A Dissertation on

MEAN PLATELET VOLUME - CORRELATION WITH HbA1C AND ITS ASSOCIATION WITH MICROVASCULAR COMPLICATIONS IN TYPE II DIABETES MELLITUS CHENNAI – 600 001.

Submitted to

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M.D. BRANCH - I

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MAY 2018

CERTIFICATE BY THE INSTITUTION

This is to certify that **Dr. ANJU ROSE GEORGE,** Post - Graduate Student (May 2015 TO May 2018) in the Department of General Medicine STANLEY MEDICAL COLLEGE, Chennai- 600 001, has done this dissertation on "**MEAN PLATELET VOLUME- CORRELATION WITH HbA1C AND ITS ASSOCIATION WITH MICROVASCULAR COMPLICATIONS IN TYPE II DIABETES MELLITUS"** under my guidance and supervision in partial fulfillment of the regulations laid down by the Tamilnadu Dr. M. G. R. Medical University, Chennai, for M.D. (General Medicine), Degree Examination to be held in April 2017.

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DECLARATION

I, **Dr. ANJU ROSE GEORGE,** declare that I carried out this work on "**MEAN PLATELET VOLUME- CORRELATION WITH HbA1C AND ITS ASSOCIATION WITH MICROVASCULAR COMPLICATIONS IN TYPE II DIABETES MELLITUS**" at the outpatient and Medical wards of Government Stanley Hospital. I also declare that this bonafide work or a part of this work was not submitted by me or any other for any award, degree, or diploma to any other university, board either in India or abroad.

This is submitted to The Tamilnadu DR. M. G. R. Medical University, Chennai in partial fulfilment of the rules and regulation for the M. D. Degree examination in General Medicine.

DR. ANJU ROSE GEORGE

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CONTENTS

ABBREVIATIONS

T2DM - Type 2 Diabetes Mellitus

HbA_{1c} - Glycosylated hemoglobin

MPV - Mean Platelet Volume

TGL - Triglyceride

HDL - High Density Lipoprotein

LDL - Light Density Lipoprotein

AGE - Advance Glycosylation Endproducts

DCCT - Diabetes Control and Complication Trial

UKPDS - United Kingdom Prospective Diabetes Study

ACCORD-Eye - Action to control cardiovascular risk in diabetes eye subgroup

ADVANCE - The Action in Diabetes and Vascular Disease: Preterax and Diamicron modified release-controlled evaluation

UAE - Urinary Albumin Excretion

MA - Microalbuminuria

INTRODUCTION

Diabetes mellitus (DM) is a global pandemic[1]. It is the most common group of metabolic disorder characterized by chronic hyperglycemia associated with secondary damage in multiple organ systems especially kidneys, eyes, peripheral nerves and blood vessels.

Increased platelet activation has been suggested to be involved in the pathogenesis of vascular complications[22]. It is being found that MPV values are high in patients with diabetes mellitus, more so in uncontrolled diabetes. Platelet volume, a marker of the platelet function and activation, is proposed as to be involved as a causative agent with respect to altered platelet morphology and function. The higher the MPV, the larger and younger the platelets are and more is the risk for thrombosis and are associated with increased risk for hyperglycemic complications.

Mean platelet volume (MPV), an important, simple, effortless, and cost-effective tool measured by hematology analyzer assess the volume and function of platelets and thus has potential to be used as indicator of presence of vascular complications .

REVIEW OF LITERATURE

Type 2 diabetes mellitus (T2DM) is a chronic disease which is posing as one of the major public health problems facing mankind[2]. Estimates by the International Diabetes Federation (IDF) states that 387 million people have diabetes worldwide in 2014 and by 2035 this number is expected to rise to 592 million. China (98.4 million) and India (65.1 million) were the countries topping the list with the largest number of individuals with diabetes in 2013. T2DM accounts for 90% of cases of diabetes globally.

Diabetes mellitus - worldwide prevalence. Regional estimates of the number of individuals with diabetes (20–79 years of age) are shown (2013)

T2DM occurs in genetically predisposed persons once they are exposed to a series of environmental influences which trigger the onset of clinical disease. Ethnic background, sex and age are important factors which determine the risk of development of T2DM[4]. Reduced secretion of insulin, decreased utilization of glucose and increased production of glucose contribute to hyperglycemia in DM depending upon the etiology. Diabetes Mellitus is a state of metabolic dysregulation and can lead to secondary pathophysiologic changes in various organ systems leading to microvascular and macrovascular complications[2]. This will impose a remarkable burden on the diabetics as well as on the health care system. Therefore it is of utmost importance to detect the complications early in a cost effective way to control and treat them. DM will be likely a leading cause of morbidity and mortality in the future.

EPIDEMIOLOGIC DETERMINANTS AND RISK FACTORS OF TYPE 2 DM[2]

GENETIC FACTORS:

- Genetic markers
- Family history
- “Thrifty genes”

DEMOGRAPHIC CHARACTERISTICS:

- Sex
- Age (>45 years)
- Ethnicity (e.g., Asian American, Latino, Native American, African American, Pacific Islander)

BEHAVIORAL AND LIFESTYLE-RELATED RISK FACTORS:

- Obesity (including duration and distribution of obesity)
- Physical inactivity
- Diet
- Stress
- Westernization, urbanization, modernization

METABOLIC DETERMINANTS AND INTERMEDIATE-RISK CATEGORIES OF TYPE2 DIABETES[2]

- Impaired glucose tolerance
- Insulin resistance
- Pregnancy-related determinants
- Parity
- Gestational diabetes
- Diabetes in offspring of women with diabetes during pregnancy

– Intrauterine malnutrition or overnutrition

ETIOLOGIC CLASSIFICATION OF DIABETES MELLITUS[1]

I. Type 1 diabetes (absolute insulin deficiency resulting from beta cell destruction)

A. Immune-mediated

B. Idiopathic

II. Type 2 diabetes (may have predominant insulin resistance with relative insulin deficiency or predominant insulin secretory defect with insulin resistance)

III. Other specific types of diabetes

A. Genetic defects of beta cell development or function characterized by mutations in:

1. Hepatocyte nuclear transcription actor (HNF) 4α - MODY 1

2. Glucokinase - MODY 2

3. HNF-1α - MODY 3

4. Insulin promoter factor-1 (IPF-1) - MODY 4

5. HNF-1β - MODY 5

6. NeuroD1 - MODY 6

7. Mitochondrial DNA

8. Subunits of ATP-sensitive potassium channel

9. Proinsulin or insulin

10. Other pancreatic islet regulators/proteins such as KLF11, PAX4, BLK, GATA4, GATA6, RFX6, GLIS3, SLC2A2 (GLUT2).

B. Genetic defects in insulin action

1. Type A insulin resistance

2. Leprechaunism

3. Rabson-Mendenhall syndrome

4. Lipodystrophy syndromes

C. DISEASES OF THE EXOCRINE PANCREAS—pancreatitis, pancreatectomy, neoplasia, fibrocalculous pancreatopathy, cystic fibrosis, hemochromatosis, mutations in carboxyl ester lipase

D. ENDOCRINOPATHIES — acromegaly, Cushing's syndrome, glucagonoma, pheochromocytoma, somatostatinoma, hyperthyroidism, aldosteronoma.

E. Drug or chemical induced—glucocorticoids, vacor (a rodenticide), pentamidine, nicotinic acid, diazoxide, β-adrenergic agonists, thiazides, calcineurin and mTOR inhibitors, hydantoins, asparaginase, α-intereron, protease inhibitors, antipsychotics (atypicals and others), epinephrine.

F. INFECTIONS—congenital rubella, cytomegalovirus, coxsackievirus.

G. Uncommon forms of immune-mediated diabetes -"stif-person" syndrome, anti- insulin receptor antibodies.

H. Genetic syndromes sometimes associated with diabetes—Wolfram's syndrome, Down's syndrome, Klinefelter's syndrome, Turner's syndrome, Friedreich's ataxia, Huntington's chorea, Laurence-Moon-Biedl syndrome, myotonic dystrophy, porphyria, Prader-Willi syndrome

IV. Gestational diabetes mellitus

CRITERIA FOR THE DIAGNOSIS OF DIABETES MELLITUS[3]

• Symptoms of diabetes with random blood glucose concentration ≥200 mg/dL

or

• Fasting plasma glucose ≥1 2 6 m g / d L

or

• Haemoglobin A1c ≥6.5%

or

• 2-h plasma glucose ≥ 200 mg/dL during an oral glucose tolerance test

SCREENING FOR TYPE 2 DIABETES MELLITUS - MAJOR RECOMMENDATIONS[3]

Screening to detect T2DM and assessment of future risk for diabetes is to be considered in asymptomatic adults who are overweight or obese (BMI ≥25 kg/m2or ≥23 kg/m2 in Asian Americans) and who possess one or more additional risk factors for diabetes.

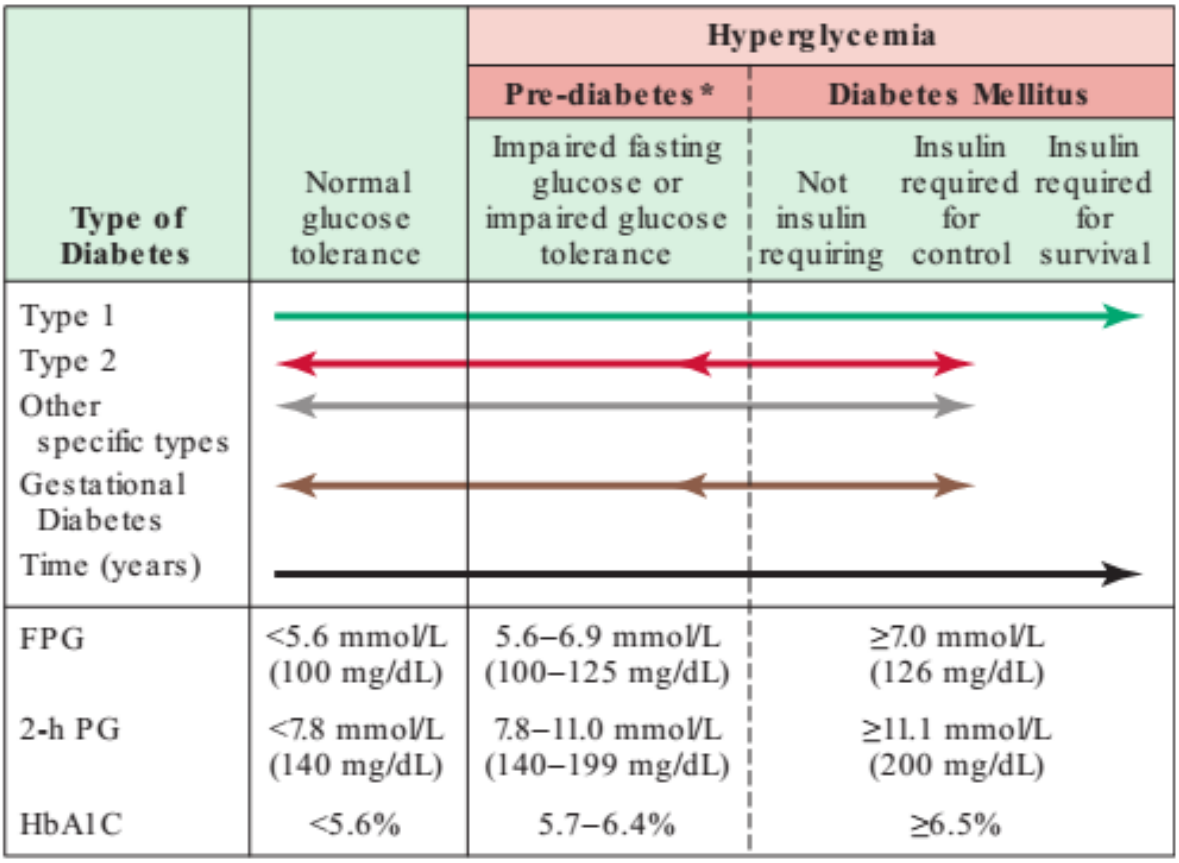
• Testing should begin at the age of 30-45 yr, in those without risk factors for T2DM

• Repeat testing is to be carried out at 3- to 5-yr intervals, if the test results are normal.

• Any of the test is appropriate: HbA1c, FPG, 2-hr 75-g OGTT.

• Identify and treat other CVD risk factors, in those who are found to have increased risk for future diabetes.

SPECTRUM OF GLUCOSE HOMEOSTASIS AND DM



American Diabetes Association, 2014.

PATHOGENESIS OF TYPE 2 DIABETES MELLITUS[3]

Interaction of certain genetic abnormalities with adverse environmental factors results in the development of type 2 diabetes.

GENETIC FACTORS

There is a 70 - 90% concordance of type 2 DM in between identical twins. Increased risk of type 2 DM is observed among individuals with a parent with type 2 DM. The risk approaches 40% if both the parents have type 2 DM. It is interesting to note that the in utero environment also plays a significant role in the development of this disease. Either increased or reduced birth weight can increase the risk of type 2 DM in adult life.

The various genes that predispose to type 2 DM are not completely identified. Most prominent gene is a transcription factor 7- like 2 gene variant; this has been associated with type 2 DM in numerous populations and with impaired glucose tolerance (IGT) in one population at high risk of diabetes. The genes encoding the inward rectifying potassium channel, peroxisome proliferator–activated receptor γ, zinc transporter, IRS, and calpain 10 also revealed genetic polymorphisms associated with type 2 DM. The exact mechanisms by which these genetic loci lead to increased susceptibility to type 2 DM are still not clear; but most are believed to alter islet cell function or development or secretion of insulin.

ENVIRONMENTAL FACTORS

Well recognised environmental factors leading to development of type 2 diabetes are physical inactivity and excessive caloric intake. These factors can either independently produce the disease in a genetically susceptible person, or can do so via production of obesity and metabolic syndrome. Visceral or central obesity (as evidenced by the waist- hip ratio), is very common in type 2 DM (≥80% are obese). Metabolic syndrome occurs when type 2 DM and obesity occur together with other anomalies like high triglycerides, low HDL-cholesterol and hypertension. Estimates suggest that 25% to 30% of the Indian population suffers from the metabolic syndrome.

Type 2 DM is characterized by insulin resistance, impaired secretion of insulin, excessive glucose production from liver and abnormal fat metabolism. Glucose tolerance remains near-normal in the early stages of the disease despite insulin resistance, as the pancreatic beta cells try to compensate by increasing insulin output. The pancreatic islets in certain individuals will be unable to sustain the hyperinsulinemic state as compensatory hyperinsulinemia and insulin resistance progress. The person then develops elevations in postprandial glucose, characteristic of IGT. Overt diabetes with fasting hyperglycemia results with further decline in insulin secretion and an increase in hepatic glucose production. Beta cell failure ensues ultimately.

METABOLIC ABNORMALITIES

Abnormal muscle and fat metabolism:

The reduced ability of insulin to act effectively on target tissues (especially fat, liver and muscle); insulin resistance, is the prominent feature of type 2 DM. The combination of obesity and genetic susceptibility results in development of insulin resistance. This leads to impaired glucose utilization by insulin-sensitive tissues and increased hepatic glucose output. Both effects lead to the hyperglycemia. Increased FPG levels is predominantly the result of increased hepatic glucose output, on the other hand, decreased peripheral glucose utilization accounts for postprandial hyperglycemia. There is a greater impairment in glycogen formation (nonoxidative glucose usage) than in glycolysis (oxidative glucose metabolism) in skeletal muscle. In type 2 DM, glucose metabolism is not altered in insulin independent tissues.

Phosphorylation/ dephosphorylation regulated by insulin appear to play the major role in insulin resistance. There will be accumulation of lipid within skeletal myocytes, which can lead to impairment of mitochondrial oxidative phosphorylation and reduction in insulin-stimulated ATP production in mitochondria. Generation of reactive oxygen species such as lipid peroxides results from impaired fatty acid oxidation and accumulation of lipid within the skeletal myocytes.

Obesity, particularly in a central or visceral location, accompanying type 2 DM, is thought to be a part of the pathogenic process. The increase in adipocyte mass leads to increase in circulating levels of free fatty acids and other fat cell products. Adipocytes secrete numerous biologic products (nonesterified free fatty acids, leptin, NF-α, retinol-binding protein 4, resistin, IL-6 and adiponectin). The free fatty acids impair glucose utilization in skeletal muscle, impair beta cell function and promote glucose production by the liver. The production of an insulin-sensitizing peptide by adipocytes; adiponectin, in contrast, is reduced in obesity, and this might contribute to hepatic insulin resistance. Adipokines and adipocyte products may also produce an inflammatory state.

Impaired insulin secretion

Initially insulin secretion increases in response to insulin resistance in type 2 DM to maintain normal glucose tolerance. Hence, the insulin secretory defect is initially mild and involves glucose stimulated insulin secretion only, including a great reduction in the first secretory phase.

The decrease in Beta cell mass in individuals with long-standing type 2 DM is by approximately 50%. In individuals with long-standing type 2 DM, the amyloid fibrillar deposit found in the islets is formed by amylin or Islet amyloid polypeptide, co-secreted by the beta cell. Chronic hyperglycemia, in turn,

paradoxically leads to impaired islet function ("glucose toxicity") and causes worsening of hyperglycemia[18]. Glycemic control improvement is often associated with improved islet function. In addition, increased dietary fat and elevation of free fatty acid levels ("lipotoxicity") may also worsen islet function. Reduced GLP-1 action may also play a role in the reduction of insulin secretion.

Increased hepatic glucose and lipid production:

In the liver, insulin resistance reflects the failure of hyperinsulinemia to suppress gluconeogenesis, which results in decreased glycogen storage by the liver in the postprandial state and in fasting hyperglycemia. Insulin resistance in adipose tissue leads to increase in lipolysis as well as free fatty acid flux from adipocytes, which in turn leads to increased lipid (triglyceride and very-low-density lipoprotein[VLDL]) synthesis in hepatocytes[12]. Thus there is lipid storage or steatosis in the liver which may lead to nonalcoholic fatty liver disease (NAFLD) and liver function test abnormalities. This is another factor causing the dyslipidemia found in type 2 DM.

INSULIN RESISTANCE SYNDROMES

Also called as syndrome X, the metabolic syndrome and the insulin resistance syndrome[8]

This includes a spectrum of metabolic derangements which includes insulin resistance, dyslipidemia (decreased HDL and elevated triglycerides), hypertension, central or visceral obesity, IGT /IFG or type 2 DM and accelerated cardiovascular disease. In adults, two distinct severe insulin resistance syndromes have been described:

(1) Type A: It affects young females and is characterized by severe hyperinsulinemia, features of hyperandrogenism and obesity.

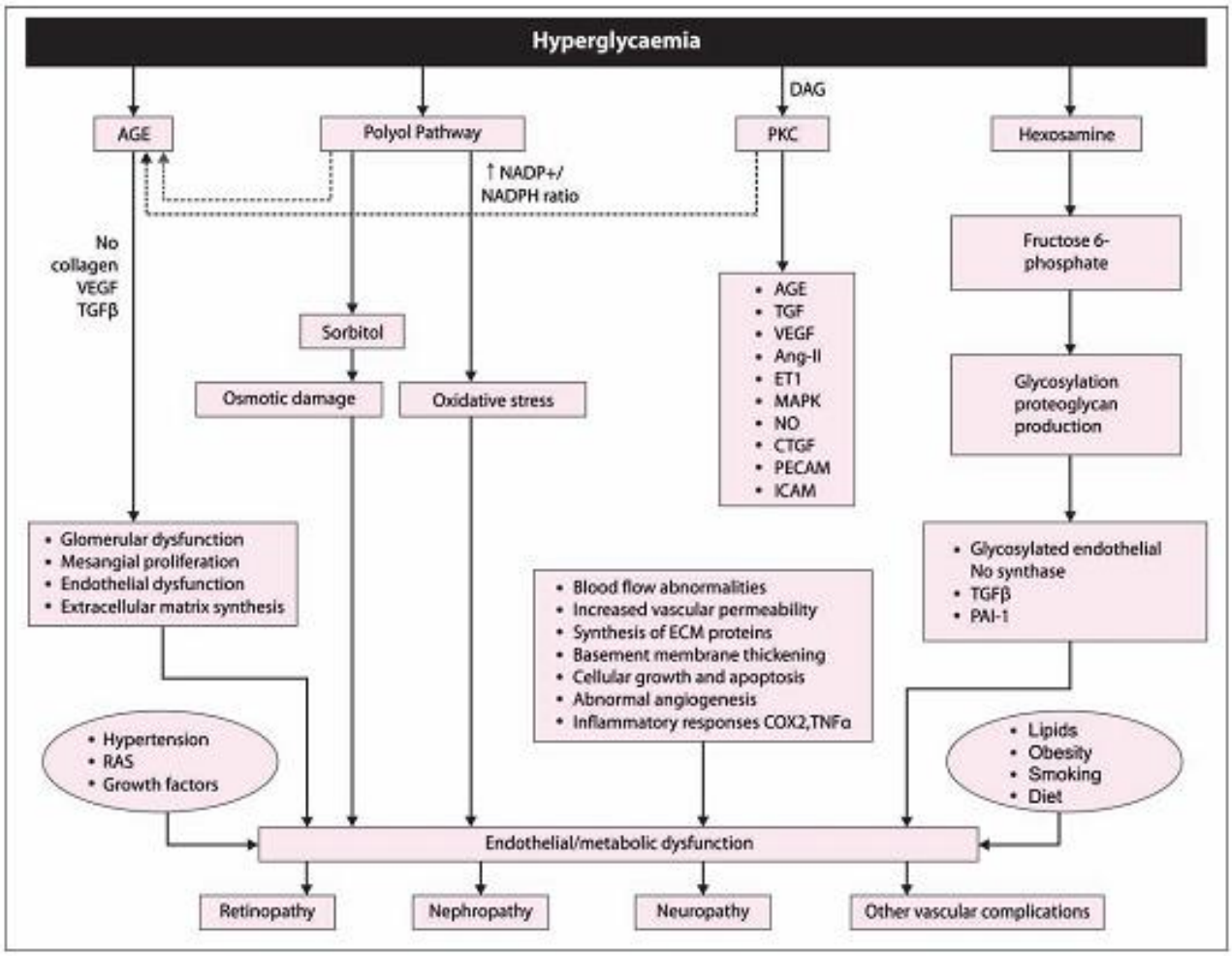
(2) Type B: It affects middle-aged women. They are found to have autoantibodies directed at the insulin receptor. These receptor autoantibodies may stimulate the insulin receptor, hence leading to intermittent hypoglycaemia or may block insulin binding. It is characterized by features of hyperandrogenism, severe hyperinsulinemia and autoimmune disorders.

CHRONIC COMPLICATIONS OF HYPERGLYCAEMIA

Complications are divided into: (a) Microvascular Complications - diabetic retinopathy, neuropathy and nephropathy.

b) Macrovascular Complications - coronary artery disease (CAD), cerebrovascular stroke and peripheral artery disease (PAD).

Chronic hyperglycemia plays a major role in the development of these complications but some individuals may possess genetic susceptibility for development of particular complications. Large randomised clinical trials conducted among individuals with type-1 or type-2 diabetes have demonstrated conclusively that a reduction in chronic hyperglycaemia delays or prevents nephropathy, neuropathy and retinopathy.



Pathways leading to microvascular diseases in diabetes mellitus

PATHOGENESIS OF MICROVASCULAR DISEASES[4]

The exact mechanism behind the development of such diverse organ and cellular dysfunction is unknown. Four major theories have been proposed.

1) **AGES THEORY:** elevated intra-cellular glucose causes interaction of glucose with amino acid groups on proteins thus resulting in nonenzymatic glycosylation of intra- and extra-cellular proteins. Hence, it results in formation of advanced glycosylation end products (AGEs). Non-enzymatic glycation products are reversible in the early stages. With continuing hyperglycaemia, poorly reversible intermediate products are formed and irreversible AGEs are produced later. Serum levels of AGEs increases with the level of hyperglycaemia. These products mount up with decline in glomerular filtration rate. AGEs have been found to crosslink proteins (e.g. extracellular matrix protein, collagen), promote glomerular dysfunction, accelerate atherosclerosis, induce endothelial dysfunction, reduce nitric oxide (NO) synthesis and alter composition and structure of extra-cellular matrix. AGEs have receptors in mesangial cells, interaction with which leads to increase in expression of transforming growth factor-β(TGF-β) and synthesis of extra-cellular matrix.

2**) SORBITOL PATHWAY (POLYOL PATHWAY)**[15]

Glucose in the cell is metabolised pre-dominantly by phosphorylation and subsequent glycolysis. Aldose reductase enzyme converts excess glucose to

sorbitol when the level of glucose exceeds. Increase in sorbitol concentration is believed to alter the redox potential, generates reactive oxygen species, increases cellular osmolality and increases AGEs formation, and hence likely lead to other types of cellular dysfunction. Retinopathy, cataract formation and neuropathy have been linked to this pathway, to some extent. Newer aldose reductase inhibitors like epelrestat have been approved for use. Fidarestat is undergoing clinical trials.

3) PROTEIN KINASE C PATHWAY

There is increased formation of diacylglycerol in the context of Hyperglycaemia, which leads to protein kinase C (PKC) activation. PKC belongs to a family of serine threonine kinases that can lead to alteration in the transcription of genes for type IV collagen, fibronectin, extracellular matrix proteins and contractile proteins in neurons and endothelial cells. Hence, diverse vascular functions like blood flow, contractility, cellular proliferation and vascular permeability is believed to be regulated by PKC. PKC also mediates angiotensin-II, TGF-β1and vascular endothelial growth factor (VEGF). PKC, in addition, modulates mitogen activated proteinkinase (MAPK), which further mediates sclerosis.

4) HEXOSAMINE PATHWAY

Hexosamine pathway, generates fructose-6-phosphate which serves as a substrate for O-linked glycosylation and proteoglycan production. Hyperglycaemia causes increased flux through the hexosamine pathway. The proteoglycans may alter function by glycosylation of proteins like endothelial nitric oxide synthase, or by causing changes in gene expression of plasminogen activator inhibitor-1 (PAI-1) or transforming growth factor β(TGF-β).

In DM related complications, growth factors appear to play a major role and most of these proposed pathways increase their production. In diabetic proliferative retinopathy; VEGF-A is increased locally and after laser photocoagulation, their level decreases. Reduction of VEGF is also caused by inhibition of angiotensin-II, which could thus be one the basis for the beneficial effects of angiotensin II receptor blockers on microangiopathic diseases.

In diabetic nephropathy; TGF-β is increased and stimulation of production of collagen by basement membrane and of fibronectin by mesangial cells. Other growth factors have been suggested to play a role in DM related complications are platelet derived growth factor, insulin like growth factor, epidermal growth factor, basic fibroblast growth factor, growth hormone, connective tissue growth factor and even insulin.

ANGIOTENSIN-II-

In diabetes, abnormal activation in the renin-angiotensin system (RAS) occurs in the kidney. Direct binding of Angiotensin-II to receptors in renal cells occur and this causes deposition of matrix in mesangial and tubular cells via TGF-β1. Angiotensin-II causes stimulation of VEGF production in mesangial cells and results in impairment of glomerular structure and function. In the retina also, similar effects are observed.

A definitive proof showing that a reduction in chronic hyperglycaemia can lead to prevention of many early complications of type 1 DM was obtained from the diabetes control and complications trial (DCCT)[32]. This was a large multicentre clinical trial which randomised 1,400 type 1 diabetics to either conventional or intensive diabetes management and evaluated prospectively for the development of nephropathy, retinopathy and neuropathy. A substantially lower HbA1c (7.3%) was achieved by the intensive group rather than the conventionally managed group (9.1%). DCCT also showed that a reduction in microalbuminuria by 39% and clinical nephropathy by 54%, non-proliferative and proliferative retinopathy by 47% and neuropathy by 60% resulted from improvement of glycemic control. The benefit of the improved glycaemic control achieved during the DCCT persisted even after the study concluded and the glycaemic control worsened.

The United Kingdom Prospective Diabetes Study (UKPDS) demonstrated that a 35% reduction in microvascular complications was achieved with each percent point decrease in HbA1c. Improvement in lipoprotein risk profiles such as increased HDL and reduced triglycerides resulted with improved glycaemic control. A significant finding of this study was that strict blood pressure control considerably reduced both macrovascular and microvascular complications. The favourable effects of blood pressure control were found to be greater than those of control of glycaemic status.

A trial on 4065 type 2 diabetics from 77 centres over Canada and US; Action to control cardiovascular risk in diabetes eye subgroup (ACCORD-Eye) trial showed with intensive glycaemic control and lipid control especially with fenofibrate lead to a 40% reduction in retinopathy within a short period of 4 years.

Another large randomised-controlled trial was conducted at 215 centres in 20 countries in Asia, Europe, Australia and US; The Action in Diabetes and Vascular Disease: Preterax and Diamicron modified release-controlled evaluation (ADVANCE) trial. This involved 11140 type2 diabetics and they were studied over 5 years. This study demonstrated a 21% reduction in new onset microalbuminuria and reduction in occurrence of new or worsening nephropathy, with intensive glycaemic control (HbA1c 6.5%) as opposed to standard glycaemic control (HbA1c 7.3%). These two large trials also prove the

importance of control of glycaemic status in the prevention of microvascular disease among diabetics.

OTHER RISK FACTORS

HYPERTENSION causes dysfunction of endothelium, impaired availability of NO, renal vasoconstriction, decrease in glomerular filtration, impairment in tubuloglomerular feedback, decrease in medullary flow and worsened pressure natriuresis and progressive proteinuria[40]. This causes rise in intraglomerular pressure and leads to a series of changes in matrix. Thus hypertension is an important determinant of macroangiopathy and microangiopathy in diabetes. Along with glycaemic control, strict lowering of blood pressure with angiotensin receptor blockers (ARBs) or ACE-inhibitors (ACE-I) causes reduction in intraglomerular pressure and blockage of the RAS. This has been shown to result in reduced incidence of nephropathy.

DYSLIPIDAEMIA has been believed to result in increase in urinary excretion of albumin and causes damage to the endothelium by oxidative stress in diabetics. In large multicentric trials, improvement in retinopathy and maculopathy was shown with treatment of dyslipidemia with atorvastatin [collaborative atorvastatin diabetes study (CARDS)] and fenofibrate [fenofibrate intervention in event lowering in diabetes (FIELD)].

OBESITY: Non-esterified fatty acids (NEFAs), leptin, tumour necrosis factor-α (TNF-α) and adiponectin are mediators, directly secreted by adipocytes. These modulate microvascular changes, endothelial function, NO production and inflammation and thus contribute to microangiopathy.

SMOKING: Induces oxidative stress, endothelial damage and vasoconstriction and thus aggravates microangiopathy. DIETS containing excessive amounts of trans and saturated fats result in oxidative stress. In clinical studies, targeting hyperglycaemia, hypertension, dyslipidemia, obesity, smoking, diet and lifestyle modification is demonstrated to have definitive benefit in microangiopathic disease.

DIABETIC KIDNEY DISEASE (DKD)

Excessive urinary excretion of albumin (UAE) followed by loss of kidney function is the characteristic feature of DKD[16]. The hallmark of DKD is proteinuria. Classical definition of DKD is the presence of persistent albuminuria (≥300 mg/24 hours or ≥200 μg/min) or proteinuria ≥0.5 gm/24 hours with the fulfilment of one of the following additional criteria: the absence of evidence (clinical or laboratory) of other kidney and renal tract disease and the presence of diabetic retinopathy. In type 2 diabetes, microalbuminuria i.e; persistent albuminuria in the range of 30 to 299 mg/24 hours, has been

demonstrated to be the marker for incidence of nephropathy as well as cardio vascular disease (CVD) risk and in type 1 diabetes,it is shown to be the earliest stage of diabetic nephropathy (DN).

Stages of Diabetic Nephropathy Based on Urinary Albumin Excretion

Stages	Urine with Marked Time (µg/min)*	24 hours Urine (mg/24 h)*	Random Urine Sample	
			Albumin Concentr-ation (mg/Lit)**	Albumin Concentr-ation Ratio (mg/g)*
Normoalbuminuria	<20	<30	<17	<30
Microalbuminuria	20 to 199	30 to 299	17 to 173	30 to 299
Macroalbuminuria	≥200	≥300	≥174	≥300

** Values according to the American Diabetic Association.*

AETIOPATHOGENESIS OF DKD

Increased glomerular filtration, inadequate tubular absorption and increased tubular secretion are believed to be important mechanisms responsible for development of proteinuria, with glomerular mechanism being the dominant one.

SCREENING AND DIAGNOSIS

For microalbuminuria (MA) screening, measurement of the albumin-to-creatinine ratio in an early morning or random spot collection (preferred method) may be performed. Within a 3 to 6 months period, two out of the three

specimens collected should be abnormal. In all adults with diabetes, measurement of Serum creatinine should be done at least annually, regardless of the UAE. Estimated GFR (eGFR) can then be estimated using the modifications of diet in renal disease (MDRD) study equation. The chronic kidney disease (CKD) may be graded into five grades based on the GFR.

Stages	Description	GFR (mL/ min/1.73 m^2)
1	Renal damage with GFR* normal increased	≥90
2	Renal damage with GFR* slightly decreased	60 to 89
3	GFR moderately decreased	30 to 59
4	GFR severely decreased	15 to 29
5	End-stage chronic renal failure	<15 or dialysis

MANAGEMENT OF DIABETIC KIDNEY DISEASE

Multiple risk factor interventional approach is believed to be the best treatment for DKD. The aim should be to retard the development or progression of DKD and to cause reduction in cardiovascular risk and mortality.

Hyperglycaemia is a major risk factor for the development of microalbuminuria. A 37% decrease in microvascular endpoints can be achieved from a reduction of haemoglobin A1c by 1%.

Arterial Hypertension is probably the best known relevant factor for the development and progression of DKD. Analysis of UKPDS showed that a 13% reduction in the risk of microvascular complications occurs with every 10 mm Hg reduction in systolic BP. The patients with systolic BP <120 mm Hg posses the smallest risk of complications. ARBs and ACEI should be used initially along with a diuretic, since they have known renoprotective effect. Even in normotensive subjects, they could used for the prevention of development of microalbuminuria. The use of ACE inhibitors and ARBs in addition to diminishing the risk for the development of microalbuminuria and macro-albuminuria, also contribute to the reduction in occurrence of cardiovascular events.

Dietary protein restriction has shown to slow the decline of renal function and reduce albuminuria.

Dyslipidaemia: there is no available data to show that the treatment of dyslipidaemia will lead to prevention of the development or progression of DKD. Anyhow for patients with DM, the desired of low density lipoprotein (LDL) target is <100 mg/ dL and <70 mg/dL when cardiovascular disease is present

Advanced renal disease in diabetic patients may require renal replacement therapy to reduce renal failure symptoms and restore their body homeostasis.

DIABETIC RETINOPATHY (DR)

It is a retinal microvasculopathy which results from prolonged hyperglycaemia. Increase in the duration of diabetes increases the prevalence of DR. After about 20 years, retinopathy develops in almost all patients with type 1 and in 60% with type 2 DM. In the Indian subcontinent, the prevalence of DR is 12% to 37% among type 2 DM. The major risk factors for the onset and progression of DR are duration of diabetes, degree of glycaemic control, hypertension, and hyperlipidaemia.

AETIOPATHOGENESIS

Hyperglycaemia-induced vascular injury heralds the onset of diabetic retinopathy (DR). Thickening of the basement membrane and loss of pericytes of the retinal capillaries are found to be the earliest histological lesions. Various biochemical, endocrine and hemodynamic factors result in these changes. All factors ultimately cause incompetence/closure of the retinal capillaries.

CLINICAL COURSE OF DIABETIC RETINOPATHY

DR is asymptomatic in the initial stages. Hence, it is recommended that annual dilated fundus examination be performed in all patients with type 1 DM till they develop signs of retinopathy[22]. But fundus examination should be performed at the time of diagnosis among patients with type 2 DM since the

onset and duration of the disease is unknown. Thereafter depending on the severity of the DR, fundus examination may be done.

EARLY TREATMENT DIABETIC RETINOPATHY STUDY (ETDRS) LEVELS OF DIABETIC RETINOPATHY[4]

NON-PROLIFERATIVE DIABETIC RETINOPATHY (NDPR)

Mild NPDR

At least one microaneurysm

Moderate NPDR

Haemorrhages or microaneurysms (H/Ma), venous beading (VB), Soft exudates and intraretinal microvascular abnormalities (IRMAs) definitely present

Severe NPDR

H/Ma in all 4 quadrants, VB in 2 or more quardrants, Intra-retinal microvascular abnormalities (IRMA) in at least 1 quadrant

Very Severe NPDR

Any two or more of severe NPDR levels

PROLIFERATIVE DIABETIC RETINOPATHY (PDR)

Early PDR

New vessels on the retina and definition for high-risk PDR not met

High-Risk PDR

New vessels on the disc of 1/4 to 1/3 or more of the disc area or any new vessel and vitreous or preretinal or vitreous haemorrhage

CLINICALLY SIGNIFICANT MACULAR OEDEMA (any one of the following):

- Thickening of the retina located 500 μm or less from the centre of the macula.
- Hard exudates at 500 μm or less from the centre of the macula with thickening of the adjacent retina.
- A zone of retinal thickening, one disc area or larger in size, any portion of which is one disc diameter or less from the centre of the macula.

DIAGNOSIS AND MANAGEMENT

DR is essentially a clinical diagnosis made by using a direct ophthalmoscope. However, to plan treatment and to monitor the response to treatment, the ophthalmologists may use stereoscopic biomicroscopic fundus examination, fundus fluorescein angiography and optical coherence

tomography. Ultrasonography may be useful in patients with obscure media to plan for surgery.

TREATMENT

As in the management of diabetic nephropathy, a multimodal approach is employed in the management of DR also. Treatment goals should include an optimal control of hyperglycaemia, HbA1c levels, dyslipidaemia, hypertension, nephropathy and anaemia. Diabetic macular oedema (DME) management includes local treatment with laser photocoagulation and systemic control of risk factors. UKPDS established that a 1% reduction in HbA1c leads to microvascular complications reduction (including DR) by as much as 37%.

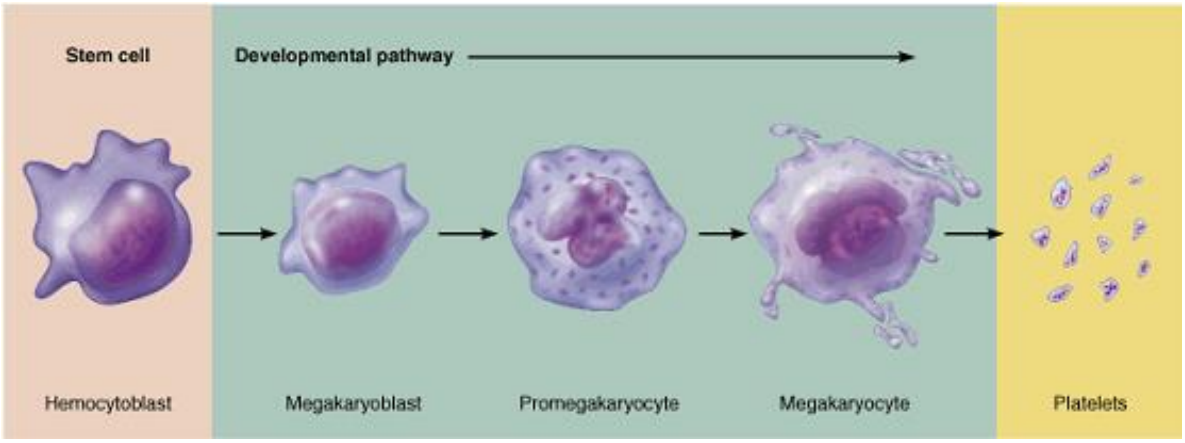
PROGNOSIS AND PREVENTION

The keys to prevention of visual impairment/blindness from DR are early detection, education and research. The most commonly used screening technique for DR is direct ophthalmoscopy. It is crucial to identify patients who are in urgent need of laser photocoagulation.

PLATELETS

Platelets are small anucleate cell fragments. They circulate in blood and play a crucial role in regulating hemostasis and managing vascular integrity. They are involved in the fundamental process of chronic inflammation, associated with disease pathology[29]. Platelets usually remain in an inactive state and they get activated only when blood vessel damage occurs. However, hemostasis is not the sole function of platelets. It is rather employed in several processes that monitor the homeostasis of the body. It has high sensitivity to different disease states. It serves as an important inflammatory marker through its interactions with leukocytes and endothelial cells.

Developmental pathway of platelets



Megakaryocytes of bone marrow are the site of platelet formation. Megakaryocytes are giant cells with the nucleus containing multiple copies of DNA[30]. Platelets are cell fragments which are generated when the edges of

megakaryocytes break off. 5000–10000 platelets are produced from each megakaryocyte.

ULTRASTRUCTURE

A mature platelet has a diameter of is 2-3μm. The normal platelet count is (150-400) $\times 10^3$ per microliter of blood. 2/3rd of the platelets circulate in the blood and 1/3rd is stored in the spleen approximately. Their lifespan is usually 5–9 days. 10^{11} platelets are produced by an average healthy adult every day.

Platelets have a unique structural assembly. They are anucleate cells with distinct mitochondria. A phospholipid bilayer, which constitutes the platelet plasma membrane is the site where various surface receptors and lipid rafts are expressed. They help in intracellular trafficking and signalling. The markers include CD9, CD36, CD63, IIbIIIa, GPCR and GLUT-3. These receptors also cause the release of α granules. Among the surface receptors, GPCR has been shown to play an important role in ADP secretion from dense granules. During non procoagulant state, asymmetrically arranged phospholipids (e.g., phosphatidylinositol and phosphatidylserine) which are present in the inner layer of the plasma membrane helps in maintaining the stability of its surface.

A “tunnel” system is present throughout the platelet cell called the open canalicular system (OCS). This remains connected with the plasma

membrane. Its major role is to release its granule contents to the exterior and give entry of external elements into the platelets. It helps in the formation of filopodia during activation of platelet in addition to being a major site of storage for plasma membrane glycoproteins.

The highly specialized cytoskeleton of platelets has three major components: (1) the spectrin based membrane skeleton, (2) the marginal microtubule coil and (3) the actin cytoskeleton. This helps to maintain its discoid structure and also guards the cell from getting sheared in bloodstream.

PLATELET GRANULES - two major storage granules are present in platelets, namely, α and dense granules. Their function is to store biologically active molecules which are involved in initiation of coagulation and recruitment of other cells during inflammation

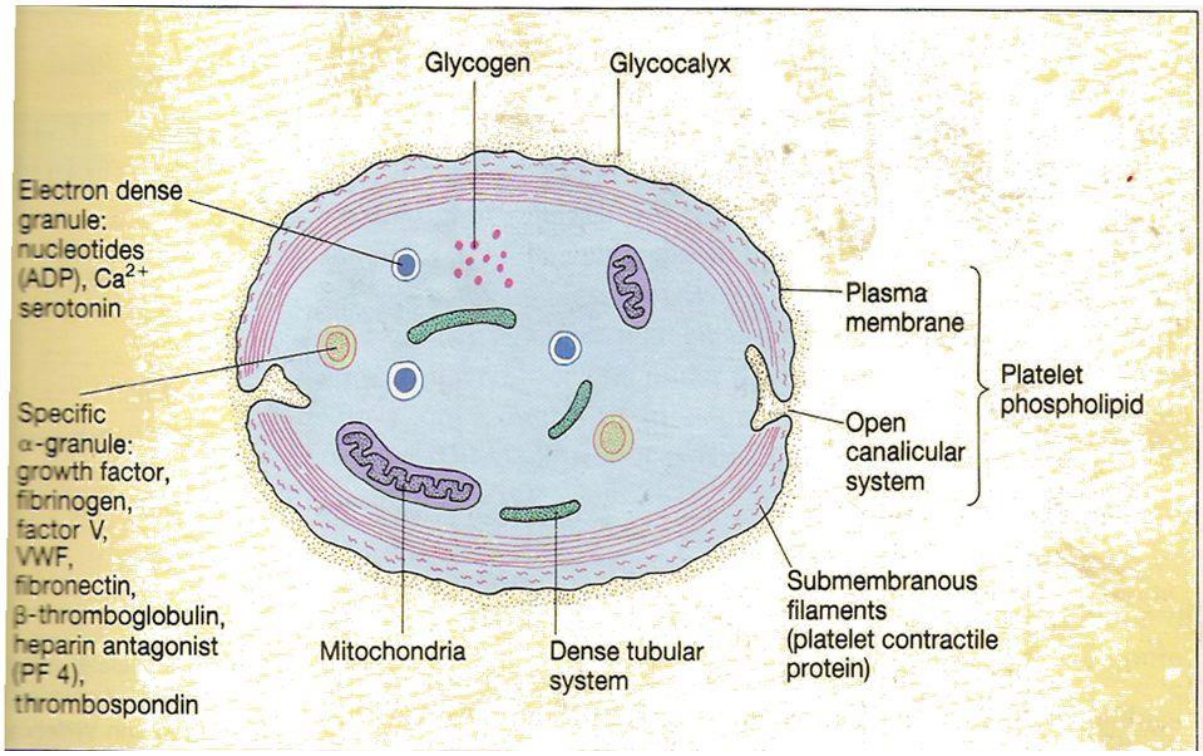
Alpha granules: contain platelet specific proteins like PF-4, thromboglobulin, PDGF, thrombospondin, homologs of plasma proteins like fibrinogen, fibronectin, albumin, GPIIbIIIa, fibrinogen, vWf and also P-selectin (CD62P) and CD36.

GPIIbIIIa, fibrinogen, and vWf, initiate the coagulation cascades. P-selectin via Pselectin glycoprotein ligand (PSGL1) has been reported to recruit neutrophils

Dense tubular system: Prostaglandin converting enzymes, contractile calcium, peroxisomes (containing Catalase), lysosomes (containing acid hydrolase).

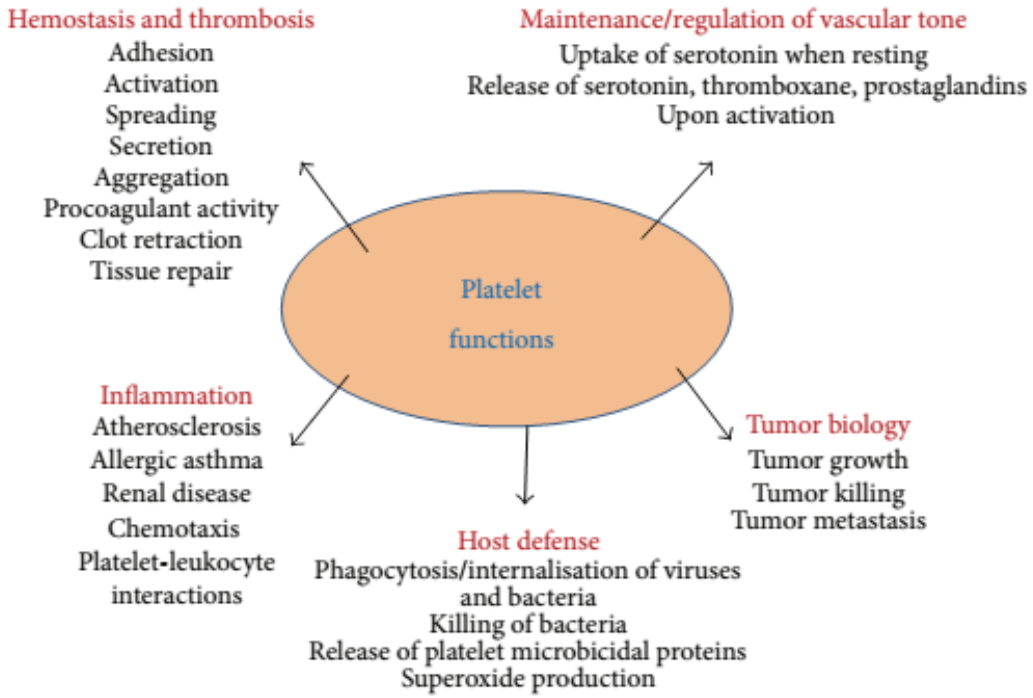
Dense granules are the storage site for a variety of hemostatically active molecules which include catecholamines, calcium, serotonin, adenosine-5-diphosphate (ADP), and adenosine-5-triphosphate (ATP).. These are secreted during platelet activation. ADP is a weak agonist of platelets. It causes change in platelet shape, release of granules and result in platelet aggregation.

Platelets structure and function:



PLATELET FUNCTIONS

Platelets are concerned with many pathophysiological processes. Functions include haemostasis and thrombosis, inflammation, maintenance of vascular tone, host defence and tumour biology.

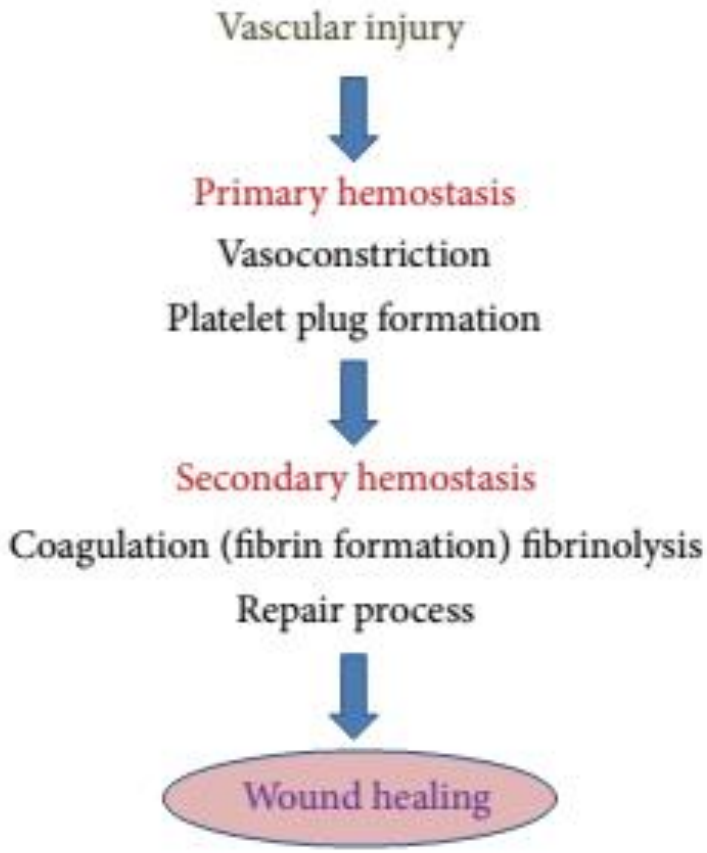


HEMOSTASIS AND THROMBOSIS:

Formation of hemostatic plug occurs due to adhesviness to damaged lining of blood vessels (promoted by Ca^{++} ions & ADP) and aggregation of platelets leading to formation of white bodies or micro thrombi which may grow until they almost fill the lumen of a small vessel. Initially, the aggregation is reversible. Later, it becomes irreversible. There will be discharge of platelet granules, adherence of leukocytes to platelets and deposition of fibrin. Upon

adherence of platelet to collagen in damaged vessels wall, ATPase converts ATP to ADP. ADP thus released promotes aggregation of passing platelets leading to formation of platelet plug. Thrombin formation is triggered by the tissue factor & phospholipids released from damaged vessels and platelets respectively. Firm clot seals the vessel permanently.

The damaged vessels constrict for about 20 minutes after the initial dilatation. 5-HT plays an important in this vasoconstriction. Prostaglandins (PG) are also involved in platelet aggregation. Phospholipase releases arachadonic acid from the platelet cell membrane. It is immediately oxidized by the enzyme cyclo-oxygenase to PGG2& PGH2. PGG2 is converted to thromboxane A2 (TX A2) in the platelets. TX A2 is a highly potent platelet aggregator and arterial muscle constrictor. PGG2 & PGH2 are converted to prostacyclin in the arterial endothelium. prostacyclin is a vasodilator and potent inhibitor of platelet aggregation. Platelets also play a major role in intrinsic clotting pathway by causing release of platelet factor 3 (PF 3) which brings about conversion of prothrombin to thrombin by factor X and V.



PHAGOCYTOSIS

Carbon particles, viruses and immune complexes are phagocytosed by platelet.

STORAGE & TRANSPORT

Store 5-HT, histamine, epinephrine & potassium. They can also take up 5-HT by active transport.

NON HEMATOLOGICAL FUNCTIONS OF PLATELETS

INFLAMMATION:

Platelets possess 4 features common to all inflammatory cells.

1, they contain a wide range of infammatory mediators.

2, they have receptors for other inflammatory cells.

3, they are able to respond to noxious stimuli.

4, they are cooperative with other inflammatory cells.

Platelet & growth factors

PDGF, EGF, FGF are released from alpha granules of platelets

Platelets & neurotransmitters

It has been shown that platelets behave like serotonergic neurons in CNS, despite being of different embryological origin. Thus they offer a neuronal model for studying neurons.

Platelets & metastasis

By protecting tumour cells from attack by NK cells, platelets promote tumour survival in blood stream.

Platelets in asthma

Bronchial hyperreactivity is caused by the release of PAF & PF4 from platelets.

STRUCTURAL, FUNCTIONAL AND METABOLIC ALTERATIONS OF PLATELETS IN DIABETES MELLITUS

A large number of structural, functional and metabolic changes are seen in platelets of diabetic patients. Increased activation of coagulation proteins and platelets along with decreased fibrinolytic activity in diabetes makes it a prothrombotic state.

ENHANCED PLATELETS ACTIVATION

In diabetic patients, there is proof for the in vivo activation of circulating platelets. There are frequent episodes of release of granules from the platelets in circulation. Augmented granule release means reduced survival of platelets due to their hastened sequestration in the circulation[32]. These increased turnovers of platelets reflect increased thrombopoiesis. Altered response to agonists, increased adhesive proteins on the surface of platelets, enhanced expression of glycoprotein receptors, decreased membrane fluidity and increased fibrinogen binding is instrumental in platelet activation**.**

PLATELET HYPERAGGREGABILITY

When compared with non-diabetic individuals, diabetic patients show increased aggregation of platelets in response to ADP, collagen, thrombin, epinephrine and arachidonic acid[38]. After stimulation with platelet agonists,

these platelets show reduced threshold for aggregation, under in vitro conditions. In diabetics with macro vascular disease, this aggregation is more apparent. Adhesiveness, spontaneous aggregation as well as aggregation on extra cellular matrix are found to be increased in platelets from diabetic subjects.

INCREASED ARACHIDONIC ACID METABOLISM

Increased activation of arachidonic acid pathway[37] leads to increased TXA2 formation, augmented phosphoinositide turnover which results in enhanced protein phosphorylation, increased inositol trisphosphate production and afterwards accelerated mobilisation of Ca^{2+}. TX A2 is one of the most potent platelet activators. In patients with diabetes, in vitro as well as in vivo TXA2 production has been reported to be increased. HbA1c directly influence thromboxane metabolism. In several studies, reduction in TX A2 has been achieved with improved glycaemic control. Increased TXA2 has been linked to micro and macro angiopathy associated with diabetes.

INCREASED CALCIUM FLUX

In patients with diabetes mellitus, it has been observed that there is increased mobilisation of calcium from intra-platelet storage pool and thus higher levels of intracellular free calcium[40]. This increased free calcium in the cell has been correlated with the decline in membrane fluidity. This results in

platelet hyperfunction. Reduction in intracellular magnesium concentrations is also seen, consistent with an enhanced platelet adhesiveness and hyperaggregability.

PLATELET NITRIC OXIDE SYNTHESIS

Prostacyclin and nitric oxide (NO) inhibit interaction between platelet and endothelium and results in endothelium mediated vasodilation. Platelets in patients with diabetes are found to produce less prostacyclin and NO. NO synthesis in platelets is stimulated by insulin.

PLATELET SECRETARY PRODUCTS

Mitogenic and chemotactic are released by activated platelets. In diabetic patients, levels of thromboglobulin and PF -4 in plasma are found to be elevated.

PLATELET MEMBRANE GLYCATION

Non-enzymatic glycation of membrane proteins in platelets is brought about by hyperglycemia[40]. This leads to alterations in the structure and conformation of proteins and dynamics of the lipid membrane. Reduced platelet membrane fluidity appears to be related to the extent of glycation of membrane proteins. This altered platelet membrane lipid dynamics leads to enhanced expression of receptors that are vital for platelet functions. Frequent episodes of

platelet activation and degranulation have been attributed to increased expression of adhesion receptors, e.g., αIIbβ3.

MEMBRANE GLYCATION OF LOW DENSITY LIPOPROTEINS

Glycation of low density lipoproteins (LDL) has been found to contribute towards increased sensitivity of platelet to aggregating agents. Hyperglycaemia causes an increase in non-enzymatically glycated LDL (glycLDL), thus making the platelets more prone to oxidative stress. GlycLDL causes inhibition of Ca2+-ATPase in the platelet membrane which may bring about increase in intracellular concentration of Ca2+ and decrease in activity of NO synthase. GlycLDL also causes inhibition of activity of platelet membrane Na+/K+-adenosine triphosphatase (Na+/K+-ATPase), thereby resulting in further platelet dysfunction. Lipoproteins also increase thromboxane generation during activation of platelet.

EXPRESSION OF INCREASED SURFACE MARKERS ON PLATELET MEMBRANE

Increase in number, adhesiveness and activity of several platelet specific glycoprotein receptors (GP) are observed in platelets from diabetic patients. Increased platelet αIIbβ3 receptor expression is consistent with enhanced aggregability and fibrinogen binding[41]. CD40-CD40 ligand system up-regulation has also been observed in diabetic patients. CD40L and P-selectin

are shed from the surface of the platelet into plasma in a soluble biologically active form. High levels of these compounds may denote a prothrombotic state and lead to accelerated atherosclerosis. In patients with diabetes mellitus, increased collagen-induced aggregation is seen additionally.

PLATELET METABOLIC ALTERATIONS

Hyperglycaemia is an established causal factor for platelet hyperactivity and in vivo activation of platelet in patients with diabetes mellitus. Glucose concentration inside the platelet parallel the extra cellular concentration as glucose entry into the platelets is insulin-independent. Increased sensitivity of platelets to agonists are due to metabolic alterations which include impaired calcium homeostasis, decreased platelet-derived nitric acid production, activation of PKC and increased of superoxide anion formation[33]. In addition, reduced levels of glutathione levels and nitric oxide synthase activity are observed in the platelets of diabetic patients.

ALTERED PLATELET SIZE AND VOLUME

Large platelets predominate the circulation in patients with diabetes mellitus. This has been considered due to an increased activation and ploidy of megakaryocytes. Larger platelets are younger are believed to be more aggregable and reactive. They have denser granules, secrete more beta thromboglobulin and serotonin and produce more TX A2 than smaller platelets.

The thromboxane synthesising capacity, the number of GP molecules on platelet membrane and platelet granule concentration of various platelet specific proteins correlates well with the mean platelet volume (MPV). Proliferative diabetic retinopathy has also been associated with increased MPV.

PLATELET LIFE SPAN

In diabetics with overt vascular complications, decreased platelet survival has been demonstrated by some studies.

PLATELET-LEUKOCYTE INTERACTION

Leukocyte activation, chemotaxis and phagocytosis may be influenced by platelets. The chemottractant PDGF enhances phagocytosis by monocytes and neutrophils. Platelets release P–selectin and nitric oxide, which inhibits leukocyte chemotaxis, adhesion and generation of superoxide**.**

EFFECTS OF INSULIN ON PLATELETS

In diabetes mellitus, insulin resistance and metabolic syndrome associated with it, correlate with the prothrombic state to put a collective effect in the progression of vascular complications. Insulin has been reported to have antiplatelet effects. Insulin-induced attenuation of the thrombin-induced Ca2+ response has been directly correlated with platelet aggregation. Ca2+ homeostasis is found to be altered in diabetic platelets. Tyrosine

phosphorylation of Ca2+ -ATPase in the platelet plasma membrane may cause inhibition of membrane Ca2+-ATPase. This results in increased intracellular calcium during platelet activation.

There have been reports which suggest that binding of PGI2 with insulin is decreased in non-diabetic patients with acute ischemic heart disease. In coronary heart diseases, administration of insulin has found to normalize platelet activity to PGI2. Vitamin E deficiency plays an important role in platelet aggregation, where supplementation of α-tocopherol in response to ADP decreases TX A2. Insulin therapy may improve sensitivity of platelets to NO, thus providing a suitable proposal in treating CVDs among diabetic patients.

AIM OF STUDY

1, To compare Mean Platelet Volume in type 2 Diabetes Mellitus patients with good glycemic control with that of poor glycemic control.

2, To investigate the association between mean platelet volume and microvascular complications of diabetes. (retinopathy and nephropathy)

3, To assess the relation between mean platelet volume, glycemic control, sex, BMI, duration of diabetes, hypertension, hypertriglyceridemia and abdominal circumference.

SAMPLE SIZE: 100

STUDY DESIGN: Cross sectional study

INCLUSION CRITERIA:

- Known type II diabetes mellitus patients on treatment with OHA/Insulin.
- Male and female patients of age >30 years.
- Newly detected type II diabetes mellitus patients.

EXCLUSION CRITERIA:

-Type I diabetes mellitus

- Gestational Diabetes Mellitus

- Male patients with Hb<12mg% and female with Hb <11mg%

- Patients on antiplatelets and antithrombotics.

- Patients with diagnosed malignancy.

- Patients with known chronic kidney disease.

- Patients with UTI, cardiac failure.

METHODOLOGY:

A total of 100 type 2 DM patients attending medicine OPD and admitted in medicine wards of Government Stanley Hospital from April 2017 to September 2017 were studied. Our patients were on treatment with insulin or OHAs. All the patients underwent a detailed clinical evaluation. Body weight , height, abdominal circumference were measured in all subjects. BMI was calculated as weight (kg) divided by height^2 (metres^2).

Systolic and diastolic blood pressures (SBP and DBP) were measured after a 5 min rest in a semisitting position with a sphygmomanometer. BP was

determined at least 3 times from the right upper arm for analysis,the mean of the 3 was used. Patients with mean blood pressure levels >/= 140/90 mm of Hg or patients already on antihypertensive medications were diagnosed as having hypertension.

LABORATORY METHODS

Venous samples were collected after 12 hours of overnight fasting at 8:30 am for Mean Platelet Volume, HbA1C, FBS, PPBS, Hb, triglyceride (TG) and serum creatinine levels.

HbA1c was measured by High Performance Liquid Chromatography.

Measurement of MPV was done using an automatic blood counter (Beckman Coulter Act5Diff).

Plasma glucose estimation (FBS and PPBS) was carried out by the glucose oxidase method in the autoanalyzer.

Hypertriglyceridemia was defined as having triglyceride levels > 150 mg/dl.

Microalbuminuria, which is the hallmark of diabetic retinopathy was examined using spot urine albumin creatinine ratio (ACR). Patients with ACR of <20 mg/g for men and <30 mg/g for women were categorized as

microalbuminuria negative and those with >20mg/g and >30mg/g respectively as microalbuminuria positive.

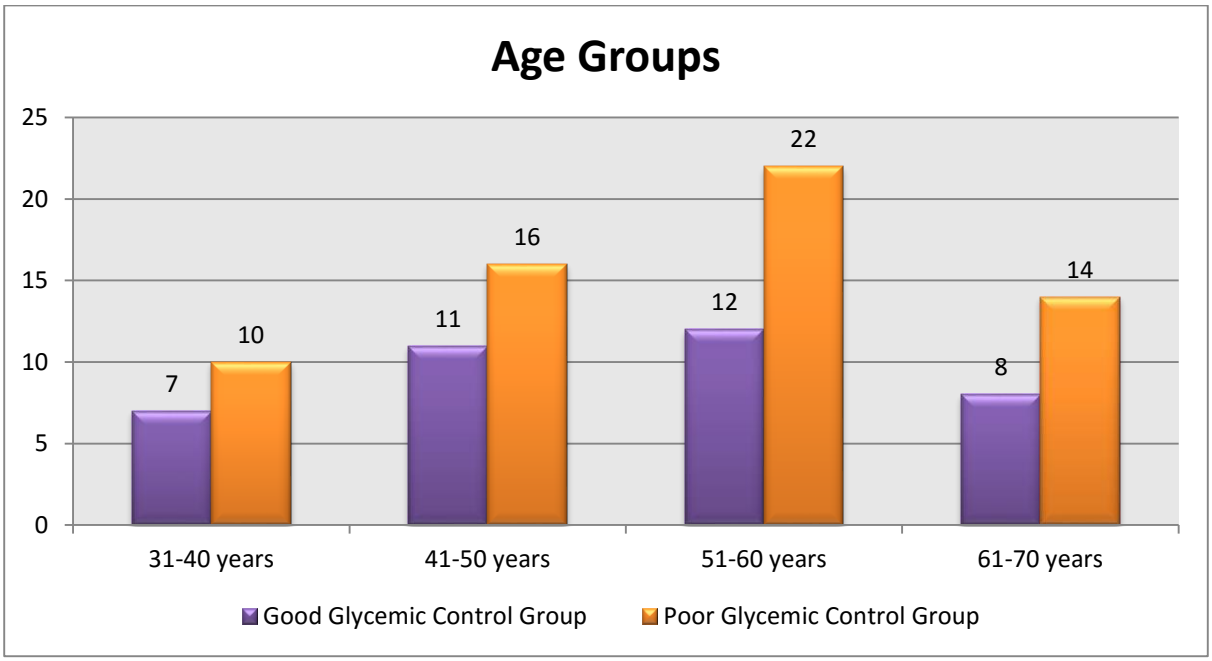
Diabetic Retinopathy was defined by direct ophthalmoscopic examination. Patients with at least 2 microaneurysms and/or retinal hemorrhage, and/or other signs of retinal damage were diagnosed as having retinopathy.

Creatinine clearance was calculated with the cockroft-Gault formulae as (140-age) * weight / 72 * serum creatinine. Multiplication of the result by 0.85 was done for female patients.

After baseline evaluation, the patients were divided into 2 groups based on HbA1C levels. Diabetics with good glycemic control (patients with HbA1c<7%) and those with poor glycemic control (patients with HbA1c >7%). All the parameters were compared between both the groups. These groups were further sub grouped based on the presence or absence of complications. The MPV in each group were compared.

RESULTS AND DISCUSSION

Age

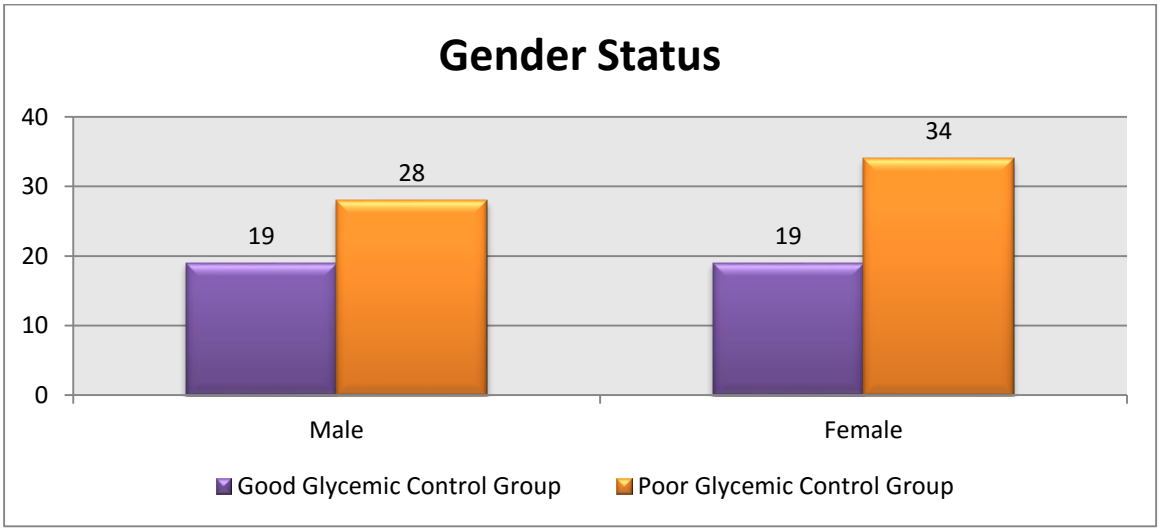


Age Groups	Good Glycemic Control Group	%	Poor Glycemic Control Group	%
31-40 years	7	18.42	10	16.13
41-50 years	11	28.95	16	25.81
51-60 years	12	31.58	22	35.48
61-70 years	8	21.05	14	22.58
Total	38	100.00	62	100.00

Age Distribution	Good Glycemic Control Group	Poor Glycemic Control Group	P value Unpaired t Test
Mean	50.84	52.73	0.3806
SD	10.79	10.12	

While analyzing age distribution, it was observed that, majority in good glycemic control group belonged to 51-60 years age class interval (31.58%) with a mean age of 50.84 years and majority in poor glycemic control group belonged to same age class interval (35.48%) with a mean age of 52.73 years (p=0.3806)

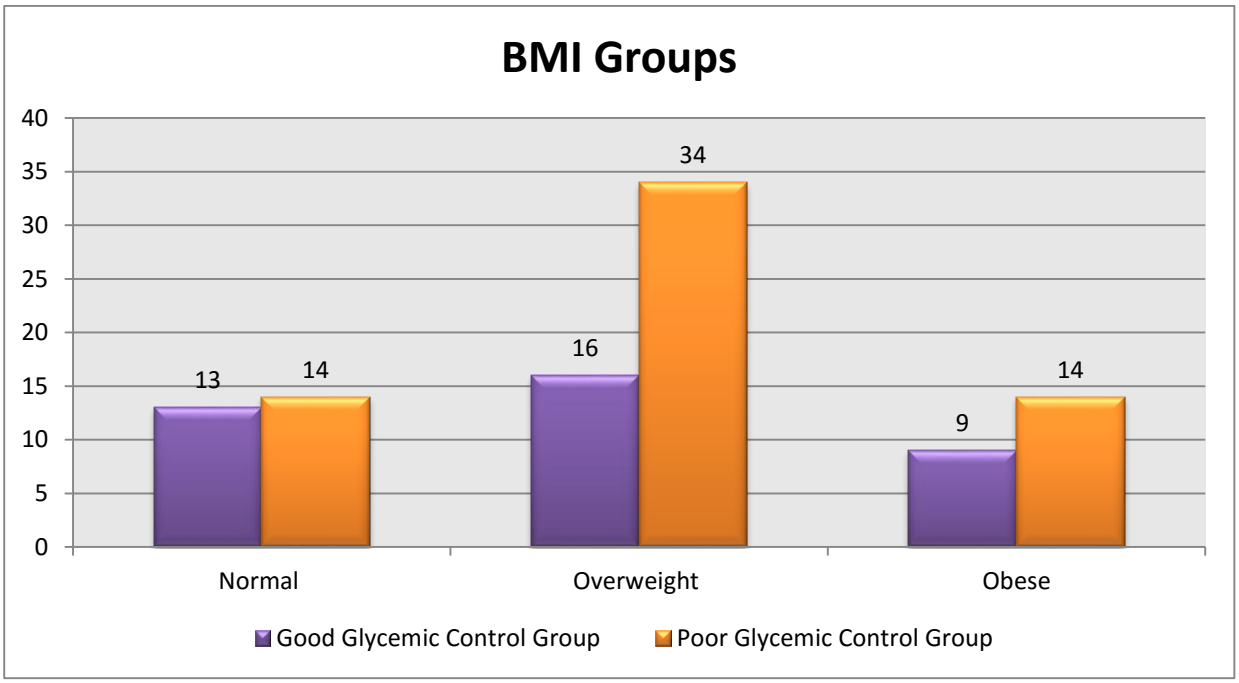
Gender



Gender Status	Good Glycemic Control Group	%	Poor Glycemic Control Group	%	P value Chi Squared Test
Male	19	50.00	28	45.16	0.6382
Female	19	50.00	34	54.84	
Total	38	100.00	62	100.00	

While analyzing gender status, it was observed that, in good glycemic control group males and females were equally distributed (50.00%) and majority in poor glycemic control group were females (54.84%) (p=0.6382)

BMI

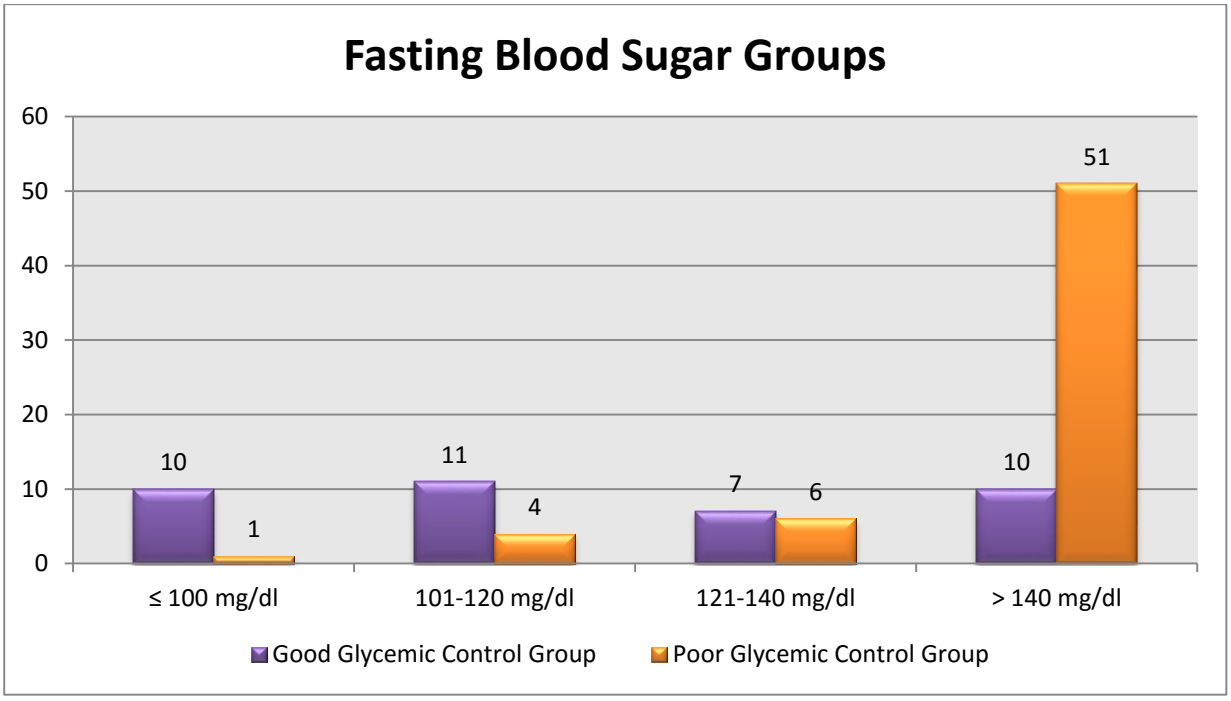


BMI Groups	Good Glycemic Control Group	%	Poor Glycemic Control Group	%
Normal	13	34.21	14	22.58
Overweight	16	42.11	34	54.84
Obese	9	23.68	14	22.58
Total	38	100.00	62	100.00

BMI Distribution	Good Glycemic Control Group	Poor Glycemic Control Group	P value Unpaired t Test
Mean	27.30	28.11	0.3507
SD	4.77	3.83	

While analyzing BMI distribution, it was observed that, majority in good glycemic control group belonged to overweight BMI class interval (42.11%) with a mean BMI of 27.30 and majority in poor glycemic control group belonged to same BMI class interval (54.84%) with a mean BMI of 28.11 (p=0.3806)

FBS



Fasting Blood Sugar Groups	Good Glycemic Control Group	%	Poor Glycemic Control Group	%
≤ 100 mg/dl	10	26.32	1	1.61
101-120 mg/dl	11	28.95	4	6.45
121-140 mg/dl	7	18.42	6	9.68
> 140 mg/dl	10	26.32	51	82.26
Total	38	100.00	62	100.00

Fasting Blood Sugar Distribution	Good Glycemic Control Group	Poor Glycemic Control Group	P value Unpaired t Test
Mean	121.63	161.21	<0.0001
SD	23.32	30.54	

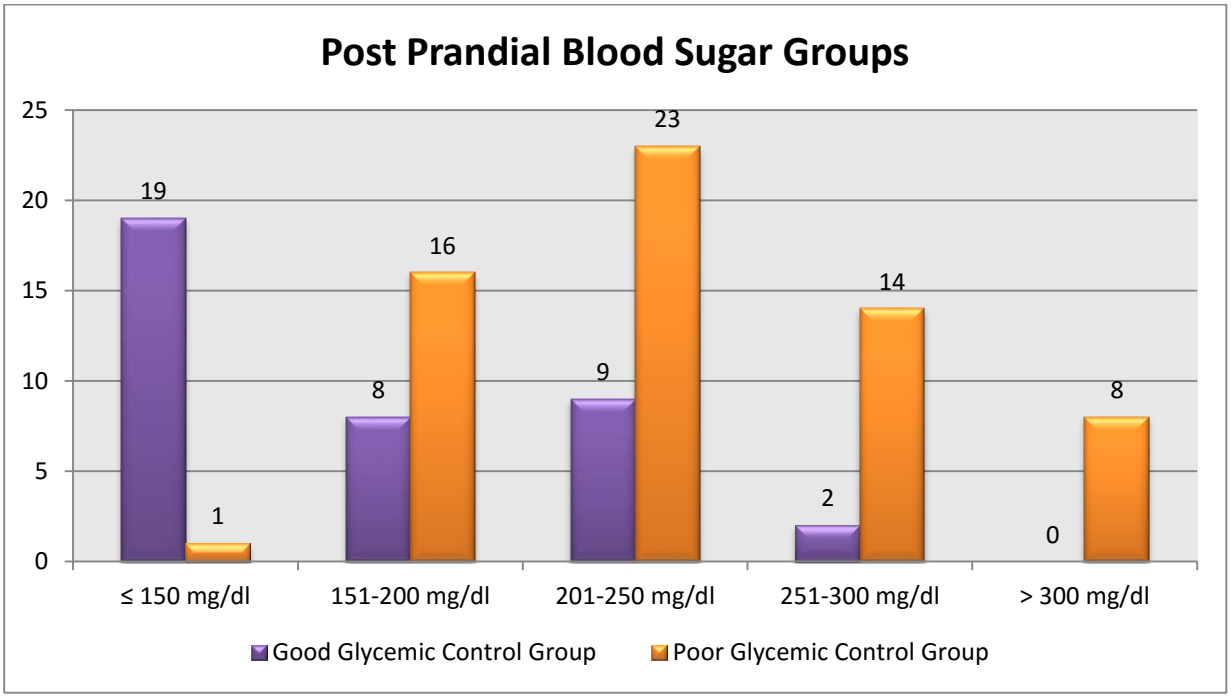
While analyzing FBS distribution, it was observed that, majority in good glycemic control group belonged to 101-120 mg/dl FBS class interval (28.95%) with a mean FBS of 121.63 mg/dl and majority in poor glycemic control group belonged to > 140 mg/dl FBS class interval (82.26%) with a mean FBS of 161.21 mg/dl (p= <0.0001)

Discussion

The data subjected to statistical unpaired t test reveals the existence of statistically significant association between FBS distribution and glycemic control based on Hba1c levels ($p < 0.05$)

This significance is exhibited by the increased mean in FBS levels of poor glycemic control group compared to good glycemic control group (39.58 mg/dl increase, 25% higher)

PPBS



Post Prandial Blood Sugar Groups	Good Glycemic Control Group	%	Poor Glycemic Control Group	%
≤ 150 mg/dl	19	50.00	1	1.61
151-200 mg/dl	8	21.05	16	25.81
201-250 mg/dl	9	23.68	23	37.10
251-300 mg/dl	2	5.26	14	22.58
> 300 mg/dl	0	0.00	8	12.90
Total	38	100.00	62	100.00

Post Prandial Blood Sugar Distribution	Good Glycemic Control Group	Poor Glycemic Control Group	P value Unpaired t Test
Mean	170.47	239.58	<0.0001
SD	45.00	53.76	

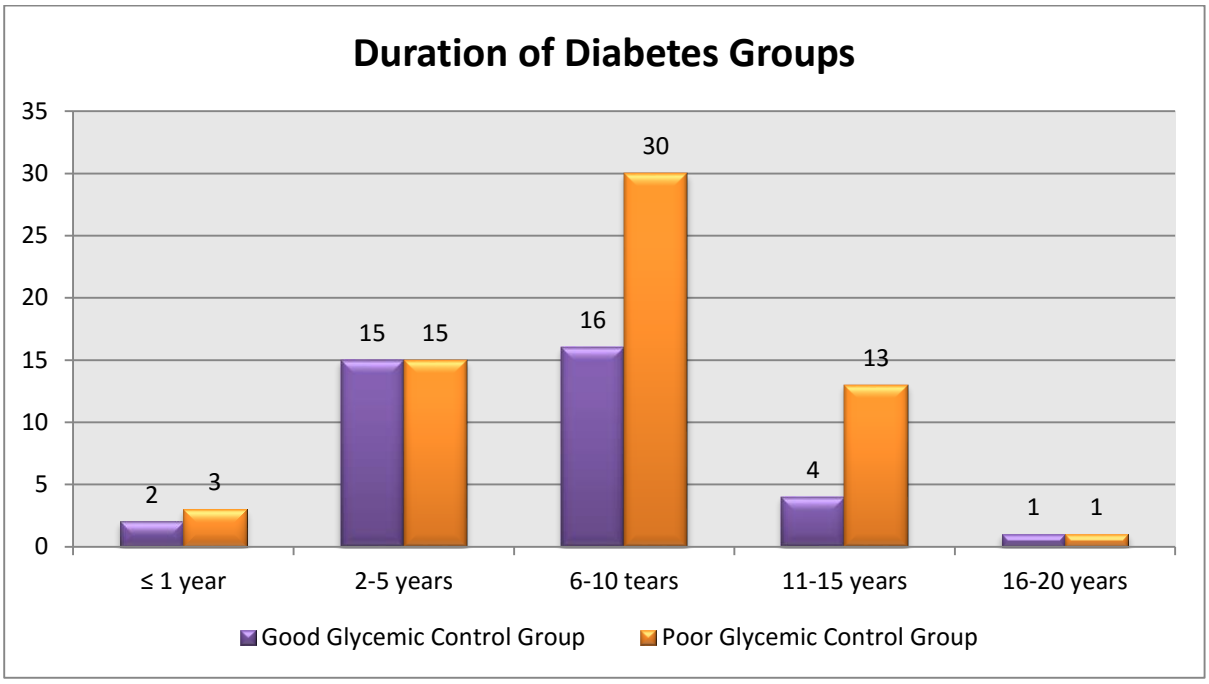
While analyzing PPBS distribution, it was observed that, majority in good glycemic control group belonged to ≤ 150 mg/dl PPBS class interval (50.00%) with a mean PPBS of 170.47 mg/dl and majority in poor glycemic control group belonged to 201-250 mg/dl PPBS class interval (37.10%) with a mean PPBS of 239.58 mg/dl (p= <0.0001)

Discussion

The data subjected to statistical unpaired t test reveals the existence of statistically significant association between PPBS distribution and glycemic control based on Hba1c levels ($p < 0.05$)

This significance is exhibited by the increased mean PPBS levels in poor glycemic control group compared to good glycemic control group (69.11 mg/dl increase, 29% higher).

Duration of Diabetes

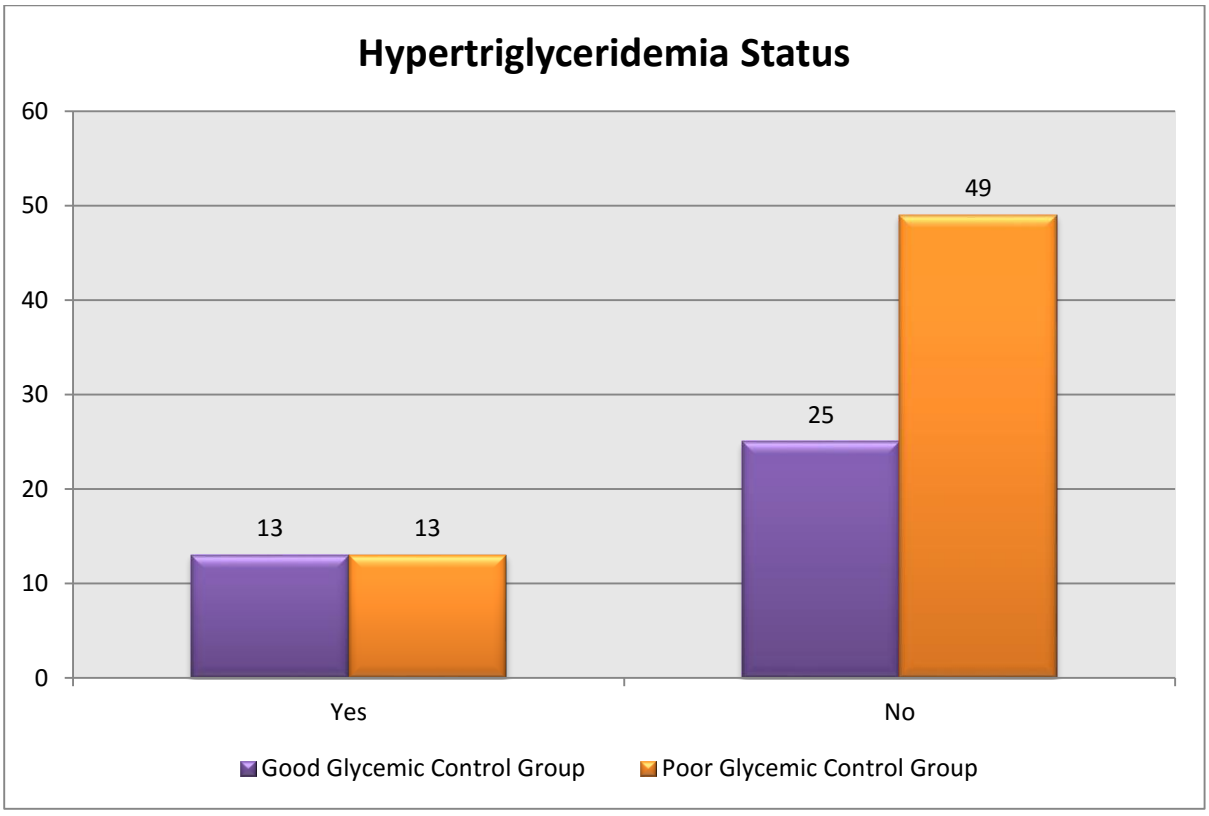


Duration of Diabetes Groups	Good Glycemic Control Group	%	Poor Glycemic Control Group	%
≤ 1 year	2	5.26	3	4.84
2-5 years	15	39.47	15	24.19
6-10 years	16	42.11	30	48.39
11-15 years	4	10.53	13	20.97
16-20 years	1	2.63	1	1.61
Total	38	100.00	62	100.00

Duration of Diabetes Distribution	Good Glycemic Control Group	Poor Glycemic Control Group	P value Unpaired t Test
Mean	6.53	7.35	0.2632
SD	3.71	3.49	

While analyzing duration of diabetes distribution, it was observed that, majority in good glycemic control group belonged to 6-10 years duration of diabetes class interval (42.11%) with a mean duration of diabetes of 6.53 years and majority in poor glycemic control group belonged to same duration of diabetes class interval (48.39%) with a mean duration of diabetes of 7.35 years (p=0.2632).

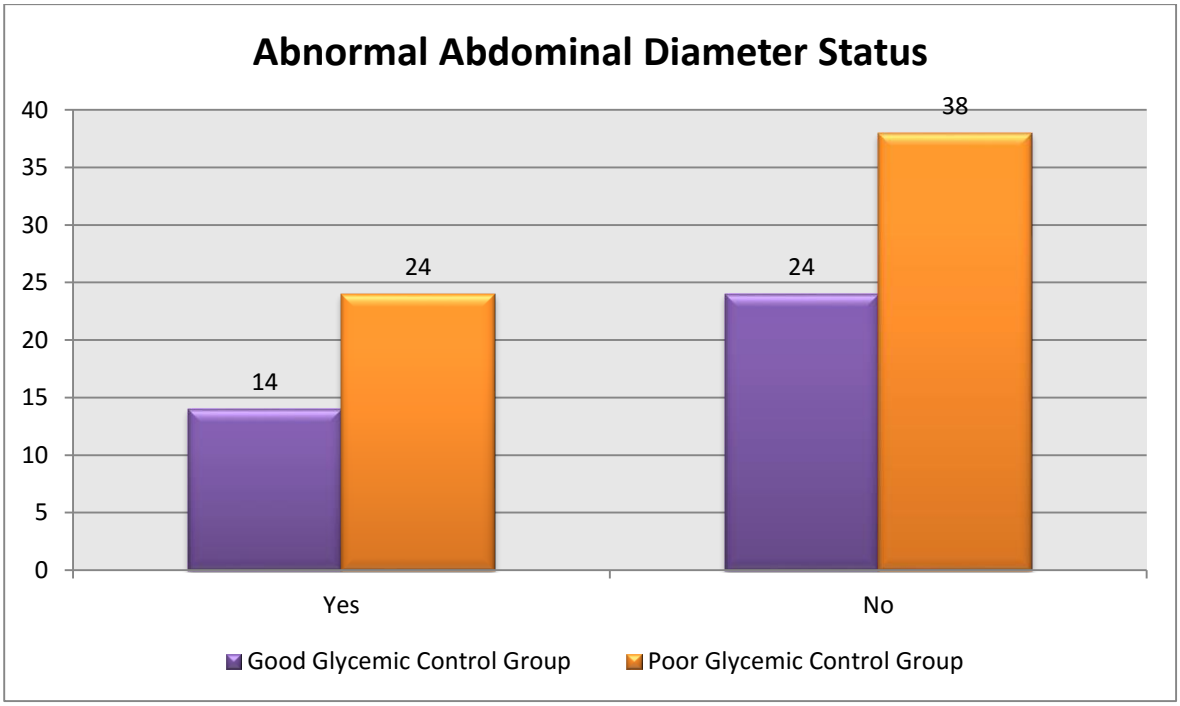
Hypertriglyceridemia



Hypertriglyceridemia Status	Good Glycemic Control Group	%	Poor Glycemic Control Group	%	P value Chi Squared Test
Yes	13	34.21	13	20.97	0.1437
No	25	65.79	49	79.03	
Total	38	100.00	62	100.00	

While analyzing hypertriglyceridemia status, it was observed that, in good glycemic control group incidence of hypertriglyceridemia was 34.21% and in poor glycemic control group incidence of hypertriglyceridemia was 20.97% (p=0.1437)

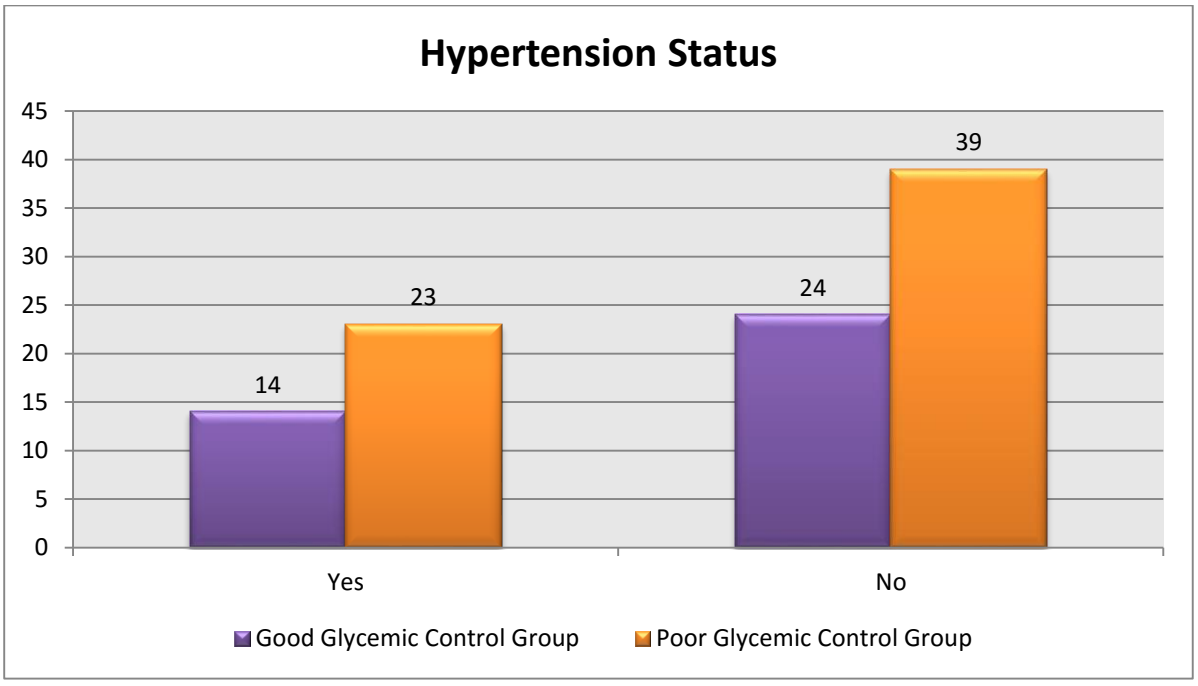
Abnormal Abdominal Diameter



Abnormal Abdominal Diameter Status	Good Glycemic Control Group	%	Poor Glycemic Control Group	%	P value Chi Squared Test
Yes	14	36.84	24	38.71	0.8522
No	24	63.16	38	61.29	
Total	38	100.00	62	100.00	

While analyzing abnormal abdominal diameter status, it was observed that in good glycemic control group incidence of abnormal abdominal diameter was 36.84% and in poor glycemic control group incidence of abnormal abdominal diameter was 38.71% (p=0.8522)

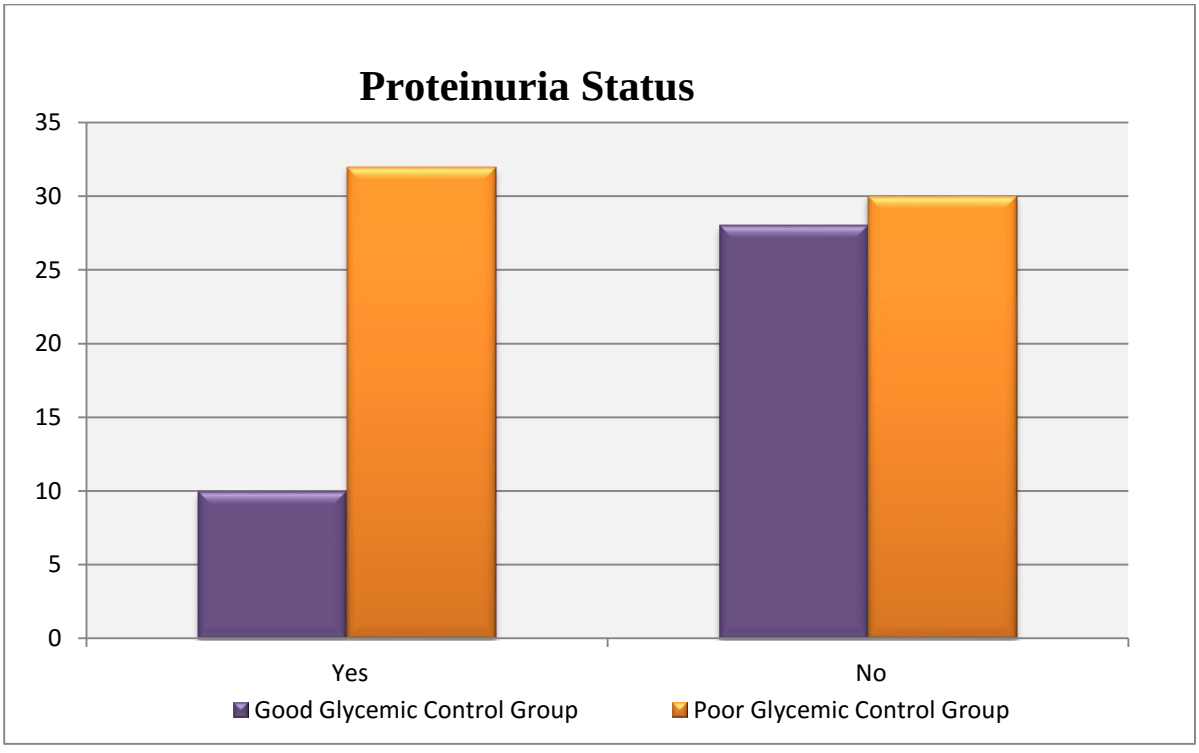
Hypertension



Hypertension Status	Good Glycemic Control Group	%	Poor Glycemic Control Group	%	P value Chi Squared Test
Yes	14	36.84	23	37.10	0.9801
No	24	63.16	39	62.90	
Total	38	100.00	62	100.00	

While analyzing hypertension status, it was observed that in good glycemic control group incidence of hypertension was 36.84% and in poor glycemic control group incidence of abnormal hypertension was 37.10% (p=0.9801)

Proteinuria



Proteinuria Status	Good Glycemic Control Group	%	Poor Glycemic Control Group	%	P value Chi Squared Test
Yes	10	26.32	32	51.61	
No	28	73.68	30	48.39	0.0132
Total	38	100.00	62	100.00	

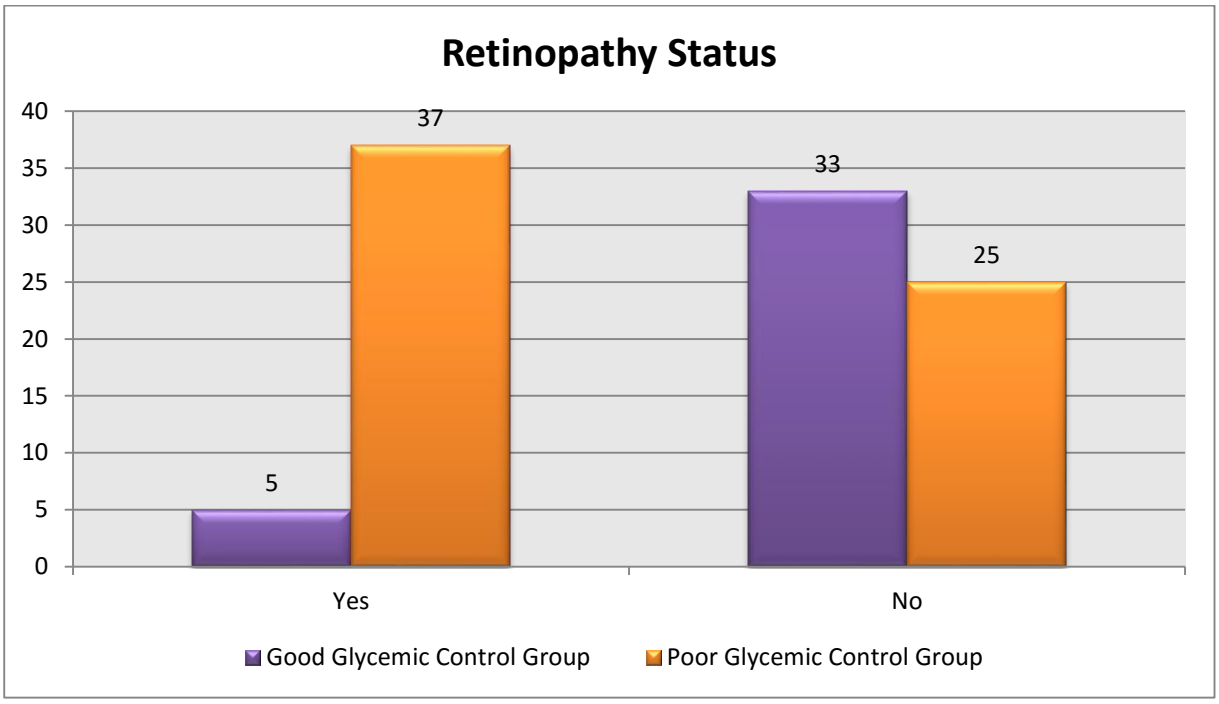
While analyzing proteinuria status, it was observed that, in good glycemic control group incidence of proteinuria was 73.68% and in poor glycemic control group incidence of proteinuria was 48.39% (p= 0.0132)

Discussion

The data subjected to statistical chi squared test reveals the existence of statistically significant association between proteinuria status and glycemic control based on Hba1c levels ($p < 0.05$)

This significance is exhibited by the increased incidence of proteinuria in poor glycemic control group compared to good glycemic control group (25.30 percentage points increase, 34% higher)

Retinopathy



Retinopathy Status	Good Glycemic Control Group	%	Poor Glycemic Control Group	%	P value Chi Squared Test
Yes	5	13.16	37	59.68	
No	33	86.84	25	40.32	<0.0001
Total	38	100.00	62	100.00	

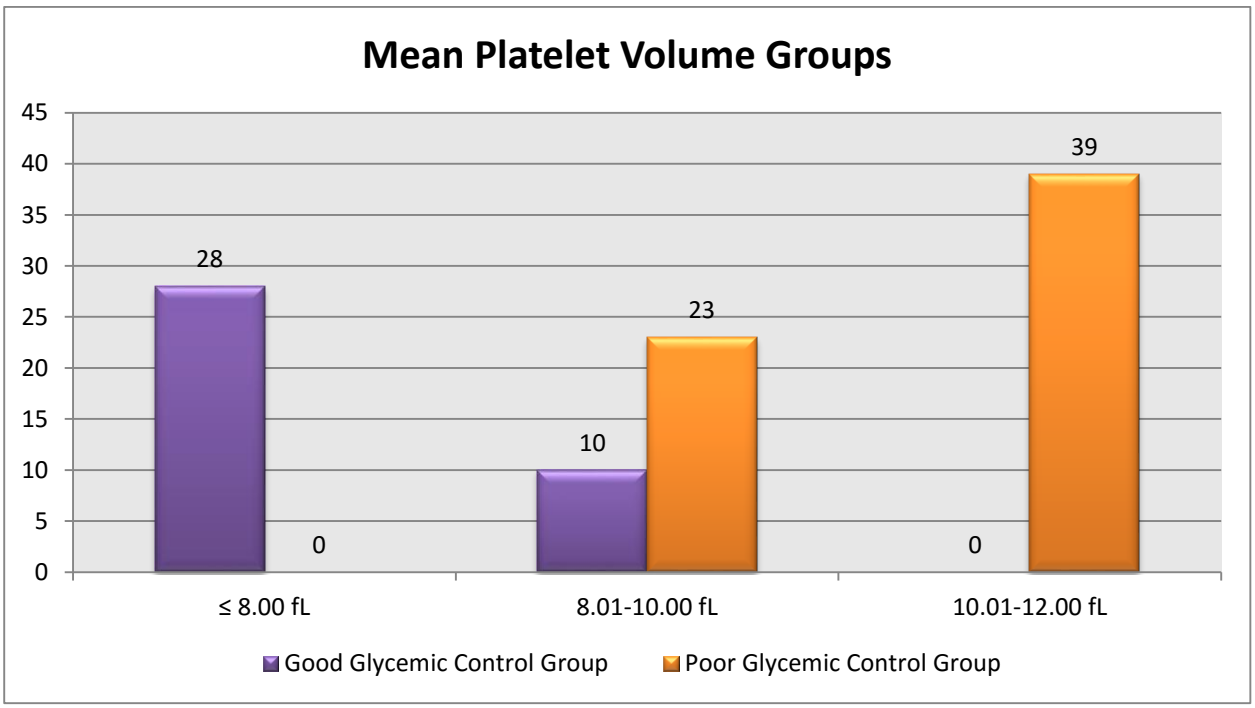
While analyzing retinopathy status, it was observed that, in good glycemic control group incidence of retinopathy was 13.16% and in poor glycemic control group incidence of retinopathy was 59.68% (p= <9.9991)

Discussion

The data subjected to statistical chi squared test reveals the existence of statistically significant association between retinopathy status and glycemic control based on Hba1c levels ($p < 0.05$)

This significance is exhibited by the increased incidence of retinopathy in poor glycemic control group compared to good glycemic control group (46.52 percentage points increase, 78% higher)

Mean Platelet Volume



Mean Platelet Volume Groups	Good Glycemic Control Group	%	Poor Glycemic Control Group	%
≤ 8.00 fL	28	73.68	0	0.00
8.01-10.00 fL	10	26.32	23	37.10
10.01-12.00 fL	0	0.00	39	62.90
Total	38	100.00	62	100.00

Mean Platelet Volume Distribution	Good Glycemic Control Group	Poor Glycemic Control Group	P value Unpaired t Test
Mean	7.82	10.21	<0.0001
SD	0.48	0.85	

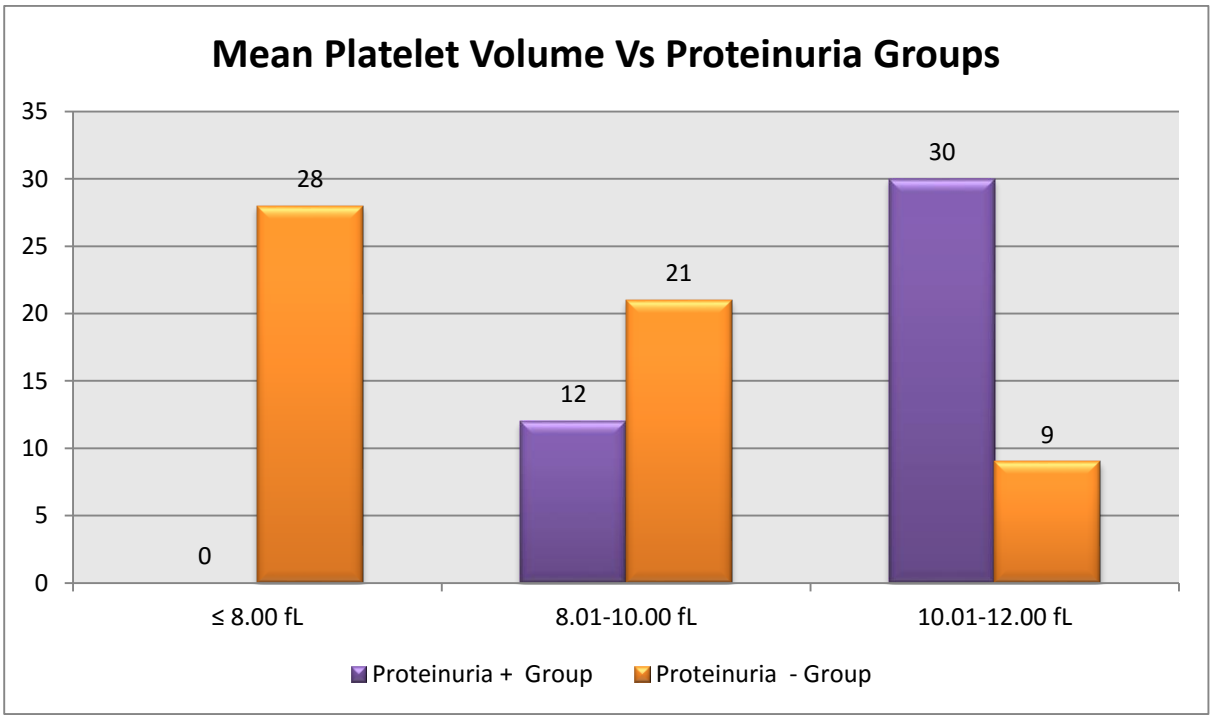
While analyzing MPV distribution, it was observed that, majority in good glycemic control group belonged to ≤ 8.00 fL MPV class interval (73.68%) with a mean MPV of 7.82 fL and majority in poor glycemic control group belonged to 10.01-12.00 fL MPV class interval (62.90%) with a mean MPV of 10.21 fL (p= <0.0001)

Discussion

The data subjected to statistical unpaired t test reveals the existence of statistically significant association between MPV distribution and glycemic control based on Hba1c levels ($p < 0.05$)

This significance is exhibited by the increased mean MPV levels in poor glycemic control group compared to good glycemic control group (2.39 fL increase, 23% higher)

Mean Platelet Volume Vs Proteinuria



Mean Platelet Volume Vs Proteinuria Groups	Proteinuria + Group	%	Proteinuria - Group	%
≤ 8.00 fL	0	0.00	28	48.28
8.01-10.00 fL	12	28.57	21	36.21
10.01-12.00 fL	30	71.43	9	15.52
Total	42	100.00	58	100.00

Mean Platelet Volume Vs Proteinuria Distribution	Proteinuria + Group	Proteinuria - Group	P value Unpaired t Test
Mean	10.26	8.62	<0.0001
SD	1.08	1.14	

While analyzing MPV distribution, it was observed that, majority in proteinuria +ve group belonged to 10.01-12.00 fL MPV class interval (71.43%) with a mean MPV of 10.26 fL and majority in proteinuria -ve group belonged to ≤ 8.00 fL MPV class interval (48.28%) with a mean MPV of 8.62 fL (p= <0.0001)

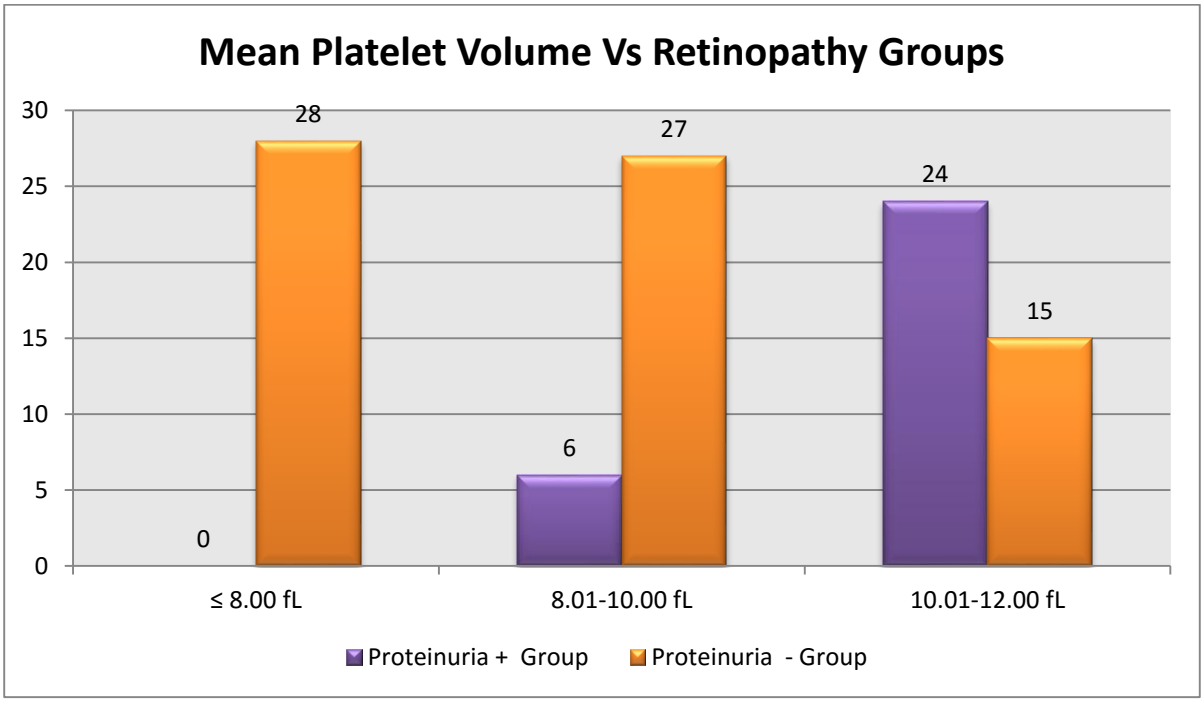
Discussion

The data subjected to statistical unpaired t test reveals the existence of statistically significant association between MPV distribution and proteinuria status ($p < 0.05$)

This significance is exhibited by the increased mean MPV levels in proteinuria +ve group compared to proteinuria -ve group (1.64 fL increase, 16% higher) The same view was echoed by studies done by Ates et al and Papanas et al. This suggested a role for the increased platelet activity in the pathogenesis of vascular complications. On the other hand, in the studies done by Hekimsoy et

al and Demirtunc et al, MPV in diabetic subjects with and without complications did not show any significant difference. They explained it to be possibly because of rapid consumption of activated platelets in diabetic patients with complications.

Mean Platelet Volume Vs Retinopathy



Mean Platelet Volume Vs Retinopathy Groups	Retinopathy + Group	%	Retinopathy - Group	%
≤ 8.00 fL	0	0.00	28	40.00
8.01-10.00 fL	6	20.00	27	38.57
10.01-12.00 fL	24	80.00	15	21.43
> 12 fL	0	0.00	0	0.00

Mean Platelet Volume Vs Retinopathy Distribution	Retinopathy + Group	Retinopathy - Group	P value Unpaired t Test
Mean	10.50	8.79	<0.0001
SD	0.95	1.20	

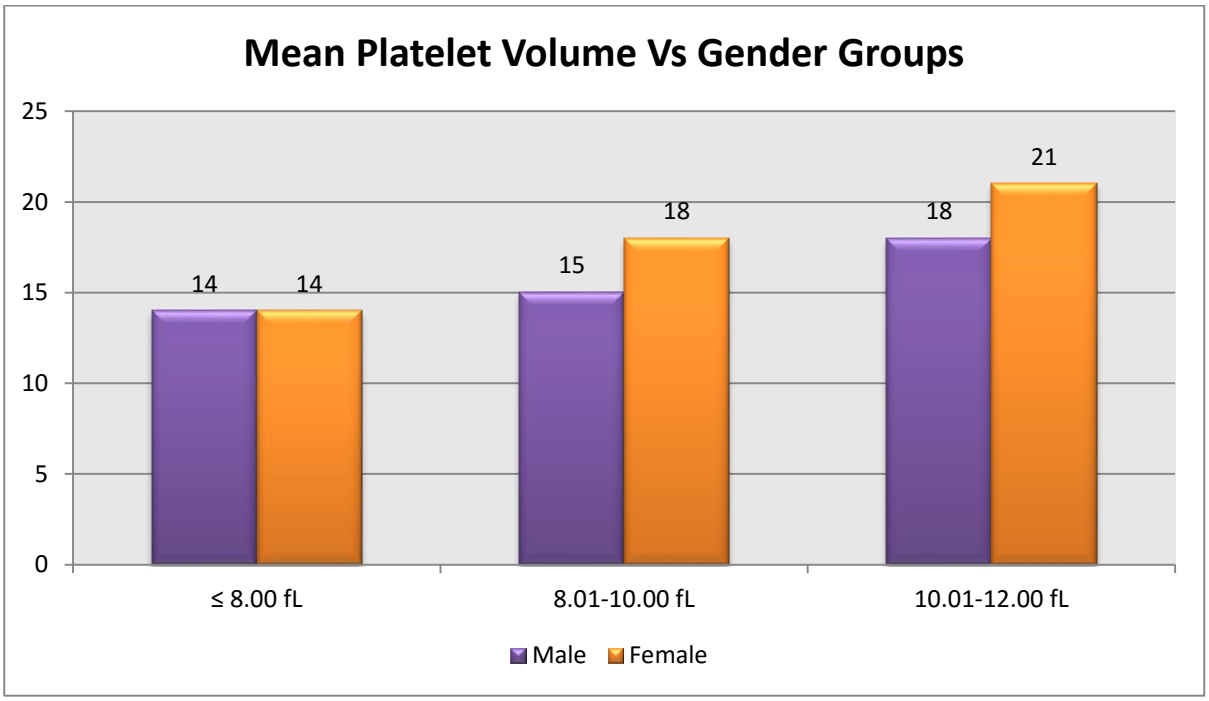
While analyzing MPV distribution, it was observed that, majority in retinopathy +ve group belonged to 10.01-12.00 fL MPV class interval (80.00%) with a mean MPV of 10.50 fL and majority in retinopathy -ve group belonged to ≤ 8.00 fL MPV class interval (40.00%) with a mean MPV of 8.79 fL (p= <0.0001)

Discussion

The data subjected to statistical unpaired t test reveals the existence of statistically significant association between MPV distribution and retinopathy status ($p < 0.05$)

This significance is exhibited by the increased mean MPV levels in retinopathy +ve group compared to retinopathy -ve group (1.71 fL increase, 16% higher). This is in agreement with the studies done by Kodiatte et al. which showed higher MPV values in diabetic patients with micro vascular complications than those without complications. Hence, its proved that platelet hyperactivity has a major role in pathogenesis of micro vascular complications.

Mean Platelet Volume Vs Gender

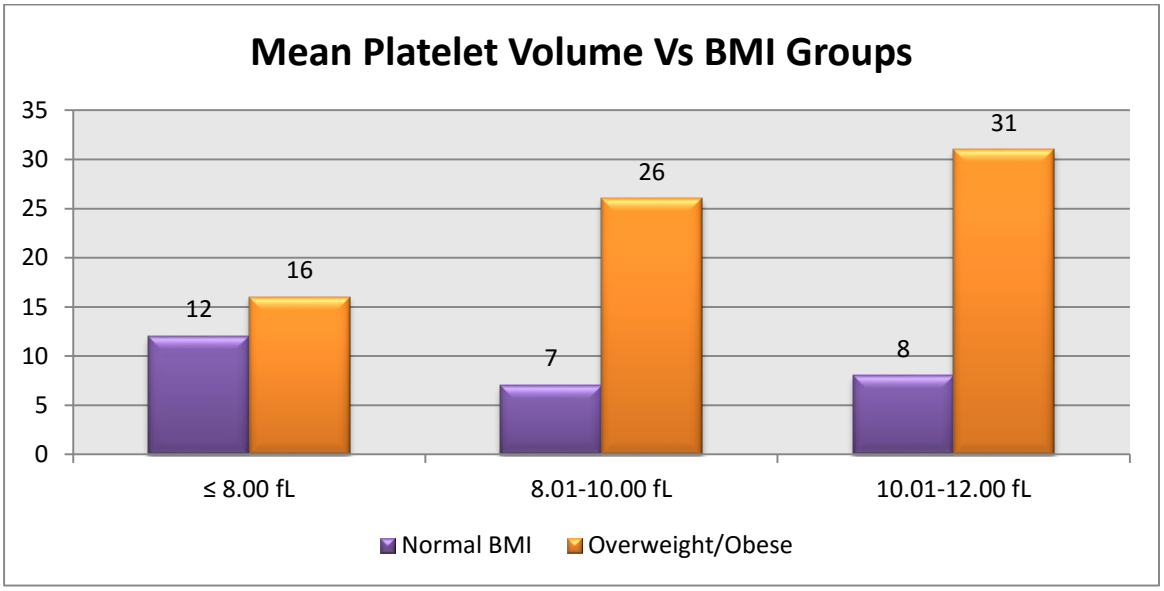


Mean Platelet Volume Vs Gender Groups	Male	%	Female	%
≤ 8.00 fL	14	29.79	14	26.42
8.01-10.00 fL	15	31.91	18	33.96
10.01-12.00 fL	18	38.30	21	39.62
Total	47	100.00	53	100.00

Mean Platelet Volume Vs Gender Distribution	Male	Female	P value Unpaired t Test
Mean	9.23	9.37	0.6146
SD	1.41	1.36	

While analyzing MPV distribution in relation to gender status, it was observed that in male group, majority belonged to 10.01-12.00 fL MPV class intervals (38.30%) with a mean MPV of 9.23 fl and in female group majority belonged to same MPV class intervals (39.62%) with a mean MPV of 9.37 fl (p=0.6146)

Mean Platelet Volume Vs BMI

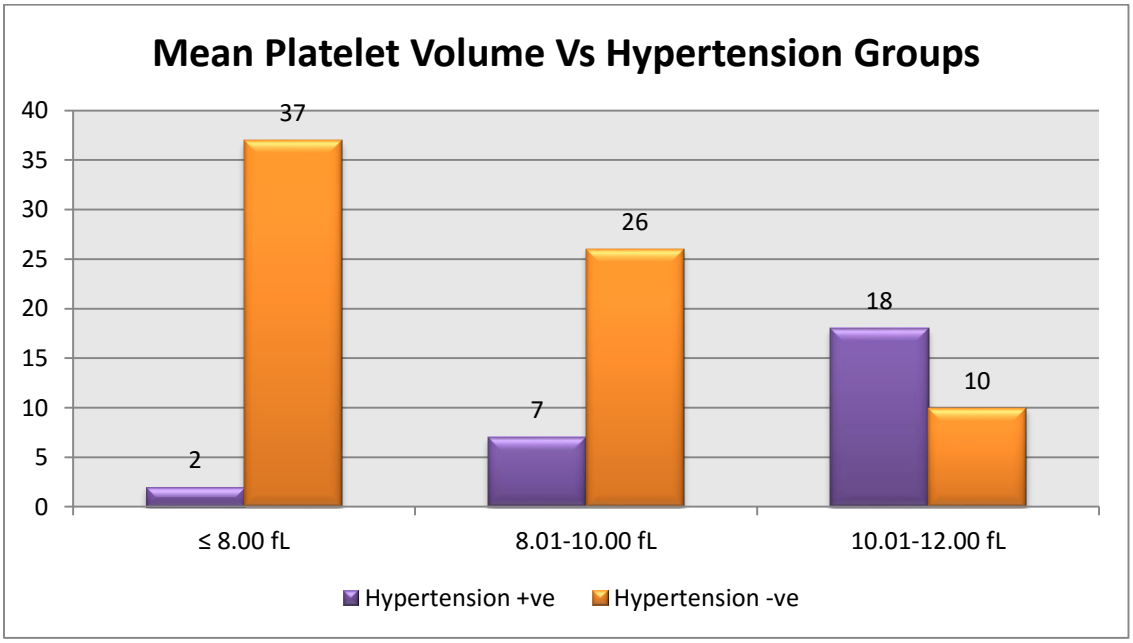


Mean Platelet Volume Vs BMI Groups	Normal BMI	%	Overweight/Obese	%
≤ 8.00 fL	12	44.44	16	21.92
8.01-10.00 fL	7	25.93	26	35.62
10.01-12.00 fL	8	29.63	31	42.47
Total	27	100.00	73	100.00

Mean Platelet Volume Vs BMI Distribution	Normal BMI	Overweight/Obese	P value Unpaired t Test
Mean	8.90	9.45	0.0752
SD	1.37	1.36	

While analyzing MPV distribution in relation to BMI, it was observed that in normal BMI group, majority belonged to ≤ 8.00 fL MPV class intervals (44.44%) with a mean MPV of 8.90 fl and in overweight/obese group majority belonged to 10.01-12.00 fL MPV class intervals (42.47%) with a mean MPV of 9.45 fl (p=0.0752)

Mean Platelet Volume Vs Hypertension



Mean Platelet Volume Vs Hypertension Groups	Hypertension +ve	%	Hypertension - ve	%
≤ 8.00 fL	2	7.41	37	50.68
8.01-10.00 fL	7	25.93	26	35.62
10.01-12.00 fL	18	66.67	10	13.70
Total	27	100.00	73	100.00

Mean Platelet Volume Vs Hypertension Status	Hypertension +ve	Hypertension -ve	P value Unpaired t Test
Mean	9.77	8.06	<0.0001
SD	1.25	0.81	

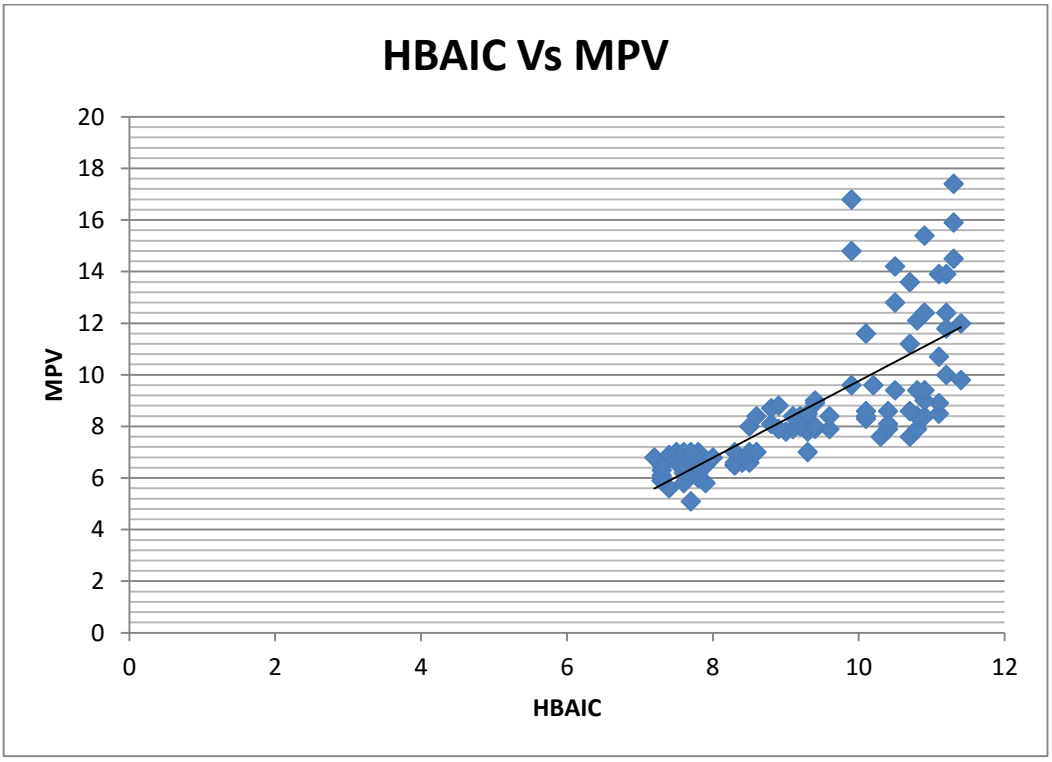
While analyzing MPV distribution, it was observed that, majority in hypertension +ve group belonged to 10.01-12.00 fL MPV class interval (66.67%) with a mean MPV of 9.77 fL and majority in hypertension -ve group belonged to ≤ 8.00 fL MPV class interval (50.68%) with a mean MPV of 8.06 fL (p= <0.0001)

Discussion

The data subjected to statistical unpaired t test reveals the existence of statistically significant association between MPV distribution and hypertension status ($p < 0.05$)

This significance is exhibited by the increased mean MPV levels in hypertension +ve group compared to hypertension -ve group (1.71 fL increase, 18% higher). This observation was similar to the study done by Coban et al[41].This may be because of the higher platelet activation in hypertensive patients.

CORRELATION



Correlation Statistics - HBAIC Vs MPV	
Pearson's R	0.75
R Square	0.56
P value	<0.0001

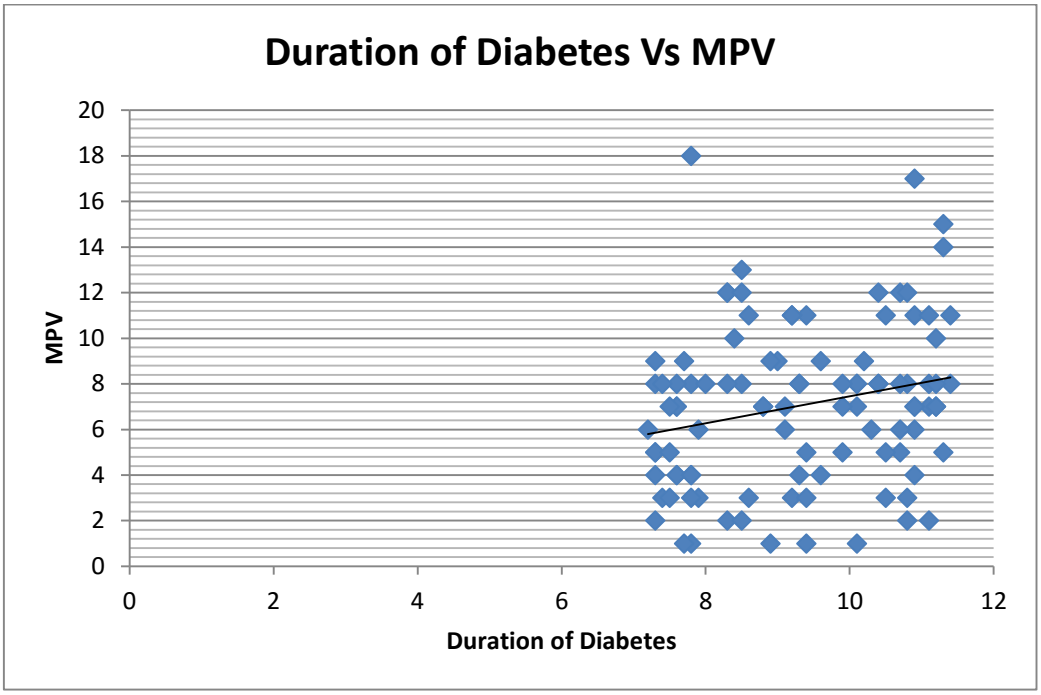
ANOVA	

There is a strong positive correlation between Hba1c levels and MPV levels. This is indicated by the Pearson's R Correlation value of 0.75with a p-value of <0.0001.

Discussion

By conventional criteria the relationship between the Hba1c levels and MPV levels is considered to be statistically significant since $p < 0.05$. This means as Hba1c levels increases MPV levels also increases in a direct and linear fashion in our study subjects. This observation was similar to the studies done by Kodiatte et al. This proves that increased platelet volume and activity results from hyperglycaemic states. In simple terms, for every 1% increase in Hba1c level there is a 5.1 fl increase in MPV among the study subjects.

Duration of Diabetes vs MPV



Correlation Statistics - Duration of Diabetes Vs MPV	
Pearson's R	0.24
R Square	0.06
P value ANOVA	0.0189

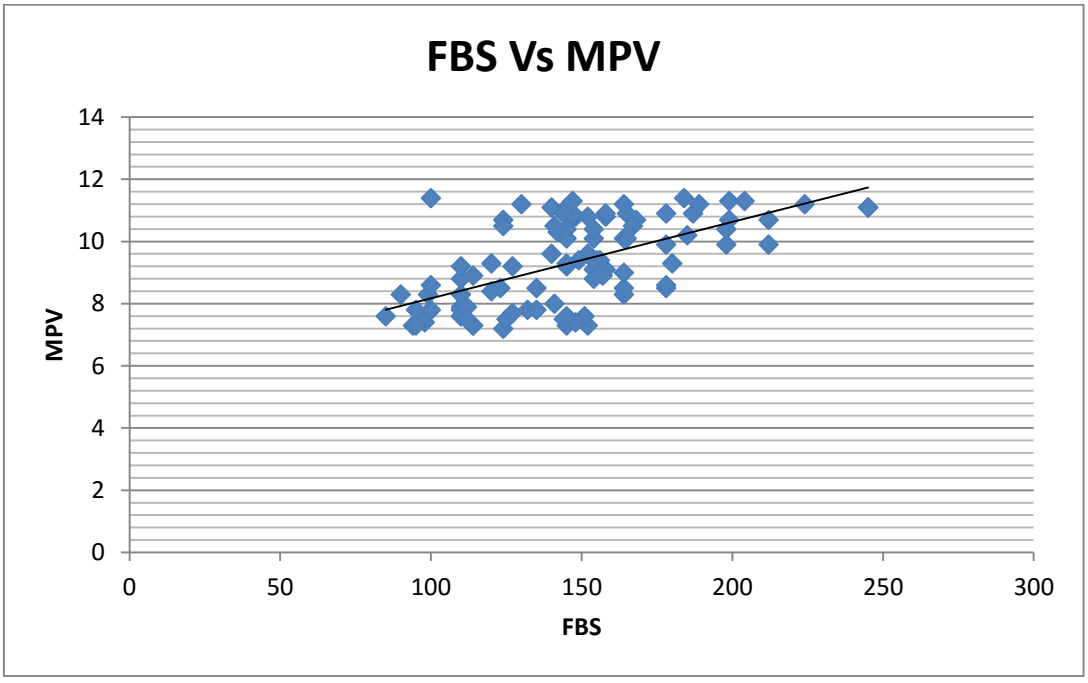
There is a positive correlation between duration of diabetes and MPV levels. This is indicated by the Pearson's R Correlation value of 0.24 with a p-value of 0.0189.

Discussion

By conventional criteria the relationship between the duration of diabetes and MPV levels is considered to be statistically significant since $p < 0.05$. This

means as duration of diabetes increases MPV levels also increases in a direct and linear fashion in our study subjects. In simple terms, for every 1 year increase in duration of diabetes there is a 9.0 fl increase in MPV among the study subjects. This is in contrast to the studies done by Demirtunc et al. and Hekimsoy et al., which showed no significant relation between MPV and duration of diabetes.

FBS vs MPV



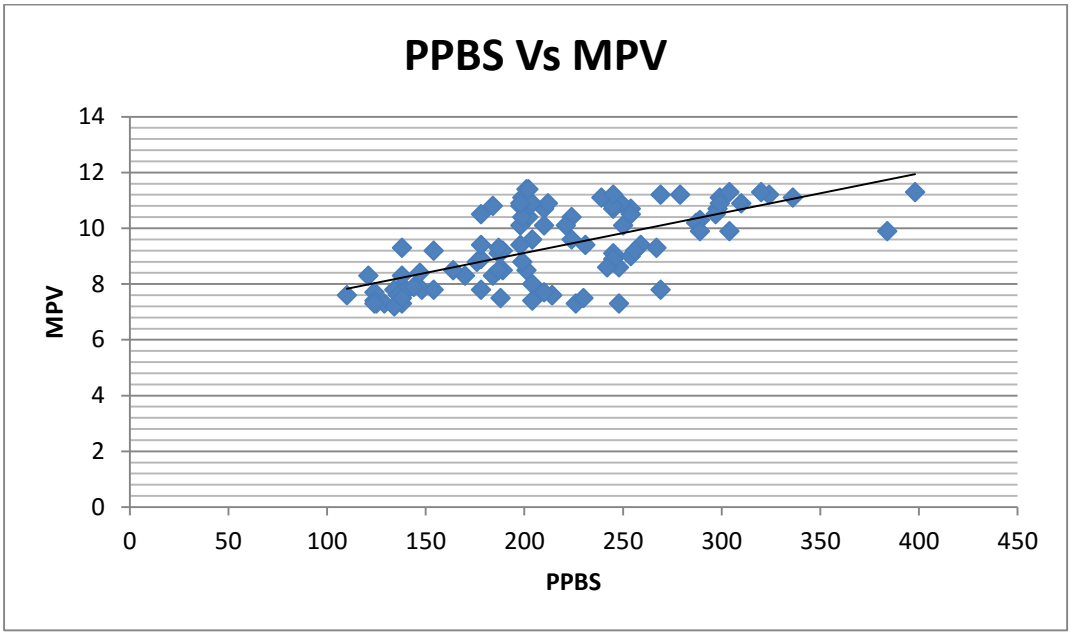
Correlation Statistics - FBS Vs MPV	
Pearson's R	0.61
R Square	0.37
P value ANOVA	<0.0001

There is a positive correlation between FBS levels and MPV levels. This is indicated by the Pearson's R Correlation value of 0.61 with a p-value of <0.0001

Discussion

By conventional criteria the relationship between the FBS and MPV levels is considered to be statistically significant since $p < 0.05$. This means as FBS increases MPV levels also increases in a direct and linear fashion in our study subjects. In simple terms, for every 100mg/dl increase in FBS there is a 7.96 fl increase in MPV among the study subjects.

PPBS vs MPV



Correlation Statistics – PPBS Vs MPV	
Pearson's R	0.63
R Square	0.39
P value ANOVA	<0.0001

There is a positive correlation between PPBS levels and MPV levels. This is indicated by the Pearson's R Correlation value of 0.63 with a p-value of <0.0001

Discussion

By conventional criteria the relationship between the PPBS and MPV levels is considered to be statistically significant since $p < 0.05$. This means as PPBS increases MPV levels also increases in a direct and linear fashion in our study subjects. In simple terms, for every 100mg/dl increase in FBS there is a 7.69 fl increase in MPV among the study subjects.

STATISTICAL ANALYSIS

Descriptive statistics was done for all data and suitable statistical tests of comparison were done. Continuous variables were analysed with the Unpaired t test/single factor ANOVA and categorical variables were analysed withchi squared test/ Fisher Exact Test. regression analysis done and odds ratio with confidence interval calculated. Statistical significance was taken as $P < 0.05$. The data was analysed using SPSS Version 16. Microsoft Excel 2010.was used to generate charts.

CONCLUSIONS

We can conclude that:

- Age, gender, BMI, duration of diabetes, hypertriglyceridemia, abdominal diameter, and hypertension, status had no statistically significant role to play on mean platelet volume while correlating it with hba1c and studying its association with microvascular complications in type ii diabetes mellitus
- On internal comparisons between good and poor glycemic control patient groups
    - Higher fasting blood sugar levels in poor glycemic control patients
    - Higher post prandial blood sugar levels in poor glycemic control patients
    - Higher incidence of proteinuria in poor glycemic control patients
    - Higher incidence of retinipathy in poor glycemic control patients
    - Higher mean platelet volume levels in poor glycemic control patients
- On internal comparisons between proteinuria based patient groups
    - Higher mean platelet volume levels in patients with proteinuria
- On internal comparisons between retinopathy based patient groups
    - Higher mean platelet volume levels in patients with retinopathy

- On internal comparisons between hypertension status patient groups
    - Higher mean platelet volume levels in patients with hypertension
- Correlation analysis results
    - For every 1% increase in Hba1c level there is a 5.1 fl increase in MPV
    - For every 1 year increase in duration of diabetes there is a 9.0 fl increase in MPV
    - For every 100mg/dl increase in FBS there is a 7.96 fl increase in MPV
    - For every 100mg/dl increase in FBS there is a 7.69 fl increase in MPV

This study is a hypothesis proving study.

Hence results have high clinical significance.

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PERFORMA

NAME:

AGE

SEX:

DURATION OF DIABETES:

TREATMENT UNDERTAKEN:

PAST H/O HYPERTENSION, DYSLIPIDEMIA, CARDIAC DISEASE, KIDNEY DISEASE, CHRONIC DRUG INTAKE

PERSEONAL HISTORY: SMOKING, ALCOHOLISM

EXAMINATION:

VITALS- PR, BP, RR, Temperature

Height, weight, BMI, abdominal diameter

SYSTEM EXAMINATION

INVESTIGATIONS:

CBC with MPV, FBS, PPBS, HbA1C, fasting lipid profile, urine protein creatinine ratio, direct ophthalmoscopy.

Informed consent- 'MEAN PLATELET VOLUME- CORRELATION WITH HbA1c AND ITS ASSOCIATION WITH MICROVASCULAR COMPLICATIONS IN TYPE II DIABETES MELLITUS '

Place of study: Govt. Stanley medical college, Chennai

I ……. have been informed about the details of the study in my own language.

I have completely understood the details of the study.

I am aware of the possible risks and benefits, while taking part in the study.

I agree to collect samples of blood/saliva/urine/tissue if study needs.

I understand that I can withdraw from the study at any point of time and even then, I can receive the medical treatment as usual.

I understand that I will not get any money for taking part in the study.

I will not object if the results of this study are getting published in any medical journal, provided my personal identity is not revealed.

I know what I am supposed to do by taking part in this study and I assure that I would extend my full cooperation for this study.

Volunteer:	Witness:
Name and address	Name and address
Signature/thumb impression:	Signature/thumb impression
Date:	Date:

Informed consent- 'MEAN PLATELET VOLUME- CORRELATION WITH Hba1c AND ITS ASSOCIATION WITH MICROVASCULAR COMPLICATIONS IN TYPE II DIABETES MELLITUS '

நான்இந்தஆராய்ச்சியில்விவரங்களைமுற்றிலும்புரிந்துகொண்டேன்.
ஆய்வில்பங்குஎடுத்துபோது,
சாத்தியமானஅபாயங்கள்மற்றும்பயன்களைபற்றிநான்அறிந்துள்ளேன்.

நான்எந்தவொருவேளையிலும்ஆய்வில்இருந்துதிரும்பமுடியும்,
அதன்பின்னர்,நான்வழக்கம்போல்மருத்துவசிகிச்சைபெறமுடியும்என்றுபுரிந்து கொள்கிறேன்.

நான்ஆய்வில்பங்குஎடுத்துபணம்எதையும்பெறமுடியாதுஎன்றுஅறிந்துள் ளேன்.
இந்தஆய்வின்முடிவுகள்எந்தமெடிக்கல்ஜர்னலில்வெளியிடப்படஇருந்தால்நான் எதிர்க்கவில்லை,என்தனிப்பட்டஅடையாளத்தைவெளிப்படுத்தப்பட்டுஇருக்கக்கூ டாது.

நான்இந்தஆய்வில்பங்கெடுப்பதன்மூலம்நான்என்னசெய்யபோகிறேன்எ ன்றுதெரியும்.
நான்இந்தஆய்வில்என்முழுஒத்துழைப்பையும்கொடுப்பேன்என்றுஉறுதியளிக்கி றேன்.

தன்னார்வளர் **சாட்சி**

பெயர்மற்றும்முகவரி பெயர்மற்றும்முகவரி

கையொப்பம் /விரல்ரேகை: கையொப்பம் / விரல்ரேகை:

ஆராய்ச்சியாளராககையொப்பம்மற்றும்தேதி

SL NO	AGE	SEX	BMI	DIABETIC				TGL
				FBS	PPBS	HBAIC	DURATION	Y OR N
1	56	F	31.8	152	204	8.4	4	Y-274
2	69	M	25.7	164	184	7	8	Y-170
3	35	M	29.2	114	124	6.1	2	Y-160
4	48	F	27.4	245	299	10.7	8	Y-210
5	41	M	32.4	120	147	6.6	10	Y-196
6	37	F	26.9	149	198	8.9	1	Y-240
7	55	M	29.4	184	201	9.8	11	N
8	69	F	38.4	199	304	14.5	15	Y-284
9	45	F	22.4	154	199	8.6	12	Y-274
10	52	F	34.7	120	138	7.8	8	Y-198
11	45	F	29.4	124	134	6.8	6	Y-274
12	52	M	28.4	110	134	6	4	Y-221
13	39	F	34.7	85	110	5.8	7	Y-246
14	35	M	28.4	178	201	8	2	Y-274
15	45	M	22.7	112	144	6.5	6	Y-245
16	56	M	31	154	199	8.7	7	Y-214
17	68	F	28.7	199	245	11.2	12	Y- 188
18	32	M	24.45	114	178	8.8	1	Y-172
19	45	M	21.6	152	184	9.4	3	Y-184
20	54	M	19.32	99	124	5.1	9	Y-199
21	46	F	28.47	147	320	17.4	5	Y-245
22	56	M	27.45	158	198	8.4	2	Y-178
23	38	F	33.4	132	148	6.4	3	Y-298
24	44	F	38.4	123	164	6.9	8	Y-210
25	54	F	27.1	245	336	13.9	11	Y-343
26	65	F	28.4	147	204	12.4	17	N
27	62	F	32.6	124	178	9.4	5	N
28	47	M	29.4	164	245	12.4	7	Y-202
29	65	F	33.1	110	138	6.5	12	Y-189
30	40	M	21.7	145	187	8.5	4	Y-175
31	58	F	28.6	135	154	6.7	18	Y=170
32	51	M	20.4	125	138	7	5	Y-198
33	60	F	33.4	110	136	6.4	4	N
34	63	F	21.4	94	125	5.9	9	N
35	54	M	27.9	212	384	14.8	5	Y-172
36	65	F	25.3	100	202	12	8	Y-200
37	61	M	37.4	99	121	6.6	12	Y-241
38	48	M	33.8	144	198	12.4	7	N
39	54	F	25.4	154	178	9	5	Y-219
40	36	F	24.6	167	254	12.8	3	Y-250
41	54	M	24.9	189	269	13.9	8	Y-244
42	45	F	29.1	178	289	16.8	8	Y-186
43	56	M	24.6	110	154	8.4	11	Y-187
44	49	F	33.3	212	298	13.6	6	Y-198
45	67	F	22.8	147	245	12.1	12	Y-166
46	37	F	31.4	145	210	11.6	1	Y-177
47	35	M	25.5	158	249	9.4	4	Y-167

48	45	F	26.8	145	239	8.9	7	Y-189
49	57	F	29.1	141	297	14.2	11	Y-168
50	48	F	36.5	178	299	15.4	7	Y-298
51	55	M	21.5	114	138	6.3	8	N
52	31	F	28.4	145	214	7	4	N
53	36	M	29.4	110	176	8.1	7	Y-214
54	47	M	27.4	141	204	6.8	8	N
55	62	F	22.8	124	210	7.6	5	Y-188
56	58	F	24.5	154	187	8.4	6	Y-140
57	49	M	26.4	140	224	7.9	9	N
58	65	F	27.5	154	198	8.4	8	Y-198
59	54	M	28.4	145	202	7.9	8	N
60	58	M	28.7	178	242	8.4	11	Y-178
61	69	F	23.4	164	187	7	13	Y-169
62	54	M	22.4	144	188	6.6	7	N
63	47	M	25.8	148	204	6.9	8	N
64	35	F	22.4	110	138	5.8	3	Y-161
65	67	M	26.3	148	202	7.9	8	Y-198
66	58	F	24.4	198	304	9.6	7	N
67	32	F	22.4	110	178	6.2	1	Y-187
68	49	M	29.4	168	254	8.6	8	Y-178
69	51	M	34.5	204	398	15.9	14	N
70	63	F	24.6	158	245	7.9	7	Y-178
71	55	F	33.4	145	189	8.4	11	Y-165
72	47	F	27.1	157	245	7.9	9	N
73	36	M	31.2	142	289	7.6	6	N
74	58	F	24.6	164	221	8.3	8	Y-158
75	67	F	27.5	155	259	7.9	11	N
76	55	M	33.6	198	224	8.1	8	Y-187
77	59	M	29.7	185	287	9.6	9	Y-184
78	68	F	34.8	224	324	11.8	10	N
79	35	F	26.5	140	199	8.5	2	Y-202
80	60	M	28.4	164	254	7.8	9	Y-187
81	34	F	24.8	98	124	5.6	3	N
82	59	M	28.7	165	212	8.4	11	Y-224
83	69	M	25.9	135	189	6.6	12	N
84	48	F	22.7	95	129	5.9	5	Y-187
85	44	F	27.9	110	138	6.2	7	N
86	69	M	28.5	165	250	8.6	7	Y-190
87	59	M	29	145	226	6.5	5	N
88	51	F	32	90	170	6.5	2	Y-200
89	47	F	25	127	256	8	3	Y-160
90	67	M	19.45	187	310	9	6	Y-163
91	58	F	22.6	151	214	6.5	8	N
92	53	M	28	100	269	7	4	Y-182
93	49	M	26.8	130	279	10	7	N
94	50	F	29.5	156	231	8	3	Y-177
95	68	M	31	180	267	7	8	Y-210
96	46	M	29	152	248	6	4	y-198
97	50	M	25	112	230	6.8	3	Y-221

98	53	F	25.8	127	210	7	1	Y-180
99	61	M	23	95	140	6.3	8	N
100	48	F	29	100	248	7	3	Y-221

AB DIAMETER Y OR N	HYPERTENSION Y OR N	PROTEINURIA Y OR N	DIABETIC RETINOPATHY Y OR NO	MPV
Y- 100	N	N	N	9.6
N	Y	Y	Y	8.3
Y-98	y	N	N	7.3
Y-90	Y	Y	Y	11.1
Y-96	Y	Y	N	8.4
Y-88	N	N	N	9.4
Y-94	Y	Y	Y	11.4
Y-109	N	Y	Y	11.3
N	Y	Y	N	10.4
Y-114	N	N	N	9.3
Y-95	Y	N	N	7.2
Y-99	Y	N	N	7.8
Y-119	N	N	N	7.6
Y-99	Y	N	N	8.5
N	Y	N	N	7.9
Y-101	N	N	N	8.8
Y-93	Y	N	Y	10.7
N	Y	N	N	8.9
N	Y	Y	N	10.8
N	Y	N	N	7.7
Y-98	Y	Y	Y	11.3
Y-94	Y	Y	N	10.8
Y=119	N	N	N	7.8
Y-110	N	Y	Y	8.5
Y-99	Y	Y	Y	11.1
Y=96	Y	Y	N	10.9
Y-110	Y	Y	N	10.5
Y-94	Y	Y	Y	11.2
Y-101	Y	Y	N	8.3
N	Y	N	N	9.3
Y-91	Y	N	N	7.8
N	Y	N	N	7.5
Y-101	Y	N	N	7.6
N	Y	N	N	7.3
Y-94	Y	N	Y	9.9
N	Y	Y	Y	11.4
Y-110	Y	Y	Y	8.3
Y-98	Y	Y	N	10.9
Y-90	N	N	N	9.4
Y-95	N	Y	Y	10.5
N	Y	Y	Y	11.2
Y-91	Y	Y	N	9.9
N	Y	N	N	9.2
Y-101	Y	N	Y	10.7
N	Y	Y	N	10.8
Y-118	N	Y	N	10.1
N	Y	Y	Y	10.9

N	Y	Y	N	11.1
Y-95	Y	N	Y	10.5
Y-104	N	Y	Y	10.9
N	N	N	N	7.3
Y-89	N	N	N	7.6
N	Y	N	N	8.8
Y-99	Y	N	N	8
Y-101	N	Y	N	10.7
Y-98	Y	N	N	9.1
N	Y	N	N	9.6
Y-98	N	N	N	10.1
N	Y	N	Y	1O.4
Y-104	N	N	N	8.6
N	N	Y	N	8.5
N	Y	N	N	7.5
Y-91	N	N	N	7.4
N	Y	N	N	7.9
N	Y	Y	Y	10.8
Y-110	Y	Y	N	9.9
Y-96	N	N	N	7.8
N	N	N	Y	10.7
Y-108	N	Y	Y	11.3
N	Y	N	N	9.1
Y-99	N	N	N	9.2
Y-104	N	N	N	8.9
Y-115	Y	N	Y	10.3
N	Y	Y	N	10.1
N	N	N	N	9.4
Y-108	Y	Y	N	10.4
N	N	N	Y	10.2
Y-98	N	Y	Y	11.2
N	Y	Y	N	11.1
N	Y	N	N	9
N	N	N	N	7.4
Y-94	N	Y	Y	10.9
N	N	Y	N	8.5
N	Y	N	N	7.3
Y-87	Y	N	N	7.6
Y-116	Y	Y	N	10.1
Y-100	N	N	N	7.3
Y-110	N	Y	N	8.3
N	Y	N	N	9.2
N	N	Y	Y	10.9
N	Y	N	N	7.6
Y-100	Y	N	N	7.8
N	N	N	Y	11.2
Y-96	N	N	N	9.4
Y -128	Y	Y	Y	9.3
y-100	Y	N	N	7.3
Y-116	N	N	N	7.5

Y-96	N	N	N	7.7
Y-88	Y	N	N	7.8
N	Y	Y	Y	8.6