Correlation of Retinal Nerve Fibre Layer and Macular Thickness with Serum Uric Acid among Type 2 Diabetes Mellitus

By

Dr Vinuthinee Naidu A/P Munisamy Naidu

MBBS (MMMC)

Dissertation Submitted in Partial Fulfillment for The Degree of Master of Medicine (Ophthalmology)

SCHOOL OF MEDICAL SCIENCES

UNIVERSITI SAINS MALAYSIA

2016

2. DISCLAIMER

I hereby certify that the work in this dissertation is my own except for the quotations and summaries which have been duly acknowledged.

Date: 24.11.2016 …….…….

Dr. Vinuthinee Naidu A/P Munisamy Naidu

P-UM0157/10

ACKNOWLEDGEMENT

First and above all, I praise God, the almighty for providing me this opportunity and granting me the capability to proceed successfully.

First and foremost, I would like to express my sincere gratitude to my supervisor, Associate Professor Datin Dr Zunaina Embong, for all her support, guidance and encouragement during the duration of this study. I might have lost the direction of my journey without her support. Her valuable suggestions and exceptional guidance allowed me to gain and advance my ophthalmology knowledge.

A special thanks to Professor Syed Hatim Noor, from Department of Biostatistics and Research Methodology for his assistance and advice during the statistical analysis and presentation of my data.

My sincere thanks to Professor Dr. Liza Sharmini Ahmad Tajudin, Head of Ophthalmology Department, Universiti Sains Malaysia and all the lecturers for their continuous guidance, assistance, encouragement and cooperation throughout this period.

I would like to thank all the members of my family; my parents and my mother-in-law for their endless love and encouragement. Last but not least, I greatly thank my lovely husband Dr Kantha Rao for his immeasurable support and utmost understanding throughout this journey. I would like to say "Love" to my adorable daughter, Sanjhana Rao.

TABLE OF CONTENTS

ABSTRAK

Latar Belakang

Serum urik asid adalah produk pecahan akhir katabolisma purin pada manusia. Ia adalah satu antioksidan yang kuat, yang menyebabkan tekanan oksidatif pada sel-sel vaskular endothelial, dengan itu mengalakkan perkembangan penyakit yang berkaitan dengan kencing manis. Terdapat pelbagai bukti epidemiologi dan eksperimen yang menunjukkan bahawa serum urik asid memainkan peranan dalam etiologi kencing manis jenis 2. Kami telah menjalankan kajian keratan rentas mengaitkan ketebalan lapisan saraf retina mata (LSRM) dan ketebalan makula dengan serum urik asid di kalangan pesakit kencing manis jenis 2 yang tiada diabetik retinopati dan dengan diabetik retinopati tidak proliferasi (DRTP).

Metodologi

Satu kajian keratan rentas telah dijalankan di Kelinik Mata, Hospital Universiti Sains Malaysia, Kelantan antara tempoh Ogos 2013 hingga Julai 2015 melibatkan pesakit diabetes jenis 2 yang tiada diabetik retinopati dan dengan DRTP. Semua pesakit telah menjalani ujian pengukuran ketebalan LSRM dan ketebalan makula menggunakan mesin Spectralis Domain Heidelberg optikal koherens tomografi. Sebanyak 6 mls darah telah diambil daripada pesakit untuk ujian serum urik asid dan HbA1c.

Keputusan

Seramai 180 pesakit kencing manis telah terlibat (tiada diabetik retinopati: 90 pesakit dan DRTP: 90 pesakit) dalam kajian ini. Tahap purata serum urik asid untuk kedua-dua kumpulan berada dalam julat normal dan tiada perbezaan yang signifikan. Tahap purata serum urik asid untuk kedua-dua jantina yang tiada diabetik retinopati adalah tinggi yang signifikan berbanding dengan mereka yang DRTP ($p = 0.004$ masing-masing). Purata serum urik asid adalah lebih tinggi yang signifikan dalam pesakit dengan HbA1c <6.5% ($p < 0.031$). Pesakit dalam kumpulan NPDR mempunyai ketebalan LSRM dan makula yang lebih tebal berbanding dengan pesakit dalam kumpulan yang tiada diabetik retinopati ($p = 0.038$). Walau bagaimanapun, hanya ketebalan LSRM dalam kuadran temporal dan ketebalan makula dalam sub-bidang luar superior, sub-bidang luar inferior dan sub-bidang luar temporal menunjukkan perbezaan yang signifikan ($p = 0.038$, $p = 0.004$, 0.033 dan <0.001 masing-masing). Di dapati korelasi yang lemah di antara ketebalan RNFL dan makula dengan serum urik asid di kalangan pesakit di kedua-dua kumpulan.

Kesimpulan

Serum urik asid menunjukkan korelasi yang lemah dengan LSRM dan ketebalan makula di kalangan pesakit kencing manis jenis 2 yang tiada retinopati diabetik dan DRTP .

ABSTRACT

Background

Serum uric acid is a final breakdown product of purine catabolism in humans. It's a potent antioxidant, that induces oxidative stress on the vascular endothelial cells, thus mediating progression of diabetic related diseases. Various epidemiological and experimental evidence suggest that uric acid has a role in the etiology of type 2 diabetes mellitus. We conducted a cross-sectional study to evaluate the correlation of RNFL and macular thickness with serum uric acid in type 2 diabetic patients.

Methodology

A cross-sectional study was conducted in the Eye Clinic, Hospital Universiti Sains Malaysia, Kelantan between the period of August 2013 till July 2015 involving type 2 diabetes mellitus patients with no diabetic retinopathy and with NPDR. An evaluation for RNFL and macular thickness using Spectralis Heidelberg optical coherence tomography was done and 6 mls of venous blood was taken for the measurement of serum uric acid and $HbA1_C$.

Results

A total of 180 diabetic patients were recruited (no diabetic retinopathy: 90 patients and NPDR: 90 patients) into the study. The mean level of serum uric acid for both the groups were within normal range and there was no significance difference between the two groups. The mean level of serum uric acid for both gender was significantly higher in no diabetic retinopathy group ($p = 0.004$ respectively). The mean serum uric acid was significantly higher in patient with $HbA1_C < 6.5\%$ ($p < 0.031$). Patients with NPDR have thicker RNFL and macular thickness compared

to patient with no diabetic retinopathy. However, only the RNFL thickness of the temporal quadrant and the macular thickness of the superior outer, inferior outer and temporal outer subfields were statistically significant ($p = 0.038$, $p = 0.004$, 0.033 and <0.001 respectively). There was poor correlation between RNFL and macular thickness with serum uric acid in both the groups.

Conclusion

Serum uric acid showed a poor correlation with RNFL and macular thickness among type 2 diabetic patients.

CHAPTER 1

INTRODUCTION

1.1 INTRODUCTION

Retina, is part of the integral component of central nervous system and plays a pivotal role in the radial flow of visual signals. The quality of vision deteriorates early in diabetes, before the vascular abnormalities becomes evident, probably indicating the early signs of neuronal dysfunction.

Diabetic retinopathy is termed as a neurovascular disease which affects both the neuroretinal and microvascular component, and the former is said to be compromised early in the course of diabetic retinopathy followed by microvascular changes (Stitt et al, 2015). The hallmark of retinal neurodegeneration are neural apoptosis and reactive gliosis.

Retinal nerve fibre layer (RNFL) is an important neuronal structure in the retina layer which is often shown to be affected in the early pathogenesis of diabetic retinopathy. The functional changes that are noticed before the vascular pathology develops are due to direct effect of diabetes on the neural retina instead of breakdown of the blood retinal barrier. The retinal ganglion cell and glial cells are the major cells in the neuronal component to be compromised (Antonetti et al, 2015) reflecting neurodegenerative changes in the diabetic retina with loss of neuronal cell bodies and axons (van Dijk et al, 2012). The prominent risk factors for RNFL degeneration in a diabetic patient are hyperglycemia, oxidative stress and advanced glycation end products. It's very important to understand the neurodegenerative process of the RNFL as it plays a role as an active mediator of the microvascular impairment and used as an index in a diabetic patient to predict the development of new microvascular disease in near future. Early detection of RNFL thinning allows the ophthalmologist to provide effective treatment, thus reducing vision loss.

Optical coherence tomography (OCT), has emerged as an excellent non-invasive ocular light-based imaging modality that can be used in biological system to study tissues in vivo with detailed information about the histological changes (Drexler et al, 2008). OCT is widely accepted and has been integrated into our everyday clinical practice, has become an essential gold standard tool in the diagnosis and monitoring most retinal diseases.

Chronic hyperglycemia contributes to the development and progression of diabetic retinopathy and the most important determinants are the disease duration and the degree of glycemic control. The $HbA1_C$ has been regarded as the gold standard indicator for glycemic control in diabetic patients, reflecting the status of the blood glucose control over 3-6 months.

Serum uric acid is a product breakdown of the endogenous and exogenous purine metabolism and its derivatives. Increase serum uric acid has been proposed as a biochemical link between diabetes and the development of microvascular and macrovascular complications (Goldberg, 2009). Goldberg (Goldberg, 2009) in his studies have attributed proatherogenic properties of uric acid that includes activation of endothelial cells, platelets activation and increase platelet adhesiveness which all contribute to the pathogenesis of diabetic retinopathy and other diabetic vascular complications. To date, there is still lack of data concerning the role of serum uric acid as a predictor in the development and progression of diabetic retinopathy, whether there is any significant correlation with RNFL and macular thickness.

This is a cross-sectional study involving patients with no diabetic retinopathy and non-proliferative diabetic retinopathy (NPDR) among type 2 diabetes mellitus. We evaluated the RNFL and macular thickness among both the groups and studied the correlation of RNFL and macular thickness with serum uric acid.

To the best of our knowledge and following an extensive literature search, there is no studies has been published on the correlation of RNFL and macular thickness with serum uric acid involving no diabetic retinopathy and NPDR among type 2 diabetes mellitus. This study is the first of its kind to be conducted.

CHAPTER 2

OBJECTIVES OF THE STUDY

2.1 GENERAL OBJECTIVE

To correlate the RNFL and macular thickness with serum uric acid among type 2 diabetes mellitus patients with NPDR and without diabetic retinopathy.

2.2 SPECIFIC OBJECTIVES

i. To determine the correlation between RNFL thickness and serum uric acid among type 2 diabetes mellitus patients with NPDR.

ii. To determine the correlation between RNFL thickness and serum uric acid among type 2 diabetes mellitus patients with no diabetic retinopathy.

iii. To determine the correlation between macular thickness and serum uric acid among type 2 diabetes mellitus patients with NPDR.

iv. To determine the correlation between macular thickness and serum uric acid among type 2 diabetes mellitus patients with no diabetic retinopathy.

CHAPTER 3

MANUSCRIPT

Correlation of Retinal Nerve Fibre Layer and Macular Thickness with Serum Uric Acid among Type 2 Diabetes Mellitus

Retinal Nerve Fibre Layer and Macular Thickness with Serum Uric Acid

Munisamy-Naidu Vinuthinee-Naidu[1,2,3], Embong Zunaina[1,2], Anuar Azreen-Redzal[3], Naing Nyi-Nyi[2,4]

[1]Department of Ophthalmology, School of Medical Sciences, Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia

[2]Hospital Universiti Sains Malaysia, Jalan Raja Perempuan Zainab II, 16150 Kubang Kerian, Kelantan, Malaysia

[3]Department of Ophthalmology, Hospital Sultanah Bahiyah, 05460 Alor Setar, Kedah, Malaysia

[4]Unit of Biostatistics and Research Methodology, School of Medical Sciences, Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia

Correspondence to:

Embong Zunaina
Department of Ophthalmology, School of Medical Sciences, Universiti Sains Malaysia, 16150 Kubang Kerian, Kelantan, Malaysia.
Email: zunaina@usm.my
Tel: +6 09 767 6362
Fax: +6 09 765 3370

Correlation of Retinal Nerve Fibre Layer and Macular Thickness with Serum Uric Acid among Type 2 Diabetes Mellitus

3.2 ABSTRACT

Background

Serum uric acid is a final breakdown product of purine catabolism in humans. It's a potent antioxidant, that induces oxidative stress on the vascular endothelial cells, thus mediating progression of diabetic related diseases. Various epidemiological and experimental evidence suggest that uric acid has a role in the etiology of type 2 diabetes mellitus. We conducted a cross-sectional study to evaluate the correlation of RNFL and macular thickness with serum uric acid in type 2 diabetic patients.

Methods

A cross-sectional study was conducted in the Eye Clinic, Hospital Universiti Sains Malaysia, Kelantan between the period of August 2013 till July 2015 involving type 2 diabetes mellitus patients with no diabetic retinopathy and with NPDR. An evaluation for RNFL and macular thickness using Spectralis Heidelberg optical coherence tomography was done and 6 mls of venous blood was taken for the measurement of serum uric acid and $HbA1_C$.

Results

A total of 180 diabetic patients were recruited (no diabetic retinopathy: 90 patients and NPDR: 90 patients) into the study. The mean level of serum uric acid for both the groups were within normal range and there was no significance difference between the two groups. The mean level of serum uric acid for both gender was significantly higher in no diabetic retinopathy group (p

= 0.004 respectively). The mean serum uric acid was significantly higher in patient with $HbA1_C$ < 6.5% ($p < 0.031$). Patients with NPDR have thicker RNFL and macular thickness compared to patient with no diabetic retinopathy. However, only the RNFL thickness of the temporal quadrant and the macular thickness of the superior outer, inferior outer and temporal outer subfields were statistically significant ($p = 0.038$, $p = 0.004$, 0.033 and <0.001 respectively). There was poor correlation between RNFL and macular thickness with serum uric acid in both the groups.

Conclusion

Serum uric acid showed a poor correlation with RNFL and macular thickness among type 2 diabetic patients.

Keywords

Diabetic retinopathy; retinal nerve fibre layer thickness; macular thickness; serum uric acid; glycosylated haemoglobin.

3.3 INTRODUCTION

Diabetes mellitus is regarded as a pandemic, representing one of the most challenging and major public health problems of the 21st century. It has become a global alarming disease not sparing any country thus posing a serious threat to its economy (1). It has been shown that, prevalence of diabetes is rapidly rising and is a major cause of morbidity and mortality (2).

Diabetic retinopathy is the commonest microvascular complication of diabetes mellitus. It remains the leading cause of preventable blindness across all age-groups and places a significant burden on health services (3). It's termed as a neurovascular disease which affects both the neuroretinal and microvascular component, and the former is said to be compromised early in the course of diabetic retinopathy followed by microvascular changes (4). The hallmark of retinal neurodegeneration is neural apoptosis of the retinal ganglion cells which are located in the inner retina layer and reactive gliosis involving the astrocytes and Muller cells (5). It is suggested that the functional changes noticed before the vascular pathology develops are due to direct effect of diabetes on the neural retina instead of breakdown of the blood retinal barrier (4).

Several studies have used electroretinogram (ERG) on the retinal nerve fibre layer (RNFL). An abnormal result suggests early retinal neural dysfunction which in later stages progress to neurodegeneration (6,7). It's also accompanied by deficits in contrast sensitivity, loss of dark adaptation and colour vision disturbances (8). Risk factors for RNFL degeneration in a diabetic patient are hyperglycemia, oxidative stress and advanced glycation end products (9). Macular thickness in the presence of diabetic retinopathy has been shown to be thicker compared to

normal population. This is also seen in diabetic patient with no demonstrable evidence of retinopathy or macular oedema (10).

Serum uric acid is the product of endogenous and exogenous purine metabolism and its derivatives (11). In the purine metabolic pathway (Figure 1), adenosine has vasoactive properties that play a role in retinal blood flow. It generates superoxide nitric oxide which affect the retinal circulation by causing capillary occlusion, apoptosis of pericytes and basement membrane thickening (12). Xanthine, a substrate of xanthine oxidase, enhances superoxide generation, causing microvascular dysfunction and exert tissue damage resulting in lipid and protein peroxidation (12). This changes are seen in pathogenesis of diabetic retinopathy. Various epidemiological and experimental evidence suggest that uric acid has a role in the aetiology of type 2 diabetes mellitus (11-13).

From as early as 1950, Griffiths M (14) reported the diabetogenic action of serum uric acid and suggested that it's levels are associated with an increased risk of type 2 diabetic complications. Several other studies have also observed similar findings (15,16). Goldberg RB (17), attributed proatherogenic properties of serum uric acid to be responsible for the pathogenesis of diabetic retinopathy and other diabetic vascular complications. These include activation of endothelial cells & platelets and increased platelet adhesiveness. Navin S et al (11) also concluded that poor glycemic control in type 2 diabetes mellitus is associated with an increased serum uric acid level and dyslipidemia, which could be the initial ongoing biochemical change in the complication of diabetes.

In view of numerous evidences suggesting the role of uric acid in diabetic patients, our study was conducted to correlate the RNFL thickness and macular thickness with serum uric acid

among type 2 diabetes mellitus patients. To the best of our knowledge, there are no studies done that correlates serum uric acid with RNFL and macular thickness in diabetic patients.

3.4 MATERIALS and METHODS

A cross-sectional study was carried out on 180 patients with type 2 diabetic mellitus (no diabetic retinopathy: 90 patients and non-proliferative diabetic retinopathy (NPDR): 90 patients) who attended the Eye Clinic, Hospital Universiti Sains Malaysia, Kelantan, Malaysia from August 2013 to July 2015. The study followed the tenets of the declaration of Helsinki and was approved by the local ethical board [USM/ JEPeM/ 276.2. (4)].

Type 2 diabetes mellitus patients aged between 40-65 years old with clear media were included. Those patients with optic nerve, retina and macular pathology, previous history of laser, history of intraocular surgery, renal disease and hyperuricemia were excluded from this study.

The demographic data (patients age, gender and ethnicity) was obtained either from the patient or their medical record. A thorough slit lamp and fundus examination was performed by one identified ophthalmologist to confirm the diagnosis. The classification of diabetic retinopathy was based on the International Diabetic Retinopathy Severity scales (18). Only one eye of the worst severity was selected.

This was followed by measurement of the RNFL and macular thickness using the Spectralis-Domain Optical Coherence Tomography (SD-OCT) (Heidelberg Engineering, Heidelberg, Germany).

For measurement of RNFL thickness, a 3.45 mm diameter peripapillary ring was measured (Figure 2A). All the four quadrants were taken for analysis in this study; superior, inferior, temporal and nasal (Figure 2B). The macular thickness was determined by using the '6 mm fast macular mapping' scanning pattern. It consists of a high resolution 19 raster line scan protocol that was applied on an area centered on the fovea with the horizontal lines spaced 240 µm apart (Figure 3A). The Early Treatment Diabetic Retinopathy Study (ETDRS) grid was applied dividing the macula into 9 subfields, encircled by rings of 1, 3 and 6 mm in diameter. All the subfields were taken for analysis in this study: fovea; superior inner, inferior inner, temporal inner, nasal inner in the second ring; superior outer, inferior outer, temporal outer and nasal outer in the outer ring (Figure 3B).

Six mls of venous blood was drawn from the median cubital vein for measurement of serum uric acid and $HbA1_C$. The reference normal range for serum uric acid measured in Hospital Universiti Sains Malaysia differs in each gender: Male 180 - 420 µmol/L; Female 150 - 360 µmol/L.

The statistical analysis was carried out using Statistical Package for Social Sciences (SPSS) Version 22. All values were tested for normal distribution and equal variances. Chi Square test was used for comparison of gender and ethnicity. Independent t-test was used for comparison of age, $HbA1_C$, serum uric acid, RNFL thickness and macular thickness. Significance of difference in values was determined by the 'p' value < 0.05. The correlation between RNFL thickness and macular thickness with serum uric acid was tested using Pearson correlation. Significance of correlation was decided based on 'r' (19) and 'p' values.

3.5 RESULTS

Demographic Data

The distribution of demographic data is shown in Table 1. The male and female gender were not equally distributed among both groups, and statistically not significant ($p = 0.101$). The percentage of male was higher in no diabetic retinopathy and vice versa in NPDR.

Majority of the patients were Malay in both the groups, 73% with no diabetic retinopathy and 75% with NPDR. There was no statistically significant difference among the ethnicity ($p = 0.138$).

Both the groups showed a poor diabetic control with mean $HbA1_C$ more than 6.5%. The $HbA1_C$ was significantly higher in NPDR (9.59%, SD 2.27) compared to no diabetic retinopathy (7.84%, SD 2.01) ($p < 0.001$).

Serum Uric Acid

The mean level of serum uric acid for both the groups were within normal range but at the upper limit. There was no statistically significant difference between the groups ($p = 0.220$) (Table 2). However, based on gender, diabetic patients with no diabetic retinopathy showed a significantly higher level of mean serum uric acid compared to those in NPDR group ($p = 0.004$ respectively). Generally, the male gender had a higher mean serum uric acid level compared to female gender in both the groups.

The mean level of serum uric acid was significantly higher in those patient with HbA1c < 6.5% compared to those with HbA1c ≥ 6.5% (p = 0.031) (Table 3).

Retinal Nerve Fibre Layer and Macular Thickness

Patients with NPDR generally have thicker RNFL and macular thickness compared to patient with no diabetic retinopathy. However, only the RNFL thickness of the temporal quadrant and the macular thickness of the superior outer, inferior outer and temporal outer subfields were statistically significant (p = 0.038, p = 0.004, 0.033 and <0.001 respectively) (Table 4).

Correlation between RNFL Thickness and Serum Uric Acid

The RNFL thickness showed a poor negative correlation with serum uric acid in all the quadrants for both the groups except the RNFL thickness of the temporal quadrant of no diabetic retinopathy group showed a poor positive correlation (Table 5).

Correlation between Macular Thickness and Serum Uric Acid

All the macular subfields in the no diabetic retinopathy group showed a poor negative correlation with serum uric acid except the fovea subfield showed poor positive correlation. Whereas, all the macular subfields in the NPDR group showed a poor positive correlation except superior outer subfield showed a poor negative correlation (Table 5).

3.6 DISCUSSION

Serum uric acid is a final breakdown product of purine catabolism in humans. It's a potent antioxidant, that induces oxidative stress on the vascular endothelial cells, thus mediating progression of diabetic related diseases (20). Various epidemiological and experimental evidence suggest that uric acid has a role in the etiology of type 2 diabetes mellitus (11-13)**.** We conducted a cross-sectional study to evaluate the correlation of RNFL and macular thickness with serum uric acid in type 2 diabetic patients.

The mean age of our study participants ranged between 51 to 52 years old. This was fairly consistent with several other studies (21,22). In contrast, Eydis Olafsdottir et al (23) showed the prevalence of no diabetic retinopathy and those with retinopathy in diabetic patients was slightly in the older age group. However, Hansson-Lundblad et al. (24), reported there is no association between age and retinopathy. Perhaps, duration of diabetes, is an independent risk factor for occurrence of retinopathy (21,23). Both the gender was not equally distributed in our study. This was similar to previous studies that reported no significant differences between retinopathy and gender (21,25).

The majority of the participants in our study were Malay. Malaysia has a multiethnic population with three main races; Malay, Chinese and Indian. However, Kelantan is a predominantly Malay village in the north-eastern state of the Peninsular. This perhaps explains the preponderance of Malay participants in our study. From our findings, the NPDR group had poor glycemic control. This was similar to other studies done at various places (26,27).

There are several studies that have linked serum uric acid with the pathogenesis and progression of diabetic retinopathy (13,28). In our study, total serum uric acid was higher in

no diabetic retinopathy group, but not statistically significant. The serum uric acid was significantly higher in patients with $HbaA1_C$ < 6.5%. This in accordance with previous studies that stated elevated serum uric acid levels during the early stage of impaired glucose metabolism is said to predict the onset of the type 2 diabetes and has been linked to both micro and macrovascular complications (29,30). Bonakdaran S et al (31), noted a significant correlation between hyperuricemia and $HbA1_C$. Dehghan A et al (16) reported high serum uric acid level precedes hyperinsulinemia and diabetes inducing endothelial dysfunction and oxidative stress.

Two studies involving type 2 diabetic patients were conducted in China, showed serum uric acid levels correspond to the severity of diabetic retinopathy (32,33). The serum uric acid level was showed to increase gradually with increase of severity of diabetic retinopathy. Causevic A et al (34) reported that the serum uric acid level was increased in type 2 diabetes mellitus and associated with insulin resistant syndrome. Moreover, Ashakiran S. et al (35), noticed that the compensatory hyperinsulinemia (insulin resistant) showed an antiuricosuric effect on the kidneys which lead to increase serum uric acid level.

In contrast, Nan H et al (36) mentioned that the serum uric acid and fasting plasma glucose level increases in non-diabetic individuals but showed lower level in diabetic patient. However, Olukoga AO et al (37) and Segato T et al (38), reported no significant association between serum uric acid and diabetic retinopathy. Pfister R et al (39), tested eight common genetic variants which were identified as determinant of serum uric acid level on diabetic patients involving a large cohort of European descent. Their results do not support the association of serum uric acid in the development of type 2 diabetes mellitus.

The male gender had higher level of serum uric acid compared to the female gender. We also observed, the mean serum uric acid in both the gender were statistically significant in no diabetic retinopathy than in NPDR. Our findings were in parallel to another study that showed serum uric acid level was much higher in male than female (40). Evidence regarding estrogen promotes uric acid excretion (41) supports the findings of hyperuricemia among the males. Choi H.K et al (41), reported that serum uric acid was significantly higher in diabetic male than female. However, this sex predilection it's still controversial. In contrary Causevic A et al (34), demonstrated there is no effect of gender on serum uric acid levels in diabetic patient.

As we know, there are various studies that regards diabetic retinopathy as a neurovascular disease whereby the retinal neurodegeneration antedates the microvascular abnormalities (4,42). The retinal nerve fibre loss was contributed by retinal ganglion cell death and axonal degeneration. In our study, the mean RNFL thickness in all the 4 quadrants were thinner in no diabetic retinopathy compared to NPDR. Only the temporal quadrant was statistically significant. Our findings were parallel to some studies (26,42) and in contrast to few other studies (43,44). All 9 subfields of the macular region showed similar changes as the RNFL. However, only 3 subfields were statistically significant (superior outer, inferior outer and temporal outer). These results of our studies were consistent to some studies (13,45) and contradictory to few other studies (46,47).

Several studies, including ours, showed presence of neuronal abnormalities at the early stage of diabetes irrespective of the type of OCT used (25,48-49). This explains the thinner retina in no diabetic retinopathy and the increase vascular permeability leads to increased retinal thickness. In our study, we have pooled all severity of NPDR grades, gender and age. Similar to findings found by other studies (25,49), our study made an assumption that increase in the

retinal thickening observed in the NPDR group was due to the insult from hard exudates and retinal hemorrhages leading to accumulation of intra-retinal fluids. This subsequently results in an increased thickness of the RNFL. Although we have excluded patients with clinically apparent and extensive retinal oedema, a possible presence of subclinical retinal oedema in our diabetic patients can interfere the OCT measurement. Subclinical macular oedema in which clinically shows absence of macular oedema on slit lamp examination but on OCT shows abnormally increased macular thickness (49).

We found that only the temporal RNFL quadrant showed a mean significant difference. The superior RNFL quadrant also showed a change in the retinal thickness between the no diabetic retinopathy and NPDR groups however it was not significant. The inferior RNFL quadrant showed a minimal change which is almost negligible between the two groups. In the macular thickness, only the outer most ring including the superior, temporal and inferior subfield showed a significant difference. There are several studies that observed the superior and temporal quadrants to be thinner than the inferior quadrants (50,51) showing that the early events of diabetic retinal disease (microaneurysms and acellular capillaries) occur preferentially in the superior temporal quadrant rather than in inferior areas. Chung et al (52) demonstrated that blood flow in the superior temporal retina increased in response to hypercapnia, but did not decrease in response to hyperoxia. In contrast, hyperoxia led to a decrease in blood flow to the inferior retina, whereas hypercapnia did not result in an increased blood flow within this area (52). The lack of normal vasoconstrictor response in this superior quadrant could explain why this region is more susceptible to micro aneurysms and acellular capillaries in diabetes mellitus and also why the retinal fibres are preferentially lost in this region even before clinically detectable diabetic retinopathy (53). The superior quadrant was more susceptible to undergoing damage compared with other areas and may have a tendency

for higher rates of cell death, which results in RNFL thinning (26). We also noticed that the thickest RNFL in nasal quadrant might be due to the lack of micro aneurysm presence in this area and therefore less RNFL damage occurred in this quadrant. We assume the differences in the vascular hemodynamics in the macular region might be also the contributing factors to these phenomena of asymmetrical retinal thickness.

Serum uric acid pathogenesis, has been associated with the development and worsening of the diabetic retinopathy (31). Chien KL et al (54) and Ishizaka N et al (55), suggested that increase level of serum uric acid acts as a predictor for diabetic vascular complications. Oxidative stress is a culprit for the progression of diabetic retinopathy (29). Serum uric acid conversely can act as an antioxidant (31). However, Jianfei Xia et al (56) demonstrated that there was significantly increased concentration of uric acid among diabetic patients and serum uric acid might be a risk factor for diabetic retinopathy. High serum uric acid level was said as a risk factor of type 2 diabetes mellitus. Lowering the serum uric acid with xanthine oxidase inhibitors can reduce the incidence of type 2 diabetes mellitus and its complication but this is still controversial. Anju G et al (57) in her studies concluded that serum uric acid levels increase in newly diagnosed diabetic patients, thus it can serve as potential biomarker of the glucose metabolism.

We did a correlation between the serum uric acid with RNFL thickness and macular thickness among no diabetic retinopathy and NPDR. Based on our Medline research, our study is the first study to be conducted. Uric acid inspite being an antioxidant in the circulation, it induces oxidative stress in the vascular endothelial cells, thus mediating progression of disease related to diabetic. Oxidative stress is believed to play an important role in the development of vascular complications in type 2 diabetes mellitus (21).

From our study, we did not find any momentous correlation between the RNFL and macular thickness with serum uric acid in both the groups. However, in patient with no diabetic retinopathy, only the inferior outer macular subfield which had poor negative correlation with serum uric acid showed a statistically significant relationship. This negative relationship indicates that the higher serum uric acid level, the patient is likely to have minimal thinning of the macular region but the relationship is at a lower rate. This has to be interpreted cautiously as only one macular subfield showed a significant relationship. From our findings, we postulate that, in no diabetic retinopathy group, the serum uric acid level is higher which is said to precede the type 2 diabetes, and inversely causes thinning of the macular region secondary to retinal neurodegeneration.

In patient with NPDR, only the central subfield and inferior inner macula showed a fair positive correlation with a significant relationship. This positive significant relationship indicates that the higher serum uric acid level, the patient is likely to have thicker macular but the relationship is at a lower rate. From our findings, we postulate that, in NPDR group, there will be high likely for nephropathy to set in, thus affecting the renal excretion, increases rates of renal reabsorption, increases the excretion of purine metabolism subsequently raises the production of uric acid. The increase in serum uric acid level via the inflammatory process results in progression of diabetic vascular complication, thus causing vascular leakage and indirectly causing an impact to the macular thickness especially in the posterior pole.

There are several limitations found in our studies. The pooling of the severity of NPDR would have led to potential spurious comparisons. Other limitations include not taking into account the duration of diabetes, no follow up given, distribution of race and gender were not equal.

We also did not include parameters for kidney dysfunction or diabetic nephropathy that could alter the excretion of uric acid.

3.7 CONCLUSION

Serum uric acid showed a poor correlation with RNFL and macular thickness among diabetic patients. A large population cohort study with well distributed ethnicity and gender is needed to observe if there is presence of correlation between serum uric acid with RNFL and macular thickness among type 2 diabetic patients.

3.8 REFERENCES

1. Hu FB. Globalization of diabetes: the role of diet, lifestyle, and genes. *Diabetes Care*. 2011;34(6):1249-1257.

2. Guariguata L, Whiting D, Weil C, Unwin N. The International Diabetes Federation Diabetes Atlas methodology for estimating global and national prevalence of diabetes in adults. *Diabetes Res Clin Pract*. 2011; 94(3):322-332.

3. Tarr JM, Kaul K, Chopra M, Kohner EM, Chibber R. Pathophysiology of diabetic retinopathy. *ISRN Ophthalmol*. 2013;2013:343560.

4. Barber AJ, Gardner TW, Abcouwer SF. The significance of vascular and neural apoptosis to the pathology of diabetic retinopathy. *Invest Ophthalmol Vis Sci*. 2011;52(2):1156-1163.

5. Kern TS, Barber AJ. Retinal ganglion cells in diabetes. *J Physiol*. 2008;586(18):4401-4408.

6. Bronson-Casstain KW, Bearse MA Jr, Neuville J, Jonasdottir S, King-Hooper B, Barez S, et al. Adolescent with type 2 diabetes: early indications for focal retinal neuropathy, retinal thinning, and venular dilation. *Retina*. 2009;29(5):618-626.

7. Bearse MA Jr, Adams AJ, Han Y, Schneck ME, Ng J, Bronson-Castain K, et al. A multifocal electroretinogram model predicting the development of diabetic retinopathy. *Prog Retin Eye Res*. 2006;25(5):425-448.

8. Jackson GR, Scott IU, Quillen DA, Walter LE, Gardner TW. Inner retinal visual dysfunction is a sensitive marker of non-proliferative diabetic retinopathy. *Br J Ophthalmol*. 2012;96(5):699-703.

9. Nakamura M, Barber AJ, Antonetti DA, LaNoue KF, Robinson KA, Buse MG, et al. Excessive hexosamines block the neuroprotective effect of insulin and induce apoptosis in retinal neurons. *J Biol Chem*. 2001;276(47):43748-43755.

10. Lattanzio R, Brancato R, Pierro L, Bandello F, Iaccher B, Fiore T, et al. Macular thickness measured by optical coherence tomography (OCT) in diabetic patients. *Eur J Ophthalmol*. 2002;12(6):482-487.

11. S N, N K, S A, Dayanand CD. The association of hypomagnesia, high normal uricemia and dyslipidemia in the patients with diabetic retinopathy. *J Clin Diagn Res*. 2013;7(9):1852-1854.

12. Xia J, Wang Z, Zhang F. Association between related purine metabolites dan diabetic retinopathy in Type 2 Diabetic Patients. *Int J Endocrinol*. 2014;2014:651050.

13. Kushiyama A, Tanaka K, Hara S, Kawazu S. Linking uric acid metabolism to diabetic complications. *World J Diabetes*. 2014;5(6):787-795.

14. Griffiths M. The mechanism of the diabetogenic action of uric acid. *J Biol Chem*. 1950;184(1):289-298.