“CORRELATION OF BLOOD INFLAMMATORY MARKERS TO ANKLE BRACHIAL INDEX IN ASYMPTOMATIC ATHEROSCLEROSIS IN TYPE 2 DIABETES MELLITUS PATIENTS – A CROSS SECTIONAL STUDY”

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This is to certify that the dissertation entitled **"CORRELATION OF BLOOD INFLAMMATORY MARKERS TO ANKLE BRACHIAL INDEX IN ASYMPTOMATIC ATHEROSCLEROSIS IN TYPE 2 DIABETES MELLITUS PATIENTS – A CROSS SECTIONAL STUDY"** is a bonafide research work done by **THE CANDIDATE REG NO. BG0108007** in the Department of General Medicine, Jawaharlal Nehru Medical College, Nehru Nagar, Belgaum – 590 010.

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LIST OF ABBREVIATIONS USED

ABI	-	Ankle Brachial Index
AD	-	After death
ADP	-	Adenosine diphosphate
AGE	-	Advanced glycosylated end products
AMI	-	Acute myocardial infarction
ATP	-	Adenosine triphosphate
CAD	-	Coronary artery disease
cAMP	-	Cyclic adenosine monophosphate
CRP	-	C-reactive protein
CV	-	Cardiovascular
CVD	-	Cardiovascular disease
d	-	Deci
Da	-	Dalton
DCCT	-	Diabetes Control and Complication Trial
DM	-	Diabetes mellitus
DNA	-	De-oxyribo nucleic acid
ECG	-	Electrocardiogram
ELISA	-	Enzyme linked immunosorbent assay
ESRD	-	End stage renal disease
FPG	-	Fasting plasma glucose
GDM	-	Gestational diabetes mellitus
GLUT	-	Glucose transporter
GrB	-	Growth factor receptor bound protien
HbA_1c	-	Glycated haemoglobin

HDL	-	High density lipoprotein
HNF	-	Hepatocyte nuclear transcription factor
hs CRP	-	High sensitivity C-reactive protein
IDL	-	Intermediate density lipoprotein
IFG	-	Impaired fasting glucose
IGF	-	Insulin like growth factor
IGT	-	Impaired glucose tolerance
IL	-	Interleukin
IPD	-	In patient department
IPF	-	Insulin promoter factor
IRS	-	Insulin receptor substrate
JUPITER	-	Justification for use of statins in primary prevention
L	-	Litre
LDL	-	Low density lipoprotein
mg	-	Milligram
MODY	-	Maturity onset diabetes of the young
mRNA	-	Messenger ribonucleic acid
NGT	-	Normal glucose tolerance
NHANES III	-	Third National Health and Nutrition Examination Survey
NPH	-	Neutral protamine hagedorn
NPV	-	Negative predictive value
OGTT	-	Oral glucose tolerance test
OPD	-	Out patient department
PAI	-	Plasminogen activator inhibitor

PI3kinase	-	Phophatidyl inositol – 3 kinase.
PKC	-	Protein kinase C
PVD	-	Peripheral vascular disease
S. No.	-	Serial number
SOS	-	Son-of-sevenless
SUR	-	Sulfonyl urea receptor
T2	-	Type 2
TAFI	-	Thrombin activatable fibrinolysis inhibitor
TGF	-	Transforming growth factor
t-PA	-	Tissue plasminogen activator
VEGF	-	Vascular endothelial growth factor
VLDL	-	Very low density lipoprotein

ABSTRACT

Background and objectives

Atherosclerosis remains the major cause of death and premature disability worldwide. Diabetes mellitus is a major risk factor for the evolution of atherosclerosis. The objective of the present study was to compare the efficacy of blood inflammatory markers, hs-CRP and fibrinogen with ABI in detecting asymptomatic atherosclerosis in type 2 DM patients.

Methodology

The present One year cross sectional study was conducted in Department of Medicine, Jawaharlal Nehru Medical College, Belgaum on 38 patients with T2 DM, attending KLES Dr. Prabhakar Kore Hospital and Medical Research Centre during the period between January 2009 and December 2009. Investigations such as FBS, serum hs-CRP, serum fibrinogen, lipid profile, HbA1c were done. ABI was determined as the ratio of ankle systolic blood pressure to the brachial systolic blood pressure, with both determined using a hand held Doppler.

Results

In this study, 29 patients (76.32%) were males whereas 9 patients (23.68%) were females. Majority of the patients (34.21%) were between 35 and 50 years of age and were diabetic for a period of less than 5 years (63.16%). The HbA1c levels were between 6.4 to 7.5 gm% in 57.89% patients and 28.95% had hs CRP levels above 12 mg/L. The fibrinogen levels in 42.11% patients were between 401 to 600 mg/dL. The sensitivity of hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis was 0.88 and 0.73 respectively whereas their NPV

was 0.86 and 0.63 respectively. Comparison of the sensitivities of hs-CRP, fibrinogen, LDL, non-HDL and total cholesterol to HDL ratio showed hs-CRP had the best sensitivity (88%) towards detecting asymptomatic atherosclerosis followed by non-HDL.

Interpretation and conclusion

This study found a statistically significant difference between the sensitivities of hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis. (p=0.017).

Keywords

Atherosclerosis; Ankle brachial index; Diabetes mellitus; Fibrinogen; High sensitivity C-reactive protein; Lipid markers;

CONTENTS

LIST OF TABLES

LIST OF GRAPHS

LIST OF FIGURES

INTRODUCTION

Atherosclerosis remains the major cause of death and premature disability worldwide. Current estimations predict that by year 2020, cardiovascular disease, notably atherosclerosis will become the leading global cause of total disease burden.[1] The incidence of atherosclerosis in patients with Diabetes Mellitus (DM) is many times more than those without DM.[2] Diabetes mellitus is a major risk factor for the evolution of atherosclerosis.[3]

The clinical benefit of screening for sub clinical peripheral atherosclerosis is that it helps in identifying patients in whom timely intervention can reduce both mortality and morbidity. Peripheral atherosclerosis is a powerful predictor of future cerebrovascular and cardiovascular events such as Acute Myocardial Infarction (AMI) and Stroke.[4]

Ankle Brachial Index (ABI) can be used as a non invasive method of assessing sub clinical peripheral atherosclerosis. When compared to angiography, the sensitivity of ABI in detecting peripheral atherosclerosis is 90% and the specificity is 98%.[5] Results of one of the studies states that ABI is the gold standard in screening for atherosclerosis.[6] Organizations such as the American Heart Association and the Society of Interventional Radiology recommend the use of the ABI in the evaluation of asymptomatic atherosclerosis.[6]

Fibrinogen, an acute phase reactant, participates in early atherosclerotic plaque formation and in thrombus formation by conversion of fibrinogen to fibrin

through the action of thrombin.[7] Elevated levels of plasma fibrinogen have been associated with cardiovascular mortality and morbidity.[8]

C Reactive Protein (CRP) is one of the acute phase proteins that increase during systemic inflammation. Evidence is accumulating that serum concentrations of CRP are particularly associated with future coronary events.[9]

The present study was conducted to compare the efficacy of blood inflammatory markers, hs-CRP and fibrinogen with Ankle Brachial Index (ABI) in detecting asymptomatic atherosclerosis in type 2 DM patients.

OBJECTIVES

The objective of the present study was to compare the efficacy of blood inflammatory markers, hs-CRP and fibrinogen with ABI in detecting asymptomatic atherosclerosis in type 2 DM patients.

REVIEW OF LITERATURE

Diabetes Mellitus

Diabetes mellitus refers to a group of common metabolic disorders that share the phenotype of hyperglycemia. Several distinct types of DM exist and are caused by a complex interaction of genetics and environmental factors. Depending on the etiology of the DM, factors contributing to hyperglycemia include reduced insulin secretion, decreased glucose utilization, and increased glucose production.[1]

The metabolic dysregulation associated with DM causes secondary pathophysiologic changes in multiple organ systems that impose a tremendous burden on the individual with diabetes and on the health care system. DM is the leading cause of end-stage renal disease (ESRD), nontraumatic lower extremity amputations, and adult blindness. It also predisposes to cardiovascular diseases. With an increasing incidence worldwide, DM will be a leading cause of morbidity and mortality for the foreseeable future.[1]

History of Diabetes Mellitus

For 2,000 years diabetes has been recognized as a devastating and deadly disease. In the first century A.D. a Greek, Aretaeus, described the destructive nature of the affliction which he named "diabetes" from the Greek word for "siphon."[10]

Eugene J. Leopold in his text Aretaeus the Cappodacian describes Aretaeus' diagnosis "….for fluids do not remain in the body, but use the body only as a channel through which they may flow out. Life lasts only for a time, but not very long. For they urinate with pain and painful is the emaciation. For no essential part of the drink is absorbed by the body while great masses of the flesh are liquefied into urine".[10]

Physicians in ancient times, like Aretaeus, recognized the symptoms of diabetes but were powerless to effectively treat it. Aretaeus recommended oil of roses, dates, raw quinces, and gruel. And as late as the 17th century, doctors prescribed gelly of viper's flesh, broken red coral, sweet almonds, and fresh flowers of blind nettles.[10]

In the 17th century a London physician, Dr. Thomas Willis, determined whether his patients had diabetes or not by sampling their urine. If it had a sweet taste he would diagnose them with diabetes mellitus- "honeyed" diabetes. This method of monitoring blood sugars went largely unchanged until the 20th century.[10]

In the early 20th century, diabetologists such as Dr. Frederick Allen prescribed low calorie diets-as little as 450 calories per day for his patients. His diet prolonged the life of people with diabetes but kept them weak and suffering from near starvation.[10]

In 1921, in Ontario, Canada, a young surgeon Frederick Banting, and his assistant Charles Best, kept a severely diabetic dog alive for 70 days by injecting it with a murky concoction of canine pancreas extract. With the help of Dr. Collip

and Dr. Macleod, Banting and Best administered a more refined extract of insulin to Leonard Thompson, a young boy dying of diabetes. Within 24 hours, Leonard's high blood sugars had dropped to near normal levels.[10]

Since insulin's discovery, medical breakthroughs continued to prolong and ease the lives of people with diabetes. In 1935 Roger Hinsworth discovered there were two types of diabetes: "insulin sensitive" (type I) and "insulin insensitive" (type II). By differentiating between the two types of diabetes, Hinsworth helped open up new avenues of treatment.[10]

Starting in the late 1930s, new types of pork and beef insulin were created to better manage diabetes. Protamine zinc insulin, a longer acting insulin, was created in 1936. In 1938 NPH insulin was marketed, and in 1952 Lente, containing high levels of zinc which promotes a longer duration of action was invented.[10]

In the 1950s, oral medications-sulfonylureas were developed for people with T2 DM.[10]

The HbA1c test was devised in 1979 in order to create a more precise blood sugar measurement. With the A1c, hemoglobin, the oxygen-carrying pigment in red blood cells, is used to track glucose changes over a period of four months, the life span of the cell. Hemoglobin links with the glucose in blood; the more glucose present, the greater amount of hemoglobin linked with glucose.[10]

The A1c became a standard measurement for blood sugar control in the comprehensive ten-year study from 1983 to 1993 the Diabetes Control and

Complications Trial (DCCT). With the conclusion of the DCCT in 1993, studies showed that people who were able to keep their blood glucose levels as close to normal as possible had less chance of developing complications associated with diabetes.[10]

Before this, many doctors had not put much emphasis on tight control of blood glucose levels. The common belief for decades was that diligent monitoring of blood sugars and intensive insulin therapy had little consequence for people with diabetes. Since the DCCT's findings, statistics have proven that tight blood glucose control can be extremely beneficial for people with diabetes.[10]

CLASSIFICATION OF DIABETES MELLITUS

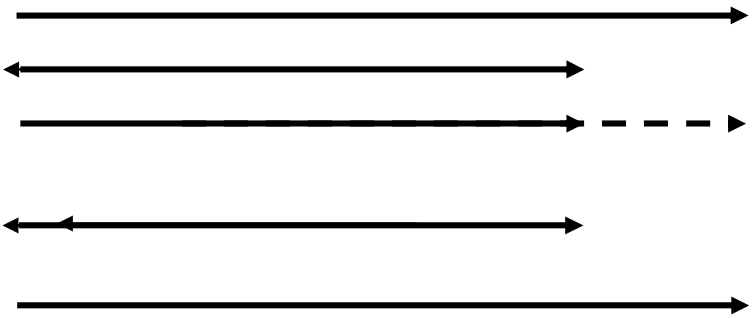
DM is classified on the basis of the pathogenic process that leads to hyperglycemia, as opposed to earlier criteria such as age of onset or type of therapy. The two broad categories of DM are designated as[1]

- Type 1
- Type 2

Both types of diabetes are preceded by a phase of abnormal glucose homeostasis as the pathogenic processes progresses. Type 1 diabetes is the result of complete or near-total insulin deficiency. Type 2 DM is a heterogeneous group of disorders characterized by variable degrees of insulin resistance, impaired insulin secretion, and increased glucose production. Distinct genetic and metabolic defects in insulin action and/or secretion give rise to the common

phenotype of hyperglycemia in type 2 DM and have important potential therapeutic implications now that pharmacologic agents are available to target specific metabolic derangements. Type 2 DM is preceded by a period of abnormal glucose homeostasis classified as impaired fasting glucose (IFG) or impaired glucose tolerance (IGT).[1]

Table 1. Spectrum of glucose homeostasis and diabetes mellitus[1]

			Hyperglycemia		
Type of diabetes	**Normal glucose tolerance (NGT)**	**Impaired fasting glucose or impaired glucose tolerance**	**Diabetes mellitus**		
			Not insulin required	**Insulin required for control**	**Insulin required for survival**
Type 1					
Type 2					
Other Specific types					
Gestational diabetes					
Time (years)					
FPG (mg/dl)	< 100	100-125		≥ 126	
2-h plasma glucose (mg/dl)	< 140	140 – 199		≥ 200	

Etiologic classification of diabetes mellitus[1]

I. Type 1 diabetes (-cell destruction, usually leading to absolute insulin deficiency)

A. Immune-mediated

B. Idiopathic

II. Type 2 diabetes (may range from predominantly insulin resistance with relative insulin deficiency to a predominantly insulin secretory defect with insulin resistance)

III. Other specific types of diabetes

A. Genetic defects of β-cell function characterized by mutations in :

1. Hepatocyte nuclear transcription factor (HNF) 4α maturity onset diabetes of young (MODY) 1
2. Glucokinase (MODY 2)
3. HNF – 1α (MODY 3)
4. Insulin promoter factor (IPF) 1 (MODY 4)
5. HNF – 1β (MODY 5)
6. Neuro D1 (MODY 6)
7. Mitochondrial deoxyribo nucleic acid (DNA)
8. Sub units of adenosine triphosphate (ATP) – sensitive potassium channel.
9. Proinsulin or insulin conversion

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, cystic fibrosis, hemochromatosis, fibrocalculouspancreatopathy.

D. Endocrinopathies – acromegaly, Cushing's syndrome, glucagonoma, pheochromocytoma, hyperthyroidism, somatostatinoma, aldosteronoma

E. Drug or chemical induced – Vacor, pentamidine, nicotinic acid, glucocorticoids, thyroid hormone, diazoxide, beta-adrenergic agonists, thiazides, phenytoin, α– interferon, protease inhibitors, clozapine, beta blockers.

F. Infections – congenital rubella, cytomegalovirus, coxsackie.

G. Uncommon forms of immune-mediated diabetes – "stiff-man" syndrome, anti-insulin receptor antibodies.

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

IV. Gestational diabetes mellitus (GDM)

EPIDEMIOLOGY

The worldwide prevalence of DM has risen dramatically over the past two decades, from an estimated 30 million cases in 1985 to 177 million in 2000. Based on current trends, more than 360 million individuals will have diabetes by the year 2030. Although the prevalence of both type 1 and type 2 DM is increasing worldwide, the prevalence of type 2 DM is rising much more rapidly because of increasing obesity and reduced activity levels as countries become more industrialized. This is true in most countries, and 6 of the top 10 countries with the highest rates are in Asia. The prevalence of type 2 DM and its harbinger, IGT, is highest in certain Pacific islands, intermediate in countries such as India and the United States, and relatively low in Russia. This variability is likely due to genetic, behavioral, and environmental factors.[1]

In India it is estimated that presently 19.4 million individuals are affected by this deadly disease, which is likely to go up to 57.2 million by the year 2025.[11]

The prevalence of diabetes is four to six fold lower in rural areas, which is probably attributed to a conventional lifestyle which has beneficial effect on glucose tolerance (IGT). National Urban Diabetes Survey done in six cities, found age standardized prevalence rates of 12% for diabetes; with a slight male preponderance and 14% for impaired glucose tolerance. Subjects under the age of 40 years, had a prevalence of five percent for DM and 13% prevalence of impaired glucose tolerance.[12]

Epidemiologic determinants and risk factors of type 2 diabetes[13]

Genetic Factors

- Genetic markers
- Family history
- "Thrifty gene(s)"

Demographic characteristics

- Sex
- Age
- Ethnicity

Behavioral and lifestyle-related risk factors

- Obesity (including distribution of obesity and duration)
- Physical inactivity
- Diet
- Stress

Metabolic determinants and intermediate-risk categories of type 2 diabetes

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

- Intrauterine malnutrition or overnutrition

DIAGNOSIS OF DIABETES

Criteria for the Diagnosis of Diabetes Mellitus[1]

- Symptoms of diabetes plus random blood glucose concentration more than or equal to 11.1 mmol/L (200 mg/dL)[a] *or*
- Fasting plasma glucose more than or equal to 7.0 mmol/L (126 mg/dL)[b] *or*
- Two-hour plasma glucose more than or equal to 11.1 mmol/L (200 mg/dL) during an oral glucose tolerance test[c]

[a]Random is defined as without regard to time since the last meal.

[b]Fasting is defined as no caloric intake for at least 8 h.

[c]The test should be performed using a glucose load containing the equivalent of 75 g anhydrous glucose dissolved in water; not recommended for routine clinical use.

In the absence of unequivocal hyperglycemia and acute metabolic decompensation, these criteria should be confirmed by repeat testing on a different day.

Glucose tolerance is classified into three categories based on the FPG:

1. FPG less than 5.6 mmol/L (100 mg/dL) is considered normal;
2. FPG equal to 5.6–6.9 mmol/L (100–125 mg/dL) is defined as IFG; and
3. FPG more than or equal to 7.0 mmol/L (126 mg/dL) warrants the diagnosis of DM.

Oral glucose tolerance test

The test uses the following procedures.

- It first employs an FPG test.
- A blood test is then taken two hours after drinking a 75 g anhydrous glucose solution.

Based on the OGTT, IGT is defined as plasma glucose levels between 7.8 and 11.1 mmol/L (140 and 199 mg/dL). Diabetes is defined when plasma glucose is more than 11.1 mmol/L (200 mg/dL), 2 h after a 75 g oral glucose load.

The current criteria, for the diagnosis of DM emphasize that the FPG is the most reliable and convenient test for identifying DM, in asymptomatic individuals. A random plasma glucose concentration more than or equal to 11.1 mmol/L (200 mg/dL) accompanied by classic symptoms of DM (polyuria, polydipsia, weight loss) is sufficient for the diagnosis of DM.

Hemoglobin A1C (HbA1c) was advocated as a diagnostic test for DM. Though there is a strong correlation between elevations in the plasma glucose and the A1C, the relationship between the FPG and the A1C in individuals with normal glucose tolerance or mild glucose intolerance is less clear, and thus the use of the A1C is not currently recommended to diagnose diabetes.[1]

PATHOPHYSIOLOGY

Insulin biosynthesis

Insulin is produced in the beta cells of the pancreatic islets. It is initially synthesized as preproinsulin. Subsequent proteolytic processing removes the amino terminal signal peptide, giving rise to proinsulin. Cleavage of an internal 31-residue fragment from proinsulin generates the C peptide and the A (21 amino acids) and B (30 amino acids) chains of insulin, which are connected by disulfide bonds. The mature insulin molecule and C peptide are stored together and co-secreted from secretory granules in the beta cells.[1]

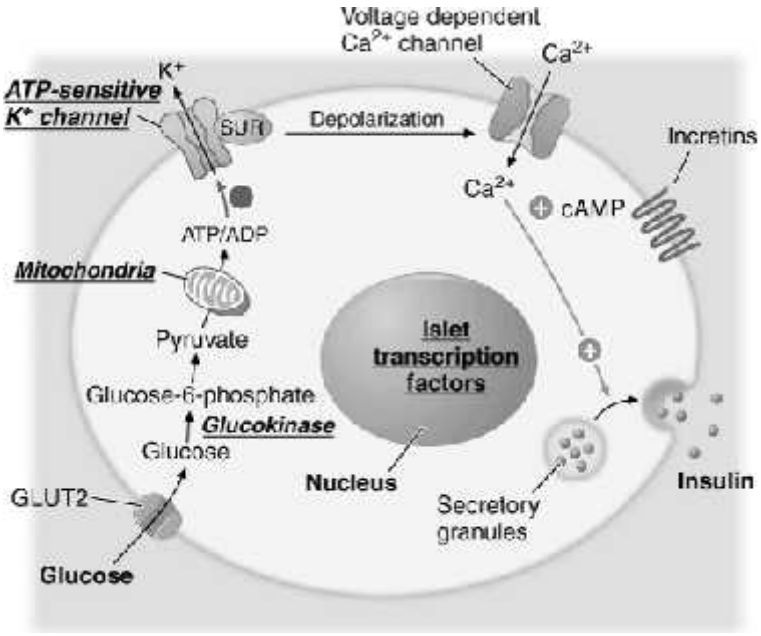


Fig 1. Diabetes and abnormalities in glucose-stimulated insulin secretion

Glucose and other nutrients regulate insulin secretion by the pancreatic beta cell. Glucose is transported by the GLUT2 glucose transporter; subsequent glucose metabolism by the beta cell alters ion channel activity, leading to insulin

secretion. The SUR receptor is the binding site for drugs that act as insulin secretagogues. Mutations in the events or proteins underlined are a cause of maturity onset diabetes of the young (MODY) or other forms of diabetes.[1]

Secretion

Glucose is the key regulator of insulin secretion by the pancreatic beta cell, although amino acids, ketones, various nutrients, gastrointestinal peptides, and neurotransmitters also influence insulin secretion. Glucose levels > 3.9 mmol/L (70 mg/dL) stimulate insulin synthesis, primarily by enhancing protein translation and processing. Glucose stimulation of insulin secretion begins with its transport into the beta cell by the GLUT2 glucose transporter. Glucose phosphorylation by glucokinase is the rate-limiting step that controls glucose-regulated insulin secretion. Further metabolism of glucose-6-phosphate via glycolysis generates ATP, which inhibits the activity of an ATP-sensitive K^+ channel. This channel consists of two separate proteins: one is the binding site for certain oral hypoglycemics (e.g., sulfonylureas, meglitinides); the other is an inwardly rectifying K^+ channel protein. Inhibition of this K^+ channel induces beta cell membrane depolarization, which opens voltage-dependent calcium channels, and stimulates insulin secretion. Insulin secretory profiles reveal a pulsatile pattern of hormone release, with small secretory bursts occurring about every 10 min.[1]

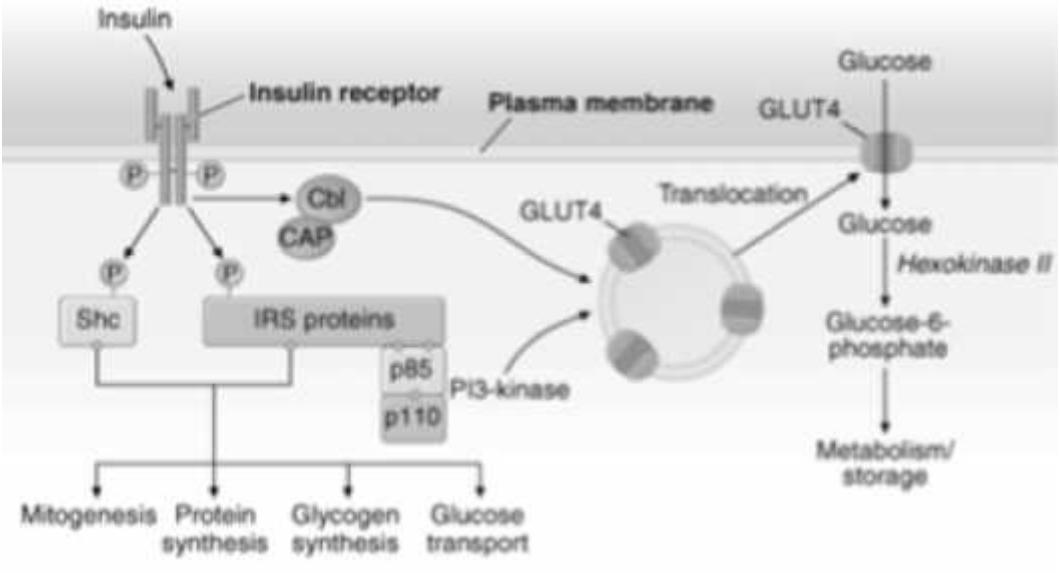


Fig. 2. Insulin signal transduction pathway in skeletal muscle

The insulin receptor has intrinsic tyrosine kinase activity and interacts with insulin receptor substrates (IRS and Shc) proteins. A number of "docking" proteins bind to these cellular proteins and initiate the metabolic actions of insulin [GrB-2, SOS, SHP-2, p65, p110, and phosphatidylinositol-3'-kinase (PI-3-kinase)]. Insulin increases glucose transport through PI-3-kinase and the Cbl pathway, which promotes the translocation of intracellular vesicles containing GLUT4 glucose transporter to the plasma membrane.[1]

Action

Once insulin is secreted into the portal venous system, about 50% is degraded by the liver. Unextracted insulin enters the systemic circulation where it binds to receptors in target sites. Insulin binding to its receptor stimulates intrinsic tyrosine kinase activity, leading to receptor autophosphorylation and the recruitment of intracellular signaling molecules, such as insulin receptor

substrates (IRS). IRS and other adaptor proteins initiate a complex cascade of phosphorylation and dephosphorylation reactions, resulting in the widespread metabolic and mitogenic effects of insulin.[1]

Glucose homeostasis reflects a balance between hepatic glucose production and peripheral glucose uptake and utilization. Insulin is the most important regulator of this metabolic equilibrium, but neural input, metabolic signals, and other hormones result in integrated control of glucose supply and utilization. In the fasting state, low insulin levels increase glucose production by promoting hepatic gluconeogenesis and glycogenolysis and reduce glucose uptake in insulin-sensitive tissues, thereby promoting mobilization of stored precursors such as amino acids and free fatty acids. Glucagon, secreted by pancreatic alpha cells when blood glucose or insulin levels are low, stimulates glycogenolysis and gluconeogenesis by the liver and renal medulla. Postprandially, the glucose load elicits a rise in insulin and fall in glucagon, leading to a reversal of these processes.[1]

Type 2 Diabetes mellitus

Insulin resistance and abnormal insulin secretion are central to the development of type 2 DM. Although the primary defect is controversial, most studies support the view that insulin resistance precedes an insulin secretory defect but that diabetes develops only when insulin secretion becomes inadequate.[1]

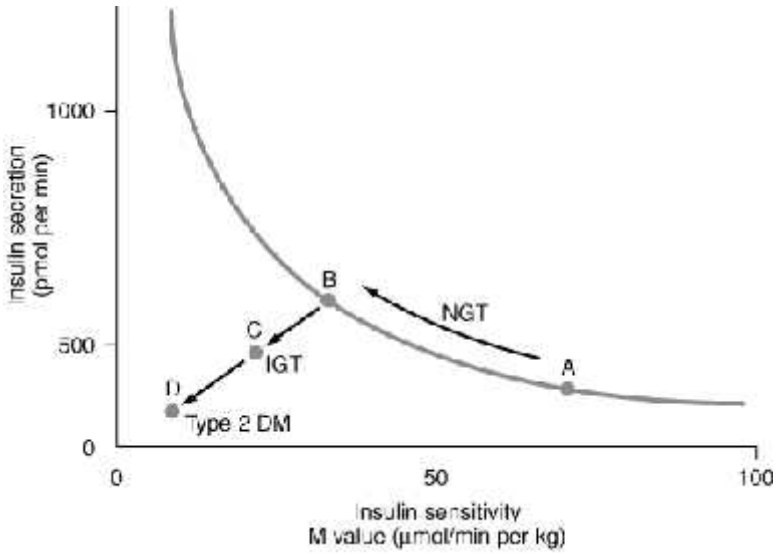


Fig 3. Metabolic changes during the development of type 2 diabetes mellitus

Insulin secretion and insulin sensitivity are related, and as an individual becomes more insulin resistant (by moving from point A to point B), insulin secretion increases. A failure to compensate by increasing the insulin secretion results initially in impaired glucose tolerance (IGT; point C) and ultimately in type 2 DM (point D).[1]

Pathophysiology of type 2 DM

Type 2 DM is characterized by impaired insulin secretion, insulin resistance, excessive hepatic glucose production, and abnormal fat metabolism. Obesity, particularly visceral or central, is very common in type 2 DM. In the early stages of the disorder, glucose tolerance remains near normal, despite insulin resistance, because the pancreatic beta cells compensate by increasing insulin output. As insulin resistance and compensatory hyperinsulinemia progress, the pancreatic islets in certain individuals are unable to sustain the hyperinsulinemic state. IGT, characterized by elevations in postprandial glucose,

then develops. A further decline in insulin secretion and an increase in hepatic glucose production lead to overt diabetes with fasting hyperglycemia. Ultimately, beta cell failure may ensue.[1]

Complications of type 2 diabetes mellitus[1]

Acute

- Diabetic Ketoacidosis
- Hyperglycemic Hyperosmolar State

Chronic

- Microvascular
    - Eye disease
        - Retinopathy (nonproliferative/proliferative)
        - Macular edema
    - Neuropathy
        - Sensory and motor (mono- and polyneuropathy)
        - Autonomic
    - Nephropathy
- Macrovascular
    - Coronary artery disease
    - Peripheral vascular disease
    - Cerebrovascular disease
- Other
    - Gastrointestinal

- Genitourinary
- Dermatologic
- Cataracts
- Glaucoma
- Infectious
- Periodontal disease

Chronic complications

The risk of chronic complications increases as a function of the duration of hyperglycemia; they usually become apparent in the second decade of hyperglycemia. Since type 2 DM often has a long asymptomatic period of hyperglycemia, many individuals with type 2 DM have complications at the time of diagnosis.[1]

The microvascular complications of both type 1 and type 2 DM result from chronic hyperglycemia. Large, randomized clinical trials of individuals with type 1 or type 2 DM have conclusively demonstrated that a reduction in chronic hyperglycemia prevents or delays retinopathy, neuropathy, and nephropathy. Other incompletely defined factors may modulate the development of complications.[1]

Evidence implicating a causative role for chronic hyperglycemia in the development of macrovascular complications is less conclusive. However, coronary heart disease events and mortality are two to four times greater in patients with type 2 DM. These events correlate with fasting and postprandial

plasma glucose levels as well as with the A1C. Other factors like dyslipidemia and hypertension also play important roles in macrovascular complications.[1]

Mechanisms of complications

Four prominent theories, which are not mutually exclusive, have been proposed to explain how hyperglycemia might lead to the chronic complications of DM.[1]

One theory is that increased intracellular glucose leads to the formation of advanced glycosylation end products (AGEs) via the nonenzymaticglycosylation of intra- and extracellular proteins. Nonenzymaticglycosylation results from the interaction of glucose with amino groups on proteins. AGEs have been shown to cross-link proteins (e.g., collagen, extracellular matrix proteins), accelerate atherosclerosis, promote glomerular dysfunction, reduce nitric oxide synthesis, induce endothelial dysfunction, and alter extracellular matrix composition and structure. The serum level of AGEs correlates with the level of glycemia, and these products accumulate as glomerular filtration rate declines.[1]

A second theory is based on the observation that hyperglycemia increases glucose metabolism via the sorbitol pathway. Intracellular glucose is predominantly metabolized by phosphorylation and subsequent glycolysis, but when increased, some glucose is converted to sorbitol by the enzyme aldosereductase. Increased sorbitol concentration alters redox potential, increases cellular osmolality, generates reactive oxygen species, and likely leads to other types of cellular dysfunction.[1]

A third hypothesis proposes that hyperglycemia increases the formation of diacylglycerol leading to activation of protein kinase C (PKC). Among other actions, PKC alters the transcription of genes for fibronectin, type IV collagen, contractile proteins, and extracellular matrix proteins in endothelial cells and neurons.[1]

A fourth theory proposes that hyperglycemia increases the flux through the hexosamine pathway, which generates fructose-6-phosphate, a substrate for O-linked glycosylation and proteoglycan production. The hexosamine pathway may alter function by glycosylation of proteins such as endothelial nitric oxide synthase or by changes in gene expression of transforming growth factor β (TGF-β) or plasminogen activator inhibitor-1 (PAI-1).[1]

Growth factors appear to play an important role in DM-related complications, and their production is increased by most of these proposed pathways. Vascular endothelial growth factor A (VEGF-A) is increased locally in diabetic proliferative retinopathy and decreases after laser photocoagulation. TGF- β is increased in diabetic nephropathy and stimulates basement membrane production of collagen and fibronectin by mesangial cells. Other growth factors, such as platelet-derived growth factor, epidermal growth factor, insulin-like growth factor I, growth hormone, basic fibroblast growth factor, and even insulin, have been suggested to play a role in DM-related complications. A possible unifying mechanism is that hyperglycemia leads to increased production of reactive oxygen species or superoxide in the mitochondria; these compounds may activate all four of the pathways described above.[1]

Atherosclerosis

Atherosclerosis is a disease affecting arterial blood vessels. It is a disease of large and medium sized arteries and is characterized by endothelial dysfunction, vascular inflammation and the buildup of lipids, cholesterol and various other kinds of debris within the intima of the vessel walls. This buildup results in plaque formation, acute and chronic luminal obstruction and diminished oxygen supply to target organs.

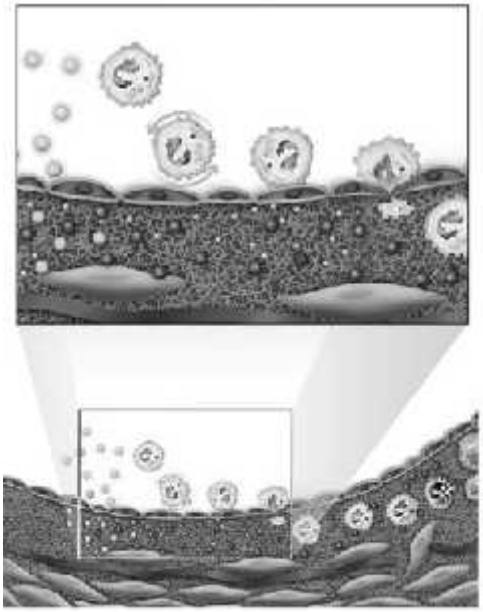


Fig 4. Cross-sectional view of an artery depicting steps in development of an atheroma from left to right[1]

The *upper panel* shows a detail of the boxed area below. The endothelial monolayer overlying the intima contacts blood. Hypercholesterolemia promotes accumulation of LDL particles (light spheres) in the intima. The lipoprotein particles often associate with constituents of the extracellular matrix, notably proteoglycans. Sequestration within the intima separates lipoproteins from some

plasma antioxidants and favors oxidative modification. Such modified lipoprotein particles (darker spheres) may trigger a local inflammatory response responsible for signaling subsequent steps in lesion formation. The augmented expression of various adhesion molecules for leukocytes recruits monocytes to the site of a nascent arterial lesion.[1]

Once adherent, some white blood cells will migrate into the intima. The directed migration of leukocytes probably depends on chemoattractant factors including modified lipoprotein particles themselves and chemoattractant cytokines depicted by the smaller spheres, such as the chemokine macrophage chemoattractant protein-1 produced by vascular wall cells in response to modified lipoproteins. Leukocytes in the evolving fatty streak can divide and exhibit augmented expression of receptors for modified lipoproteins (scavenger receptors). These mononuclear phagocytes ingest lipids and become foam cells, represented by a cytoplasm filled with lipid droplets. As the fatty streak evolves into a more complicated atherosclerotic lesion, smooth-muscle cells migrate from the media (*bottom of lower panel*), through the internal elastic membrane (*solid wavy line*), and accumulate within the expanding intima where they lay down extracellular matrix that forms the bulk of the advanced lesion (*bottom panel, right-hand side*).[1]

Pathophysiology

A complex and incompletely understood interaction exists between the critical cellular elements of the atherosclerotic lesion. These cellular elements are endothelial cells, smooth muscle cells, platelets, and leucocytes. Vasomotor

function, the thrombogenicity of the blood vessel wall, the state of activation of the coagulation cascade, the fibrinolytic system, smooth muscle cell migration and proliferation, and cellular inflammation are complex and interrelated biological processes that contribute to atherogenesis and the clinical manifestations of atherosclerosis.[14]

The "response-to-injury" theory is the most widely accepted mechanism for atherosclerosis. Endothelial injury causes vascular inflammation and a fibroproliferative response ensues. Probable causes of endothelial injury include oxidized low-density lipoprotein (LDL) cholesterol; infectious agents; toxins, including the byproducts of cigarette smoking; hyperglycemia; and hyperhomocystinemia. Circulating monocytes infiltrate the intima of the vessel wall, and these tissue macrophages act as scavenger cells, taking up LDL cholesterol and forming the characteristic foam cell of early atherosclerosis. These activated macrophages produce numerous factors that are injurious to the endothelium.[14]

Elevated serum levels of LDL cholesterol overwhelm the antioxidant properties of the healthy endothelium and result in abnormal endothelial metabolism of this lipid moiety. Oxidized LDL is capable of a wide range of toxic effects and cell/vessel wall dysfunctions that are characteristically and consistently associated with the development of atherosclerosis. These dysfunctions include impaired endothelium-dependent dilation and paradoxical vasoconstriction. These dysfunctions are the result of direct inactivation of nitric oxide by the excess production of free radicals, reduced transcription of nitric

oxide synthase messenger RNA (mRNA), and posttranscriptional destabilization of mRNA.[14]

The decrease in the availability of nitric oxide also is associated with increased platelet adhesion, increased plasminogen activator inhibitor, decreased plasminogen activator, increased tissue factor, decreased thrombomodulin, and alterations in heparin sulfate proteoglycans. The consequences include a procoagulant milieu and enhanced platelet thrombus formation. Furthermore, oxidized LDL activates inflammatory processes at the level of gene transcription by up-regulation of nuclear factor kappa-B, expression of adhesion molecules, and recruitment of monocytes/macrophages.[14]

The earliest pathologic lesion of atherosclerosis is the fatty streak. The fatty streak is the result of focal accumulation of serum lipoproteins within the intima of the vessel wall. Microscopy reveals lipid-laden macrophages, T lymphocytes, and smooth muscle cells in varying proportions. The fatty streak may progress to form a fibrous plaque, the result of progressive lipid accumulation and the migration and proliferation of smooth muscle cells. Platelet-derived growth factor, insulinlike growth factor, transforming growth factors alpha and beta, thrombin, and angiotensin II are potent mitogens that are produced by activated platelets, macrophages, and dysfunctional endothelial cells that characterize early atherogenesis, vascular inflammation, and platelet-rich thrombosis at sites of endothelial disruption. The relative deficiency of endothelium-derived nitric oxide further potentiates this proliferative stage of plaque maturation.[14]

These smooth muscle cells are responsible for the deposition of extracellular connective tissue matrix and form a fibrous cap that overlies a core of lipid-laden foam cells, extracellular lipid, and necrotic cellular debris. Growth of the fibrous plaque results in vascular remodeling, progressive luminal narrowing, blood-flow abnormalities, and compromised oxygen supply to the target organ. Human coronary arteries enlarge in response to plaque formation, and luminal stenosis may only occur once the plaque occupies greater than 40% of the area bounded by the internal elastic lamina. Developing atherosclerotic plaques acquire their own microvascular network called vasavasorum, which are prone to hemorrhage and contribute to progression of atherosclerosis.[14]

Denudation of the overlying endothelium or rupture of the protective fibrous cap may result in exposure of the thrombogenic contents of the core of the plaque to the circulating blood. This exposure constitutes an advanced or complicated lesion. The plaque rupture occurs due to weakening of the fibrous cap. Inflammatory cells localize to the shoulder region of the vulnerable plaque. T lymphocytes elaborate interferon gamma, an important cytokine that impairs vascular smooth muscle cell proliferation and collagen synthesis. Furthermore, activated macrophages produce matrix metalloproteinases that degrade collagen. These mechanisms explain the predisposition to plaque rupture and highlight the role of inflammation in the genesis of the complications of the fibrous atheromatous plaque. A plaque rupture may result in thrombus formation, partial or complete occlusion of the blood vessel, and progression of the atherosclerotic lesion due to organization of the thrombus and incorporation within the plaque.

Epidemiology

The frequency of clinical manifestations of atherosclerosis in Great Britain and Scotland in particular, is especially high. The same is true of Finland and Scandinavia. Russia and many of the former states of the Soviet Union have recently experienced an exponential increase in the frequency of coronary heart disease that likely is the result of widespread economic hardship and social upheaval, a high prevalence of cigarette habituation, and a diet high in saturated fats.

The frequency of coronary heart disease in the Far East is significantly lower than that documented in the West. Ill-defined genetic reasons for this phenomenon may exist, but significant interest surrounds the role of diet and other environmental factors in the absence of clinical atherosclerotic vascular disease in these populations. Atherosclerotic cardiovascular disease is also rare on the African continent, although growing evidence indicates that this too is changing as a result of rapid westernization and urbanization of the traditionally rural and agrarian African populations. The prevalence of coronary heart disease is also increasing in the Middle East, India, and Central and South America.

Mortality/Morbidity

Atherosclerosis is the leading cause of death in the developed world, and atherosclerosis is predicted to be the leading cause of death in the developing world within the first quarter of the next century.

- In 2005, cardiovascular disease was responsible for 864,5000 deaths, or 35.3% of all deaths that year. They included 151,000 deaths from myocardial infarction and 143,600 deaths from stroke.[15]

- An encouraging decrease in mortality due to coronary heart disease in the developed world has occurred. This decrease has not occurred in the developing world, and an exponential increase in tobacco habituation and the adoption of a Western diet high in saturated fats likely predicts the continued increase in death and disability due to coronary heart disease.

Sex

Atherosclerosis is more common among men than women. The higher prevalence of atherosclerosis in men is thought to be due to the protective effects of the female sex hormones. This sex effect is absent after menopause in women.

Age

Most cases of atherosclerotic vascular disease become clinically apparent in patients aged 40 and older.

Causes and risk factors

A number of large epidemiological studies in North America and Europe have identified numerous risk factors for the development and progression of atherosclerosis.

The risk factors can be divided into modifiable and nonmodifiable risk factors and include hyperlipidemia, hypertension, cigarette habituation, diabetes mellitus,

age, and sex. More recently, a number of novel risk factors have been identified that add to the predictive value of the established risk factors and may prove to be a target for future medical interventions.

Hyperlipidemia

Hyperlipidemia and dyslipidemia are established risk factors for atherosclerosis. Convincing evidence exists that lowering serum cholesterol reduces the risk of subsequent coronary heart disease events and overall mortality.

Hypertension

Hypertension is a risk factor for the development of atherosclerosis, atherosclerotic cardiovascular disease, and stroke. The mechanism by which hypertension causes these effects is not known, and some uncertainty exists as to what the primary and secondary factors are in a typically multifactorial syndrome. These factors may include hyperlipidemia, hypertension, diabetes mellitus, obesity, and physical inactivity.

Hypertension is associated with morphologic alterations of the arterial intima and functional alterations of the endothelium that are similar to the changes observed in hypercholesterolemia and established atherosclerosis. Endothelial dysfunction is a feature of hypertension, hyperlipidemia, and atherosclerosis and is known to represent and contribute to the procoagulant, proinflammatory, and proliferative components of atherogenesis. Hypertension has been shown, in both epidemiologic and experimental studies, to accelerate

atherosclerotic vascular disease and increase the incidence of clinical complications.

Diabetes mellitus

An important risk factor for hyperlipidemia and atherosclerosis and commonly associated with hypertension, abnormalities of coagulation, platelet adhesion and aggregation, increased oxidative stress, and functional and anatomic abnormalities of the endothelium and endothelial vasomotion.

Cigarette smoking

Cigarette smokers are two to four times more likely to develop coronary heart disease than non-smokers and they have double the risk for stroke.[15] The mechanisms are complex and likely multifactorial and result in endothelial dysfunction and a relatively hypercoagulable state. It is known that after smokers give up smoking, their risk of mortality and future cardiac events declines, although whether cardiovascular risk for former smokers ever reaches that of never smokers. Using data from the Third National Health and Nutrition Examination Survey (NHANES III), researchers found that the smoking-associated inflammatory response subsides within 5 years after smoking cessation, suggesting that the cardiovascular risk subsides gradually with reduced exposure.[16]

The other risk factors are *obesity* and *metabolic syndrome*.

In recent years, air pollution has gained increasing recognition as a contributing modifiable risk factor in the urban communities. The mechanism is thought to be through the participation of combustion-derived nanoparticles acting through proinflammatory or alternatively direct cardiac toxic pathways.

Novel risk factors

The established risk factors noted above successfully predict future cardiac events in about 50-60% of patients. In recent years, a concerted effort to identify and validate new markers of future risk of the clinical consequences of atherosclerosis has been made.

The novel risk factors that have gained importance in the recent years are

- C-reactive protein
- Fibrinogen

C-reactive protein

C-reactive protein is a protein found in the blood, the levels of which rise in response to inflammation (an acute-phase protein). Its physiological role is to bind to phosphocholine expressed on the surface of dead or dying cells (and some types of bacteria) in order to activate the complement system via the C1Q complex.[17] CRP is synthesized by the liver[18] in response to factors released by adipocytes.[19] It is a member of the pentraxin family of proteins.

History

CRP was originally discovered by Tillett and Francis in 1930 as a substance in the serum of patients with acute inflammation that reacted with the *C* polysaccharide of pneumococcus.[20] Initially it was thought that CRP might be a pathogenic secretion as it was elevated in people with a variety of illnesses including cancer,however discovery of hepatic synthesis demonstrated that it is a native protein.

Genetics

The *CRP* gene is located on the first chromosome (1q21-q23). CRP is a 224-residue protein with a monomer molar mass of 25106 Da. The protein is an annular pentameric disc in shape and a member of the small pentraxins family.

Function

CRP is a member of the class of acute-phase reactants, as its levels rise dramatically during inflammatory processes occurring in the body. This increment is due to a rise in the plasma concentration of IL-6, which is produced predominantly by macrophages as well as adipocytes. CRP binds to phosphocholine on microbes. It is thought to assist in complement binding to foreign and damaged cells and enhances phagocytosis by macrophages, which express a receptor for CRP. It is also believed to play another important role in innate immunity, as an early defense system against infections.

CRP rises up to 50,000-fold in acute inflammation, such as infection. It rises above normal limits within 6 hours, and peaks at 48 hours. Its half-life is

constant, and therefore its level is mainly determined by the rate of production and hence the severity of the precipitating cause.

Diagnostic use

CRP is used mainly as a marker of inflammation. Apart from liver failure, there are few known factors that interfere with CRP production.[18] Measuring and charting CRP values can prove useful in determining disease progress or the effectiveness of treatments. Various analytical methods are available for CRP determination, such as ELISA, immunoturbidimetry, rapid immunodiffusion, and visual agglutination.

A high-sensitivity CRP (hs-CRP) test measures low levels of CRP using laser nephelometry. Normal concentration in healthy human serum is usually below 3 mg/L. Higher levels are found in late pregnant women, inflammation, viral infections, bacterial infections and burns.[21]

Baseline C-reactive protein (CRP) levels add to the predictive value of lipid parameters in determining the risk of first myocardial infarction in apparently healthy men and women without a history of coronary heart disease. Baseline CRP levels also were found to be predictive of symptomatic peripheral vascular disease in a cohort of healthy men. CRP reflects systemic inflammation, and these results support the hypothesis that chronic inflammation may play a role in the pathogenesis and progression of atherosclerosis.

Fibrinogen

Fibrinogen (*factor I*) is a a soluble plasma glycoprotein, synthesized by the liver, that is converted by thrombin into fibrin during blood coagulation. Processes in the coagulation cascade activate the zymogenprothrombin to the serine protease thrombin, which is responsible for converting fibrinogen into fibrin. Fibrin is then cross linked by factor XIII to form a clot. Factor XIIIa stabilizes fibrin further by incorporation of the fibrinolysis inhibitors alpha-2-antiplasmin and TAFI (thrombin activatable fibrinolysis inhibitor, procarboxypeptidase B), and binding to several adhesive proteins of various cells.[22] Both the activation of Factor XIII by thrombin and plasminogen activator (t-PA) are catalyzed by fibrin.[22] Fibrin specifically binds the activated coagulation factors factor Xa and thrombin and entraps them in the network of fibers, thus functioning as a temporary inhibitor of these enzymes which stay active and can be released during fibrinolysis.[23] Recent research has shown that fibrin plays a key role in inflammatory response.

Diagnostic use

Fibrinogen levels can be measured in venous blood. Normal levels are about 150 to 400 mg/dL, depending on the method which is used. Higher levels are, amongst others, associated with cardiovascular disease. It may be elevated in any form of inflammation, as it is an acute phase protein.

Fibrinogen may be elevated in association with risk factors for atherosclerosis, including smoking, age, and diet. However, recent evidence suggests that elevated levels of fibrinogen are a strong independent predictor of

future cardiovascular events in apparently healthy patients and patients with a prior history of cardiovascular disease. This association may be as strong as the established association between hypercholesterolemia and coronary heart disease.

One of the studies recommends that fibrinogen is a major cardiovascular risk factor and that all the future studies of cardiovascular morbidity and mortality should include this variable.[8]

Ankle brachial index (ABI)

The ABI is the ratio of the blood pressure in the legs to the blood pressure in the arms. The ABI is calculated by dividing the higher systolic blood pressure in either the dorsalis pedis or posterior tibial arteries by the higher of the two systolic blood pressures in the arms. It can be performed in primary care settings without expensive or elaborate equipment or extensive training or experience.

Method

A Doppler ultrasound blood flow detector, commonly called Doppler Wand or Doppler probe, and a sphygmomanometer (blood pressure cuff) are usually needed. The blood pressure cuff is inflated proximal to the artery in question. Measured by the doppler wand, the inflation continues until the pulse in the artery ceases. The blood pressure cuff is then slowly deflated. When the artery's pulse is re-detected through the doppler probe the pressure in the cuff at that moment indicates the systolic pressure of that artery.

The higher systolic reading of the left and right arm brachial artery is generally used in the assessment. The pressures in each foot's posterior tibial

artery and dorsalis pedis artery are measured with the higher of the two values used as the ABI for that leg.

$$ABPI_{Leg} = \frac{P_{Leg}}{P_{Arm}}$$

Where, P_{Leg} is the systolic blood pressure of dorsalis pedis or posterior tibial arteries

P_{Arm} is the highest of the left and right arm brachial systolic blood pressure

Ankle Brachial Index can be used as a non invasive method of assessing sub clinical peripheral atherosclerosis. When compared to angiography, the sensitivity of ABI in detecting peripheral atherosclerosis is 90% and the specificity is 98%.[5] Results of one of the studies states that ABI is the gold standard in screening for atherosclerosis.[6] Organizations such as the American Heart Association and the Society of Interventional Radiology recommend the use of the ABI in the evaluation of asymptomatic atherosclerosis.[6] A study has even recommended that ABI should be incorporated into routine cardiovascular screening methods and that the potential of its inclusion into cardiovascular scoring systems should be examined.[24]

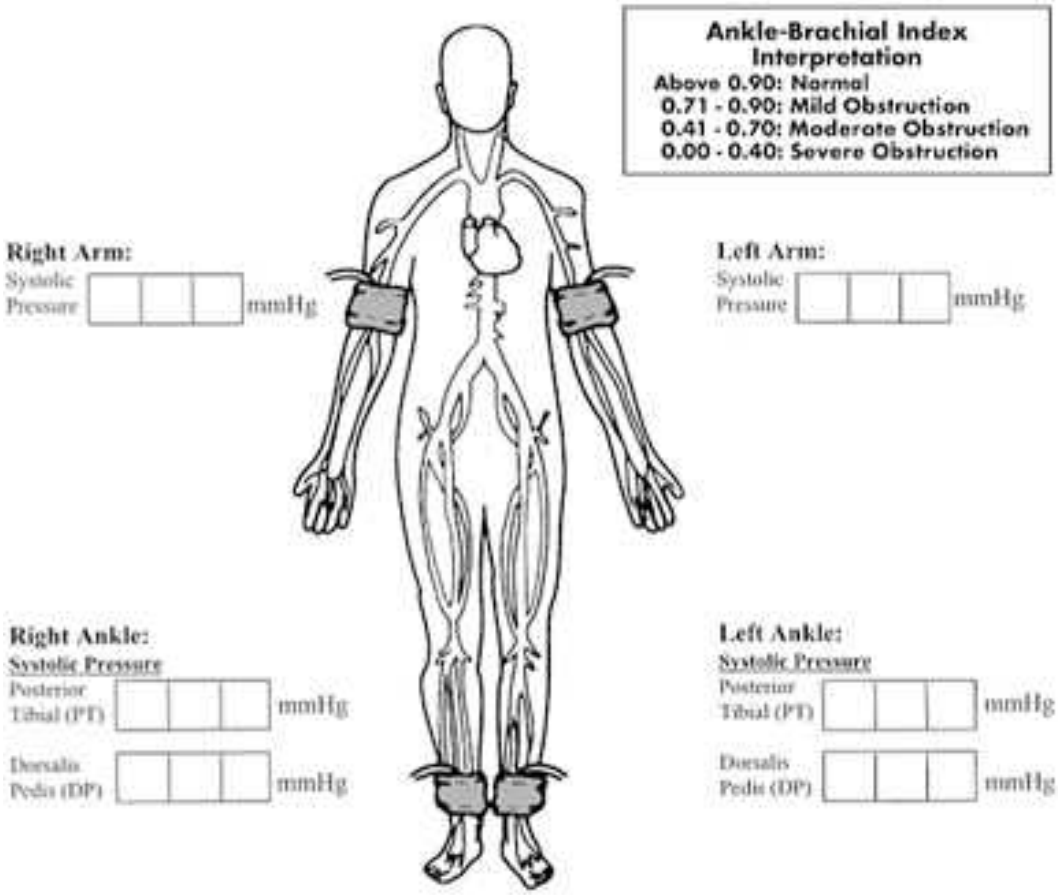


Fig 5. Ankle-Brachial Index (ABI) Worksheet

Interpretation of results

ABI scores should be interpreted as follows:

- Greater than 0.90 = normal
- 0.71 – 0.90 = mild obstruction
- 0.41 – 0.70 = moderate obstruction
- Less than 0.40 = severe obstruction

METHODOLOGY

The present study, was conducted in the Department of Medicine, KLES Dr. Prabhakar Kore Hospital and Medical Research Centre, Belgaum on patients with type 2 DM during the period of January 2009 to December 2009.

Study design

One year cross-sectional study.

Study period

The present study was conducted during January 2009 to December 2009.

Method of collection of data

Source of Data

Patients who attend OPD and/or admitted in the wards of KLES Dr. Prabhakar Kore Hospital and Medical Research Centre, Belgaum.

Sample size

A total of 38 type 2 diabetic patients were studied.

Sampling procedure

The sample size (n) was calculated using the following formula:

$$n = 4\,pq/d^2$$

Where,

n = Sample size

p = Prevalence

q = 100 – p

d = Error

The prevalence (p) of atherosclerosis in asymptomatic diabetic patients was taken as 91.4%.[25] The error (d) was taken as 10% of the prevalence. Hence, by substituting these values in the above given formula a sample size of 38 was obtained.

Selection criteria

Inclusion Criteria

- Type 2 DM patients

Exclusion Criteria

- Type 1 DM patients.
- Coronary Artery Disease: CAD was defined as a history of myocardial infarction, coronary artery bypass grafting or an abnormal ECG or coronary angiogram in the past.
- Stroke: Stroke defined as a history of stroke and/or confirmed by cerebral computed tomography or magnetic resonance imaging studies
- Peripheral Vascular Disease: PVD was defined as history of symptom of intermittent claudication, peripheral artery reconstruction, amputation of leg, lower extremity vascular surgery.

- All confounding factors for increased serum levels of hs CRP like inflammatory disorders, tissue injury or necrosis, post surgery, burns, malignancies, viral and bacterial infections

Procedure

The study was approved by the Ethical and Research Committee of Jawaharlal Nehru Medical College, Belgaum. During the study period, all patients presenting with and fulfilling the selection criteria were included in this study after obtaining informed written consent (Annexure–I).

Detailed relevant history and clinical examination was done according to predesigned and pretested proforma (Annexure-II).

Investigations like fasting blood sugar or random blood sugar, lipid profile and ECG were done. Using the lipid profile values, total cholesterol to HDL ratio and non HDL were calculated.

Others tests such as serum hs CRP, serum fibrinogen and HbA_1c were done.

Ankle brachial index was determined as the ratio of ankle systolic blood pressure to the brachial systolic blood pressure, with both determined using a hand held Doppler. The higher of left and right brachial pressures was used for the calculation of the ABI.

Statistical methods

The results were tabulated and the data was analysed using rates, ratios and percentages. From the data obtained, sensitivities and NPV were calculated using Fisher's probability test. The comparison of variables was done using binomial population test and simple regression analysis.

RESULTS

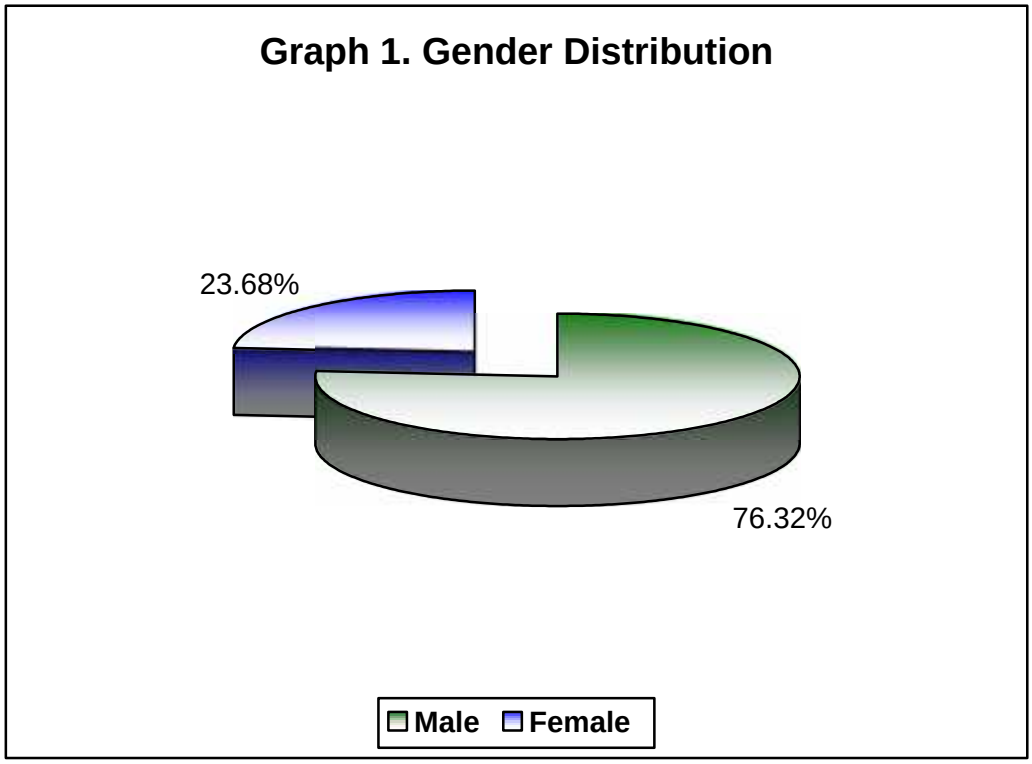
The present study was conducted in the Department of Medicine, Jawaharlal Nehru Medical College, Belgaum on patients with T2DM, who attended the out-patient department as well as those who were admitted in the wards of KLES Dr. Prabhakar Kore Hospital and Medical Research Centre during the period between January 2009 and December 2009.

A total of 38 patients were included in the study over a period of one year. The observations and findings were recorded and analyzed as follows.

BASIC DEMOGRAPHIC CHARACTERISTICS

Table 2. Gender Distribution

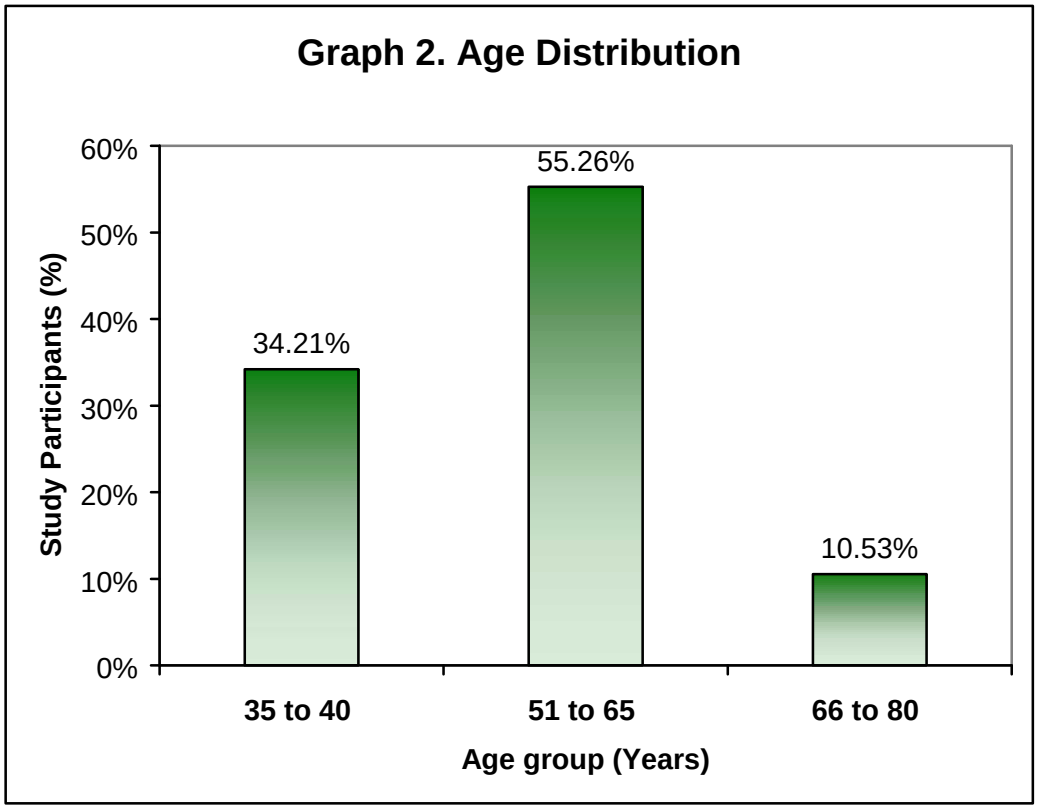
Gender	Patients (n = 38)	
	Number	**Percentage**
Male	29	76.32
Female	9	23.68
Total	**38**	**100.00**



The gender of the patients was not taken into consideration while selecting the study population. Of the 38 patients, 29 patients (76.32%) were males, whereas 9 patients (23.68%) were females.

Table 3. Age Distribution

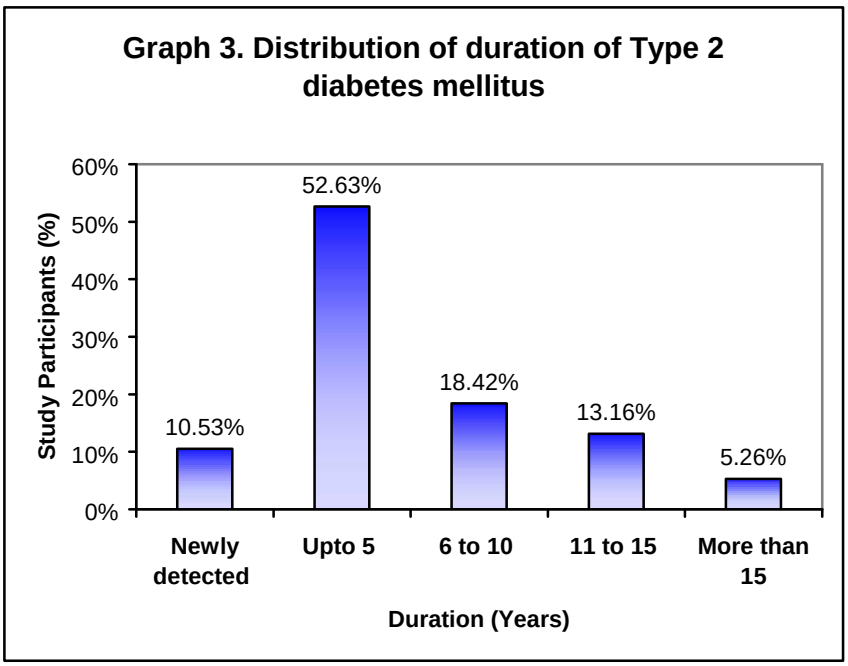
Age Group (Years)	Patients (n = 38)	
	Number	**Percentage**
35 – 50	13	34.21
51 – 65	21	55.26
66 – 80	4	10.53
Total	**38**	**100.00**



The ages of the patients included in the study ranged between 35 to 80 years. 13 patients (34.21%) were between 35 and 50 years of age, 21 patients (55.26%) were between 51 to 65 years and 4 patients (10.53%) were between 66 to 80 years. The mean age of all the 38 patients was 55.08 ± 8.86 years.

Table 4. Distribution of duration of Type 2 diabetes mellitus

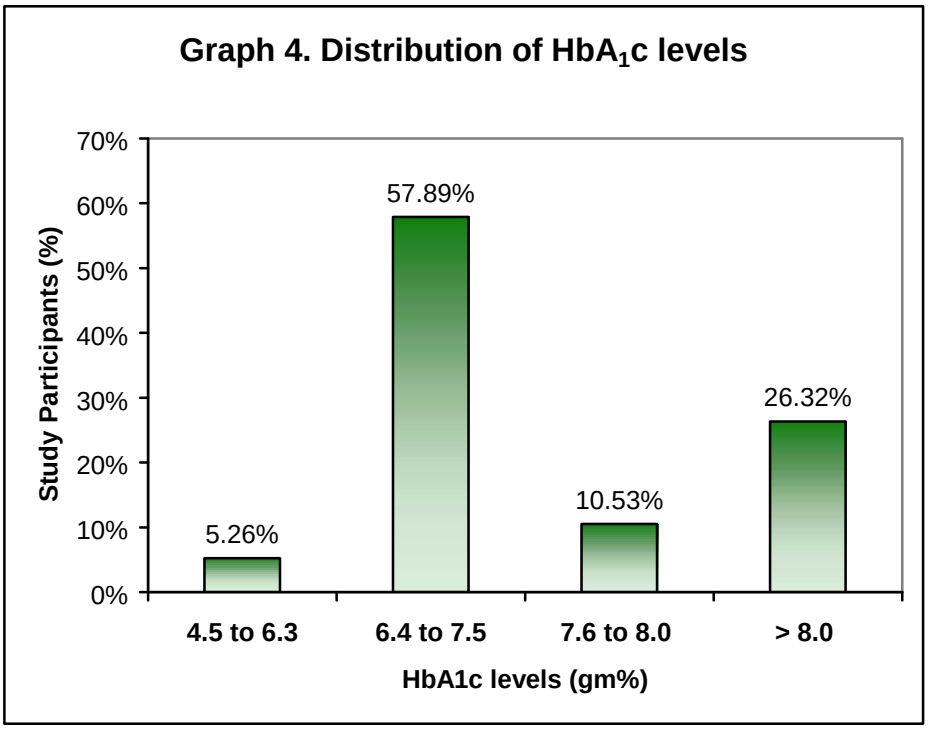
Duration (Years)	Patients (n = 38)	
	Number	**Percentage**
Newly detected	4	10.53
Up to 5	20	52.63
6 – 10	7	18.42
11 – 15	5	13.16
>16	2	5.26
Total	**38**	**100.00**



The history of duration of T2DM in the study was noted. 24 (63.16%) of the 38 subjects were diabetic for a period of less than 5 years, 7 patients (18.42%) had T2DM for a period ranging between 6 to 10 years, 5 patients (13.16%) between 11 to 15 years and 2 patients (5.26%) had T2DM for a period of more than 15 years. The duration of T2DM of all the 38 patients was 5.82 ± 4.91 years.

Table 5. Distribution of HbA_1c levels

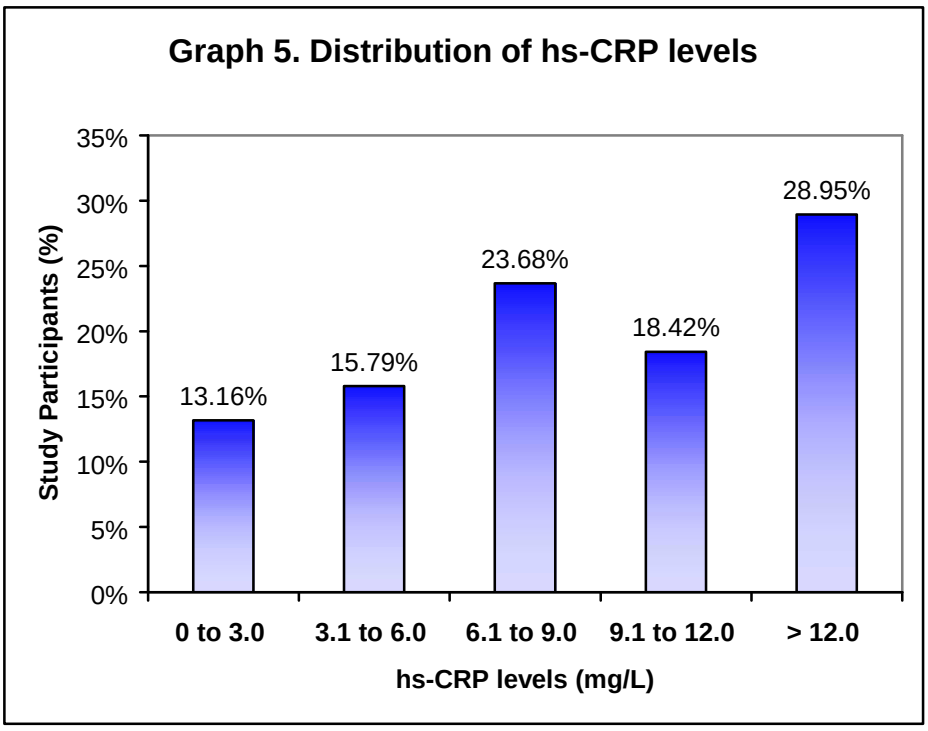
HbA_1c Levels (gm%)	Patients (n = 38)	
	Number	**Percentage**
4.5 – 6.3	2	5.26
6.4 – 7.5	22	57.89
7.6 – 8.00	4	10.53
> 8	10	26.32
Total	**38**	**100.00**



In our study, 2 patients (5.26%) had HbA_1c levels between 4.5 gm% and 6.3 gm%, 22 (57.89%) had levels between 6.4 gm% and 7.5 gm%, 4 (10.53%) had levels between 7.6 gm% and 8.0 gm% and 10 patients (26.32%) had levels in excess of 8 gm%. The mean HbA_1c of all the 38 patients was 7.51 ± 1.12.

Table 6. Distribution of hs-CRP levels

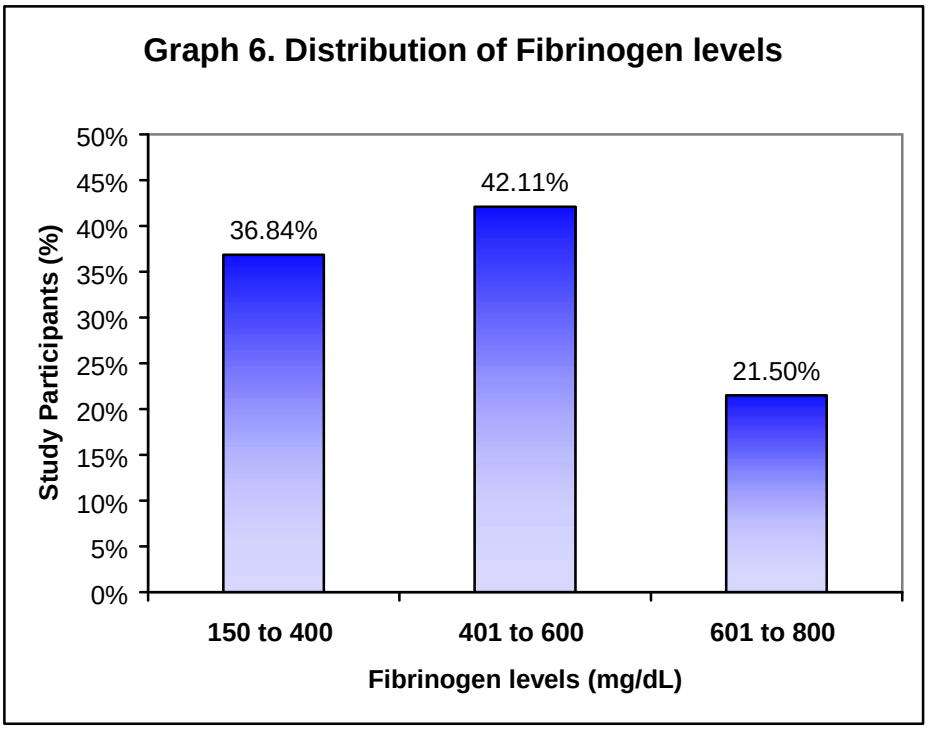
hs-CRP Levels (mg/L)	Patients (n = 38)	
	Number	**Percentage**
0 – 3.0	5	13.16
3.1 – 6.0	6	15.79
6.1 – 9.0	9	23.68
9.1 – 12.0	7	18.42
> 12	11	28.95
Total	**38**	**100.00**



In our study, 5 patients (13.16%) had normal hs-CRP levels, whereas 6 patients (15.79%) had levels between 3.1 mg/l and 6.0 mg/l, 9 patients (23.68%) between 6.1 mg/l and 9.0 mg/l, 7 patients (18.42%) between 9.1 mg/l and 12.0 mg/l and 11 of the 38 patients had levels above 12 mg/l.

Table 7. Distribution of Fibrinogen levels

Fibrinogen levels (mg/dL)	Patients (n = 38)	
	Number	Percentage
150 – 400	14	36.84
401 – 600	16	42.11
601 – 800	8	21.05
Total	**38**	**100.00**



Of the 38 subjects included in the study, 14 patients (36.84%) had normal fibrinogen levels, 16 patients (42.11%) had levels between 401 mg/dl and 600 mg/dl and 8 patients (21.05%) had levels between 601 mg/dl and 800 mg/dl.

Table 8. Comparison of duration of T2 DM

	hsCRP (Normal)	hsCRP (Increased)
Mean	6.0	4.6
S.D.	± 4.96	± 4.93

The mean duration of diabetes in patients who had raised levels of hs-CRP was 6.0 ± 4.96 years, whereas the duration of diabetes in patients with normal hs-CRP levels was 4.6 ± 4.93 years.

Table 9. Comparison of HbA1c levels

	hsCRP (Normal)	hsCRP (Increased)
Mean	7.58	7.02
S.D.	± 1.15	± 0.74

The comparison of HbA1c levels between patients who had increased levels of hs-CRP and those who had normal levels did not show a statistically significant difference. (p=0.3)

Table 10. Sensitivity of hs-CRP

Total Number of subjects	38
Subjects with normal hsCRP levels (0-3mg/l)	5
Subjects with increased hsCRP levels (>3mg/l)	33
Negative predictive value	0.86
Sensitivity	0.88

In our study, the sensitivity of hs-CRP in detecting asymptomatic atherosclerosis was 0.88 whereas its negative predictive value was 0.86.

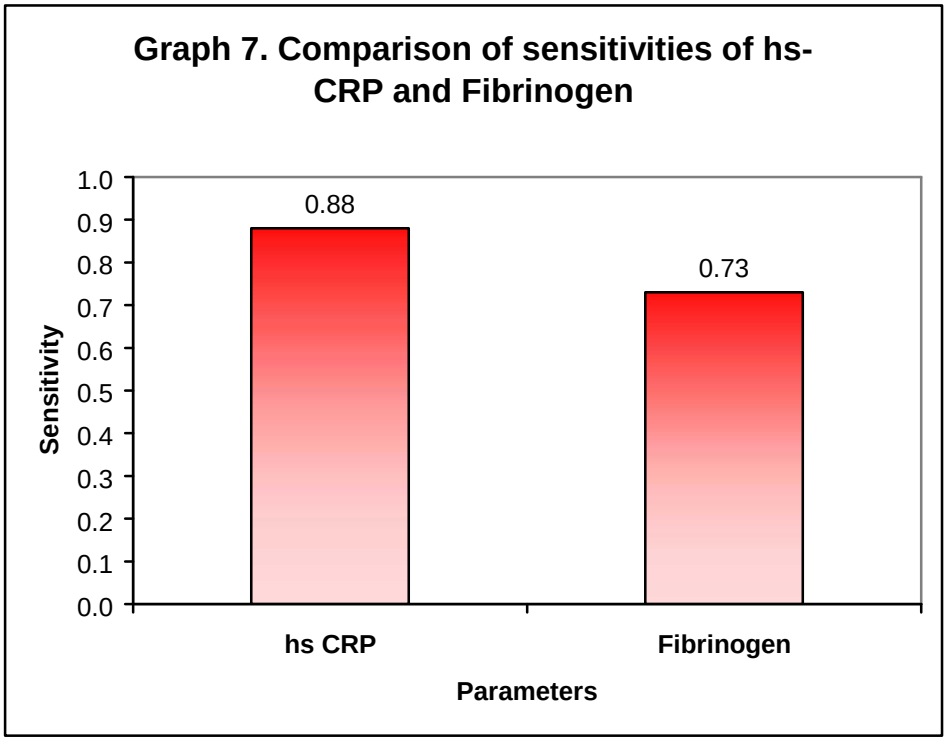
Table 11. Sensitivity of Fibrinogen

Total Number of subjects	38
Subjects with normal fibrinogen levels (150 – 400mg/dl)	14
Subjects with increased fibrinogen levels (>400mg/dl)	24
Negative predictive value	0.63
Sensitivity	0.73

The above table shows the sensitivity and negative predictive value of fibrinogen, which are 0.73 and 0.63 respectively.

Table 12. Comparison of sensitivities of hs-CRP and Fibrinogen

Marker	Sensitivity
hs CRP	0.88
Fibrinogen	0.73
P Value	0.017



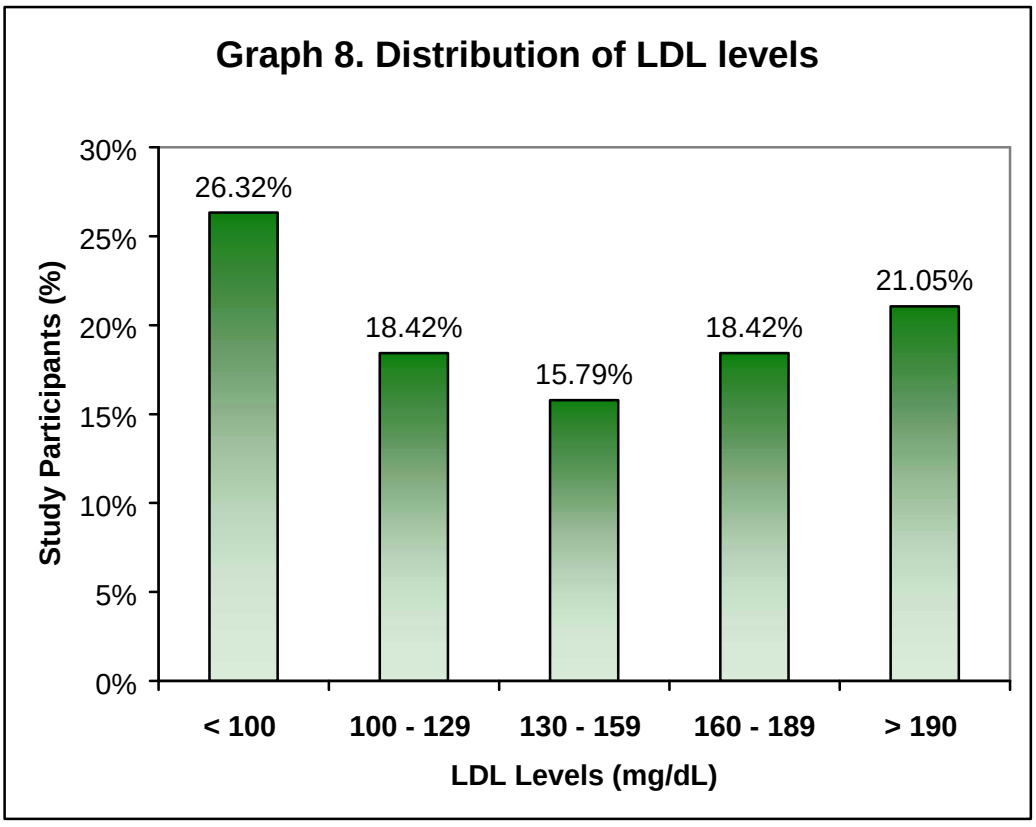
Our study found a statistically significant difference between the sensitivities of hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis. (p=0.017)

DISTRIBUTION OF LIPID PARAMETERS

The blood samples of the 38 subjects were also analyzed for the levels of total cholesterol, LDL, HDL and triglycerides and the following data was obtained.

Table 13. Distribution of LDL levels

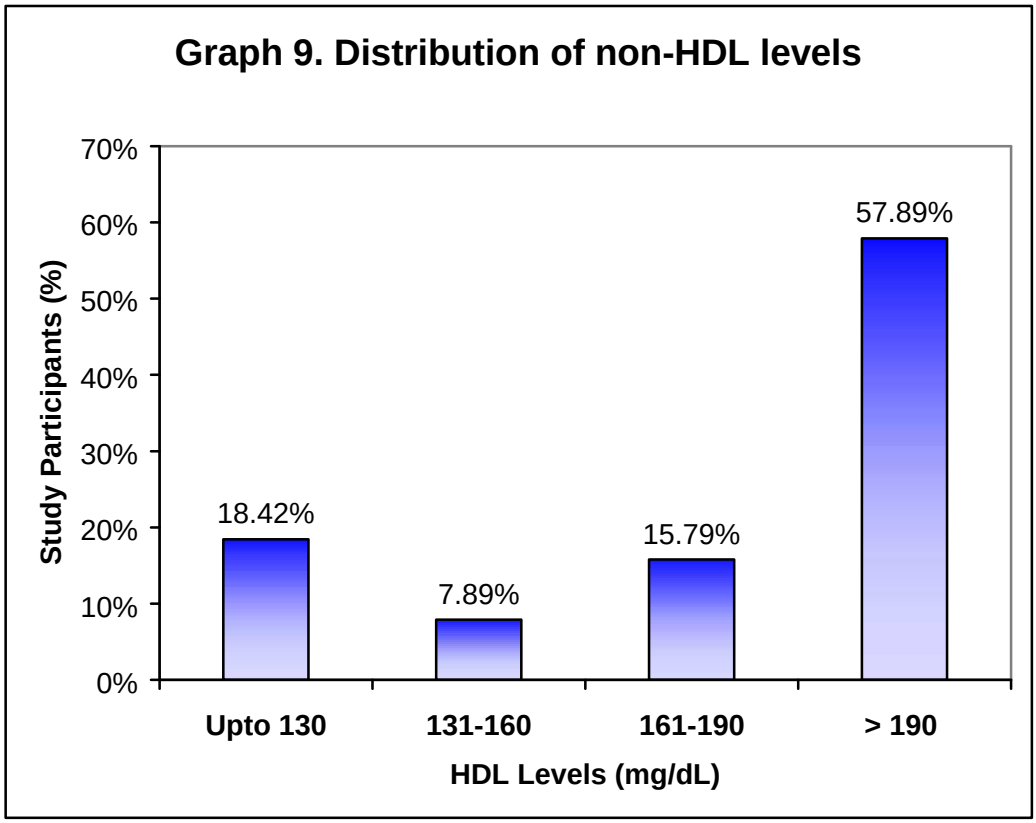
LDL Levels (mg/dL)	Patients (n = 38)	
	Number	**Percentage**
<100	10	26.32
100 – 129	7	18.42
130 – 159	6	15.79
160 – 189	7	18.42
> 190	8	21.05
Total	**38**	**100.00**



10 patients (26.32%) had normal LDL levels, whereas 28 patients (73.68%) had LDL levels above 100 mg/dl.

Table 14. Distribution of non-HDL levels

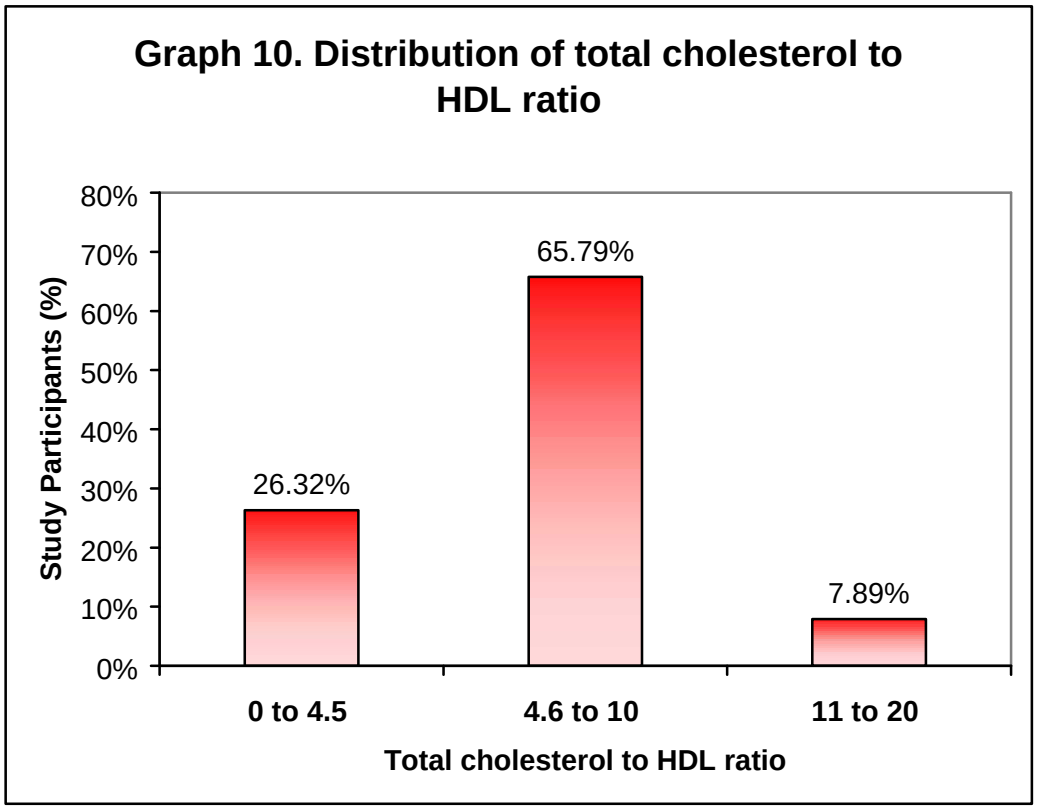
Non HDL Levels (mg/dL)	Patients (n = 38)	
	Number	**Percentage**
Upto 130	7	18.42
131 – 160	3	7.89
161 – 190	6	15.79
> 190	22	57.89
Total	**38**	**100.00**



Of the 38 patients, 7 (18.42%) had normal non-HDL levels, whereas 3 (7.89%) had levels between 131 mg/dl to 160 mg/dl, 6 (15.79%) were between 161 mg/dl to 190 mg/dl and 22 (57.89%) had levels in excess of 190 mg/dl.

Table 15. Distribution of total cholesterol to HDL ratio

Total cholesterol to HDL ratio	Patients (n = 38)	
	Number	Percentage
0 – 4.5	10	26.32`
4.6 – 10	25	65.79
11 – 20	3	7.89
Total	**38**	**100.00**



The total cholesterol to HDL ratio of all the 38 patients was calculated. 9 (23.68%) had a ratio of less than or equal to 4.5. Most of the patients (65.79%) had a ratio between 4.6 and 10, and only 4 patients had a ratio of more than 10.

Table 16. Sensitivity of LDL

Total Number of subjects	38
Subjects with normal LDL levels (<100mg/dl)	10
Subjects with increased LDL levels (≥100mg/dl)	28
Negative predictive value	0.74
Sensitivity	0.79

The sensitivity of LDL was 0.79 whereas its negative predictive value was 0.74

Table 17. Sensitivity of non-HDL

Total Number of subjects	38
Subjects with normal non HDL levels (≤130mg/dl)	7
Subjects with increased fibrinogen levels (>130mg/dl)	31
Sensitivity	0.84

Among the 38 patients, 31 patients had raised non-HDL levels and hence, its sensitivity in detecting asymptomatic atherosclerosis was 0.84.

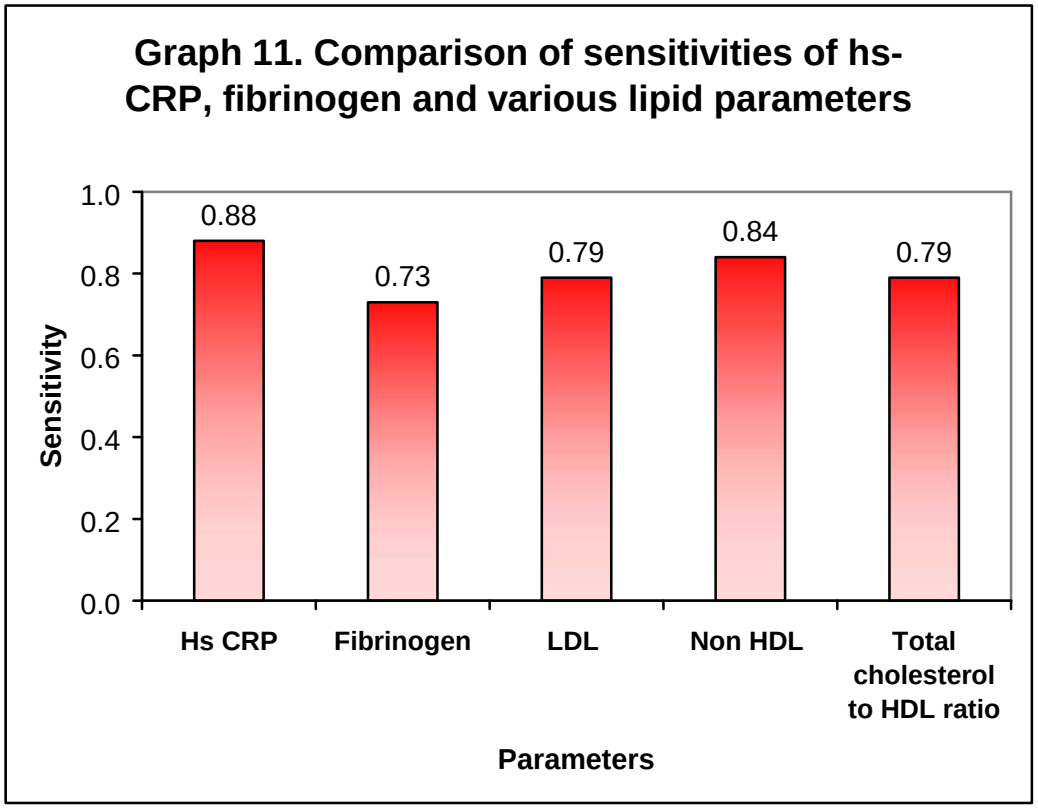
Table 18. Sensitivity of total cholesterol to HDL ratio

Total Number of subjects	38
Subjects with normal TC to HDL ratio (≤4.5)	10
Subjects with increased TC to HDL ratio (>4.5)	28
Negative predictive value	0.74
Sensitivity	0.79

The sensitivity of total cholesterol to HDL ratio was 0.79 whereas its negative predictive value was 0.74

Table 19. Comparison of sensitivities of hs-CRP, fibrinogen and various lipid parameters

Marker	Sensitivity
Hs CRP	0.88
Fibrinogen	0.73
LDL	0.79
Non HDL	0.84
Total Cholesterol to HDL ratio	0.79



The above table compares the sensitivities of hs-CRP, fibrinogen, LDL, non-HDL and total cholesterol to HDL ratio. Of the five, hs-CRP had the best sensitivity towards detecting asymptomatic atherosclerosis followed by non-HDL.

Table 20. Correlation coefficient between hs-CRP and various variables

Variable	Correlation coefficient (r)	P value
Duration of T2 DM (Years)	0.0003	> 0.05
HbA1C (gm%)	0.302	>0.05
Fibrinogen (mg/dL)	0.435	<0.01
HDL (mg/dL)	- 0.227	>0.05
LDL (mg/dL)	0.563	<0.001
Non HDL (mg/dL)	0.440	<0.01
Total Cholesterol to HDL ratio	0.472	<0.005

The above table shows the correlation co-efficient between hs-CRP levels and various variables. It was found that there was a statistically significant association between hs-CRP and LDL, non-HDL and total cholesterol to HDL ratio.

Table 21. Correlation coefficient between fibrinogen and various variables

Variable	Correlation coefficient (r)	P value
Duration of T2 DM (Years)	0.13	>0.05
HbA1C (gm%)	0.31	> 0.05
HDL (mg/dL)	- 0.24	> 0.05
LDL (mg/dL)	0.31	> 0.05
Non HDL (mg/dL)	0.28	> 0.05
Total Cholesterol to HDL ratio	0.32	< 0.05

Fibrinogen had a positive association with total cholesterol to HDL ratio but did not correlate with the other variables mentioned above in the table.

DISCUSSION

Atherosclerosis is a pervasive and malignant disease of the arterial circulation. It is by far the most frequent cause of angina, AMI, and peripheral arterial disease and is responsible for many cases of stroke. Yet, many individuals, even those with advanced disease, are unaware because they have no symptoms. In 30 to 50% of these individuals, the first indicator of atherosclerosis is an AMI, which often is fatal (sudden cardiac death).[26]

The determination of asymptomatic atherosclerosis in type 2 diabetics has a great clinical and epidemiological importance.

Since there are many pharmacologic and non-pharmacologic therapies to reduce the risk of AMI and stroke, early detection of atherosclerosis itself, before symptoms occur, can provide a major opportunity to prevent many such events.[26] Most of the mortality and morbidity associated with atherosclerotic manifestations can be prevented by early detection and aggressive treatment of asymptomatic atherosclerosis. Atherosclerosis begins to develop early in life and progresses with time, but the speed of progression is unpredictable and varies markedly among different subjects. People with traditional risk factors such as smoking, hypertension, hypercholesterolemia, and diabetes are more prone to the hazards associated with atherosclerosis.[26] The risk of cardiovascular morbity and mortality is magnified by the presence of T2DM. The traditional therapeutic approaches emphasize glycemic control, which limits microvascular disease but lacks an established benefit in macrovascular disease. Understanding

atherosclerosis in diabetes and instituting therapy guided by emerging evidence should improve outcomes in patients.[27]

At every level of risk factor exposure, there is substantial variation in the amount of evolved atherosclerosis, probably because the individual susceptibility to atherosclerosis and its risk factors varies greatly, thereby explaining the limited ability to predict clinical outcomes based on risk factor assessment alone.

The poor predictive power of major traditional risk factors was clearly demonstrated by Weissler[28] who calculated a weak likelihood ratio of 1.03 to 1.42 for prediction of coronary events in men and women, despite the high frequency of presence of risk factors in the population with cardiovascular disease.[28]

A newer understanding of the natural history of atherosclerosis, from endothelial dysfunction and activation, to intimal accumulation of lipids, inflammatory cells, and growth of smooth muscle cells, to plaque formation, disruption, and thrombosis, provides the opportunity to identify the high-risk or vulnerable patient at various stages of the process. Refinement of diagnostic methods should greatly enhance our ability to identify individuals with early-onset atherosclerosis that is likely to progress, thereby offering the opportunity for preventive intervention.[26]

Although many trials have documented the benefits of lowering plasma LDL cholesterol levels for the primary and secondary prevention of CVD, about two thirds of CVD cases cannot be prevented. As CVD morbidity and mortality rates continue to increase in developed and developing societies, despite several

improvements in CVD management, this observation suggests that other risk factors beyond LDL cholesterol and other traditional CVD risk factors may yield new insights into the assessment and management of CVD risk.[29,30,31]

Since inflammatory processes accompany all stages of atherosclerosis, measurement of plasma/serum concentrations of circulating inflammatory biomarkers might aid in identifying individuals at high risk for CVD. In particular, such biomarkers might add to the predictive value of the atherogenic lipoprotein phenotype to further improve assessment of future global cardiovascular (CV) risk,[32-36] since many of these molecules can be measured systemically by sensitive assays, and elevated concentrations in the circulation have been shown to be associated with future CV events. This study compared two such biomarkers, namely hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis.

Effective screening for asymptomatic atherosclerosis could confer great public health benefit. It is surprising that screening for subclinical atherosclerosis has not yet been incorporated into national and international clinical guidelines. In our study, ABI was used to screen for the presence of asymptomatic atherosclerosis. Previous studies have demonstrated the ABI to be a valid and reproducible method for detecting asymptomatic peripheral arterial disease.[37] Since it is simple, inexpensive, and non- invasive, the ABI is suitable for screening asymptomatic individuals and in community-based studies. As the measurement is typically highly reproducible, this technique can also be used in the clinical setting.

The present study consisted of 38 T2DM patients who had an ABI of less than 0.9. Of these, 29 patients were male and 9 patients were female.

The lifetime risk of developing cardiovascular disease after the age of 40 years is 49% for men and 32% for women.[38] The risk of developing cardiovascular disease also increases with increasing age beyond 40 years.[38] Most of the patients enrolled in our study were aged between 40 and 65 years (89.47%) with the mean age of all the patients being 55.08 ± 8.86 years.

In our study, 31 patients (81.58%) had a history of duration of T2DM of less than or equal to 10 years and only 7 patients (18.42%) had a duration of diabetes exceeding 10 years. This is in accordance with a previous study, which had stated that the incidence of symptomatic atherosclerosis increases with increasing duration of T2 DM.[39] The mean duration of T2DM of all the patients inour study was 5.82 ± 4.91 years.

The mean HbA1c level of all the patients in our study was 7.51 ± 1.12. The HbA1c of patients with increased levels of hs-CRP was 7.58 ± 1.15 whereas that of patients with normal levels was 7.02 ± 0.74. These results suggest a positive correlation between raised hs-CRP levels and poor glycemic control but the difference was not statistically significant. (p=0.3)

A study done in Columbia by Dennis et al. found a statistically significant, positive correlation between HbA1c levels and hs-CRP levels ($p<0.05$).[40]

In this study, the sensitivity of hs-CRP in detecting atherosclerosis was 0.88 whereas the sensitivity of fibrinogen was 0.73 (p=0.017). This shows that hs-CRP is a better marker of atherosclerosis than fibrinogen in T2DM patients.

A study done by Kuller et al, concluded that there was a significant association between the rise of hs-CRP levels and the subsequent development of coronary heart disease.[41] Several other studies in the past have also revealed a strong positive correlation between hs-CRP and future cardiovascular events.[42,43]

Our study has shown the positive role of both hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis but the sensitivity and negative predictive value of hs-CRP has been shown to be statistically superior to fibrinogen.

A study analyzed data from the Reykjavik Study, a prospective cohort study of 19,000 middle-aged men and women without a history of myocardial infarction. Patients with a hs-CRP value in the top one third had a relative risk of coronary heart disease of 1.92, as compared with patients whose values were in the bottom one third.[44] Regarding fibrinogen the study showed a moderate association between the plasma fibrinogen level and the risk of cardiovascular events, stroke, other vascular mortality, and nonvascular mortality in a wide range of circumstances in healthy middle aged adults. The author concluded that assessment of any causal relevance of elevated fibrinogen levels to disease needed additional research.[44]

In our study, the sensitivities of lipid measures such as LDL, total cholesterol to HDL ratio and non-HDL were calculated. Of all the lipid measures,

non-HDL had the best sensitivity in detecting asymptomatic atherosclerosis followed by both LDL and total cholesterol to HDL ratio.

This result is in accordance with a study done by Ridker et al., which concluded that non-HDL is as good as or better than the standard lipid parameters in the prediction of future cardiovascular events.[45]

These results indicate a causative role of apolipoproteins other than LDL in the development of atherosclerosis. Few studies have reported increased levels of intermediate density lipoproteins (IDL), dense LDL and dense very low-density lipoproteins (VLDL) and low high-density lipoprotein-2 in majority of patients with coronary artery disease. Hence, these lipoproteins along with LDL may have a role in the development of atherosclerosis.[26]

The results of the JUPITER trial recommend the need for therapeutic intervention in patients with normal LDL levels but increased levels of hs-CRP, irrespective of the patient's diabetic status.[46]

Risk prediction algorithms such as the Reynold's risk score that take into consideration the levels of hs-CRP along with the traditional risk factors, can be used to improve the efficacy of predicting future atherosclerotic events.

CONCLUSION

- Among T2DM patients detected to have asymptomatic atherosclerosis by ABI, levels of both the blood inflammatory markers, hs-CRP and fibrinogen were raised.

- When compared, hs-CRP was significantly superior to fibrinogen in detecting asymptomatic atherosclerosis.

- hs-CRP was also superior to lipid parameters such as LDL, non-HDL and total cholesterol to HDL ratio which are traditionally used in the risk stratification of atherosclerosis.

- Among the lipid parameters, non-HDL was superior to others in detecting asymptomatic atherosclerosis.

- hs-CRP had a positive correlation with lipid measures such as LDL, total cholesterol, non-HDL and total cholesterol to HDL ratio but did not correlate with HDL levels and with duration of T2DM.

- On the other hand, fibrinogen did not show any significant correlation with the various lipid parameters.

- hs-CRP can be used as a potent tool for risk stratification as well as detection of asymptomatic atherosclerosis in T2DM patients.

- hs-CRP levels can play a significant role in guiding therapy in patients who have raised hs-CRP levels, but whose lipid parameters including LDL are within normal range.

- hs-CRP can be used as a part of a risk prediction algorithm but further studies are needed to validate its use in such a panel.

SUMMARY

Atherosclerosis remains the major cause of death and premature disability worldwide. Fibrinogen, participates in early atherosclerotic plaque formation and in thrombus formation by conversion of fibrinogen to fibrin through the action of thrombin. C Reactive Protein is one of the acute phase proteins that increase during systemic inflammation. Ankle Brachial Index can be used as a non invasive method of assessing sub clinical peripheral atherosclerosis. The present study was conducted to compare the efficacy of blood inflammatory markers, hs-CRP and fibrinogen with ABI in detecting asymptomatic atherosclerosis in type 2 DM patients.

The present One year cross sectional study was conducted in the Department of Medicine, Jawaharlal Nehru Medical College, Belgaum on 38 patients with T2 DM, who attended the out-patient department as well as those who were admitted in the wards of KLES Dr. Prabhakar Kore Hospital and Medical Research Centre during the period between January 2009 and December 2009. ABI was determined as the ratio of ankle systolic blood pressure to the brachial systolic blood pressure, with both determined using a hand held Doppler. The higher of left and right brachial pressures were used for the calculation of the ABI.

In this study, 29 patients (76.32%) were males whereas 9 patients (23.68%) were females. Majority of the patients (34.21%) were between 35 and 50 years of age and were diabetics for a period of less than 5 years (63.16%). The HbA1c levels were between 6.4 to 7.5 gm% in 57.89% patients and 28.95% had

hs CRP levels above 12 mg/L. The fibrinogen levels in 42.11% patients were between 401 to 600 mg/dL. The sensitivity of hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis was 0.88 and 0.73 respectively whereas their NPV was 0.86 and 0.63 respectively. Comparison of the sensitivities of hs-CRP, fibrinogen, LDL, non-HDL and total cholesterol to HDL ratio showed hs-CRP had the best sensitivity (88%) towards detecting asymptomatic atherosclerosis followed by non-HDL.

This study found a statistically significant difference between the sensitivities of hs-CRP and fibrinogen in detecting asymptomatic atherosclerosis. ($p=0.017$).

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ANNEXURE I – CONSENT FORM

"CORRELATION OF BLOOD INFLAMMATORY MARKERS TO ANKLE BRACHIAL INDEX IN ASYMPTOMATIC ATHEROSCLEROSIS IN TYPE 2 DIABETES MELLITUS PATIENTS" – A ONE YEAR CROSS SECTIONAL STUDY

Objective and purpose of the study

This research is intended to study the correlation of blood inflammatory markers such as hs-CRP and fibrinogen to ankle brachial index in asymptomatic atherosclerosis in type 2 diabetes mellitus patients. The principal investigator of the study is Dr. ***** ****** under the guidance of Dr. **** *****. My co-operation will be of great help to patients with asymptomatic atherosclerosis in type 2 diabetes mellitus patients in future.

Procedure

If you agree to be part of the research study you will be asked the relevant history and will be subjected to relevant clinical examination and investigations. You will also have to give blood and urine samples for the necessary investigations

Risk and Benefits

The only risk and possible discomfort you might get is while taking blood from arm for the investigations. It may cause swelling, pain, redness, bruising or infection (rarely happens) at the site from where the blood is drawn.

Alternatives

Taking part in this study is voluntary. You may choose not to take part in this study, or if you decide to take part you can later change your mind and withdraw from the study. Your decision will not change the present or future health care or other services that you receive. The study doctor or sponsor may stop your participation in this study any time. If you choose not to take part in the study you will receive the standard treatment for patients with your condition.

Privacy and Confidentiality

All information collected about you during the course of this study will be kept confidential to the extent permitted by law. The code numbers will identify you in this research record. Information from this study may be published but your identity will be confidential in any publication.

Institution / Sponsor's policy

Does not apply to this research

Financial incentives for participation

You will not be paid / offered any gifts /incentives for participating in the study.

Authorization to publish the results

The results of the study would be forwarded to the KLE University, Belgaum as part of requirement towards the completion of MD degree, review and publishing.

If you have any questions about my rights as a participant you may call Principal and Chairman, J.N.M.C Ethical Committee for Human Research.

Consent Statement

I voluntarily agree to take part in this study by signing below. I may withdraw at any time. I am not giving up any of my legal rights by signing this form. My signature below indicates that I have read, or it has been read to me, this entire consent form, and have had all my questions answered.

Name of the Participant:_____ Signature / Thumb print _____

In case of queries during study or in future you may contact following person

Principal investigator : Dr. ****** *********

Guide : Dr. ****** *********

Name of the Witness _____ Signature _____

Investigator Name_____ Signature _____

Date: Place:

ANNEXURE II – PROFOMA

Name	S. No.
Age	Case/Control
Sex	OPD/IPD No.
Address	
Occupation	
Date of enrollment	

HISTORY

Presenting Complaints

History of presenting illness

Significant past history

⇒ Coronary artery disease

⇒ Cerebrovascular disease

⇒ Peripheral Vascular disease

DIABETIC HISTORY

⇒ Polyuria

⇒ Polydipsia

⇒ Polyphagia

⇒ Unexplained weight loss

Duration of diabetes

Age at onset of diabetes

Family history of diabetes

Treatment history of diabetes

⇒ Drug name

⇒ Dosage

⇒ Frequency

⇒ Duration of treatment

Other drugs if any

Significant personal history

Significant family history

GENERAL PHYSICAL EXAMINATION

MEASUREMENT OF ANKLE BRACHIAL INDEX

VITAL SIGNS

Pulse

Blood Pressure

⇒ Supine position

⇒ Standing position

ABI

Respiratory Rate

Temperature

Any significant findings (Pallor, Icterus)

SYSTEMIC EXAMINATION

Respiratory System

Cardiovascular System

Per Abdominal Examination

Central Nervous System

EVALUATION OF DIABETIC COMPLICATIONS

MICROVASCULAR COMPLICATIONS

Diabetic Neuropathy Symptoms

⇒ Tingling/Numbness/Burning

⇒ Neuropathic pain

⇒ Diabetic polyradicular pain

⇒ Mononeuropathy

Diabetic Neuropathy Examination

⇒ Sensory loss

⇒ Ankle reflex

⇒ Abnormal position sense

DIABETIC RETINOPATHY

Fundus examination

MACROVASCULAR COMPLICATIONS

Coronary artery disease

Cerebrovascular disease

Peripheral vascular disease

GENITOURINARY COMPLICATIONS

Diabetic vesicopathy

Erectile dysfunction

Retrograde ejaculation

DERMATOLOGICAL MANIFESTATIONS

Diabetic dermopathy

Scleroderma

Lipoatrophy/Hypertrophy

Xerosis/Pruritis

INVESTIGATIONS

- Fasting blood sugar
- Serum hs-CRP (Fasting)
- Serum fibrinogen (Fasting)
- Lipid Profile
- HbA_1c
- ECG
- Special investigations if any

_____

Signature of the guide

ANNEXURE III - KEY TO MASTER CHART

Sl. No	IP No./OP No.	Demography		History		Investigations											
										Lipid profile							
		Age (Years)	Sex	Duration of T2 DM (Years)	Age at onset (Years)	hs CRP (mg/L)	Fibrinogen (mg/dL)	Fasting blood sugar (mg/dL)	HbA_{1C}	Total cholesterol (mg/dL)	HDL (mg/dL)	LDL (mg/dL)	Non HDL (mg/dL)	Triglycerides (mg/dL)	Total to HDL ratio	Echocardiogram	Ankle brachial index
1	1330885	67	M	7	60	6	450	122	7.1	234	36	155	198	204	6.5	WNL	0.83
2	1304274	51	F	2	49	9	520	110	6.8	284	42	194	242	254	6.8	WNL	0.88
3	1318447	52	M	5	47	4	480	90	6.5	220	20	164	200	196	11.0	WNL	0.81
4	1315896	45	M	4	41	11	280	104	7.0	260	30	180	230	234	8.7	WNL	0.83
5	1177658	65	M	15	50	8	540	144	8.6	302	36	200	266	310	8.4	WNL	0.86
6	1314458	55	F	15	40	4	460	114	8.0	226	56	124	170	200	4.0	WNL	0.87
7	305232	75	F	15	60	14	420	98	6.8	236	36	150	200	230	6.6	WNL	0.81
8	362956	46	F	4	42	18	600	86	6.6	272	40	174	232	306	6.8	WNL	0.89
9	302125	50	M	ND	50	10	220	130	7.4	312	34	202	278	406	9.2	WNL	0.82
10	314071	75	M	17	48	14	500	100	7.0	396	24	276	372	496	16.5	WNL	0.8
11	322478	55	M	3	52	7	540	90	6.1	250	32	162	218	304	7.8	WNL	0.81
12	1094747	59	M	1	58	12	180	108	6.4	190	36	112	154	226	5.3	WNL	0.86
13	321992	50	M	ND	50	17	580	113	6.9	292	32	200	260	312	9.1	WNL	0.82
14	1062034	54	M	3	51	1	310	127	7.3	160	38	96	306	132	4.2	WNL	0.79
15	1314468	55	M	8	47	15	640	181	11.3	160	38	96	248	132	4.2	WNL	0.77
16	305202	55	M	5	50	13	320	89	7.0	320	34	200	286	424	9.4	WNL	0.85
17	366831	60	F	8	52	16	730	97	6.9	346	40	210	306	450	8.7	WNL	0.89
18	366793	45	M	4	41	8	620	131	8.2	250	36	142	214	356	6.9	WNL	0.81
19	316318	60	M	12	48	3	370	79	6.5	168	44	94	248	150	3.8	WNL	0.76
20	312734	65	F	16	49	10	380	115	7.9	224	40	140	184	212	5.6	WNL	0.87
21	365858	47	M	3	44	6	400	110	7.0	264	30	162	234	354	8.8	WNL	0.81
22	307029	62	M	5	57	11	520	220	9.6	324	38	200	286	424	8.5	WNL	0.88
23	306173	53	M	7	46	14	800	190	8.4	286	26	176	260	400	11.0	WNL	0.79
24	364885	42	M	3	39	7	610	130	8.0	190	28	120	162	228	6.8	WNL	0.84
25	302126	40	F	ND	40	1	310	90	7.0	142	36	82	106	110	4.0	WNL	0.88
26	309466	43	M	ND	43	16	460	170	7.8	242	30	150	212	312	8.1	WNL	0.8
27	365802	65	M	5	60	10	640	210	8.1	202	32	120	170	276	6.3	WNL	0.86
28	302143	55	M	6	49	9	590	160	8.8	170	30	108	140	172	5.7	WNL	0.83
29	311147	63	M	12	51	8	610	130	6.8	180	30	112	150	202	6.0	WNL	0.82
30	361936	50	M	2	48	18	430	230	10.2	282	36	182	246	326	7.8	WNL	0.76
31	362962	45	F	2	43	12	540	170	8.5	212	38	122	174	252	5.6	WNL	0.81
32	310272	50	M	3	47	15	630	120	7.2	208	30	140	178	202	6.9	WNL	0.8
33	362977	46	M	1	45	4	500	100	6.4	124	40	60	84	112	3.1	WNL	0.87
34	305278	68	M	5	63	8	380	110	7.2	140	34	84	106	126	4.1	WNL	0.85
35	322463	52	F	1	51	2	180	80	6.2	136	42	76	94	102	3.2	WNL	0.89
36	1089474	52	M	7	45	3	155	104	8.1	156	40	66	116	256	3.9	WNL	0.86
37	1107986	56	M	10	46	9	300	126	6.8	118	38	60	80	130	3.1	WNL	0.76
38	365244	65	M	5	60	6	380	110	7.0	156	34	94	122	140	4.6	WNL	0.8

KEY TO MASTER CHART

dL	-	Decilitre
DM	-	Diabetes mellitus
F	-	Female
HbA_1c	-	Glycated haemoglobin
HDL	-	High density lipoprotein
hs CRP	-	High sensitivity C-reactive protein
IP No.	-	In patient number
LDL	-	Low density lipoprotein
M	-	Male
mg	-	Milligram
ND	-	Newly detected
OP No.	-	Outpatient number
Sl. No.	-	Serial Number
T2	-	Type 2
WNL	-	Within normal limits