates with parameters obtained using optical coherence tomography angiography (OCTA).

Materials and Methods: OCT and OCTA images were collected from 116 POAG eyes and 40 normal eyes in a prospective, cross-sectional observational study. Glaucomatous eyes were further divided into three groups according to a Glaucoma Staging System. Measurements of macular vessel density, ganglion cell complex (GCC), and disc retinal nerve fibre layer (RNFL) thickness were compared among groups.

Results: The macular vessel density, GCC, and RNFL are significantly reduced in POAG compared to normal eyes that also corresponds to the severity of glaucoma (Kruskal-Wallis test with Dunnett's correction; $p < 0.0001$). Visual field mean deviation correlates significantly with macular vessel density ($p = 0.0028$, r = 0.3), GCC ($p < 0.0001$, r = 0.6), and RNFL ($p = 0.008$, r = 0.36) in POAG. There are significant correlations between GCC and RNFL ($p < 0.0001$, r = 0.76) as well as macular vessel density ($p < 0.0001$, r = 0.48). Increased age also correlates with reduced macular vessel density in both normal ($p = 0.0002$, r = 0.49) and glaucomatous eyes ($p < 0.0001$, r = 0.48), but a greater proportionate reduction of vessel density is seen in glaucomatous eyes.

Conclusion: Reduced macular vessel density occurs in POAG despite of age-related changes, which also correlates with reductions in RNFL and GCC measurements. OCTA can detect microstructural defects and offers potential to facilitate diagnosis of glaucoma.

Key words: glaucoma, intraocular pressure, macula, field of vision, optic nerve, OCTA

INTRODUCTION

Glaucoma is a prevalent blinding disease affecting 60.5 million people worldwide in 2010 and predicted to rise to 79.6 million by 2020, of which bilateral blindness will be present in 11.2 million people.[1] To date, the only consistent evidence-based treatment for glaucoma is to lower intraocular pressure (IOP), however, many patients continue to lose vision despite adequately controlled IOP. Glaucoma is generally regarded as a family of optic neuropathies with multifactorial risk factors dependent or independent of IOP. The association of glaucoma with age, ethnicity and genetic associations, hypotension, diabetes mellitus, migraine, sleep apnoea and primary vasospasm has been reported.[2-4] Elevated IOP is important but not the sole factor responsible for retinal ganglion cell (RGC) death and optic nerve damage.[5]

Many studies have revealed reduced ocular blood flow and perfusion pressure as risk factors for glaucoma,[6, 7] with underlying mechanisms including endothelial cell dysfunction and impaired neurovascular unit.[8] Maintained health of retinal (and CNS) neurons depends on functional interactions between neurons, glia and blood vessels, termed the "neurovascular unit". Understanding neurovascular regulatory mechanisms and retinal haemodynamics in glaucoma, may provide a scientific rationale for novel strategies to treat or prevent glaucoma.[9] In this prospective cross-sectional study, we aim to explore how retinal blood flow changes in different stages of glaucoma using Optical Coherence Tomography Angiography (OCTA) and provide preliminary evidence for a potential to increase our sensitivity to detect early microstructural changes prior to functional loss of vision. Specifically, we wish to answer whether OCTA macular vessel density correlates with currently used clinical parameters, including OCT derived retinal nerve fibre layer (RNFL) thickness, ganglion cell complex (GCC) and visual field (VF).

OCT is a non-invasive imaging modality commonly used to measure RNFL and macular thickness parameters for glaucoma diagnosis. OCTA is a high-speed OCT that compares the decorrelation signals between sequential OCT B-scans taken at precisely the same cross-section in order to construct a map of blood flow. OCTA offers opportunity for clinicians to visualise and assess retinal capillaries non-invasively with acceptable reproducibility, sensitivity and specificity.[7, 10] There is precedence in the use of OCTA in glaucoma focusing on optic disc or parapapillary perfusion.[7, 11] Glaucomatous optic neuropathy is characterized by loss of retinal ganglion cells (RGCs). The primary insult in glaucoma is believed to occur

at the optic nerve head, however, an increasing body of evidence has shown that functional visual loss correlates well with macular inner retinal thinning.[12] Therefore we wished to determine whether the macular microvasculature, as part of the neurovascular unit, is impaired in glaucoma which may explain inner retinal thinning observed by others.

Here we demonstrate that OCTA can detect early defects in disc rim area, RNFL thickness, GCC thickness, and macular vessel density in glaucomatous eyes. There was significant correlation between these measurements and the severity of Primary Open Angle Glaucoma (POAG).

METHODS

Study participants

This is a prospective cross-sectional study conducted at the Clinical Research Unit of the Bristol Eye Hospital, UK. The study protocol and ethics were approved by South West Frenchay Research Ethics Committee and Health Research Authority in England (Ref: 16/SW/0277). The study was performed in accordance with and adhered to the tenets of the Declaration of Helsinki. Written informed consent was obtained from all patients.

A total of 148 eyes from 81 subjects were imaged with the AngioVue™ (OptoVue) platform with 35 to 40 eyes in each group. Each patient underwent best-corrected visual acuity testing, intraocular pressure (IOP) measurement with Goldmann applanation tonometry, static perimetry and OCT scanning of peripapillary RNFL, macular GCC, and macular OCTA. Data including age, gender, blood pressure and co-existing medical conditions, such as diabetes, were collected from patients' medical records.

Inclusion criteria for POAG were: 1. At least 18 years of age; 2. Evidence of optic disc neural rim loss on clinical examination, and/or a glaucomatous pattern of visual field defect on Humphrey visual field (HVF) testing. 3. The IOP has been well controlled for more than 14 days, maximum IOP in individual's medical history is less than 50 mmHg, or IOP within normal range on the day of OCTA scanning. Inclusion criteria for normal subjects were 1. At least 18 years of age; 2. IOP ≤ 21 mmHg with no history of elevated IOP; 3. Normal clinical appearance of the optic disc; 4. No RNFL loss; 5. No history of visual field defect or no documented ocular disease history. Exclusion criteria for all participants included: 1. Any retinal pathology, non-glaucomatous optic neuropathy, uveitis, or ocular trauma; 2. Vision worse than 6/60; 3. Any comorbidity contributing to visual field loss; 4. Significantly poor mobility; 5. Vulnerable individuals or those lacking capacity. 6. A history of intraocular surgery, except for uncomplicated cataract or glaucoma surgery.

Humphrey visual field test

All the visual field tests were performed by a Humphrey Field Analyzer 740i (Carl Zeiss) Swedish Interactive Threshold Algorithm (SITA) 24-2 test. Only reliable results of visual field tests were included in this study with less than 20% fixation loss, 15% false-negative errors, and 15% false-positive errors. All tests were completed without rim and eyelid artefacts, evidence of inattention or fatigue effects. Any loss of visual field caused by a disease other

than glaucoma was excluded from this study. The glaucoma stage was determined based on Glaucoma Staging System (GSS).[13] In brief, the GSS stage assignment based primarily on Humphrey visual field parameters. MD was decided on as the primary measure for assigning stage 0 to 5. Here in this paper we recruited participants as follows: stage 0 (no glaucoma) was determined when MD is more than 0 dB, stage 1 (mild glaucoma) with MD between -0.01 dB and -5.00 dB, stage 2 (moderate glaucoma) with MD between -5.01 dB and -12.00 dB, and stage 3 (advanced glaucoma) with MD is between -12.01 dB and -20dB. and stage 4 (severe glaucoma) with MD is more than -20 dB. In this study, we pooled the advanced with severe glaucoma patients.

OCTA image acquisition and processing

The AngioVue provides non-invasive visualisation of vascular structures of the retina and choroid based on low-coherence interferometry. AngioVue uses an 840-nm light source and has an A-scan rate of 70,000 scans per second with dual orthogonal volumetric imaging of the retina to minimize motion artefacts. All eyes underwent four volumetric raster scans using AngioVue™ OCTA, including two horizontal priority (x-fast) and two vertical priority (y-fast) scans, obtained consecutively volumetric scans covering an area of 3 × 3 mm field size for macular regions to observe the most capillary detail allowed on OCTA. The pupil was dilated with 1% tropicamide 15 minutes prior to all imaging.

An orthogonal registration algorithm is used to produce merged 3-dimensional OCT angiograms. In order to quantify macular circulation, *en face* retinal angiograms from OCTA acquired images were processed and vessel density was calculated using the AngioAnalytics algorithm. AngioVue OCTA system uses a split spectrum amplitude-decorrelation angiography (SSADA) algorithm to minimise scanning time and to capture the motion of blood flow by measuring the variation of the OCT signal amplitude among consecutive cross-sectional scans. The SSADA algorithm compares the consecutive B-scans at the same location to detect flow using motion contrast. Vessel densities are calculated over the entire scan area, which is the whole *en face* macular. Vessel density is defined as the percentage area occupied by the large vessels and microvasculature in a particular region. Macular vessel density analysed in this study included superficial vascular plexus present in the inner layers of retina, extending from internal limiting membrane to inner plexiform layer. Macular vessel density was analysed over a 1.5mm-wide parafoveal, circular annulus centered on the macular.

Measurement of peripapillary RNFL and macular GCC

The peripapillary RNFL thickness and macular GCC thickness were acquired using RNFL 3.45 scanning mode and the GCC scan protocol within the AngioVue™ OCTA. The RNFL mode measures RNFL thickness along a circle 3.45 mm in diameter around the optic disc and the GCC scan covered a 6 × 6 mm scan area centred on the fovea. GCC scan includes the nerve fibre layer, the ganglion cell layer and the inner plexiform layer. The OptoVue algorithm was used for automatic segmentation of the RNFL and GCC for calculations. Average, superior and inferior hemi-retinal RNFL and GCC measurements were included for analysis.

Statistical analysis

Descriptive statistics were used to calculate mean and median of retinal nerve fibre layer (RNFL), vessel density, flow index and to calculate standard deviations. Student's *t* tests were used to compare the average values of measurements between normal and glaucomatous eyes. The Chi-square test was used for frequency data on gender and number of diabetes. Univariate analysis with Pearson correlation test and multivariate analysis with ANOVA testing were performed to determine the correlation between macular vessel density and other variables: age, RNFL thickness, rim area, MD 24-2, and PSD 24-2 in visual fields. Statistical significance was taken as $p < 0.05$. However, for multiple comparisons among groups, a Kruskal-Wallis test with Dunn's correction was applied with resultant significance level set at $p < 0.01$.

RESULTS

Demographic characteristics of POAG and normal subjects

Table 1 summarises the clinical characteristics and ophthalmic measurements of each group. There was no statistically significant difference between normal and POAG groups for age and proportion of diabetes mellitus. As expected, VF mean deviation (MD) was significantly lower and maximum IOP was significantly higher in the POAG group compared to normal.

Progressive structural and perfusional changes are detected in the macular of glaucomatous eyes

Table 2 is an overview of the visual field MD, pattern standard deviation (PSD), macular and disc structural measurements for four groups (normal, mild, moderate and advanced glaucoma) according to the Glaucoma Staging System. There were significant differences in all measurements amongst the groups, including the whole, superior and inferior areas of RNFL thickness, GCC thickness, macular vessel density, rim area and C/D area ratio ($p < 0.0001$).
In the *en face* OCTA image of the superficial vascular complex (SVC), there are centripetally branching vessels terminating in the central foveal avascular zone (FAZ) (**Figure 1A**). The GCC is absent at the fovea, but gradually becomes thicker and reaches the thickest point at the parafoveal annulus. The red reflections in the cross-section of retinal layers represent blood flow in retinal vascular plexuses, which are denser in normal eyes than those with glaucoma. Reduced macular GCC thickness and blood flow in glaucoma was detected and a statistically significant correlation with glaucoma stages exists (**Figure 1B-C**). In addition, the data suggests correspondence between GCC thinning and loss of capillaries in the SVC.

Reduced RNFL thickness is detected at the optic disc in glaucoma

As expected, the rim area was significantly smaller with higher rim/disc area ratios in glaucomatous eyes corresponding to the severity of disease (**Figure 2A).** In agreement with the traditional diagnosis of glaucoma, there was a significant reduction in RNFL thickness across the whole area of the disc in glaucomatous eyes compared to normal eyes, which included both superior and inferior of optic disc regions (**Figure 2B**).

OCTA measurements correlate with visual function in glaucoma

There was no correlation between the visual field MD and GCC, RNFL, macular vessel density in normal eyes. In contrast, the visual field MD was significantly correlated with GCC ($p <$

0.0001, r = 0.6), RNFL (p = 0.008, r = 0.36) and macular vessel density (p = 0.0028, r = 0.3) in POAG (**Figure 3).** The correlation with VF was also regionally correlated to VF sensitivity in the corresponding hemisphere, which is in agreement with traditional glaucoma diagnostic measurements.[14]

The correlation between macular vessel density and RNFL or GCC thickness were statistically significant within the glaucoma group, while there was no correlation within normal subjects (**Table 3**). The data suggested that, in addition to RNFL thickness, macular vessel density could also be considered in diagnosis of glaucoma, with OCTA imaging providing direct and visualized measurement of all these parameters.

Increasing age is correlated with reduced macular vessel density in both normal and glaucoma subjects

Age was not correlated with GCC or RNFL in either the normal or POAG group. However, age was suggested to be an important factor correlating to macular blood flow. There was a significant correlation between increasing age and reducing macular vessel density in both normal (p = 0.008, r = -0.41) and glaucomatous eyes (p < 0.0001, r = 0.-48) (**Table 4**), indicating aged related change may affect macular vessel density. In addition, the degree of reduction of macular vessel density was also significantly related to glaucoma. The decreased macular vessel density in eyes with glaucoma was much higher than normal when compared in same age group (**Table 5**). There was a 1.8% and 3.9% greater reduction in macular vessel density in glaucomatous eyes compared to normal in age group of 66-75 and above 75 years old respectively. The data indicates that macular vessel density decreases significantly more in glaucoma even when age-related changes are accounted for (p < 0.05 in all age groups).

DISCUSSION

We have demonstrated that macular vessel density is reduced in glaucomatous eyes using OCTA. Additionally, we have shown the positive correlation of macular vessel density with retinal GCC thickness, RNFL thickness and visual field MD. In the past decades, a few innovative technologies have been developed for the assessment of blood vessel diameter, ocular blood flow, and oxygen saturation such as retinal vessel analyser, video angiography, bidirectional laser Doppler velocimetry, and colour Doppler imaging. None of these techniques however provide direct visualisation and accurate measurement of retinal capillaries.[15] OCTA provides new and important information on retinal haemodynamics using non-invasive and fast acquisition of images.

IOP plays an important role in RGC damage in glaucoma, but a critical role of vascular insufficiency in the initiation and progression of glaucoma has also been suggested.[16] This is supported by the observation that POAG and normal tension glaucoma (NTG) are often associated with migraine, peripheral vascular abnormality, vasospasm, and nocturnal hypotension.[17, 18] Further evidence of vascular insufficiency in glaucoma is re-enforced by magnetic resonance imaging revealing a greater extent of cerebral small-vessel ischemic changes and infarcts in NTG compared to age-matched controls.[19,20] Here we detected macular vascular insufficiency in POAG patients using OCTA compared to normal. This is in agreement with a recent study, which has demonstrated that reduced blood perfusion of the macular superficial vascular complex in glaucoma,[14] and other studies describing dropout of the peripapillary microvasculature in glaucomatous eyes.[10, 21,22]

It has long been debated whether reduced blood perfusion in glaucomatous eyes is secondary to loss of ganglion cells or is indeed a causative factor. The process of damage to the retinal vessels and RGCs, the cause of ultimate visual loss in glaucoma is not the principal subject of this study. Here we tested the tenet that structural outcome measures of macular GCCs and RNFL correlates with blood flow and combined parameters may better facilitate the detection of glaucoma progression to be tested. As shown in **Table 3** both GCCs and RNFL thickness are significantly correlated with macular vessel density in measured areas, and this need to be undertaken in a longitudinal study to validate the temporal relationship between GCC and superficial capillary density.

Studies have explored structure-function relationships between various glaucoma outcome measures and threshold sensitivity in visual field testing.[23, 24] We have evidence here that correlations of macular GCC thickness with VFs are comparable to those of peripapillary RNFL with VFs. Strictly GCC does not solely reflect loss of ganglion cells as the measure of GCC is a combination of RNFL, ganglion cell layer and inner plexiform layer. However, previous attempts to map macular substructures with OCT in glaucoma concludes that inner plexiform layer is currently the best macular parameter for glaucoma diagnosis, as inner nuclear layer and outer retinal thinning in glaucoma is negligible compared to normal eyes.[25] Therefore, a more accurate measurement is required to detect the minor changes in sub-regions of retina. OCTA allows measurements of vessel density and flow index in 6 areas: foveal, perifoveal, superior, inferior, temporal and nasal areas. Here we analysed GCC and macular vessel density, which showed significant correlation between these two parameters in both whole area and hemifield analyses. Although it has been demonstrated that macular vessel area density decreases at a rate of 0.4% per year with aging,[26] the reduction of macular vessel density was also significantly related to glaucoma. Our data has shown that the decreasing rate of macular vessel density in glaucoma was 9.7% and 13.3% more in glaucoma eyes than normal in age group of 66-75 and above 75, respectively. The finding indicates that macular vessel density significantly decreases in glaucoma despite age as a factor, which suggests that macular vessel density may be an important diagnostic parameter of glaucoma.

There are limitations to this study. Normalised data with much larger group of individuals and ethnicity subgroups are required to discriminate normal subjects from glaucoma patients. Here in this study, we attempted to recruit equal numbers for each group but recognise that a smaller number of normal participants were recruited than glaucoma as whole, and therefore there is a potential bias in interpretation. Longitudinal study will be required to provide more information of how macular vessel density changes in glaucoma progression and aging population with OCTA. When performing OCTA for participants, a fixed gaze at a certain angle for dozens of seconds is required, which remains a challenge for those with poor vision. Therefore, the image quality was suboptimal in clarity compared to the majority.

In conclusion, our study demonstrates that OCTA can provide objective and quantitative measurements to detect vascular defects of the macula in POAG patients. There is significant difference between normal and POAG eyes in all parameters measured, including RNFL thickness, GCC thickness and macular vessel thickness. Visual field MD significantly

correlates with the reduction of GCC, RNFL and macular vessel density in POAG in accordance to the severity of glaucoma. Macular vessel density significantly reduces in glaucoma despite correction for age, and correlates with reduction of RNFL and GCC. Therefore, OCTA can detect microstructural defects, which make it a new tool for facilitating the diagnosis, staging and estimated visual function in glaucoma.

Declaration of interest

The authors report no conflicts of interest. The authors alone are responsible for the contest and writing of the paper.

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Ethics approval and consent to participate: The study protocol and ethics were approved by South West Frenchay Research Ethics Committee and Health Research Authority in England (Ref: 16/SW/0277).

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Table 1 Characteristics of the study subjects

	Normal (40 eyes, 21 patients)	Glaucoma (108 eyes, 60 patients)	*P* Value
Age (y)	68.50 ± 1.317	71.06 ± 0.9452	0.15 [a]
Gender, n (Male/Female)	8/13	37/23	0.06 [b]
VF			
MD (dB)	-1.862 ± 0.4253 N=33	-8.620 ± 0.7161 N=108	< 0.0001 [a]
PSD (dB)	1.826 ± 0.1131 N=33	6.565 ± 0.3713 N=108	< 0.0001 [a]
Max IOP (mmHg)	17.93 ± 0.7324 N=40	28.72 ± 0.7988 N=96	< 0.0001 [a]
Diabetics	3/21	5/60	0.43 [b]
Topical glaucoma medication, N			
0	null	39	
1	null	29	
>1	null	48	
Glaucoma surgery history, N	null	50	

dB = decibels; IOP = intraocular pressure; MD= mean deviation; PSD = pattern standard deviation VF = visual field.

[a] Statistical significance tested with Student's *t* test.

[b] Statistical significance tested with the Chi-square test

Table 2 Results of diagnositc measurements

Variables	Normal (40 eyes)	Mild (36 eyes)	Moderate (37 eyes)	Advance (35 eyes)	*P* Value [a]
VF					
MD (dB)	-1.326 ± 0.2714	-2.372 ± 0.2674	-8.164 ± 0.3500	-19.43 ± 1.029	< 0.0001
PSD (dB)	1.826 ± 0.1131	3.288 ± 0.3186	7.881 ± 0.5374	10.10 ± 0.4492	< 0.0001
Structural measurements					
Whole RNFL thickness (μm)	98.52 ± 2.255	81.62 ± 2.294	77.47 ± 5.864	73.24 ± 4.005	< 0.0001
Superior RNFL thickness (μm)	99.29 ± 2.478	82.92 ± 2.914	76.53 ± 6.594	75.37 ± 4.368	< 0.0001
Inferior RNFL thickness (μm)	97.67 ± 2.598	80.32 ± 2.275	78.40 ± 5.900	71.11 ± 3.913	< 0.0001
Whole GCC thickness (μm)	92.25 ± 2.452	84.62 ± 2.466	74.87 ± 1.987	72.14 ± 2.586	< 0.0001
Superior GCC thickness (μm)	92.93 ± 2.804	84.81 ± 2.252	72.23 ± 1.883	74.80 ± 3.015	< 0.0001
Inferior GCC thickness (μm)	91.58 ± 2.269	84.44 ± 3.024	77.52 ± 3.249	69.48 ± 2.496	< 0.0001
Optic disc					
Rim area (mm^2)	1.478 ± 0.1396	0.9974 ± 0.05734	0.9060 ± 0.1388	0.7928 ± 0.09938	< 0.0001
R/D area ratio	0.6750 ± 0.04002	0.5338 ± 0.03742	0.4066 ± 0.05839	0.3373 ± 0.04488	< 0.0001
Macular vessel density (%)					
Whole	50.04 ± 0.7950	48.22 ± 0.7811	45.87 ± 1.052	44.54 ± 0.9713	< 0.0001
Superior	52.12 ± 0.7632	50.83 ± 0.7639	48.15 ± 1.419	46.81 ± 0.9984	< 0.0001
Inferior	51.99 ± 0.8205	50.85 ± 0.8758	48.64 ± 1.255	46.68 ± 0.8899	< 0.0001

GCC = ganglion cell complex; MD = mean deviation; PSD = pattern standard deviation; R/D = rim area/disc; RNFL = retinal nerve fiber layer; VF = visual field. Numbers displayed are mean ± SEM.

[a] Differences between groups were tested with Kruskal-Wallis test with Dunnett's correction.

Table 3 Correlation between macular vessel density and RNFL and GCC

Parameters	Macular Vessel Density (%) *			
	Normal		Glaucoma	
RNFL (μm)				
Whole	p = 0.5998	r = 0.1215	p = 0.0085	r = 0.3681
Superior	p = 0.9378	r = -0.0182	p = 0.0063	r = 0.3742
Inferior	p = 0.4018	r = 0.1931	p = 0.0628	r = 0.2599
GCC (μm)				
Whole	p = 0.6270	r = 0.1097	p = 0.0257	r = 0.3092
Superior	p = 0.1799	r = 0.2967	p = 0.0995	r = 0.2265
Inferior	p = 0.9992	r = 0.0002	p = 0.0247	r = 0.3055

RNFL = retinal nerve fiber layer; GCC = ganglion cell complex.
* Correlation between parameters was tested with Pearson correlation test.

Table 4 Correlation between age and other variables in healthy and POAG subjects

Parameters	Age *			
	Normal		Glaucoma	
GCC	$p = 0.5$	r = -0.1	$p = 0.7$	r = -0.05
macular vessel density	$p = 0.008$	r = -0.41	$p < 0.0001$	r = -0.48
RNFL	$p = 0.68$	r = -0.07	p = 0.1	r = -0.21

GCC = ganglion cell complex; RNFL = retinal nerve fiber layer.

* Correlation between parameters was tested with Pearson correlation test.

Table 5 The decreasing rate of macular vessel density

Age	average macular VD (μm)		P Value [a/*]	decreasing rate (%)	
	Normal (N)	glaucoma (N)		Normal	glaucoma
≤ 65	53.11 (13)	50.93 (30)	0.04		
66-75	48.86 (17)	45.98 (41)	0.03	7.90%	9.70%
>75	48.08 (10)	44.17 (37)	0.002	9.40%	13.30%

VD = vessel density; N = n number

[a] Statistical significance tested with Student's *t* test.

* The comparisons are for the average macular vessel density.

LEGENDS

Figure 1: **Progressive structural and perfusional changes are seen in the macula of glaucomatous eyes.** (A) These are representative images of 3 x 3 mm *en face* angiograms generated by OCTA. Images in the first column are retinal thickness maps. The corresponding OCT sections of retina are in the second column, with heat maps of retina in the third column. (B-C) The quantitative graphs demonstrate that a significant reduction of GCC and macular vessel density was observed in glaucoma eyes compared to normal eyes. Comparison performed using Kruskal-Wallis test with Dunn's correction. * $p < 0.05$, *** $p < 0.0001$.

Figure 2: **Reduced RNFL thickness is seen at the optic disc in glaucoma.** (A) Retinal nerve fibre layer density images are in the first and third column. The other images are corresponding OCT sections of the RNFL. (B) There is significant reduction of RNFL in glaucomatous eyes compared to normal eyes. Comparison was performed by using Kruskal-Wallis test with Dunn's correction. **$p < 0.001$, *** $p < 0.0001$.

Figure 3: **OCTA measurements correlate with visual function in glaucoma**. The correlation plots demonstrate that visual field MD significantly correlate with GCC, RNFL and macular vessel density in glaucoma eyes. Comparison was performed using univariate analysis with the Pearson correlation test.