RETINAL NERVE FIBRE LAYER THICKNESS IN A NORMAL BLACK SOUTH AFRICAN POPULATION

Dr Sarah Ismail

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

A research report submitted to the Faculty of Health Sciences, University of the Witwatersrand, in partial fulfilment of the requirements for the degree of Master of Medicine in Ophthalmology.

Supervisor:

Dr HD Alli FCOphth (SA) MMed MBBCh

Co Supervisor:

Dr N Ally FCOphth (SA) MMed MBBCh

Department of Neurosciences, Division of Ophthalmology

University of the Witwatersrand, South Africa

Johannesburg

17 May 2020

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

CONTENTS

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

DECLARATION
DEPARTMENT OF NEUROSCIENCES

Neurology, Neurological Surgery, Ophthalmology,

Otorhinolaryngology

School of Clinical Medicine, Faculty of Health Sciences,

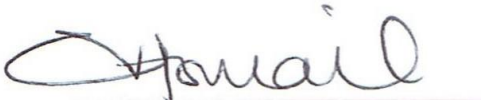
7 York Road, Johannesburg 2193, South Africa

Tel: +27 11 717-2774 · Fax: +27 11 717 2775

Plagiarism declaration for written work

I … **Sarah Ismail** … as a postgraduate student registered for an MMed at the University of the Witwatersrand declare the following:

- I am aware that plagiarism is the use of someone else's work without their permission and or without acknowledging the original source.
- I am aware plagiarism is wrong.
- I confirm that this written work is my own work except where I have stated otherwise.
- I have followed the required conventions in referencing the thoughts and ideas of others.
- I understand that the University of the Witwatersrand may take disciplinary action against me if there is a belief that this is not my own unaided work or if I have failed to acknowledge the ideas or writing of others.

Signature 

Date … **17-05-2020**…

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

CO- AUTHORS

University of the Witwatersrand

Department of Neurosciences

Division of Ophthalmology

11/01/2020

To Whom It May Concern

We, the undersigned, confirm that the article "***Retinal Nerve Fibre Layer Thickness in a Black South African Population"*** published in the journal Eye is the original work of Dr Sarah Ismail. She is the first author and was the principal investigator of the study. She actively conceived, designed, executed, analysed and wrote up all aspects of the article. We give consent for the article to be used for the purposes of her MMed degree.

Yours faithfully

Dr Hassan D. Alli

FCOphth (SA) MMed MBBCh

Dr Naseer Ally

FCOphth (SA) MMed MBBCh

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

ACKNOWLEDGEMENTS

All Gratitude to The Almighty for Everything

My Parents; For all the sacrifice past and present, support, love and care

My Siblings; For always being there and looking out for me

My daughter; My light in the dark

My husband; For the unwavering support, encouragement, sacrifice and love

This would not have been possible without the participants in the study, my supervisor and co- supervisor.

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

PUBLISHED ARTICLE

PDF re-print of article

https://doi.org/10.1038/s41433-019-0677-7

Eye
https://doi.org/10.1038/s41433-019-0677-7

The ROYAL COLLEGE of OPHTHALMOLOGISTS

ARTICLE

Retinal nerve fibre layer thickness in a normal black South African population

Sarah Ismail[1] · Naseer Ally[1] · Hassan Dawood Alli[1]

Received: 23 June 2019 / Accepted: 24 October 2019

Abstract

Background The measurement of retinal nerve fibre layer (RNFL) thickness on spectral domain OCT (SD-OCT) are compared with built-in age— and gender—matched European normative databases and this difference is used to assist with glaucoma diagnosis. However, there are differences in RNFL thickness between population groups. Therefore, using the built in European normative database as a comparison across all population groups could lead to erroneous results, due to the basic assumption that the normative values for non-European populations are the same as their European counterparts.

Methods Cross-sectional study of RNFL thickness in normal black South African patients.

Results One hundred and thirty-two eyes of 132 patients were enroled in this study. The mean (SD) age of patients in this study was 41.3 (12.5) years. Males comprised 40.9% ($n = 54$; $p = 0.0367$). All RNFL sectors except the temporal sector were significantly thicker than the reference database. The RNFL sectors measured as follows: global (108.7 µm, $p < 0.001$), superotemporal (152.4 µm, $p < 0.001$), superonasal (132.6 µm, $p < 0.001$), inferotemporal (150.1 µm, $p < 0.001$), inferonasal (129.2 µm, $p < 0.001$), nasal (77.7 µm, $p < 0.001$), temporal 74.8 µm, $p = 0.9534$).

Conclusion The RNFL thickness of normal black South Africans is significantly thicker than that of the European database on the Spectralis SD-OCT. This needs to be taken into account when performing RNFL thickness measurements on black patients.

Introduction

Worldwide, glaucoma is known to be the leading cause of irreversible blindness with the number of people above 40 years of age living with the disease projected to increase from 76 million in 2020 to 111.8 million in 2040 [1]. In African patients, glaucoma is 4–5 times more prevalent than Europeans, occurs ~10 years earlier and is more aggressive [2, 3]. Multiple imaging modalities are used to detect early structural glaucomatous damage.

Technological advances in imaging quality and accuracy have resulted in older modalities (Heidelberg Retinal Tomogram and Time Domain Optical Coherence Tomography) becoming obsolete and newer modalities such as Spectral Domain Optical Coherence Tomography (SD-OCT) and Optical Coherence Tomography Angiography (OCTA) being at the forefront in detecting pre-perimetric glaucoma. SD-OCT can be used in several ways to assist in glaucoma diagnosis; such as measurements of peripapillary retinal nerve fibre layer (RNFL) thickness, macular thickness and measurement of Bruch's-Membrane Opening-Minimum Rim Width (BMO-MRW) [4].

The measurement of RNFL thickness on SD-OCT are compared to built-in age— and gender—matched European normative databases and this difference is used to assist with glaucoma diagnosis [5]. However, there are differences in RNFL thickness between population groups [6–11]. Therefore, using the built in European normative database as a comparison across all population groups could lead to erroneous results, due to the basic assumption that the normative values for non-European populations are the same as their European counterparts. In addition, these databases are also machine-specific and not universally applicable between different types of SD-OCT machines (e.g. Spectralis™, Cirrus™, OptoVue™,Topcon™) [12–15].

Studies using the Spectralis SD-OCT (Heidelberg Engineering, Germany) to ascertain the normal RNFL thickness compared to the built-in European normative database, have been conducted in numerous countries including Germany,

✉ Naseer Ally
naseerally@gmail.com

[1] Department of Neurosciences, Division of Ophthalmology, University of the Witwatersrand, Johannesburg, South Africa

Published online: 13 November 2019

India, Brazil, Nepal and in the French Alienor population [6, 8–10, 16]. These studies have shown a thicker RNFL in non-European populations [8–10]. In two Nigerian popu- lations and South Africa, a thicker RNFL has been shown using the Stratus™ OCT and the Ivue-100™ SD-OCT machines respectively [11, 17–20].

The aim of this study was to compare the normal retinal nerve fibre layer thickness in a healthy black population to the European normative database on the Spectralis SD-OCT (Heidelberg engineering). To the authors knowledge, this is the first study conducted in Africa using the Spectralis SD-OCT (Heidelberg engineering).

Materials and method

We conducted a cross-sectional study at St John Eye Hospital – Chris Hani Baragwanath Academic Hospital in Soweto, Johannesburg, South Africa, from January 2017 to December 2018. The Human Research Ethics Committee of the University of the Witwatersrand, Johannesburg, gave ethical clearance for this study to be performed. The study adhered to the tenets of the Declaration of Helsinki. Staff, patients and their family members were invited to participate in the study. A convenient sample was taken from the outpatients' department at St John Eye Hospital. Each participant in the study gave written informed consent. Participants who met the following inclusion criteria were enroled in the study:

- Self-reported identity of black South African ethnicity,
- 18 years or older,
- Uncorrected Snellen visual acuity of 6/9 or better in the study eye,
- Normal intraocular pressure (10–21 mmHg) with either Perkins or Goldmann applanation tonometry,
- Clinically normal optic disc,
- Absence of other fundus pathologies,
- Fovea to Disc (FoDi) alignment within 7°.

Exclusion criteria included patients with:

- Systemic diseases such as diabetes mellitus and neurological disease with optic disc abnormalities.
- Ocular conditions such as glaucoma, tilted discs, peripapillary atrophy, clinically significant vitreal, retinal or choroidal diseases; history of cataract or posterior segment surgery and laser procedures; prior or current history of eye trauma or uveitis; and grade three and four hypertensive changes were not included in the study.
- Axial length >26.0 mm.

The participants' demographic (age and gender) and medical history were documented. A Snellen visual acuity measurement at 6 m; slit lamp examination; intra-ocular pressure measurement; and dilated fundoscopy (to assess cup:disc (C:D) ratio and exclude other pathologies) were performed on each participant. The Nidek optical AL Scan (Nidek co., Ltd, Japan) was used to measure the axial length. Only one normal eye per participant was included in the study.

The retinal nerve fibre layer and vertical disc height was measured using the Spectralis SD-OCT, Heidelberg eye explorer version 1.10.15.0, (Heidelberg Engineering, Germany). When performing the scan, the disc was identified with the aid of the built-in eye fixating system. The image was then tracked ensuring it remained within the image frame while maintaining a good quality scan. The scanning circle was manually placed over the optic nerve head and the images acquired. The participant values of each of the seven sectors (global, superotemporal, inferotemporal, superonasal, inferonasal, temporal, nasal) of the RNFL thickness were generated by the built-in software of the Heidelberg engineering. The participant values were displayed alongside the values of the European database which are automatically age- and gender-matched. Each sector is considered either "normal", "borderline", or "outside normal limits", if thicker than 5th centile, thinner than the 5th centile, or thinner than the 1st centile respectively, when compared to the European database of that sector [5]. Only good quality OCT scans (≥21 dB) and scans with the Fovea to Disc (FoDi) alignment within 7°, that were normal as defined by the age and gender matched European database, were included in the study. The reference value and participant results for each sector, as well as the quality of the scan and degree of cyclotorsion were documented. Patients whose FoDi alignment was outside 7° on either side of the fovea, were excluded from the study. Vertical disc height (VDH) was measured manually by and agreed upon by two of the investigators (SI and NA). Data collected was captured on the REDCap database. Data was then exported to STATA 15.1 (STATACorp, USA) and analysed.

Statistical analysis

A sample size of hundred and twenty-eight patients was calculated using a one-sample mean test with a power of 80% and an α-level of 0.05 to detect a difference of 2.5 µm (SD = 10) between the measured and reference values. The unpaired *t*-test was used to compare the mean RNFL thickness in the study population to the reference value per sector of the retina measured. A one sample proportion test was used to test the proportions of males and females within the study population. Univariate and multivariate

Table 1 Comparison of RNFL thickness (μm) in the study population vs reference database

RNFL sector	Mean RNFL thickness (SD) (μm) [95% CI]	Mean reference RNFL thickness (SD) (μm) (95% CI)	*p* value[a]
Global	108.7 (10.7) [106.9–110.6]	97.1 (1.0) [96.9–97.2]	<0.001
Superotemporal	152.4 (20.3) [148.9–155.9]	134.6 (2.1) [134.3–135.0]	<0.001
Superonasal	132.6 (23.9) [128.5–136.7]	102	<0.001
Inferotemporal	150.1 (19.7) [146.7–153.5]	142.5 (2.8) [141.9–143.0]	<0.001
Inferonasal	129.2 (27.9) [124.4–134.0]	105.9 (1.1) [105.7–106.0]	<0.001
Nasal	77.7 (14.6) [75.2–80.2]	72	<0.001
Temporal	74.8 (10.3) [73.0–76.5]	74.7 (1.9) [73.9–75.6]	0.9534

[a]Unpaired *t*-test (Bonferroni-corrected *p* value = 0.0024)

linear regression models were used to assess the change in RNFL thickness with increasing age. The univariate and multivariate equations are shown below:

Univariate equation: $E(RNFL) = ß0 + ß1 \cdot age$
Multivariate equation: $E(RNFL) = ß0 + ß1 \cdot age + ß2 \cdot sex + ß3 \cdot vdh$

Suitability for using a linear regression model was checked using the residual versus fitted plots. Since a multitude of statistical tests were performed on the same dataset, a Bonferroni correction was applied, and statistical significance was set at a *p* value of 0.0024.

Table 2 Comparison between the 1st and 5th percentile of the Heidelberg database reference values [5] (45 years) and our study patients

Sector	1st percentile[a] [μm]	1st percentile[b] [μm]	5th percentile[a] [μm]	5th percentile[b] [μm]
Global	87	76.0	93	82.1
Temporal	57	46.9	60	54.9
Superotemporal	111	96.3	119	107.4
Inferotemporal	117	99.1	122	111.6
Nasal	50	38.3	56	48.1
Superonasal	81	57.8	97	70.7
Inferonasal	84	53.6	90	68.8

[a]Study values

[b]Heidelberg database values

Results

One hundred and thirty-two patients were enroled in this study. The mean(SD) age of patients in this study was 41.3 (12.5). The age range of patients in our study was 19–74 years. Males comprised 40.9% ($n = 54$; $p = 0.0367$) of patients in this study. The mean RNFL thickness in all sectors was significantly thicker when compared with the European reference RNFL thickness (Table 1). (Where no reference range is given in a reference RNFL sector, the Spectralis OCT machine gave a single value irrespective of gender and age). The mean (SD) [95% CI] axial length and VDH were 23.5 (0.76) mm [23.3–23.6] and 1881.8 (185.2) μm [1849.9–1913.6] respectively.

In our study population the 1st centile was thicker than the Heidelberg database 5th centile (Table 2). The univariate and multivariate regression co-efficients showed the decrease in RNFL thickness per decade increase in age (Table 3, Figs. 1, 2).

Discussion

Our study shows that the RNFL in the black South African population is significantly thicker than that of the European normative database on the Spectralis SD-OCT (Heidelberg Engineering). The significance was maintained after applying a Bonferroni correction to the data. Although used primarily for glaucoma diagnosis, the RNFL thickness also has implications for the diagnosis of other neurological diseases such as multiple sclerosis and Alzheimer's disease [21]. In our study, the significant mean thickness difference, ranges from 5 μm in the nasal sector to 30 μm in the superonasal sector, with the higher differences being more clinically significant when used for glaucoma diagnosis. The Spectralis SD-OCT displays either a normal RNFL thickness; or "borderline" if it is thinner than the 5th percentile; or "outside normal limits" if it is thinner than the 1st percentile [5]. The Spectralis SD-OCT gives normative values at age 45 and 60 years of age [5]. Using the Spectralis normative database values at age 45 years (mean age of our study population is 41.3 years), this difference results in the 1st percentile for our study eyes being thicker than the 5th percentile in the database (Table 3). This means that a patient who has structural damage that should be flagged as "outside normal limits" by the machine, will display as normal using the current normative database. We are potentially underdiagnosing a large proportion of patients with glaucoma.

The global RNFL thickness in our study population (108.7 μm) is thicker than those of other populations measured using the Spectralis SD-OCT [6, 8–10, 16]. The global reference on the Spectralis SD-OCT, which is based on the

Table 3 Change in RNFL thickness (µm) per decade increase in age

RNFL sector	Unadjusted[a] RNFL change per decade (95% CI)	*p* value	Adjusted[b] RNFL change per decade (95% CI)	*p* value
Global	−3.5 (−4.9 to −2.2)	<0.001	−3.5 (−4.8 to −2.1)	<0.001
Superotemporal	−5.2 (−7.9 to −2.5)	<0.001	−4.9 (−7.6 to −2.1)	0.001
Superonasal	−5.8 (−8.9 to −2.6)	<0.001	−5.8 (−9.0 to −2.6)	<0.001
Inferotemporal	−4.7 (−7.3 to −2.1)	<0.001	−4.5 (−7.1 to −1.8)	0.001
Inferonasal	−3.4 (−7.2 to −0.3)	0.079	−3.3 (−7.1 to 0.4)	0.080
Nasal	−2.0 (−4.0 to −0.02)	0.047	−2.4 (−4.4 to −0.3)	0.046
Temporal	−2.5 (−3.8 to −1.1)	<0.001	−2.3 (−3.7 to −0.9)	0.002

[a]Univariate linear regression (Bonferroni-corrected *p* value = 0.0024) [b]Multivariate linear regression (Bonferroni-corrected *p* value=0.0024)

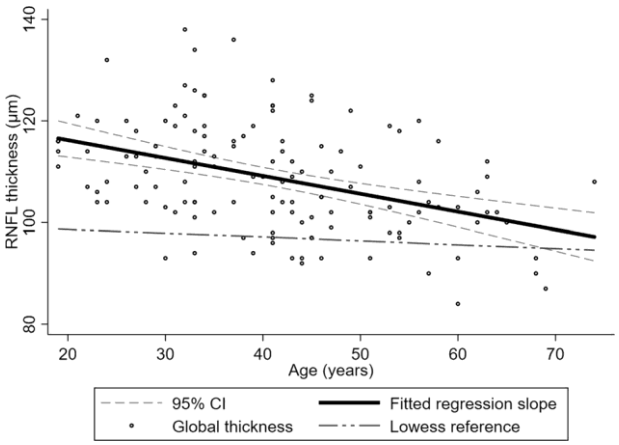


Fig. 1 Composite graph of fitted regression slope with 95% confidence interval for global RNFL thickness vs age (thick black line straddled by two dashed lines), scatterplot of measured global RNFL thickness datapoints (hollow circles), and LOWESS curve of the reference database values (dash-dots line)

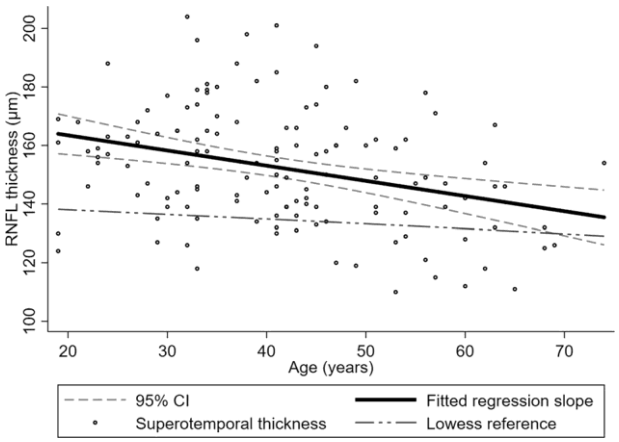


Fig. 2 Composite graph of fitted regression slope with 95% confidence interval for superotemporal RNFL thickness vs age (thick black line straddled by two dashed lines), scatterplot of measured superotemporal RNFL thickness datapoints (hollow circles), and LOWESS curve of the reference database values (dash-dots line)

European database is 97.1 µm. In Indian, Nepalese and Brazilian populations, the global RNFL thickness measured 101.43 , 102.64 and 102/103 µm respectively [8–10]. This racial difference between the European and non-European RNFL thickness is therefore not unique to African patients. Bendschneider et al. demonstrated a global RNFL thickness in a German population of 97.2 µm which is similar to the Heidelberg normative global mean thickness [6]. One of the few studies that used the Spectralis SD-OCT to measure the RNFL in a black population was conducted by Alasil et al. [7]. They measured RNFL thickness of various ethnicities in an American population [7]. Of those enroled, only 26 patients were of African-American descent and the global RNFL measured with the Spectralis SD-OCT was 99 µm which is thinner than our population findings. The study consisted of 125 Caucasian patients and no significant difference was found between the RNFL thickness of the African-American and Caucasian population groups [7]. The French Alienor study measured RNFL thickness in elderly patients with a mean age of 81 and showed a global RNFL thickness of 91 µm [16]. The mean age difference between the French Alienor study population (81 years) and ours (41.3 years), probably accounts for the large differences between mean RNFL thickness.

RNFL thickness can be influenced by factors other than the amount of nerve fibre layer tissue present in the peripapillary area, such as, vascular tissue as documented by OCTA [22]. The difference between the African RNFL thickness and European thickness could therefore be due to anatomical differences in blood vessel volume between these patients. Similarly, patients with larger disc areas have been shown to have a thicker RNFL [23]. Even though our study did not look at disc area, we have adjusted for vertical disc height in our multivariate analysis and we found it to be significant in predicting RNFL decline per decade. Black patients have also been shown to have larger disc areas when compared to their European counterparts [24]. This difference may also account for the thicker RNFL in our study.

The mean global RNFL thickness in two different Nigerian population groups conducted with the Stratus OCT were 104.1 and 107.1 µm respectively [17, 18]. Other South African studies also yielded mean values of 108–110 µm

[11, 19]. However, even though the absolute values are similar to those in our study, different OCT machines were used and thus are not directly comparable [15].

Our study consisted of a wide range of patient ages and we were able to model univariate and multivariate linear regression models. After adjusting for age, gender and VDH in the multivariate linear regression analysis, the rate of decline in global RNFL thickness in our population is −3.9 µm per decade which is higher than that of other comparable studies. We also found that although age and VDH was a significant predictor in the multivariate model, gender was not a significant predictor. The rate of decline per decade in the Indian population was −1.5 µm and the Nepalese population was −2.26 µm [9, 10]. The rate of decline in the German population was −1.9 µm per decade [6]. Only the French Alienor study had a higher rate of decline of −5.9 µm but this is probably due to the older age of the participants. The higher rate of RNFL loss among our study patients' needs to be investigated further using longitudinal studies. Since glaucoma causes accel- erated thinning of the RNFL, this high rate of loss in normal patients is probably even higher in glaucoma patients and may be the reason for increased severity of disease among African patients.

Bowd et al. recently published a prospective longitudinal study, which included participants enroled in the ADAGES and DIGS trials, comparing the RNFL thickness and BMO-MRW in African and European patients in 3 categories (Healthy eyes, glaucoma suspect, and glaucomatous eyes) [25]. Only 27 of the 124 healthy eyes studied were from patients of African descent who had a mean age of 64 years. The global RNFL thickness measured by Spectralis SD-OCT in these patients was 96.1 µm and is much lower than our African population studied. This is probably due to the older age group of the participants. The rate of decline over 3 years was measured to be −5.1 µm [25]. This is a higher rate of decline compared with our population.

A thicker RNFL has been associated with a faster rate of decline in RNFL thickness [26]. After adjusting for age and sex in the multivariate regression model, the thicker quadrants (superonasal, inferotemporal, superotemporal) were noted to have the largest rates of decline in RNFL thickness. Thus, the larger rate of decline per decade could be attributed to the thicker baseline RNFL in our population group.

The limitations of the study are that this was a hospital-based, cross-sectional study as opposed to a population-based longitudinal cohort study. There may be inherent bias in recruiting hospital patients for a normative database. However, the difference in the superior and inferior quadrants compared to the normative database are both statistically and clinically significant. BMO-MRW was not measured in this study and could be done in future studies in African patients. BMO-MRW requires more cooperation from the patient and takes longer to obtain than RNFL. Reznicek et al. showed that RNFL and BMO-MRW are both suitable for the diagnosis of glaucoma, however, BMO-MRW may be more suitable in myopic patients [27] Further longitudinal studies of African patients need to be conducted to corroborate these results.

Conclusion

RNFL thickness in a black South African population, measured by Spectralis SD-OCT is both clinically and statistically significantly thicker than the European database. The difference in the 1st and 5th percentiles of our study and the built-in database are large enough when compared, to warrant requiring African-specific databases to avoid underdiagnosis of glaucoma and other diseases causing thinning of the RNFL. The rate of decline in RNFL thickness per decade increase in age, is greater than other comparable studies. Further, multicentre, multiregional, longitudinal population-based studies are however required to confirm these findings in black patients.

Summary

What was known before

- No studies of black African population using Spectralis SD-OCT.
- African-American subjects were shown to have no difference in RNFL thickness.
- Studies on other race groups e.g. Nepalese showed a thicker RNFL compared to Caucasian counterparts.

What this study adds

- The black South African population have a significantly thicker RNFL.
- This can result in glaucoma being missed.
- There is also a higher rate of decline in RNFL thickness of the black South African population.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

References

1. Tham Y-C, et al. Global prevalence of glaucoma and projections of glaucoma burden through 2040. Ophthalmology. 2014;121: 2081–90.
2. Sample PA, et al. The African Descent and Glaucoma Evaluation Study (ADAGES): design and baseline data. Arch Ophthalmol. 2009;127:1136–45.
3. Tielsch JM, et al. Racial variations in the prevalence of primary open-angle glaucoma: the Baltimore Eye Survey. JAMA. 1991;266:369–74.
4. Tao Y-L, et al. Parameters of ocular fundus on spectral-domain optical coherence tomography for glaucoma diagnosis. Int J Ophthalmol. 2017;10:982–91.
5. Downloads—SPECTRALIS OCT Angiography Module - Non-invasive imaging | Heidelberg Engineering. https://business-lounge.heidelbergengineering.com/za/en/products/spectralis/oct-angiography-module/downloads/#downloads. Accessed 30 Mar 2018.
6. Bendschneider D, et al. Retinal nerve fiber layer thickness in normals measured by spectral domain OCT. J Glaucoma. 2010; 19:475–82.
7. Alasil T, et al. Analysis of normal retinal nerve fiber layer thickness by age, sex, and race using spectral domain optical coherence tomography. J Glaucoma. 2013;22:532–41.
8. Zangalli CES, Reis ASC, Vianna JR, Vasconcellos JPC, Costa VP. Interocular asymmetry of minimum rim width and retinal nerve fiber layer thickness in healthy Brazilian individuals. J Glaucoma. 2018;27:1136–41.
9. Appukuttan B, Giridhar A, Gopalakrishnan M, Sivaprasad S. Normative spectral domain optical coherence tomography data on macular and retinal nerve fiber layer thickness in Indians. Indian J Ophthalmol 2014;62:316–21.
10. Thapa M, Khanal S, Shrestha GB, Sharma AK. Retinal nerve fibre layer thickness in a healthy Nepalese population by spectral domain optical coherence tomography. Nepal J Ophthalmol. 2014;6:131–9.
11. Mashige KP, Oduntan OA. Retinal nerve fibre layer thickness values and their associations with ocular and systemic parameters in Black South Africans. Afr Health Sci. 2016;16:1188–94.
12. Savini G, Carbonelli M, Barboni P. Retinal nerve fiber layer thickness measurement by Fourier-domain optical coherence tomography: a comparison between Cirrus-HD OCT and RTVue in healthy eyes. J Glaucoma. 2010;19:369–72.
13. Seibold LK, Mandava N, Kahook MY. Comparison of retinal nerve fiber layer thickness in normal eyes using time-domain and spectral-domain optical coherence tomography. Am J Ophthalmol 2010;150:807–14.
14. Knight OJ, Chang RT, Feuer WJ, Budenz DL. Comparison of retinal nerve fiber layer measurements using time domain and spectral domain optical coherent tomography. Ophthalmology. 2009;116:1271–7.
15. Arthur SN, et al. Reproducibility and agreement in evaluating retinal nerve fibre layer thickness between Stratus and Spectralis OCT. Eye. 2011;25:192–200.
16. Rougier M-B, et al. Retinal nerve fibre layer thickness measured with SD-OCT in a population-based study of French elderly subjects: the Alienor study. Acta Ophthalmol. 2015;93:539–45.
17. Sani RY, Abdu L, Pam V. Retinal nerve fiber layer thickness measurements of normal Northern Nigerian adults using optical coherence tomography. Ann Afr Med. 2016;15:52–57.
18. Mahmud-Ajeigbe. Retinal nerve fibre layer measurements in normal eyes in Zaria using optical coherence tomography. http://www.ssajm.org/article.asp?issn=2384-5147;year=2015;volume=2;issue=2;spage=85;epage=88;aulast=Mahmud-Ajeigbe. Accessed 29 Jan 2019.
19. Murugan, C et al. Retinal nerve fibre layer thickness of black and Indian myopic students at the University of KwaZulu-Natal. Afr. Vis. Eye Health. 2015;74:1-6.
20. Pathai, S et al. Retinal nerve fibre layer thickness and contrast sensitivity in HIV-infected individuals in South Africa: a case-control study. PLoS ONE. 2013;8:e73694.
21. Gupta S, Zivadinov R, Ramanathan M, Weinstock-Guttman B. Optical coherence tomography and neurodegeneration: are eyes the windows to the brain? Exp Rev Neurother 2016;16: 765–75.
22. Allegrini D, et al. The volume of peripapillary vessels within the retinal nerve fibre layer: an optical coherence tomography angiography study of normal subjects. Br J Ophthalmol 2018;102:611–21.
23. Gür Güngör S, Akman A, Küçüködük A, Çolak M. Retinal nerve fiber layer thicknesses in three different optic nerve head size groups measured by cirrus spectral domain optical coherence tomography. Turk J Ophthalmol. 2016;46:58–61.
24. Oliveira C, et al. Axial length and optic disc size in normal eyes. Br J Ophthalmol 2007;91:37–39.
25. Bowd C, et al. Racial differences in rate of change of spectral-domain optical coherence tomography-measured minimum rim width and retinal nerve fiber layer thickness. Am J Ophthalmol 2018;196:154–64.
26. Leung CKS, et al. Retinal nerve fiber layer imaging with spectral-domain optical coherence tomography: a prospective analysis of age-related loss. Ophthalmology. 2012;119:731–7.
27. Reznicek L, et al. Structure-function relationship comparison between retinal nerve fibre layer and Bruch's membrane opening-minimum rim width in glaucoma. Int J Ophthalmol. 2017;10: 1534–8.

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

TURNIT IN REPORT

University of the Witwatersrand

Department of Neurosciences

Division of Ophthalmology

20/01/2020

To Whom It May Concern

The following attachment is the turnit in report submitted after the Publication of the article. The high similarity index as indicated is in line with this same publication having been submitted by the same authors to *Eye*, a nature journal.

Yours faithfully

Dr S Ismail

MBBCh (Wits) DA (SA)

Dr Hassan D. Alli

FCOphth (SA) MMed MBBCh

Dr Naseer Ally

FCOphth (SA) MMed MBBCh

0400847j:pdf_reprint_of_paper.pdf

by Sarah Ismail

Submission date: 18-Feb-2020 09:15PM (UTC+0200)
Submission ID: 1259667339
File name: ts_04e4bf66-6729-4087-bd8b-dda1c0e6fcb4_pdf_reprint_of_paper.pdf (567.72K)
Word count: 4392
Character count: 22961

0400847j:pdf_reprint_of_paper.pdf

ORIGINALITY REPORT

90%	83%	90%	13%
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

PRIMARY SOURCES

1	Sarah Ismail, Naseer Ally, Hassan Dawood Alli. "Retinal nerve fibre layer thickness in a normal black South African population", Eye, 2019 Publication	82%
2	www.nature.com Internet Source	7%
3	Submitted to University of Exeter Student Paper	<1%

Exclude quotes	On	Exclude matches	< 5 words
Exclude bibliography	On		

UNIVERSITY OF THE
WITWATERSRAND,
JOHANNESBURG

APPENDIX

Research Protocol and Ethics Clearance

Retinal Nerve Fibre Layer Thickness in a normal black South African population

Dr. S. Ismail

Registrar

Student Number: 0400847J

Department of Neurosciences

Division: Ophthalmology

University of the Witwatersrand

November 2017

Supervisor: Dr H. D. Alli

CONTENT

1. Introduction

Glaucoma is a progressive optic neuropathy that leads to a gradual decrease in visual field, starting from the periphery.[1] It is the leading cause of irreversible blindness worldwide. The global burden of disease is projected to increase from 60,5 million people in 2010, to 79,6 million people by 2020.[1] Africa has the highest burden of glaucoma as well as primary open-angle glaucoma (POAG).[1,2] Glaucoma in Africans is more aggressive and has an earlier age of onset when compared to Europeans.[3] The diagnosis of glaucoma is complex and involves both subjective and objective assessments of the eye. Clinical examination demonstrating characteristic optic disc changes, associated with a normal or raised intra-ocular pressure, raises the suspicion of glaucoma. Structural and functional assessments using various forms of technology, are used to assist in diagnosing glaucoma.[4] Multiple diagnostic modalities used in making the diagnosis and monitoring progression of this disease include standard automated perimetry (visual field test); the Heidelberg Retinal Tomography (HRT); Optical Coherence Tomography (OCT) and scanning laser polarimetry (SLP) (commercially known as Glaucoma Diagnosis (GDx)).[4] While standard automated perimetry functionally assesses the optic disc; HRT, OCT and GDx provide a structural assessment of the disc and corresponding retinal nerve fibre layer(RNFL). OCT is the newest of the latter three modalities described.

2. Literature review

The Baltimore eye survey was a large population based prevalence survey consisting of over 5000 patients with European and African ancestry.[5] African patients had a prevalence of POAG that was 4 to 5 times higher than that of Europeans.[5]

The Barbados eye study, a large population based study consisting of over 4000 patients, confirmed the higher prevalence and aggressive nature of glaucoma in the African population.[6] POAG also affects African patients 10 years earlier than Europeans.[3]

The African Descent and Glaucoma Evaluation studies (ADAGES) were set up to assist in understanding why differences occur between the African and European populations

affected with Glaucoma.[3,7] These Studies reviewed both normal and glaucomatous individuals of both population groups looking at clinical factors that differ between populations of African and European descent, such as central corneal thickness (CCT), optic disc morphology and RNFL thickness (RNFL).

The ADAGES study in 2010 found that the central corneal thickness (CCT) of Africans (534 µm) is thinner than that of Europeans(552 µm).[7] The Ocular Hypertension Treatment Study (OHTS), showed that those with thinner corneas tend to have lower intraocular pressures (IOPs), while thicker corneas, a higher intraocular pressure (IOP); the former being associated with a higher risk of developing glaucoma.[8] In the OHTS study, patients with CCT of >555 µm but ≤558 µm were 1.7 times more likely to develop POAG. Those with CCT ≤555 µm were 3.4 times more likely to get POAG.[8] The study also showed that those of African descent have on average, a 23.5 µm thinner cornea than those of Europeans.[8] Thinner corneas have lower IOPs because of the biomechanical properties of the cornea causing an underestimation of IOP readings on Goldman applanation tonometry.[9,10] Thus, the IOPs of thinner corneas are higher than what is measured and because these higher IOPs are not detected, eyes with thinner corneas are at higher risk of developing glaucomatous optic disc damage. In addition to this, the biomechanical properties of a thinner cornea, are related to the general biomechanical properties of the eye, including the lamina cribrosa of the optic disc.[9,11] Biomechanical properties of the lamina cribrosa also contribute to the optic disc damage in glaucomatous eyes.[9]

The optic disc, or optic nerve head (ONH), is the region of the eye where the ganglion cell axons of the retina converge, and leave the eye as the optic nerve. In the 2010 ADAGES study, there were different optic disc morphologies noted between the normal population groups of African and European descent.[7] The horizontal and vertical cup to disc ratio (CDR) and optic disc area in populations of African descent were larger than those of European descent.

The optic disc or ONH is a bulk representation of ganglion cell axons (which lie within the retina). These axons of ganglion cells are spread out thinly as the most superficial layer of

the retina forming the retinal nerve fibre layer (RNFL). In the ADAGES study, the average or global retinal nerve fibre layer (RNFL) thickness in the normal population of European descent was 100.6µm.[7] On OCT, people of African descent displayed a thicker superior and inferior RNFL whilst the temporal region was thinner than that of the Europeans[7]; however, there was no significant difference between the global and nasal RNFL between the two population groups. A study from Northern Nigeria by Sani et al, showed a similar mean global RNFL thickness (104.1µm) on OCT when compared to the 2010 ADAGES study (103.69µm).[7,12] There was also agreement between the two studies in terms of the RNFL thickness in the nasal and temporal quadrants i.e. the nasal RNFL is thick and the temporal RNFL is thin.

People of African descent in the ADAGES study had a thicker inferior RNFL compared to the superior RNFL.[7] However, the Nigerian population had a thicker superior RNFL compared to the inferior RNFL.[12]

In the Nigerian study females had a thicker RNFL compared to males.[12]

The Northern Nigerian Africans were found to have a similar RNFL thickness when compared to Indian and Nepalese populations.[12–14]

Two South African Studies conducted by the University of Kwazulu-Natal school of optometry measured the RNFL in Africans.[15] The study by Murugan et al compared the RNFL thickness between Indians and Africans with Myopia.[15] The RNFL of myopic African patients was found to be thicker than their Indian counterparts. Mashige et al, in their study, used the ivue SD -OCT to measure the RNFL thickness in a sample of 600 Africans.[16] This study found the global RNFL thickness to be 110µm which was higher than that found in other studies.[7,12,13,15,16] The thicker inferior RNFL compared to the superior RNFL was similar to that found in the 2010 ADAGES study.[7,16]

Multiple factors such as glaucoma, myopia, lens status and age influence the thickness of the RNFL.

On OCT, RNFL thinning is a characteristic finding in glaucoma.[11]

Normally, the peak RNFL readings are at the 11 and 7 o clock positions.[17] Studies have shown that the positions of the peak RNFL thickness differ in eyes that are myopic with

longer axial lengths. In myopic eyes, the peak superior and inferior RNFL quadrants are more temporally located, and the overall RNFL is thinner.[17] Thus the normative distribution of RNFL thickness in eyes with no myopia, cannot be applied to myopic eyes.

During OCT measurements, cataracts cause a decrease in signal strength to the retina, and the reflection back from the retina.[18] This results in a falsely thinner reading of the RNFL on OCT. This is further compounded by the fact that cataracts occur more frequently in the elderly, where increasing age alone has been shown to contribute to RNFL thinning.[19] The retinal nerve fibre layer thickness decreases 0.18% per year, or 1.53-2 µm per decade.[19]

Phacoemulsification cataract surgery has been shown to increase overall retinal nerve fibre layer thickness.[18] This is due to normalization of the signal strength, giving a more accurate RNFL thickness. A study in India by Jha et al showed an increase in signal strength of the SD-OCT post cataract surgery, as well as inferior RNFL in the inferior quadrant one month after cataract surgery; this quadrant is important in assessing glaucoma progression.[18] Conversely, the EPIC – Norfolk eye study showed a thinner RNFL in pseudophakic patients, when compared to the rest of the participants in the study[20]. This study did not stipulate the time of RNFL assessment post cataract surgery, and used the GDx to conduct these measurements.

In addition to glaucoma, age, lens status, and axial length, a thinner RNFL has also been demonstrated in the male gender and sleep apnoea.[19] Conditions that result in abnormal retinal vasculature cause RNFL thinning by neurodegeneration.[21]

Multiple techniques are available to structurally assess optic disc morphology and RNFL thickness. These are ocular coherence tomography (OCT), Heidelberg Retinal Tomography (HRT) and scanning laser polarimetry (Glaucoma diagnosis (GDx)).

Ocular coherence tomography (OCT), first described in 1991 by Huang et al, is a non-contact, non-invasive, imaging modality that can be used to scan the retina and the optic nerve.[22,23] Two types of OCTs exist, namely Time domain OCTs and Spectral domain OCTs. Spectral domain OCTs offer a faster scan speed, higher axial resolution and 3D

reconstruction of the obtained image. The faster scan speed allows for less variability in measurements due to movement artefacts.[11]

OCT provides a good structural assessment of both optic disc or optic nerve head (ONH) morphology and RNFL thickness, and can pick up early glaucomatous damage.[4,11] (The basic anatomy of the optic disc and RNFL has been described previously). OCT measures the RNFL thickness by measuring the peripapillary thickness. This is the circular area of retina surrounding the optic disc. On OCT, this circular area is divided into quadrants (temporal, superior, nasal, inferior). The superior and inferior quadrants can be further subdivided to either being temporal or nasal. RNFL thickness of the patient is compared to that of a built-in database that is specific to machine type. This is age and gender matched. A graph showing the RNFL thickness profile of each quadrant is automatically generated indicating areas that are thicker, thinner, or approximately the same. This is displayed per quadrant as being either "within normal limits (green)", "Borderline (yellow)", or "outside normal limits (red)".[7] A quadrant will be considered "borderline" if the patient's reading is outside the 95th centile, and "outside normal limits" if the quadrant reading is outside the 99th centile.[24]

The HRT device produces a 3D topographic image of the optic nerve by scanning the retinal surface with a diode laser in consecutive parallel planes.[4] The operator of the machine then draws a line manually around the optic disc and information is then obtained. The HRT allows for assessments of optic disc morphology as well as RNFL thickness.[4] The HRT 3 version derives further information about the optic disc, such as the Moorfields Regression Analysis (MRA) and the Glaucoma probability score (GPS).[4]

The retinal nerve fibre layer, like the cornea and lens, has birefringent properties. The GDx uses a polarised, near infrared light to measure the RNFL birefringence, thereby correlating it with the RNFL thickness.[4] The GDx cannot measure optic disc morphology.

A Cochrane review by Michelessi et al, concluded that all 3 devices (HRT3,OCT, and GDx) are variable but similar when aiding in the diagnosis of glaucoma.[4] i.e. As described above, OCT

and HRT can provide information of the optic disc morphology and RNFL thickness, whereas GDx is excellent in assessing RNFL thickness.

Whereas OCT, HRT, and GDx provides structural assessment of the optic disc and RNFL, functional assessment of visual field loss due to optic disc damage is achieved with standard automated perimetry. Many glaucoma patients exhibit visual field loss only once there is substantial retinal ganglion axons loss and the retinal nerve fibre layer has already thinned.[4,11,20,25]Forty percent of nerve fibre loss could occur prior to it being detected by automated perimetry.[4] Therefore to detect early glaucomatous optic disc changes, structural assessment of the optic disc and RNFL by either OCT, HRT or GDx should be carried out.

To determine if glaucoma is present or not, optic disc and RNFL thickness values on spectral domain OCT (SD-OCT) are usually compared to an age-matched European normative database which is machine-specific and not universally applicable between different types of SD-OCT machines.[12,16] As mentioned earlier, because RNFL thickness between normal Africans and Europeans differ,[7] comparing RNFL thickness in an African glaucoma patient to a European normative database can give inaccurate results. Therefore, the aim of this study is to develop a normative RNFL database in adult Africans by measuring RNFL thickness on the Heidelberg engineering spectral domain OCT; this will improve the accuracy of the results. To the best of our knowledge, no other studies were done measuring the RNFL in normal black South Africans with the Heidelberg engineering SD- OCT machine.

3. Research Question

Is the retinal nerve fibre layer thickness of black South African patients significantly different than Europeans?

4. Research Aim

The aim of this study is to measure the Retinal Nerve Fibre Layer (RNFL) thickness of normal, healthy black South African adults at St John Eye Hospital.

5. Research Objectives

To develop a normative RNFL database in black South African adults, by measuring RNFL thickness on the Heidelberg engineering spectral domain OCT. This will lead to an accurate structural assessment of the optic nerve head in patients suspected of glaucoma.

- To compare differences in RNFL thickness according to age-group and sex.
- To describe differences in RNFL thickness according to the different anatomical quadrants of the optic nerve, as measured by the spectral domain OCT (SD 0CT Heidelberg Engineering)
- To identify any associations between RNFL thickness and demographic and ocular factors (age, gender, axial length, IOP and CCT)

6. Research Method

6.1 Design

A cross-sectional, descriptive study.

6.2. Study site

The Study will be conducted at the St John Eye Hospital-Chris Hani Baragwanath Hospital in Soweto, Gauteng. Nine full time specialists, two part time consultants, fourteen registrars, and five medical officers see approximately 400 outpatients per day. Patients are referred from surrounding clinics, secondary hospitals and private general practitioners and ophthalmologists.

6.3 Study population

- Sampling

  One hundred and thirty participants will be entered into the study. This is the number (needed for a normative database) given by Heidelberg Engineering after consulting them.

One hundred and thirty adults (patients, staff and students) who attend the ophthalmology outpatients' department at St John eye hospital and who have a normal ocular examination (normal anterior segment, fundus (including normal optic disc), axial length, and intraocular pressure (IOP)) will be invited to participate in the study. Participants will be informed about

the study. An information sheet pertaining to the details of the study will be given to patients **(Appendix A).** Patients willing to participate in the study will be asked to give written consent **(Appendix B).** History (age, gender, province, language, co morbidities) will be taken and examination (visual acuity (VA), intra-ocular pressure (IOP), cup: disc (C:D) ratio) conducted on all participants; and RNFL thickness and vertical disc height on OCT will be measured **(Appendix C).** The central corneal thickness will also be measured at the time of axial length measurement.

6.4 Inclusion criteria:

All patients, doctors, nursing and medical students who attend St John eye hospital from 1 January 2018 – 30 September 2018, and meet the below criteria, will be invited to participate in the study.

- Are 18 years and older
- Are South African
- Black ethnicity
- Uncorrected visual acuity of 6/9 or better in the study eye
- Normal intraocular pressure (10–21 mmHg)
- Clinically normal optic disc (pink, well defined margins, normal vasculature in the absence of other pathologies)
- Absence of other ocular pathologies
- Good Quality OCT scans (> 22)
- Cyclotorsion of ≤ 7 degrees to fovea (to align patients' peripapillary area to the pre-set orientation of machine quadrants- superior, inferior, nasal, temporal)

6.5 Exclusion criteria:

Participants with:

1.) A history of glaucoma, intraocular surgery, a laser procedure, ocular trauma and current or previous uveitis.

2.) A history of neurological disease with optic disc abnormalities (e.g. Multiple Sclerosis)

3.) Clinically significant diabetic retinopathy

4.) Hypertensive changes (grade 3 and 4)

5.) Clinically significant vitreal, retinal or choroidal diseases

6.) Abnormal axial length (>26.0 mm)

7.) Significant cataracts interfering with fundus view and OCT quality

6.6 Measurement Tools

The SD-OCT (Heidelberg Engineering) will be used to conduct the study. A biometry machine (Nidek optical) will be used for axial length and central corneal thickness measurements. Both machines do not require any contact with the participants eyeball.

6.7 Data collection

Information collected will contain the following (Appendix C):

1. Personal information: Age, Sex, language spoken, province originally from, co-morbidities.
2. Which eye examined
3. Visual acuity
4. Intraocular pressure
5. Cup to disc ratio on clinical examination
6. Axial length
7. Central Corneal Thickness
8. White to White (WTW)
9. OCT report with RNFL thickness, vertical disc height measured, quality of the scan and degree of cyclotorsion.

6.8 Data management and analysis

Data obtained will be entered onto a data collection sheet (Appendix C).

Microsoft Excel will be used to capture and store all data collected.

- Data analysis

Statistical analysis of the data will be carried out as follows: The frequency and percentages of categorical variables will be calculated and plotted with bar charts. The mean, standard

deviation, median and interquartile range of continuous variables will be calculated, and their distribution illustrated by means of histograms.

The Kolomogorov- Smirnov test will be used to evaluate the normality of the distribution of the RNFL thickness data. The mean RNFL thickness globally as well as mean RNFL thickness in all quadrants (superior, inferior, nasal and temporal quadrants) will be determined. Significant differences in RNFL thickness between the different quadrants will be calculated using Analysis of variance (ANOVA). The t- test will be used to determine significant differences between male and female RNFL thickness. Univariate analysis will be conducted to show associations between RNFL thickness and demographic and ocular factors (age, gender, axial length, IOP and CCT). Multivariate regression analysis will further be conducted in those factors that show a strong association in the univariate analysis. A p-value of less than 0.05 will be used to define statistical significance. Stata/SE version 13.0 (Stata, College Station, Texas, USA) will be used to conduct all statistical analyses.

6.9 Time schedule

The study will commence as soon as Post-graduate approval and Ethics Committee clearance is obtained. The study will take approximately 9 months to complete.

October 2017	November 2017	December 2017	January – September 2018	October 2018	November 2018	December 2018
Protocol submission	Assessment Committee meeting and informing ethics committee of any changes	Await final ethics clearance for data collection	Data collection	Data Analysis and write up	Write up	Submission for marking

6.10 Ethical considerations:

- Participant confidentiality:

  Each participant's hospital number will be assigned a unique study number. This study number will appear on the data collection sheet together with the participant's data. The file linking the study number to the participant hospital number will be kept separate from the data collection sheet and will only be used for crosschecking purposes. Strict confidentiality will be maintained always.

- Study Approval:

  This has been obtained from the Human Research Ethics Committee of the University of the Witwatersrand. The ethics committee will be informed of any changes as suggested by the protocol committee. Data will be kept completely anonymous. No information that can constitute a breach of patient confidentiality will be published. The research will not harm the participants in any way and participation is completely voluntary.

  - Written permission from the CEO of Chris Hani Baragwanath has been obtained to conduct the study at the hospital.
  - Written permission from the head of Ophthalmology has been obtained to allow permission to conduct the study at St Johns Eye Hospital.

6.11 Costs

- Budget Breakdown
  - Consumables- paper copies of data collection sheets, information sheets, informed consent papers, pens and highlighters are the only foreseeable costs for this study.

All costs incurred during the period of the research will be borne by the principal investigator.

7.Reporting plans

The aims are to:

1. Publish an article in a scientific journal.
2. Write up a research report as part of an MMed degree.

3. Presentation of study findings at the South African Glaucoma Society (SAGS) annual congress.
4. Presentation of study findings to Heidelberg Engineering (South Africa)

8. Authorship

Dr. S. Ismail: Principal investigator

Registrar, Department of Neurosciences, Division of Ophthalmology, University of the Witwatersrand / Chris Hani Baragwanath Academic Hospital (St John Eye Hospital)

Dr H. D. Alli: Supervisor

Consultant, Division of Ophthalmology, University of the Witwatersrand / Chris Hani Baragwanath Academic Hospital (St John Eye Hospital)

Dr N. Ally

Registrar, Department of Neurosciences, Division of Ophthalmology, University of the Witwatersrand / Chris Hani Baragwanath Academic Hospital (St John Eye Hospital)

9. References

1. Quigley HA. Number of people with glaucoma worldwide. *Br J Ophthalmol.* 1996;80(5):389–93.

2. Tham Y-C, Li X, Wong TY, Quigley HA, Aung T, Cheng C-Y. Global Prevalence of Glaucoma and Projections of Glaucoma Burden through 2040. *Ophthalmology*. 2014;121(11):2081–90.

3. Sample PA, Girkin CA, Zangwill LM, Jain S, Racette L, Becerra LM, et al. The African Descent and Glaucoma Evaluation Study (ADAGES): design and baseline data. *Arch Ophthalmol Chic Ill 1960.* 2009;127(9):1136–45.

4. Michelessi M, Lucenteforte E, Oddone F, Brazzelli M, Parravano M, Franchi S, et al. Optic nerve head and fibre layer imaging for diagnosing glaucoma. *Cochrane Database Syst Rev.* 2015;11:CD008803.

5. Tielsch JM, Sommer A, Katz J, Royall RM, Quigley HA, Javitt J. Racial Variations in the Prevalence of Primary Open-angle Glaucoma: The Baltimore Eye Survey. *JAMA.* 1991;266(3):369–74.

6. Leske MC, Wu S-Y, Hyman L, Nemesure B, Hennis A, Schachat AP. Four-year incidence of visual impairment. *Ophthalmology.* 2004;111(1):118–24.

7. Girkin CA, Sample PA, Liebmann JM, Jain S, Bowd C, Becerra LM, et al. African Descent and Glaucoma Evaluation Study (ADAGES): II. Ancestry differences in optic disc, retinal nerve fiber layer, and macular structure in healthy subjects. *Arch Ophthalmol Chic Ill 1960.* 2010;128(5):541–50.

8. Gordon MO, Beiser JA, Brandt JD, Heuer DK, Higginbotham EJ, Johnson CA, et al. The Ocular Hypertension Treatment Study: baseline factors that predict the onset of primary open-angle glaucoma. *Arch Ophthalmol Chic Ill 1960*;120(6):714-720

9. ElMallah MK, Asrani SG. New ways to measure intraocular pressure. *Curr Opin Ophthalmol.* 2008;19(2):122–6.

10. Lee BL, Wilson MR, Ocular Hypertension Treatment Study (OHTS). Ocular Hypertension Treatment Study (OHTS) commentary. *Curr Opin Ophthalmol.* 2003;14(2):74–7.

11. Tao Y-L, Tao L-M, Jiang Z-X, Liu H-T, Liang K, Li M-H, et al. Parameters of ocular fundus on spectral-domain optical coherence tomography for glaucoma diagnosis. *Int J Ophthalmol*. 2017;10(6):982–91.

12. Sani RY, Abdu L, Pam V. Retinal nerve fiber layer thickness measurements of normal Northern Nigerian adults using optical coherence tomography. *Ann Afr Med*. 2016;15(2):52–7.

13. Ramakrishnan R, Mittal S, Ambatkar S, Kader MA. Retinal nerve fibre layer thickness measurements in normal Indian population by optical coherence tomography. *Indian J Ophthalmol.* 2006;54(1):11–5.

14. Thapa M, Khanal S, Shrestha GB, Sharma AK. Retinal nerve fibre layer thickness in a healthy Nepalese population by spectral domain optical coherence tomography. *Nepal J Ophthalmol*. 2014;6(2):131–9.

15. Murugan C, Golodza BZ, Pillay K, Mthembu BN, Singh P, Maseko SK, et al. Retinal nerve fibre layer thickness of black and Indian myopic students at the University of KwaZulu-Natal. *Afr Vis Eye Health*. 2015;74(1).

16. Mashige KP, Oduntan OA. Retinal nerve fibre layer thickness values and their associations with ocular and systemic parameters in Black South Africans. *Afr Health Sci*. 2016;16(4):1188–94.

17. Hong SW, Lee SB, Jee D, Ahn MD. Interocular retinal nerve fiber layer thickness difference in normal adults. *PloS One.* 2015;10(2)

18. Jha B, Sharma R, Vanathi M, Agarwal T, Sidhu T, Tomar A, et al. Effect of phacoemulsification on measurement of retinal nerve fiber layer and optic nerve head parameters using spectral-domain-optical coherence tomography. *Oman J Ophthalmol*. 2017;10(2):91–5.

19. Rougier M-B, Korobelnik J-F, Malet F, Schweitzer C, Delyfer M-N, Dartigues J-F, et al. Retinal nerve fibre layer thickness measured with SD-OCT in a population-based study of French elderly subjects: the Alienor study. *Acta Ophthalmol (Copenh)*. 2015;93(6):539–45.

20. Khawaja AP, Chan MPY, Garway-Heath DF, Broadway DC, Luben R, Sherwin JC, et al. Associations with retinal nerve fiber layer measures in the EPIC-Norfolk Eye Study. *Invest Ophthalmol Vis Sci*. 2013;54(7):5028–34.

21. Jonsson KB, Frydkjaer-Olsen U, Grauslund J. Vascular Changes and Neurodegeneration in the Early Stages of Diabetic Retinopathy: Which Comes First? *Ophthalmic Res*. 2016;56(1):1–9.

22. Huang D, Swanson EA, Lin CP, Schuman JS, Stinson WG, Chang W, et al. Optical coherence tomography. *Science*. 1991;254(5035):1178–81.

23. Hee MR, Izatt JA, Swanson EA, Huang D, Schuman JS, Lin CP, et al. Optical coherence tomography of the human retina. *Arch Ophthalmol Chic Ill 1960.* 1995;113(3):325–32.

24. Leung CK-S. Diagnosing glaucoma progression with optical coherence tomography. *Curr Opin Ophthalmol*. 2014;25(2):104–11.

25. Budenz DL, Anderson DR, Varma R, Schuman J, Cantor L, Savell J, et al. Determinants of normal retinal nerve fiber layer thickness measured by Stratus OCT. *Ophthalmology*. 2007;114(6):1046–52.

APPENDIX A- PATIENT INFORMATION SHEET

University of the Witwatersrand

Research Participant Information Sheet

Good Day

My name is Sarah Ismail. I am a doctor in the ophthalmology department of the university of the Witwatersrand. I am conducting research for a Masters in Medicine (MMed) at St John eye Hospital. I would like to invite you to participate in my study.

My objective is to measure the retinal nerve fibre layer (RNFL) of the eye, in a normal black South African population. This layer is situated in the retina which is at the back of the eye. The measurement is obtained using the ocular coherence tomography (OCT) machine. It will not hurt or cause any harm to you. No contact is made with the eyeball during this investigation. This measurement is done by taking a picture of the nerves at the back of the eye. Measuring this layer will give us an idea regarding the normal RNFL values in black South Africans.

Your participation in this study is completely voluntary, and there is no risks or direct benefit to you. However, information gained from this study could assist us in the accurate diagnosis of certain eye diseases in our black South African population. The results of the research will be made available to interested parties and your confidentiality will be maintained always. Participant details will not be displayed on the results obtained. Your details will be kept separately by the principal investigator.

You can withdraw from the study at any time you wish without any prejudice.

The study has been submitted to the Human Research Ethics Committee (Medical) at the University of the Witwatersrand. There contact details are as follows:

- Chairperson: peter.cleaton-jones1@wits.ac.za
- Administrators - Ms Zanele Ndlovu/ Mr Rhulani Mkansi/ Mr Lebo Moeng Tel 011 717 2700/2656/1234/1252
- Email: HREC-Medical.ResearchOffice@wits.ac.za

Thank you for considering my request. My contact details are below should you have any further questions.

Dr Sarah Ismail

Department of Ophthalmology

University of Witwatersrand

0724506182

Email: sarahismail77@gmail.com

APPENDIX B- PATIENT CONSENT FORM

University of the Witwatersrand

INFORMED CONSENT FORM

TITLE: Retinal nerve fibre layer thickness in a normal black South African population

Participant

I have read the research participant information sheet for this study, or someone has explained it to me in a language that I understand. All questions that I may have, have been answered and I am satisfied with the answers I have received. I voluntarily consent to be a participant in this study.

Name of Participant: _____

Date: _____

Signature (or fingerprint): _____

Investigator (Person obtaining consent)

I have explained the information in the research participation information sheet to the participant, or had it explained to the participant in a language he/she understands. I have answered all questions to the best of my ability. Consent has been given voluntarily by the research participant.

Name of Investigator: _____

Date: _____

Signature (or fingerprint): _____

Witness

I have been witness to the explanation of the research participant information sheet, and confirm that the investigator has answered all the participant's questions and that the participant has given voluntary consent.

Name of Witness: _____

Date: _____

Signature (or fingerprint): _____

APPENDIX C- DATA COLLECTION SHEET

Age: Code:

Gender: M / F

Province:

Language:

Co- Morbidities:

Eye Examined: R / L

VA:

IOP:

Fundus- C: D ratio

Axial length:

Central Corneal Thickness: WTW:

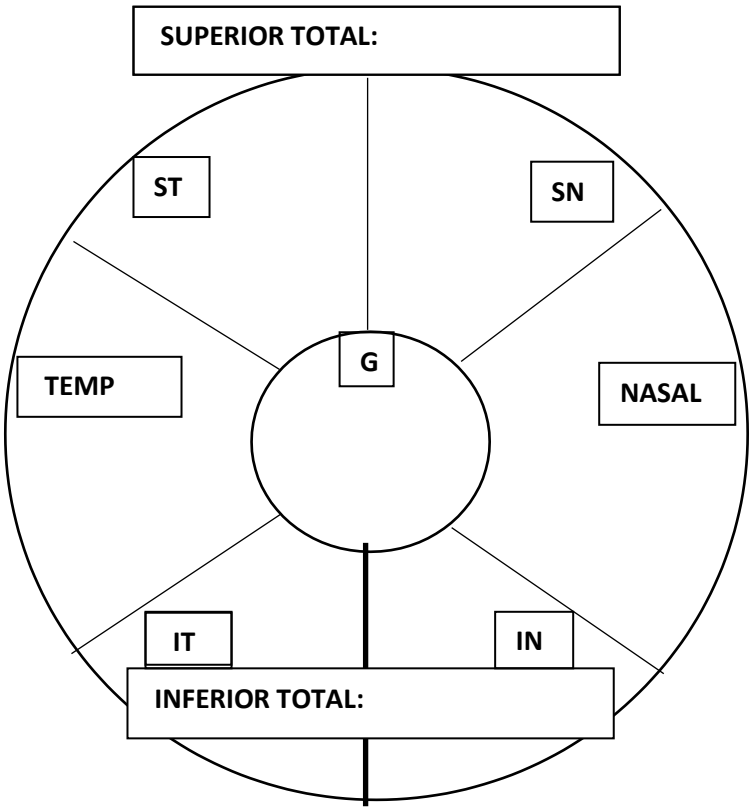
OCT report:

RNFL:

VDH: R / L

Quality:

Fovea:



R14/49 Dr Sarah Ismail

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)

CLEARANCE CERTIFICATE NO. M161114

NAME: Dr Sarah Ismail
(Principal Investigator)
DEPARTMENT: Ophthalmology
St John Eye Hospital, Outpatient Department

PROJECT TITLE: Retinal Nerve Fibre Layer Thickness in a Normal Black South African Population

DATE CONSIDERED: 25/11/2016

DECISION: Approved unconditionally

CONDITIONS:

SUPERVISOR: Akiel Asvat

APPROVED BY:
Professor P Cleaton-Jones, Chairperson, HREC (Medical)

DATE OF APPROVAL: 13/01/2017

This clearance certificate is valid for 5 years from date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Research Office Secretary in Room 301, Third Floor, Faculty of Health Sciences, Phillip Tobias Building, 29 Princess of Wales Terrace, Parktown, 2193, University of the Witwatersrand. I/we fully understand the conditions under which I am/we are authorized to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated, from the research protocol as approved, I/we undertake to resubmit the application to the Committee. **I agree to submit a yearly progress report.** The date for annual re-certification will be one year after the date of convened meeting where the study was initially reviewed. In this case, the study was initially reviewed in November and will therefore be due in the month of November each year. Unreported changes to the application may invalidate the clearance given by the HREC (Medical).

Principal Investigator Signature

Date

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES