Evaluation of carotid intima media thickness as a predictor of macrovascular complications in type 2 diabetes mellitus

Dissertation submitted in partial fulfillment of regulation for the award of M.D.

Degree in General Medicine (Branch I)

The Tamilnadu
Dr. M.G.R. Medical University
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Coimbatore Medical College & Hospital
Coimbatore - 641 014

certificate

This is to certify that the dissertation entitiled ***"Evaluation of carotid intima media thickness as a predictor of macrovascular complications in type 2 diabetes mellitus"****, herewith submitted by Dr SHANMUGAVADIVU S, post graduate in General Medicine Coimbatore Medical College Hospital is the record of a bonafide research work carried out by him under our guidance and supervision from April 2008 to December 2008.*

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Place : Coimbatore

Date: 01.12.2008

DECLARATION

I solemnly declare that the dissertation titled **"EVALUATION OF CAROTID INTIMA MEDIA THICKNESS AS A PREDICTOR OF MACROVASCULAR COMPLICATIONS IN TYPE 2 DIABETES MELLITUS"** was done by me from April 2008 to December 2008 under the guidance and supervision of **Professor Dr. S.VEERAKESARI.MD. and Professor Dr. K.UMAKANTHAN.**

This dissertation is submitted to the Tamilnadu Dr. MGR Medical University towards the partial fulfillment of the requirement for the award of MD Degree in General Medicine (Branch I).

Dr. SHANMUGAVADIVU S

Place : Coimbatore
Date : 01.12.2008

Coimbatore Medical College

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ETHICS COMMITTEE

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Period of Study : 2006 - 2009

College : COIMBATORE MEDICAL COLLEGE

Dissertation Topic : EVALUATION OF CAROTID INTIMA MEDIA THICKNESS AS A PREDICTOR OF MACROVASCULAR COMPLICATIONS IN TYPE 2 DIABETES MELLITUS

The Ethics Committee, Coimbatore Medical College has decided to inform that your Dissertation is accepted / ~~Not accepted~~ and you are permitted / ~~Not Permitted~~ to proceed with the above Study.

Coimbatore - 14.

Date : 5.3.2008

Secretary
Ethics Committee

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CONTENTS

BACKGROUND

Type 2 diabetes is known to be associated with an excessively high rate of morbidity and mortality from macrovascular disease [1].This increased risk has been attributed to high prevalence of multiple atherosclerotic risk factors among diabetic patients.

Recently, “Inflammation” and “Inflammatory” cytokines have been postulated to be important pathogenic factors in the development of insulin resistance and Type 2 diabetes [3, 4]. CRP, a non-specific marker of the inflammatory response, is most consistently associated with the development of Type 2 diabetes, (5, 6) however, a casual association remains unproven. CRP and interleukin-6 (IL-6) are associated with the risk of CHD and severity of atherosclerosis (7, 8). Whether these molecules play a causative role, or simply act as markers of the acute-phase reaction, is debatable. The molecular basis for the link between inflammation and diabetes likely relates to the action of cytokines such as IL-6 and TNF-α, which includes insulin resistance and stimulates the acute phase inflammatory response (9). The macrovascular complications are essentially due to accelerated atherosclerosis because of endothelial dysfunction linked to hyperglycemia and other factors setting up pro-atherogenic pattern (10). A non-invasive and relatively inexpensive investigation for identifying atherosclerosis, even in an asymptomatic patient, is measurement of IMT of the extra cranial carotid arteries by Doppler ultrasound. Endothelial dysfunction is an early functional marker and IMT an early morphological parameter of atherosclerosis (11).

Ethnic differences in CHD morbidity and mortality (12), in the prevalence and course of type 2 diabetes (13) and in carotid IMT have been reported (14). Asian Indians have a high risk of diabetes and have an obesity phenotype characterized by lean BMI, central obesity and high body fat percentage. Also they have high degree of insulin resistance (15). The increased predisposition of certain population of type 2 diabetes raises important question and emphasize the need for studies on inflammatory markers, insulin resistance, IMT and its determinants in different populations.

The basic pathophysiology underlying macrovascular disease is atherosclerosis. The word atherosclerosis is of Greek origin and literally means focal accumulation of lipid (ie. athero [porridge]) and thickening of intima (ie. sclerosis [hardening]) (2). It is basically a disease of large and medium sized muscular arteries and is characterized by endothelial dysfunction, vascular inflammation and build up of lipids, cholesterol, calcium and cellular debris within the intima of the vessel wall. This result in plaque formation, vascular remodeling, acute and chronic luminal obstruction, abnormalities of blood flow and diminished oxygen supply to target organs (3).

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 thrombogenecity 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 process that contribute to

atherogenesis and clinical manifestations of atherosclerosis (6). A large number of growth factors, cytokines and vasoregulatory molecules participate in this process (5).

The mechanism of atherogenesis remains uncertain. The “response-to-injury” theory is most widely accepted. Endothelial injury causes vascular inflammation and a fibro proliferative response ensues (3). Probable causes of endothelial injury include LDL cholesterol, infectious agents, toxins, including by products of cigarette smoking, hyperglycemia and hyperhomocysteinemia (2). Type 2 diabetes mellitus related macrovascular complications are primarily attributed to endothelial dysfunction and dyslipedemia. Circulating monocytes infiltrate the intima of the vessel wall and these tissue macrophages act as scavenger cells, taking up LDL and forming the characteristic foam cell of early atherosclerosis. These activated macrophages produce numerous factors that are injurious to endothelium (4).

The carotid arteries can be well visualized by ultrasonography and ultrasonographic measurements of the thickness of carotid intima media wall (IMT) have been investigated as a technique to identify and monitor subclinical atherosclerosis. B mode ultrasound is most commonly used and the intima media thickness is measured and averaged over six sites in each carotid artery.

As India has been predicted to be the world’s capital for diabetes mellitus and coronary artery disease more data availability for Indian population becomes all the more crucial. We thus are doing a comparative study and intend to get a positive correlation between the two so that Doppler can be taken as a non-invasive investigative

modality in individuals at high risk. If found significant we can predict the increase likelihood of these patients to develop a coronary event and take measures for preventing the same.

REVIEW OF LITERATURE

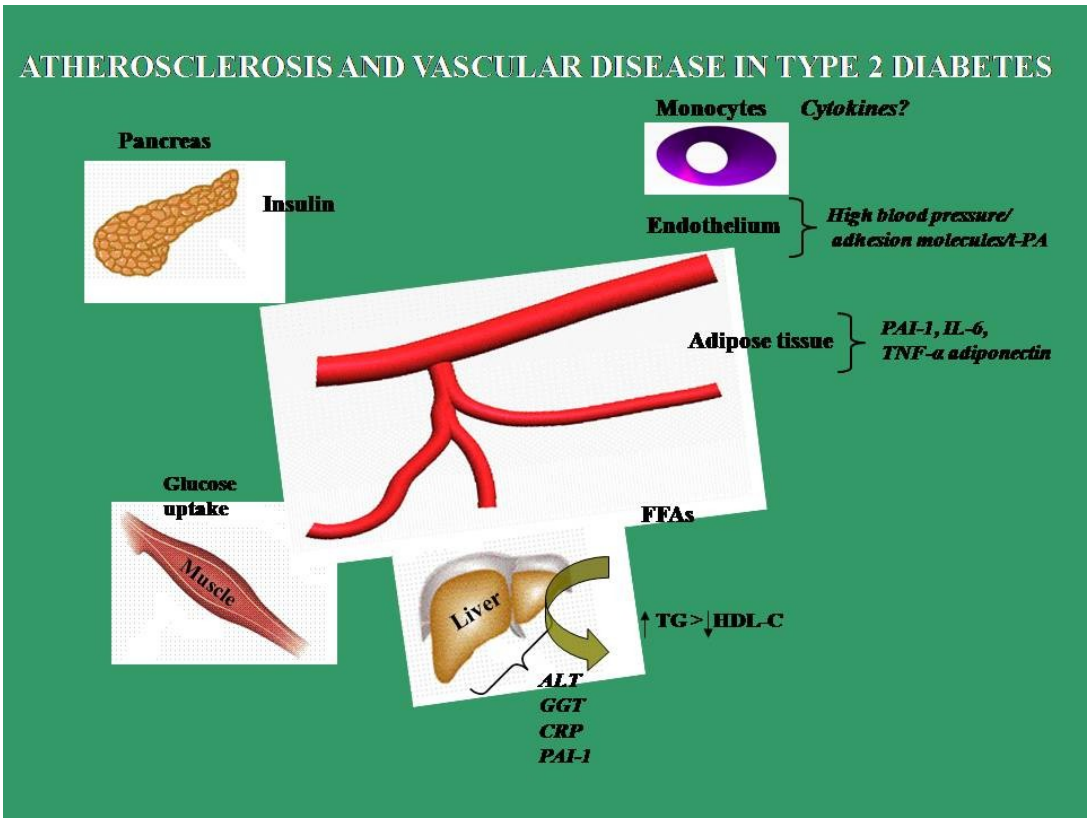
At the beginning of the 20th century, cardiovascular diseases (CVD) accounted for <10% of all the deaths worldwide. At its end, CVD accounted for nearly the deaths in the developed countries and 25% in developing world (8). By 2020, CVD will claim 25 million deaths annually and CHD will surpass infectious diseases as the world's number one cause of death.

This global rise in CVD, is the result of a dramatic shift in the health status of individuals around the world over the course of the 20th century. Before 1990, infectious diseases and malnutrition were the most common causes of death. These have gradually supplanted in (mostly developed) countries by chronic diseases such as CVD and cancer. Thanks largely to nutrition and public health measures. As this trend spreads to and continues in developing countries, CVD will dominate as the major cause of death by 2020, accounting for at least one in every three deaths (9).

The basic pathogenic mechanism underlying macro vascular disease is atherosclerosis of the vascular tree and endothelial dysfunction. The concept of endothelial dysfunction has been around for a number of years. Many researchers promote the notion that endothelial dysfunction is likely to be a critical early step in the process of atherogenesis.

Endothelial dysfunction in diabetes

There is now an ample evidence from studies that endothelial dysfunction occurs in type 2 diabetes including elevation of FFAs, characteristic lipid changes, obesity, hyper tension and low grade inflammation (Tooke and Goh, 1999, Hink *et al* 2001)



Understanding the pathogenesis of atherosclerosis firsts requires knowledge of the structure and biology of the normal artery and its indigenous cell types.

Normal arteries have a well developed trilaminar structure. The innermost layer is the tunica intima which is thin at birth in humans and many non human species.

A monolayer of endothelial cells lines the intimal surface of the entire circulatory tree, thereby representing the only stationary cell type that components of blood ever

come in contact with under normal circumstances. The endothelial surface of the adult human is enormous. It is composed of about (1 to 6 x 10^13) cells, weighs about 1 kg and covers a surface area equivalent to six tennis courts (10).

Today, we recognize that endothelium is a dynamic organ with complex metabolic capabilities, including the ability to control vascular permeability, the flow of biologically active molecules and nutrients, cell – cell and cell – matrix interactions within the vessel wall, blood flow and vascular tone, interactions of blood cells, the inflammatory response and angiogenesis.

A delicate balance exists in the capability of the endothelial cells to modulate vascular tone. An important physiological vasodilator released by endothelial cells is nitrous oxide, thus significance of nitric oxide in antiatherogenesis (26). It is a simple diatomic gas synthesized from the terminal guanidine nitrogen atoms of L arginine by the action of a group of enzymes known as nitric oxide synthases (NOS) [Vans etal, 1990; Monacada & Higgs, 1993] (11,12). The major iso form of NOS present in endothelial cells, eNOS, is constitutively active and further activated by stimuli that increase intracellular calcium including several receptor dependant agonists (e.g. Thrombin) and hemodynamic forces (shear stress and cyclic stretch). NO acts as a potent vasodilator and adhesion by stimulating soluble guanylate cyclase and thereby elevates intra cellular levels of cGMP in vascular smooth muscle cells and platelets. Prostaglandin 12 (PG12, Prostacyclin) is a major endothelium derived oxygenation product of arachidonic acid, synthesized by the sequential actions of cycloxygenase

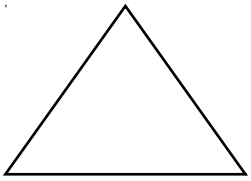
(COX) and Prostacylin synthase (13). Prostacyclin like NO is both a vasodilator and inhibitor of platelet aggregation (not adhesion); exerting these actions by stimulating adenylate cyclase and thus the intracellular CAMP in target vascular smooth muscle cells and platelets.

Endothelium derived hyperpolarizing factor (EDHF) and carbon monoxide (CO) a by-product of heme metabolism to biliverdin by heme oxygenases are the direct vasodilators elaborated by endothelial cells. Endothelial ecto-adenosine diphosphatase (ADPase), recently identified as CD39, is a membrane associated platelet inhibitor but may also indirectly promote vasodilation by generating adenosine. The vasodilatory properties of endothelium are counter balanced by endothelium – derived vasoconstrictors, including platelet activating factor, endothelin-1 and thromboxane A_2 (TXA_2)

Vaso regulation by endothelium

Nitric Oxide	PAF
PGI_2 (Prostacyclin)	Endothelin-1
Others – EDHF	Others – TxA_2
Carbon monoxide, ADPase	

Vasodilatation **Vasoconstriction**



Endothelium is also an ideal regulator of hemostasis (14). It is endowed with a remarkable repertoire of activities that permit it to rapidly transform from a potent antithrombotic to a prothrombotic surface whenever the need arises. Normal quiescent

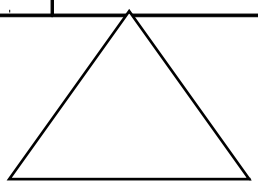
endothelium constitutively displays a potent antithrombotic (thromboresistant) surface to blood. It expresses anticoagulant, profibrinolytic and platelet inhibitory properties whenever endothelium is activated or perturbed, however, it is rapidly transformed to a prothrombotic surface that actually promotes coagulation, inhibits fibrinolysis and activates platelets. Throughout the circulatory tree, even within a single organ, there is marked heterogeneity in the phenotype of endothelial cells (15,16) with respect to hemostasis, for e.g. Endothelial cells from different tissues are heterogenous in their expressions of vWF, plasminogen activators and tissue factor. This endothelial heterogeneity is maintained by both genetic and environmental factors.

The specific antithrombotic and prothrombotic properties of endothelial Cells are as follows:-

Regulation of Hemostasis by Endothelium

Anticoagulant :	**Procoagulant :**
• GAGs/AT III • TFPI • Thrombomodulin	• Tissue factor • Binding sites for Coagulation factors and fibrin
Profibrinolytic:	**Antifibrionolytic:**
• t-PA • u-PA • Binding sites for Plasminogen • PA receptors	• PAI • TAFI

Platelet inhibitory : • PGI_2 (Prostacyclin) • Nitric Oxide • AD Pase • Carbon monoxide	**Platelet activating:** • vWF • PAF



GAgGs - lycosaminoglycans

AT III - Antithrombin III,

TFPI - Tissue factor pathway inhibitor,

t-PA - Tissue plasminogen activator,

u-PA - Urokinase type plasminogen activator,

PAI - Plasminogen activator inhibitor,

VWF - Von Willibrand factor

PAF - Platelet activating factor

The haemostatic conversion of the vessel wall is triggered by mechanical damage or by perturbation and activation of the vascular cells by agents such as cytokines, endotoxin, hypoxia and hemodynamic forces.

In many cases, endothelium derived vasodilators are also platelet inhibitors and conversely, endothelium derived vasoconstrictors can also be platelet activators. The net effect of vasodilation and inhibition of platelet function is to promote blood fluidity, whereas the net effect of vasoconstriction & platelet activation is promote Hemostasis. Thus blood fluidity and Hemostasis can be exquisitely regulated by balance of antithrombotic / prothrombotic & vasodilatory / vasoconstrictor properties of endothelial cells, which are often co-coordinately modulated by their relatives states of quiescence & activation (14),

‘’Endothelial dysfunction’’ represents a pathophysiologic state in which the collective homeostatic properties of normal endothelial cells are impaired or lost, promoting an atherogenic milieu. Abnormalities in endothelial function are believed to play a central role in the pathophysiology of common cardiovascular syndromes such as myocardial infarction, unstable angina & stroke. It is associated with a growing number of risk factors including smoking, diabetes mellitus, hypertension, hyperlipidemia, hyperhomocystinemia, menopause & advanced age. Endothelial dysfunction is thought to presage atherosclerosis and is characterized by altered permeability barrier function, enhanced adhesion molecule expression, increased leucocyte adhesion and impaired

endothelium dependent vasodilator responses {(Kupatt et al 1996) (Scalia et al 1996), (Penn et al 1997), (Rangasamy et al 1997)

Endothelial dysfunction is detectable very early in the progression of atherosclerosis, long before the appearance of visible of visible stenotic lesions. This association suggests a biological link between endothelial dysfunction and vascular disease, thus making it an ideal target for primary preventive intervention.

The etiological factors associated with endothelial dysfunction are classical and novel risk factors.

Classical risk factors

1. Diabetes
2. Hypertension
3. Smoking
4. Dyslipidemia

Novel risk factors are

5. Low grade chronic inflammation:
6. Obesity
7. Leptin
8. Homocysteine

9. Low birth weight
10. Ethnicity

Plasma Markers of Endothelial cell Function (25)

Markers for which clinical data are available

- Fibrinolytic substances
  - ✓ Plasminogen activators
  - ✓ Plasminogen activator inhibitor-1
- Procoagulant substances
  - ✓ vWF
  - ✓ Fibronectin
- Antiplatelet substances
  - ✓ Nitric Oxide
  - ✓ Prostacylin

- Metabolic substances
  - ✓ Lipoprotein lipase
- Potential markers
  - ✓ Activated protein C
  - ✓ Antithrombin III

- ✓ Angiotensin – Converting enzyme
- ✓ Heparan sulphate
- ✓ Laminin
- ✓ Thrombospondin
- ✓ Tissue factor
- ✓ Vittonecti

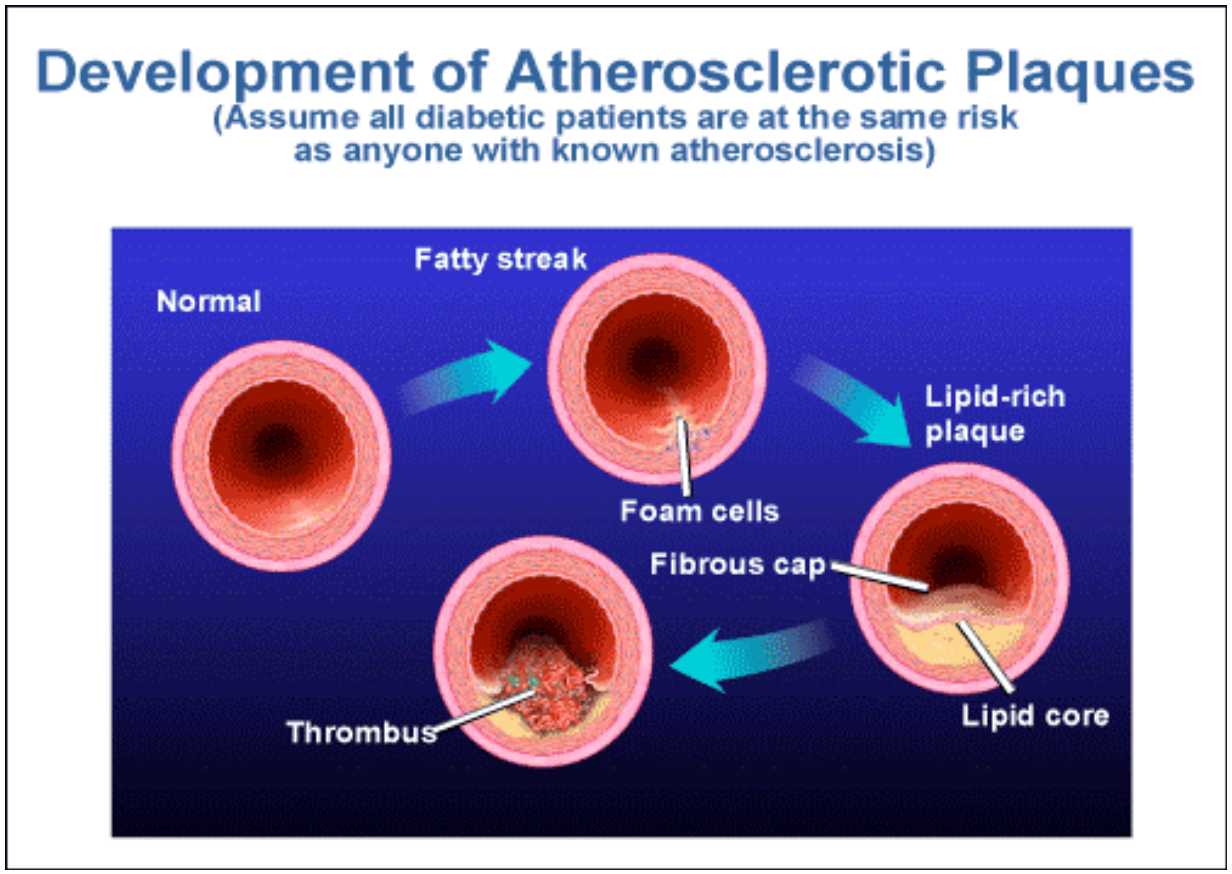
NORMAL VERSUS DYSFUNCTIONAL ENDOTHELIAL CELL (24)

(N) Endothelial Cell	Dysfunctional endothelial cell
PGI_2 Production	Decreased PGI_2 Production
EDRF release	Decreased DERF release
Ecto – ADT pase activity	Increased thromboplastin Production
Facilitation of vascular uptake & degradation of prothrombotic amines	Inhibition of endothelial cell – dependent protein C activation
Thrombin binding & inactivation	Decreased resistance to platelet adhesion
Thrombomodulin expression	Increased PAI-1 release
Glycosamino glycan expression	Impaired thrombomodulin expression
	Tissue factor expression

Clinical assessment of endothelium represents a challenging task owing to its heterogeneous functions. No single test provides a comprehensive physiological assessment of endothelial function. Several makers as described above have been used an surrogate makers of endothelial activity and positively correlate with adverse cardio vascular outcomes. Most clinical studies have focused primarily or regulation of vascular tone as a means of assessing endothelial function (2) through interrogation of

NO – mediated, endothelium – dependant vasodilator response to specific agonists, such as acetyl choline, serotonin or shear actress which normally provoke vasodilatation. Paradoxical constriction or attenuated dilator response are observed in disease states associated with atherosclerosis or risk factors for coronary artery diseases reflecting impaired vasomotor function & reduced endothelial NO bioactivity.

Because abnormal endothelial physiology is implicated in genesis and progression of vascular disease prevention or reversal of endothelial dysfunction respects attractive goal for therapeutic intervention. Indeed reversal interventions have been constantly shown to restore endothelial vasomotor function. These include among other lipid lowering therapy, ACE inhibitor therapy, L-arginine tetra hydrobiopterin, folate, anti oxidants & estrogen supplementation, smoking cessation and exercise (22, 23). Interesting by these are the very same interventions that attenuate atherosclerosis & improve morbidity & mortality outcomes from cardiovascular diseases suggesting a mechanistic link between endothelial dysfunction & atherosclerosis.



The tunica media

It lies under the intima & internal elastic lamina. The media of elastic arteries such as aorta have well developed concentric layers of smooth muscle cells inter leaved with layers of elastin rich extracellular matrix. This structure appears well adapted to the storage of kinetic energy of (Lt) ventricular systole by the walls of great arteries. The lamellar structure also contributes to the structural integrity of arterial trunks. The media of smaller muscular arteries usually have a less stereotyped organization. Smooth muscle cells in these arteries generally reside within the surrounding matrix in a more continuous than lamellary array. The smooth muscle cells in normal arteries are generally quiescent from the standpoint of growth control. That is, rates of cell division and cell death are quite low in the normal artery; a state of homeostasis of extra cellular matrix also typically prevails. Because extra cellular matrix neither accumulates nor

atrophies, rates of matrix synthesis and dissolution must balance each other under normal conditions. The external elastic lamina bounds the tunica media, forming the border with the adventitial layer.

The adventitia

The adventitia of arteries has typically received little attention, although appreciation of its role in arterial homeostasis & pathology has recently increased. The adventitia contains collagen fluids in a looser array than usually encountered in the intima Vasovasorum & nerve endings localize in this outermost layer of the arterial wall. The cellular population in the adventitia is more sparse than in other arterial layers. Cells encountered in this layer include fibroblasts in mast cells.

The initiation of atherosclerosis

The term atherosclerosis comes from the Greek work athero (porridge/gruel) and sclerosis (hardening).

Extracellular lipid accumulation

The first steps in atherogenesis in human remain largely conjectural However, integrations of tissues obtained from younger human with the results of experimental studies of atherogenesis in animal provided hints in this regard. On initiation of an atherogenic diet rich cholesterol and saturated fat, one of the first ultra structural alterations is an accumulation of small lipoprotein particle in the intima (29). These lipoprotein particles appear to decorate the proteoglycan of the arterial intima and tend to coalesce into aggregates (30). The binding of lipoprotein to proteoglycan in the intima

captures and retains these particles, accounting for their prolonged residence time. Lipoprotein particles bound to proteoglycan appear to exhibit increased susceptibility to oxidative or other chemical modifications, considered by may to be an important component of pathogenesis of early atherosclerosis. Other studies suggest that permeability of the endothelial monolayer increase at sites of lesion predilection to LDL (31). Contributors to oxidative stress in the nascent atheroma could include the NAD and NADP dependent oxidases exposed by vascular cells & lipooxygenases expressed by infiltration leucocytes. In addition of oxidation, aggregation, enzymatic processing due to sphingomyelinase & glycation can modify LDL in the intima.

Leukocyte Recruitment

The second morphologically definable event in the initiation of atheroma is leucocyte recruitment & accumulation. The normal endothelial cell generally resists adhesive interactions with leucocytes. Even in inflamed tissues, most recruitment and trafficking of leucocytes occurs in post capillary venules, not in arteries. However, very early after initiation of hypercholesterolemia, leucocytes adhere to the endothelium & diapedese between endothelial cell functions to enter the intima, where they begun to accumulate lipids & transform into foam cells (32). In addition to monocyte, T-lymphocytes also tend to accumulate in early human & animal atherosclerotic lesions (33).

The expression of certain leucocyte adhesions molecules on the surface of the endothelial cells regulates the adherence of monocytes & T cells to the endothelium.

Two broad categories of leucocyte adhesion molecules exist. Members of the immunoglobulin superfamily include structures such as vascular adhesion molecules 1 (VCAM-1) (34). This adhesion molecules has particular interest in the context of early atherosclerosis because it interacts with an integrin (very late antigen – 4 [VLA-4] characteristically expressed by only those classes of leucocytes that accumulate in nascent atheroma, monocytes and T cells moreover, studies in rabbits & mice have shown expression of VCAM-1 on endothelial cells overlying very early atheromatous lesions. Another member of the immunoglobulin super family of leucocyte adhesion molecules in intercellular adhesion molecule -1. This molecule is more promiscuous, both in the types of leucocytes it binds and because of its wide & constitutive expression at low levels by endothelial cells in many parts of the circulation. Selectin, constitute the other broad category of leucocyte adhesion molecules. The prototypical selectin, E-Selectin (E-for ‘’Endothelial’’, the cell type that selectively expresses this particular family member). Probably has little to do with early atherogenesis. E-selection preferentially recruits polymorphonuclear leucocytes, a cell type seldom found in early atheroma (but an essential protagonist in acute inflammation & host defenses against bacterial pathogens). Moreover, endothelial cells overlying atheroma do not express high levels of this adhesion molecule. Other members of this family, including P-selection, may play a greater role in leucocyte recruitment in atheroma, because endothelial cells overlying human atheroma do express this adhesion molecule. Selections tend to promote saltatory or rolling locomotion of leucocytes over the

endothelium (35). Adhesion molecules belonging to the immunoglobulin super family tend to promote trigger adhesive interaction & immobility action of leucocytes.

Once adherent to the endothelium, leucocytes must receive signal to penetrate the endothelium & enter the arterial wall. The migration of leucocytes involves action of protein molecules known as chemo attractants, cytokines or chemokines. Two groups of chemokines have particular interest in recruiting mononuclear cells characteristic of the early atheroma. One such molecule known as monocyte chemo attractant protein-1 (MCP-1) is produced by endothelium in response to oxidized lipoprotein and other stimuli. Cells intrinsic to the normal artery, including endothelium & smooth muscle, can produce this chemokine when stimulated by inflammatory mediators. MCP-1, selectively promotes the directed migration or chemotaxis of monocytes. Human atherosclerotic lesions expressed increased level MCP-1 as compared to other involved vessels. Thus MCP-1 appears causally related to monocyte recruitment during atherogenesis in vivo. Another group of chemo attractant, cytokines may attract lymphocyte accumulation in plaques. Atheromas express a trio of lymphocyte selective chemokines (interferon-inducible protein 10, interferon inducible T-cell & chemo attractant, I-TAC; monokine induced by interferon-gamma). Interferon gamma a cytokine know to be present in the atheromatous plaques induces the gene encoding this family of T-cell chemo attractants.

Atheroma typically forms focally as revealed by studies of morphology, lipid accumulation & adhesion molecule expression. Some have invoked a multicentric origin

hypothesis of atherogenesis, pointing that atheromas arises a benign leiomyomas of the arterial wall. The monotypia of various molecular markers such a G6PD isoforms in individual atheromas supports this ‘’monoclonal theory of atherogenesis’’, However, the location of sites of lesion predilection at proximal portions of arteries after branch points or bifurcation at flow divides suggests a hydrodynamic basis for early lesion development. Arteries without many branches like the internal mammary or radial arteries tend not to develop atherosclerosis.

Two concepts can help understand how local flow disturbances might render certain foci sites of lesion predilection. Locally distributed flow could induce alternations that promote the steps of early atherogenesis. Alternately, the laminar flow that usually prevails at sites that do not tend to develop early lesions may elicit antiatherogenic homeostatic mechanisms (atheroprotective function) (36). The endothelial cell experiences the laminar shear stress of normal flow and disturbed flow (usually yielding decreased shear stress) at predilection sites. This laminar shear stress can augment the expression of genes that may protect against atherosclerosis, including forms of enzymes superoxide dismutase & nitric oxide synthase.

Intercellular lipid accumulation: Foam cell formation

The monocyte, once recruited to the arterial intima, can there imbibe lipid and become a foam cell or lipid – laden macrophage. Whereas, most cells can express the classical cell surface receptor of LDL, that receptor does not mediate foam cell formation. This is evident clinically, because patients lacking functional LDL receptors

(familial hypercholesterolemia homozygotes) still develop tedious xanthomas filled with foamy macrophages. The LDL receptor does not mediate foam cell formation because of its exquisite regulation by cholesterol. As soon as a cell collects enough cholesterol from LDL capture for its metabolic needs, an elegant transcriptional control mechanism quenches expression of the receptor.

Instead of classical LDL Receptor, various molecules known as ‘’scavenger’’ receptors appears to mediate the excessive lipid uptake characteristic of foam cell formation (37). The long studies of these receptors belong to scavenger receptor – A family. These surface molecules bind modified rather than native lipoproteins & apparently participate in their internalization. Other receptors that bind modified lipoprotein and that may participate in form cell formation include CD36 & macrosialin, the latter exhibiting preferential binding specificity for oxidized forms of LDL.

Once macrophages have taken up residence in the intima and become foam cells, they not infrequently replicate. These factors that trigger macrophage cell division in the atherosclerotic plaque likely include macrophage colony stimulating factor (M – CSF). This comitogen and survival factor for mononuclear phagocytes exists in human and experimental atheromatous lesion. Other candidates for macrophage mitogens / comitogens include interleukin-3 and granulocyte macrophage colony stimulating factor.

Thus, the scenario of the evolving atheroma has involved only leucocytes, principally the macrophages. The precursor lesion, known as fatty streak, consists mainly of accumulations of such lipid – engorged leucocytes. Fatty streaks may occur in

children and in societies less affected by atherosclerosis pandemic that developed western nations. Moreover, in experimental animals withdrawal of the atherogenic diet and treatment with drugs that lower lipoprotein levels in plasma can reduce the extent of established lesions. Thus, fatty streaks, composed of primarily macrophages are likely reversible to some extent.

Further evolution of atheroma takes place as follows-

Smooth muscle cells migration & proliferation:

Whereas the early events in atheroma initiation involves primarily altered endothelial function and recruitment and accumulation of leucocytes, the subsequent evolution of atheroma into a more complex plaque involves smooth muscle cells as well (38). Smooth muscle cells in the normal arterial tunica media differ considerably form those in the intima of an evolving atheroma. Although some smooth muscle cells arrive in the arterial intima, early in life, others that have arisen in the developing atheroma likely arise from cells that have migrated form the media to the intima. The chemo attractants for smooth muscle cells likely include molecules such as platelet derived growth factor a potent smooth muscle chemo attractant secreted by the activated macrophages & over expressed in human atherosclerosis. These smooth muscle cells in atherosclerotic intima can also multiply be cell division. Estimated rates of division of smooth muscle cells in the atheromatous lesion are less than 1%. However, considerable smooth muscle cells accumulation can occur over decades of lesion evolution.

Smooth muscle cells in the atherosclerotic intima display a less mature phenotype

than the quiescent smooth muscle cells in the normal arterial media layer. Instead of expressing isoforms of smooth muscle myosin characteristic of adult smooth muscle cells, those in the intima have higher levels of embryonic isoform of smooth muscle myosin. Thus smooth muscle cells in the intima seem to recapitulate an embryonic phenotype. These intimal smooth muscle cells in the atheroma appear morphologically distinct as well. They contain more rough endoplasmic reticulum and fewer contractile fibers than do normal medial smooth muscle cells. Although replication of the smooth muscle cells in the steady state appears indolent in mature human atheromas, bursts of smooth muscle replication can occur during the life history of a given atheromatous lesion. Accumulation of smooth muscle cells during atherosclerosis and growth of intima may not occur in a continuous & linear fashion. Rather, ‘’Crisis’’ may punctuate the history of an atheroma during which bursts of smooth muscle replication and migration may occurs.

Smooth muscle cell death during angiogenesis

In addition to smooth muscle cell replication death of these cells may also participate in complication of the atherosclerotic plaque. At least some smooth muscle cells in advanced atheroma exhibit fragmentation of their nuclear DNA characteristic of programmed cell death or apoptosis – Apoptosis may occur in response to the inflammatory cytokines known to be present in the evolving atheroma. In addition to soluble cytokines that may trigger programmed cell death; the T cells in atheroma may participate in eliminating some smooth muscle cells, In particular, some T cell

populations known to accumulate in plaques can express fas ligand in their surface. Fas ligand can engage fas on the surface of the smooth muscle cells and in conjunction with pro inflammatory cytokines, lead to death of smooth muscle cells (39).

Thus, smooth muscle cells accumulation in the growing atherosclerotic plaque probably results from a tug-of-war between cell replication & cell death, Contemporary cell & molecular biological research has identified candidates for mediating both the replication & attraction of smooth muscle cells, a concept that originated from the careful, morphological observations of Virchow almost a century and half ago. Referring to the smooth muscle cells in the intima, Virchow noted that early atherosclerosis involves a ‘’multiplication of their nuclei’’ However, he recognized that cells in lesions can ‘’ hurry on to their own destruction’’ because of death of smooth muscle cells.

The arterial extracellular Matrix

Extracellular matrix rather than cells themselves makes up much of the volume of an advanced atherosclerotic plaque. Thus extracellular constituents of plaque also require consideration. The major extracellular matrix macromolecules that accumulate in atheroma include intersitital collagens (types I & III) and proteoglycans such as versican, biglycan, aggrecan and decorin. Elastin fibers may also accumulate in atherosclerotic plaques. The vascular smooth muscle cell produces these matrix molecules in disease, just as it does during development & maintenance of the normal artery. Stimuli for excessive collagen production by smooth muscle cells include PDGF and Transforming growth factor – beta (TGF – B) both constituents of platelet granules

& Products of many cell types found in lesion.

Much like the accumulation of smooth muscle cells, extracellular matrix secretion also depends on a balance. In this case, the biosynthesis of the extra cellular matrix molecules counters breakdown catalyzed in part by catabolic enzymes such as matrix metalloproteinases (CMMPs), Dissolution of extracellular matrix macromolecules undoubtedly plays a role in migration of smooth muscle cells as they penetrate in to the intima from the media through a dense extra cellular matrix, traversing the elastin-rich internal elastic lamina. In injured arteries, over expression of inhibitors of such proteins [known as tissue inhibitors of metalloproteinases (TIMP's)] can delay smooth muscle accumulation in the intima (40).

Extracellular matrix dissolution also, likely plays a role in arterial remodeling that accompanies lesion growth. During the first part of life, history of an atheromatous lesion, growth of the plaque in outward in an ab luminal direction rather than inward in a way that would lead to luminal stenosis. This outward growth of the intima leads to an increase in caliber of the entire artery. This so called positive remodeling or compensatory enlargement must involve turnover of extra cellular matrix molecules accommodate the circumferential growth of the arterial lumina. Stenosis tends to occur only after the plaque burden exceeds some 4% of the cross section of the artery.

Angiogenesis in plaque

The smooth muscle cells are not alone in its proliferation & migration within the involving atherosclerotic plaque. Endothelial migration & replication also occur as

plaques develop in microcirculation, characterized by plexus of newly formed vessels. Such plaque new vessels usually require special stains for visualization. However, histological examination with appropriate markers for endothelial cells reveals a rich neovascularisation in evolving plaques. These micro vessels likely form a response to angiogenic peptides over expressed in atheroma. This angiogenesis factors include acidic & basic fibroblast growth factors (human BGF's) vascular endothelial growth factor (VEGF) and oncostain M (41),

These micro vessels within plaques probably have considerable functional significance. For example abundant micro vessels in plaques provide a relatively large surface for the trafficking of leucocytes. Indeed, in the advanced human atherosclerotic plaque micro vascular endothelium displaces the mononuclear selective adhesion molecules such as VCAM-1 much more prominently than does the micro vascular endothelium overlying the plaque. The microvascularity of plaques may also allow growth of the plaque over coming diffusions, limitations of oxygen & nutrient supply in analogy with the concept of tumor angiogenic factors & growth of malignant lesions. Consistent with the view administration of inhibitors of angiogenesis to mice with experimentally induced atheromas limits lesion expansions. Finally, the plaque micro vessels may be friable and prone to rupture like the new vessels in the diabetic retina. Haemorrahagic thrombosis in site could promote a local round of smooth muscle cell proliferation and matrix accumulation in the area immediately adjacent to the micro vascular disruption. This scenario illustrates a special case of one of the crises described

earlier in the evolution of the atheromatous plaque. Attempt to augment myocardial perfusion by enhancing new vessel growth by transfer of angiogenic proteins or their genes might have adverse effects on lesion growth or clinical complications of atheroma by these mechanisms.

Plaque mineralization

Plaque often develops areas of calcification as they evolve. Indeed both Virchow & Rokitansky recognized morphological feature of bone formation in atherosclerotic plaques in early microscopic descriptions of atherosclerosis (42). In recent years, understanding of the mechanisms of mineralization during evolution of atherosclerotic plaques has advanced. Some subpopulations of smooth muscle cells may faster calcify by enhanced secretion of cytokines such as bone morphogenetic proteins, homologues of TGF-beta. Atheromatous plaques may also contain proteins with gamma carboxylated glutamic acid residues specialize in sequestering calcium and thus promoting mineralization.

Composition of plaques

At autopsy, the atherosclerotic plaques of patients who died of MI is composed primarily of fibrous tissues of varying density and cellularity with superimposed thrombus. Calcium, lipid laden foam cells and extra cellular lipid each constitute 5-10% of the remaining area. The atherosclerotic plaques that are associated with thrombosis and total occlusion, located in infarct related vessels, are generally more complex and irregular that those in vessels not associated with MI. Histological studies of these

lesions often reveal plaque rupture or erosion. Angiographic morphology suggestive of plaque rupture has been identified in the majority of stenosis associated with AMI or abrupt onset of unstable angina. This finding is rare in the non-infarct related vessels of patients with chronic stable angina.

Platelet rich thrombi are often associated with the surfaces of most advanced atherosclerotic lesions, called complicated plaques, which are characterized by fibro calcific degeneration, deposition of lipid, calcium, fibrous tissue, necrotic debris, extravasated blood and fibrous cap. Lumina narrowing may potentiate platelet activation through augmentation of shear forces. In pts with MI, coronary thrombi are usually superimposed on or adjacent to atherosclerotic plaques. These coronary arterial thrombi, which are approximately 1 cm in length in most cases, adhere to the luminal surface of an artery and are compressed of platelets, fibrin erythrocytes and leucocytes. The composition of the thrombus may vary at different levels; a white thrombus is composed of platelets, fibrin, platelets and leucocytes. Early thrombi are usually small and nonocclusive and are composed predominantly of platelets.

Plaque fissuring and rupture

The process of plaque fissuring is an area of intensive investigation and is likely to be multi factorial in nature. In atherosclerotic plaques prone to rupture there is an increased rate of formation of metalloproteinase enzymes such as collagenase, gelatinase and stromelysin that degrades components of the protective interstitial matrix. These proteinases may be elaborated by activated macrophages and mast cells that have been

shown to accumulate in high concentrations at the site of atheromatous erosions and plaque rupture in patients who died of AMI. Examination of specimens from atherectomy reveals a much higher content of macrophages and tissue factor in patients with unstable angina or AMI compared with patients with chronic stable angina.

In addition to these structural aspects of vulnerable plaques, stress induced by intra luminal pressure, coronary vasomotor tone, tachycardia (cyclic stretching and compression) and disruption of nutrient vessels combine to produce plaques rupture at margin of the fibrous cap near an adjacent plaque free segment of the coronary artery wall (shoulder region of plaque). A number of key physiological variables such as systolic blood pressure, heart rate, blood viscosity, endogenous tissue, plasminogen activator activity, (T-PA), plasminogen activator inhibitor – 1 (PAI-1) levels, plasma cortisol levels and plasma epinephrine levels that exhibit circadian and seasonal variations and are increased at times of stress act in concert to produce a heightened propensity for plaque rupture and coronary thrombosis, yielding the clustering of AMI in early morning hours and especially in winter and after natural disasters.

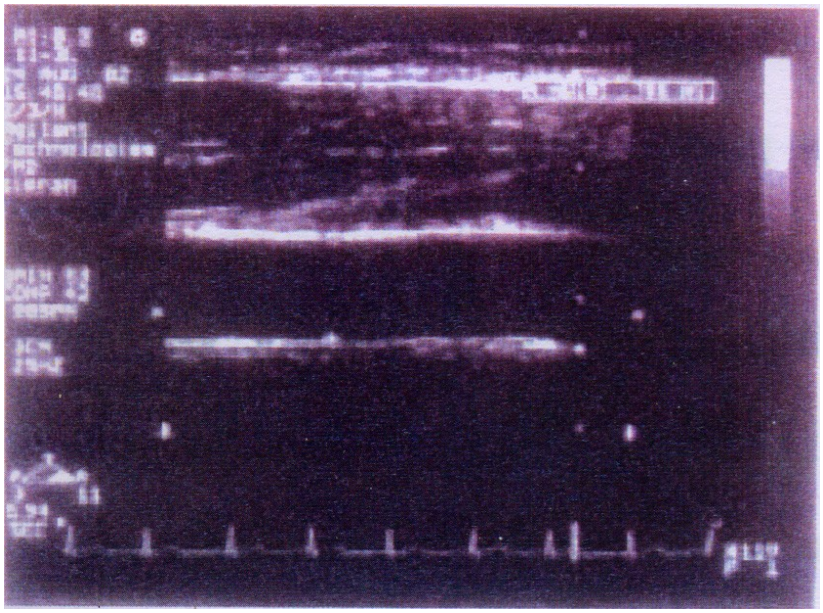
Intimal – Medial Thickness (IMT)

An early morphological change in atherosclerosis is the increase of the IMT, which can be detected non – invasively with high resolution ultrasound B mode imaging. In early stages of the atherosclerotic process, the vessel wall thickness, although the lumen maintains its internal diameter resulting in outward expansion of the vessel, a process termed positive remodeling (43). The lumen gets compromised late in

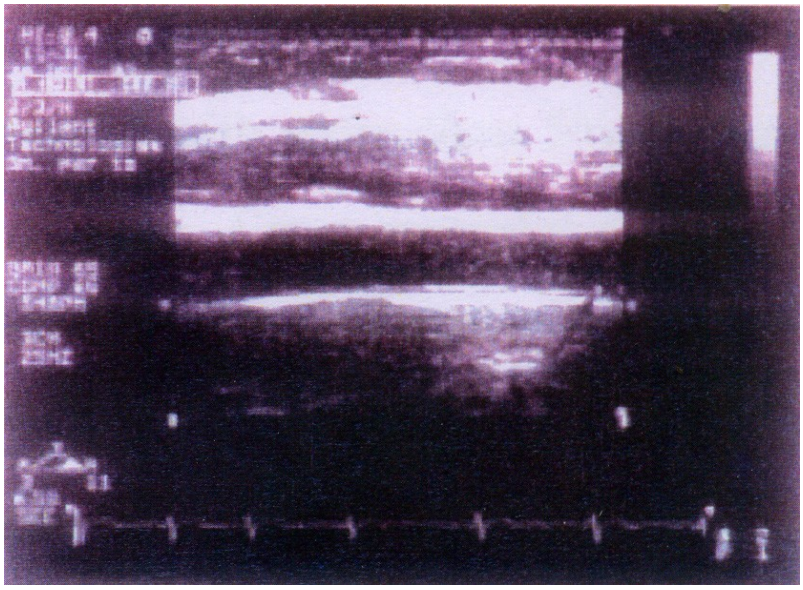
the atherosclerotic process, with the development of diffuse or focal stenosis.

Pignoli and colleagues (44) studied in vitro specimens of human aortic and common carotid arteries to determine the anatomic structures involved in ultrasound energy reflection in the arterial wall and the feasibility measurement of arterial wall thickness with B-mode real time imaging. In addition to the invitro studies, these investigators also evaluated the common carotid arteries of 10 young volunteers in vivo using the same methodology. Imaging was performed with high resolution real-time scanners equipped with 7-8 MHz probes. The vessels were grouped into macroscopically normal or with fatty streaks (class A) or vessels with atherosclerotic lesions (class B). The ultrasound pattern of class A group was characterized by two parallel echogenic lines separated by a hypo echoic or anechoic space. This seam pattern is defined as the “double line pattern”. The inner (Luminal) line was generally more regular, smooth and thin then the outer one. Correlating these findings with gross specimens, it was postulated that the inner line represents the intima, the hypo echoic line the media and the outer echogenic line the adventitia. Therefore, measuring the distance from the inner echogenic line to the interface between the hypo echoic line and the second echogenic line represents the IMT of the vascular wall. The B-mode measurements of IMT showed a significant correlation with values obtained by gross pathology and histology in both Class A and Class B specimens. Class A aortic IMT measured 1.22+0.37 mm by gross pathology; and class B aortic IMT measured 2.06+1.02 mm versus 1.93+0.84 mm by gross pathology.

Recording of Intima media thickness



Recording of common carotid artery diameter



Intima media thickness

[Intima media thickness measured by the distance between the thick and thin white lines (tip of black arrow)]

In early studies accessing the reproducibility of the technique, suboptimal

performance was reported, with low intra-observer correlation coefficients (0.72-0.77) and poor inter-observer agreement (0.48-0.65). However, in the study by O'Leary and colleagues (45), many technologists from different centers obtained images using various equipments, and the images were analyzed off line from video tape, largely accounting for the high variability observed.

More recent studies demonstrate the good reproducibility of IMT measurements (46-48). Salonen and colleagues (46) reported an inter-observer coefficient of variations of 10.5 % and an inter-observer coefficient of variation of 5.4 – 5.8%. The intra-observer variation accounted for only 4% of the total variability, whereas the remaining 96% was attributable to inter-observer variables. Espeland and Colleagues (47) also examined the reliability of longitudinal measurements of IMT from measurements obtained in the Asymptomatic Carotid Artery Progression Study (CAPS). These investigators concluded that serial IMT data were highly reliable, demonstrating the multicenter studies using B-mode measurements are feasible and valid. It was also concluded that evaluation of the common carotid artery is more reproducible than the bulbous and the internal carotid artery, likely as a result of better visualization.

Lorenz et al. (2007) (48) conducted a systematic review of the literature to provide an overview of the relevant studies, critically appraise the methods used and, where possible, to perform a meta-analysis to gain more robust estimates of the predictive value of increased IMT to predict future clinical cardiovascular end points. The review included eight observational studies with general population based samples

for which carotid IMT was measured and follow-up for clinical end points were provided. The studies represented 37,197 subjects followed for a mean of 5.5 years. The studies included either those that utilized mean carotid IMT, determined from a number of IMT measurements at specific positions or by automated software over a segment of the artery, and those that use maximal carotid IMT, with the Rotterdam study being the only one in which IMT was determined by both methods in the same population. Major sources of heterogeneity were age distribution, carotid segment definition and IMT measurement protocol. The review found that carotid IMT is a strong predictor of future vascular events. In addition, it was noted that the relative risk of increased IMT is slightly high risk of stroke than for MI. The analysis found that the age and sex-adjusted overall estimates of the relative risk of myocardial infarction were 1.26 (95% confidence interval [CI], 1.21–1.30) per 1–standard deviation common carotid artery IMT difference and 1.15 (95% CI, 1.12–1.17) per 0.10-mm common carotid artery IMT difference. Regarding the age- and sex-adjusted relative risks of stroke, the review noted they were 1.32 (95% CI, 1.27–1.38) per 1–standard deviation common carotid artery IMT difference and 1.18 (95% CI, 1.16–1.21) per 0.10 - mm common carotid artery IMT difference. The review also noted heterogeneity between the studies regarding the details of the ultrasound protocols. These details included: the precise definitions of the carotid segments investigated, the use of mean or maximal IMT, the measurement of near and far wall or IMT, and whether IMT is measured on one side or both sides. It is recommended that in future studies of IMT, ultrasound protocols should be aligned with

published studies. It appears that data for younger individuals is limited, and additional studies are required.

Kathiresan et al. (2007) (49) reported on a study of a stratified random sample of 292 participants (mean age 59.5 years; 50% women) from the offspring cohort of the Framingham Heart Study who were free of clinically apparent cardiovascular disease. The authors note that screening for subclinical atherosclerosis has been advocated for individuals at intermediate global risk for coronary heart disease (CHD); however, the distribution of subclinical atherosclerosis test values across CHD risk strata is unknown. Abdominal and thoracic aortic plaque burden was assessed by cardiovascular magnetic resonance (CMR), coronary artery calcification (CAC) and thoracic aortic calcification (TAC) by electron beam computed tomography and common carotid intima-media thickness (CIMT) by ultrasonography. The upper 20% of each measurement was classified as a high level of atherosclerosis and then these variables were evaluated across clinically relevant Framingham CHD risk score strata (low, intermediate, and high risk). In age adjusted analysis in men and women, correlations across CMR aortic plaque, CAC, TAC, and CIMT were low (maximum $r=0.30$ for CAC:TAC in women, $p<0.005$). In men and women, it was noted that the proportion of subjects with high atherosclerosis test results for any of these measurements increased significantly across the Framingham CHD risk score strata (Kruskal-Wallis test, $p<0.0001$). Regarding the intermediate Framingham CHD risk score category, 14% of men and 25% of women had a high atherosclerosis result on ¡Ý 2 measurements. It was noted that different

participants were identified as having high atherosclerosis by each modality. For example, in a comparison of the overlap across CMR aortic plaque, CAC, and CIMT, only 4% of men and 16% of women were classified as having high atherosclerosis on all three measurements. The authors concluded that correlations among subclinical atherosclerosis test results are low, and a substantial proportion has high levels of subclinical atherosclerosis detected on >= 2 imaging tests.

Application of Carotid Intimal Media thickness:

Using B-mode ultrasound imaging, Howard and Colleagues (50) examined the incidence of carotid atherosclerosis in the general population. The median wall thickness ranged from 0.5 – 1 mm at all ages, with more than 5% of the cohort having carotid wall thickness more than 2 mm. Cross-sectional analysis suggested that age-related increases in wall thickness averaged approx 0.015 mm per year in women and 0.018 mm per year in men at the carotid bifurcation, 0.010 per year in women and 0.014 per year in men at the internal carotid artery, and 0.010 mm per year in both genders at the common carotid artery.

The association of CIMT with conventional risk factors for the atherosclerosis, including diabetes, hyperglycemia and fasting insulin, but also body mass index, waist to hip circumference ratio, and physical inactivity has been reported (50). Abdominal adiposity physical inactivity and abnormal glucose metabolism are associated positively with carotid IMT, in line with their believed contribution to atherogenesis. Similarly, the atherosclerosis risk in communities (ARIC) study showed that wall thickness is strongly

associated with atherogenesic lipids, tobacco smoking and hypertension (50), suggesting that the atherosclerotic process is reflected in the IMT measurements.

The prognostic value of IMT has been prospectively evaluated and in multiple studies, increased IMT has been shown to be associated with increased cardiovascular morbidity (incidence of stroke and myocardial infarction) (51-55). In a study involving more than 4400 subjects from the Cardiovascular Health Study with age over 65 years and no known CVD, IMT was a predictor of new stroke or heart attack, even adjusting for traditional cardiovascular risk factors (55).

MATERIALS AND METHODS

Retrospective case control study

Study Interval: April 2008 to December 2008.

Population:

A random sample of 100 type 2 diabetes patients selected, 50 without macro vascular complications and 50 with macro vascular complications.

OBJECTIVE:

1. To compare the carotid intima media thickness in control group and in diseased population.
2. To find the association of increased carotid intima media thickness and CAHD/CVA
3. To assess the usage of carotid intima media thickness as a predictor of macrovascular complications in type 2 Diabetics of South India.

RESEARCH METHODOLOGY:

RESEARCH DESIGN	*Retrospective case control study*
STUDY INTERVAL	April 2008 to December 2008
POPULATION	Adult population of age 35 to 75 with Type 2
TARGET POPULATION	Those with type 2 diabetes mellitus of South India
ACCESSIBLE POPULATION	Adult population of age 35 to 75 with Type 2 Diabetes attending Coimbatore Medical College Hospital for Medical Care
SAMPLE SIZE	100

METHOD OF STUDY:

1. Informed consent obtained from patient.
2. Fasting and post prandial Blood sugar.
3. Body weight to nearest 0.5 kg.
4. Height to nearest 0.5 cm.
5. Blood pressure in sitting posture after 5 to 10 min of rest.
6. Circumference around the waist (Umbilical level) and hips (Greater trochanter level) – Waist hip ratio.
7. CAHD/CVD Diagnosis through medical history, clinical examination and appropriate investigation including Electrocardiogram, Echo Cardiograms, and Computer Tomography scan.
8. Lipid profile.
9. Intima media study using high resolution B-Mode. Ultra sonogram/Doppler

Ultra sonogram. 50 of the subjects matched by age, sex and BMI with type 2 diabetes mellitus with no evidence of Cardio Vascular/Cerebro Vascular disease are recruited as control.

ANALYSIS:

Analysis of the data obtained by the above said study is planned to be done by calculating Odds ratio.

RESULTS

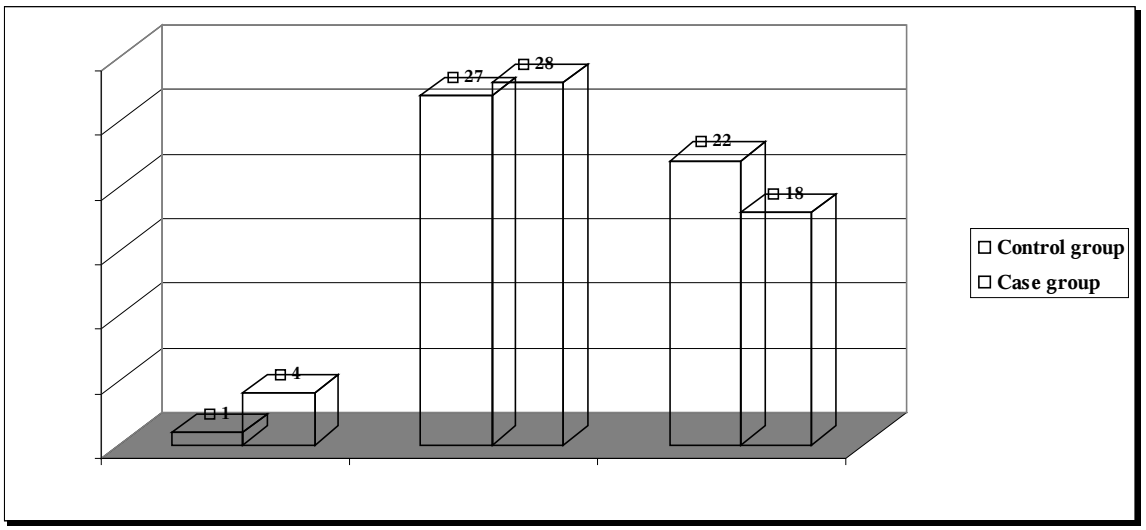
Demographic Profile

Age Distribution of Control and Study Population

Table-1 A

Age (years)	Control group	Case group
<40	1	4
41 - 59	27	28
>60	22	18

Chart – 1 A

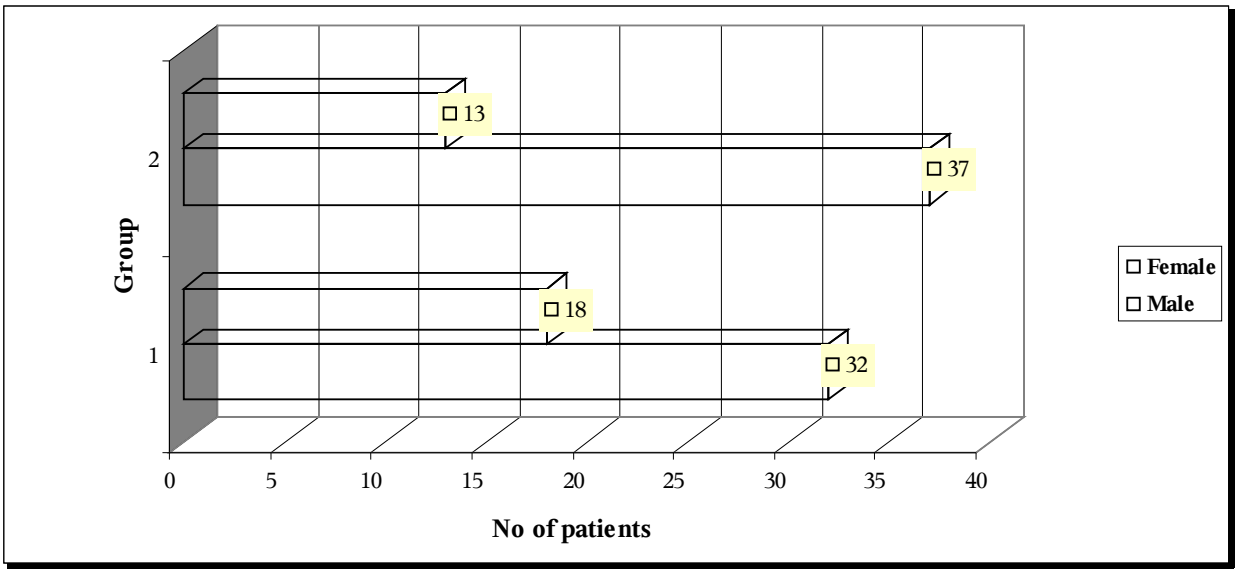


Sex Distribution of Control and Case Group

Table -1B

Sex	Control group	Case group
Male	32	37
Female	18	13

Chart – 1B

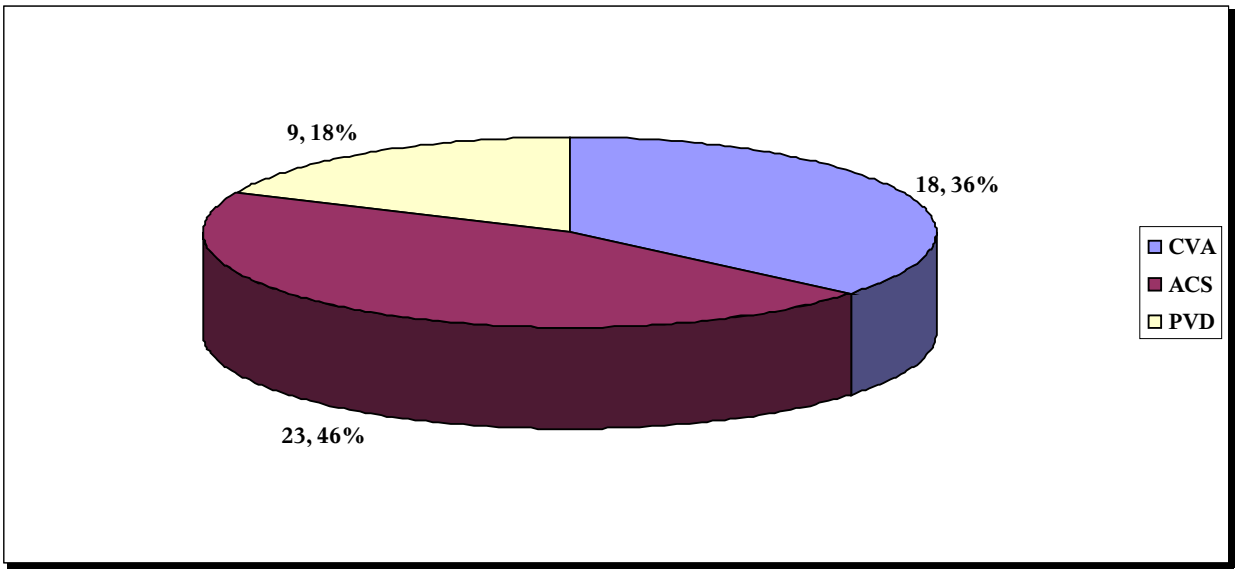


Distribution of macro vascular disease in case group

Table-2

Clinical status	No of patients
Cerebro vascular disease	**18**
Cardio vascular disease	**23**
Peripheral vascular disease	**9**

Chart – 2



Correlation of Glycemic level and macro vascular complications

Table-3

Post – prandial blood sugar (in mg/dl)	No. of patients with macrovascular complications	Percentage
150-200	1	2%
201-250	5	10%
251-300	10	20%
301-350	22	44%
351-400	12	24%

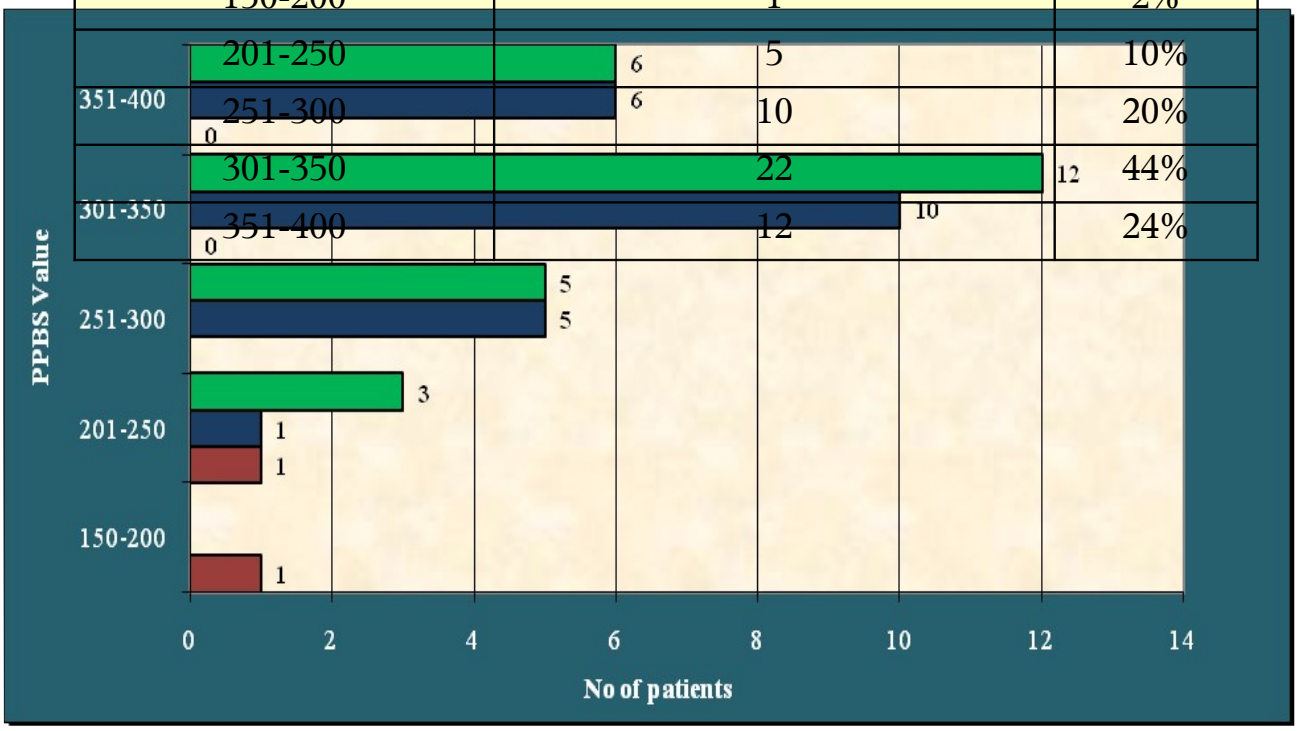


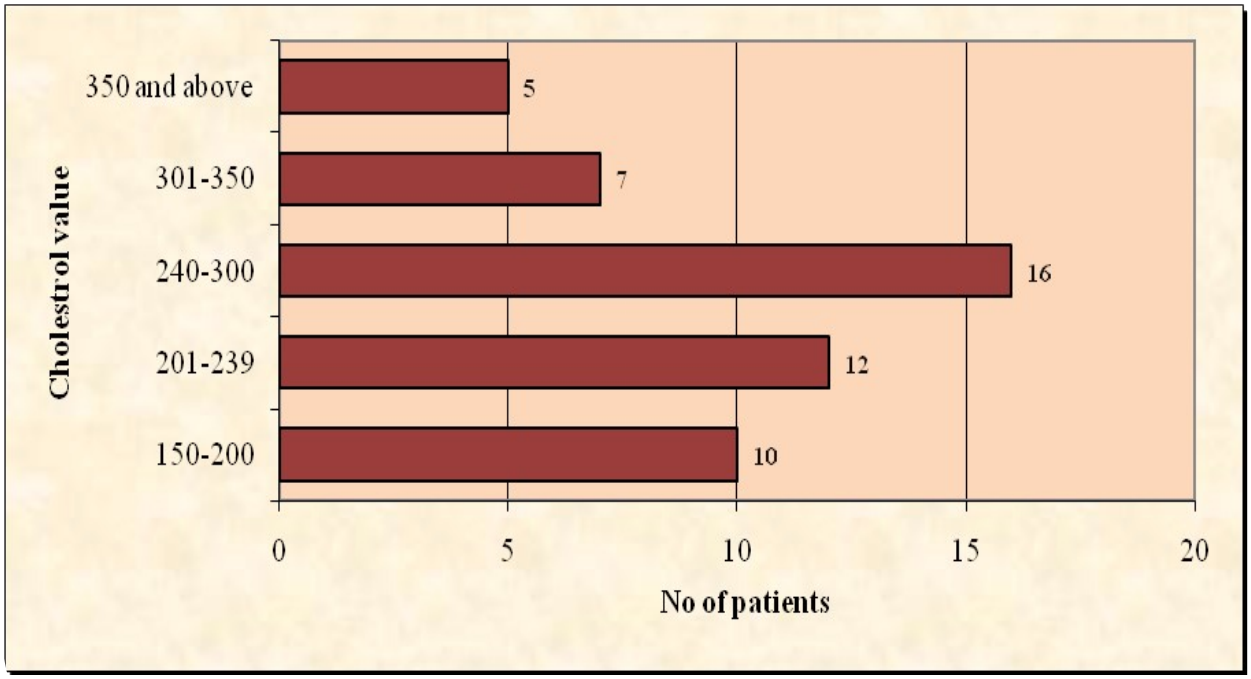
Chart – 3

Correlation between Cholesterol level and macro vascular complications

Table- 4

Cholesterol level (mg/dl)	No of cases
150-200	10
201-239	12
240-300	16
301-350	7
350 and above	5

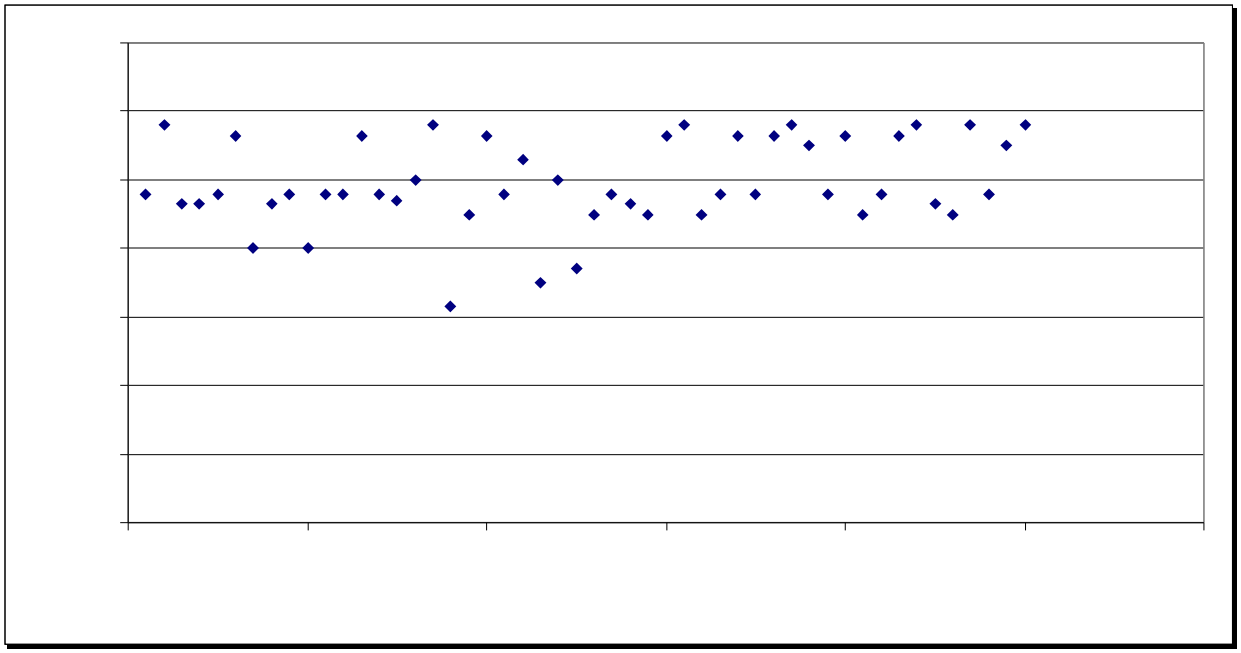
Chart – 4



Distribution of Carotid Intima Media thickness

in Case Group

Chart – 5

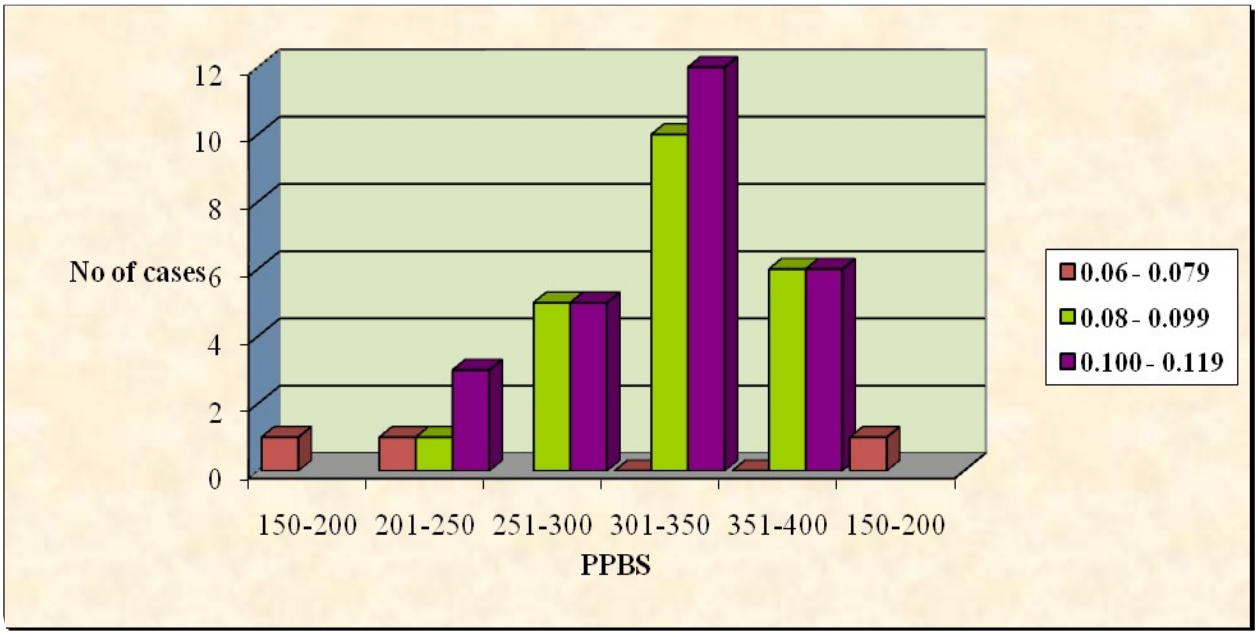


Correlation between the post prandial blood sugar levels and carotid intima media thickness

Table- 5

Post – prandial blood sugar (in mg/dl)	*Carotid intima media thickness*		
	0.06 - 0.079	*0.08 - 0.099*	*0.100 - 0.119*
150-200	1		
201-250	1	1	3
251-300		5	5
301-350	0	10	12
351-400	0	6	6
150-200	1		

Chart - 6

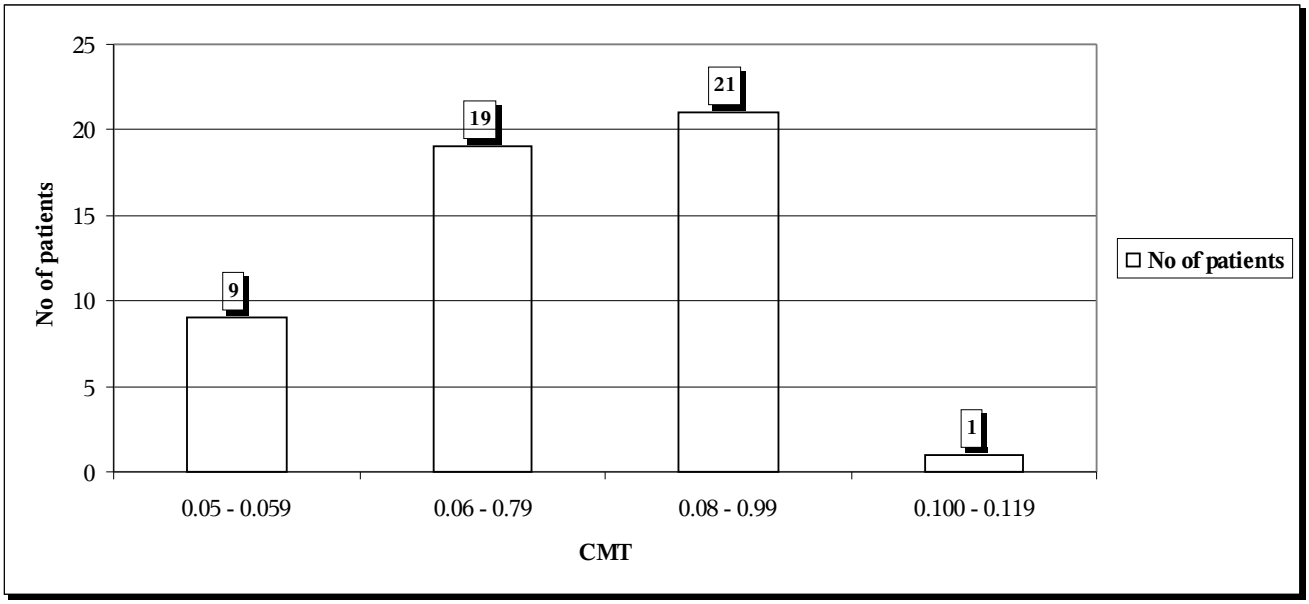


Carotid Intima Media thickness in Control Group without macro vascular complications

Table - 6

Range	No of patients
0.05 - 0.059	**9**
0.06 - 0.79	**19**
0.08 - 0.99	**21**
0.100 – 0.119	**1**

Chart – 7

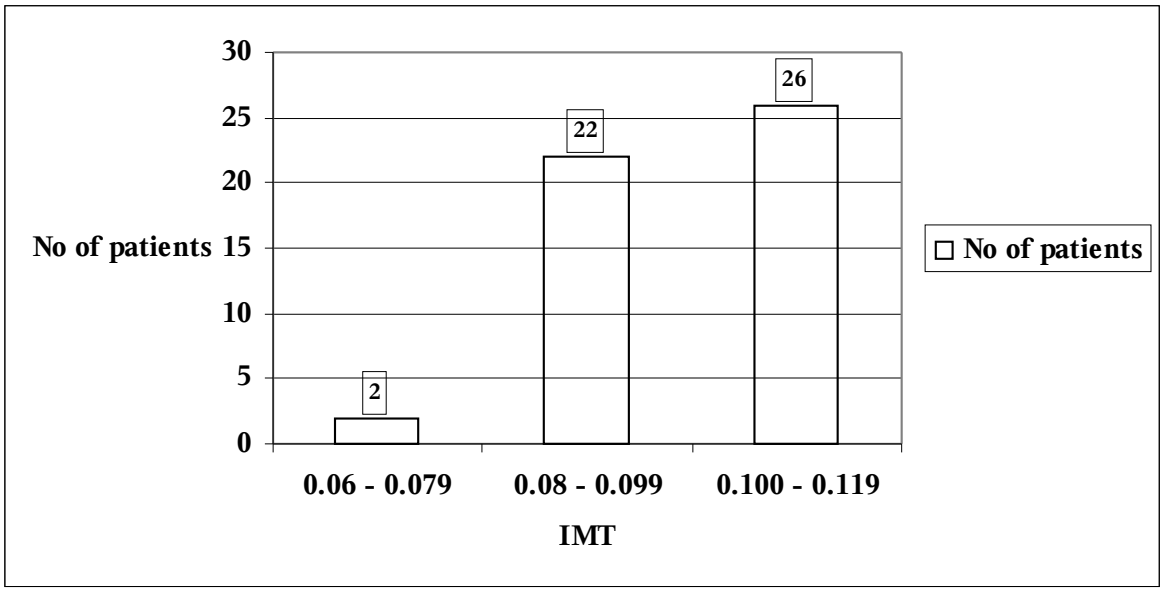


Carotid Intima Media thickness in Case Group with macro vascular complications

Table - 7

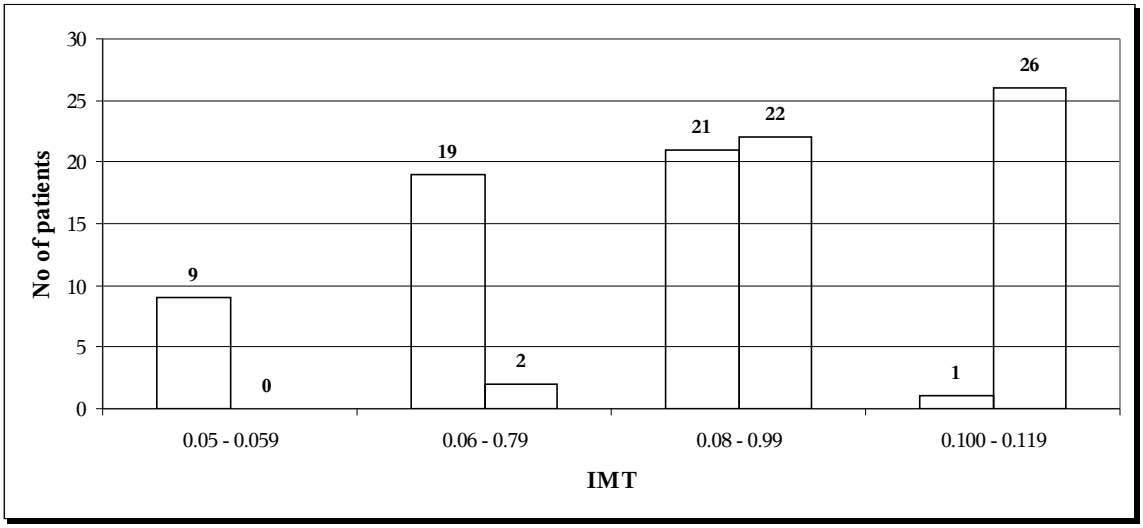
CMT in Case Group	No of cases
0.06 - 0.079	2
0.08 - 0.099	22
0.100 - 0.119	26

Chart – 8



Comparison of Intima Media thickness between Control and Case Group

Chart – 9



ANALYSIS:

Analysis of the data obtained by the above said study is done by calculating Odds ratio using SPSS Version 16.0.1.

IMT Range	Case Group	Control Group
>0.08	47	22
<0.08	3	28
Odds Ratio: **19.9**		

DISCUSSION

The identification of arterial wall changes in asymptomatic subjects indicates the need for more strict control of macrovascular risk factors, seeking to prevent future macrovascular events.

Population and hospital based studies using non-invasive techniques to evaluate early changes in the structure and function of the arterial wall, such as the measurement of intima media wall thickness, investigation of endothelial dysfunction and macrovascular calcification (63, 64, and 65) have been employed for the purpose.

Studies determined 0.8 mm as the reference values for early thickening of the intima media complex associated with an increase in macrovascular risk (54, 66-69). Grook et al conducted a study with 315 patients with familial hypercholesterolemia compared 118 controls and showed that an intima media thickening of up to 0.8 mm would be considered normal.

The present study shows that even asymptomatic patients with T2DM has increased carotid IMT value and the risk of acquiring macro vascular combination increase as IMT value increases.

This study shows that increasing CIMT is strongly associated with macrovascular disease. In the present study we used mean aggregate IMT of the extra cranial arteries (altogether 6 sites at each common carotid artery, the bifurcation and the internal carotid artery) including thickness of plaque lesion, with a view to evaluate the association between macrovascular status and grade of carotid atherosclerosis. Mean IMT aggregate

has been accessed in many previous studies, although in same of them it proved difficult to evaluate internal carotid intima media thickness precisely (50, 59). However, technological advancement in ultrasound equipment and the availability of high resolution transducer has allowed more accurate evaluation of arterial wall structure in past decade.

The present study showed carotid artery IMT was higher in patients with macrovascular complication in T2DM than in patients without macrovascular complications in T2DM. Thickening of the mean aggregate intima media complex just over 1 mm may prove to the predictive of significant macrovascular disease in nearly 95% of the patients.

The study observations are consistent with several previous studies Crouse et al (58) found a strong correlation between macrovascular disease and increased IMT in carotid artery diseases. Likewise, in my study mean aggregate IMT increases with advancing macrovascular disease.

Other investigators carried out population based studies in which IMT was evaluated with regard to the number and incidence of stroke or acute coronary syndromes (60). A strong association between IMT and risk of stroke and myocardial infection was established in Rotterdam study (60). Bots et al (53) used B mode ultrasonography to study the carotid arteries of 7983 patients aged over 55 years. Throughout 4.6 years of observation they registered 194 new myocardial infections in the study going patients who has myocardial infarction had significantly higher IMT

than others.

Furthermore, O' leary et al (55) having examined over 5800 patients (>65 years) with high resolution ultrasonography found increased IMT of carotid arteries directly associated with increased risk of myocardial infarction and stroke.

Since, both the sensitivity and specificity of Treadmill testing and Echocardiography are limited; the introduction of IMT measurements of the carotid arteries may contribute significantly to diagnose the status of patients. In these patients high aggregated IMT in carotid arteries may be adhesive factor in introduction of effective medical treatment such as statins and antihypertensive drugs along with effective blood sugar control.

IMT may be considered as another risk factor for CAD, in the same way of Diabetes, hypertension according to recommendations of European society of cardiology publications 2000.

The non-invasive assessment of common carotid intima media thickness appears to provide a promising method of study of atherosclerosis directly at the level of vessel in population at large. The use of carotid intima media thickness measurements as an indicator of generalized atherosclerosis is conditioned on the view that its measurement reflects on macrovascular disease risk.

CONCLUSION

In conclusion, the present studies based on short follow up periods:

1. Even in asymptomatic type 2 diabetic patients have increased carotid intima media thickness.
2. An increase in carotid intima media thickness strongly goes in hand with macrovascular complications.
3. Intima media thickness measurement has an intermediate or proxy end point in observational and interventional study.

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PROFORMA

NAME: AGE:

SEX: CODE NO:

ADDRESS: OCCUPATION:

CHIEF COMPLAINTS:

PAST H/O HT: DM: CAD:

FAMILY H/O HT: DM: CAD:

SMOKING: ALCOHOL: DIET:

DRUG HISTORY:

CLINICAL EXAMINATION:

PULSE: B.P:

HEIGHT: WEIGHT: BMI:

JVP: CAROTID BRUIT:

INVESTIGATIONS:

CBC: ESR: FBS:

TOTAL CHOLESTEROL: HDL: LDL:

ECG:

Master Charts

CASE GROUP - DATA

S No	Name	Age	Age Dist.	Sex	Clinical status	Past History			Personal History			Clinical examination					Investigations									IMT
						HT	DM	CAD	Smoking	Alcohol	Diet	Pulse	BP	BMI	JVP	Carotid Bruit	CBC	FBS	PPBS	PPBS Range	Cholestrol	TG	HDL	LDL	ECG	
1	Mrs P	54	2	2	CVA	No	Yes	Yes	No	No	NV	76	120/80	N	N	No	N	186	260	251-300	354	116	28	118	Ishaemia	0.096
2	Mr L	66	3	1	ACS	No	Yes	No	Yes	No	V	74	150/100	N	N	No	N	170	302	301-350	360	121	23	123	ASMI	0.116
3	Mr S	45	2	1	ACS	No	Yes	No	Yes	Yes	NV	80	148/96	OW	N	Yes	N	190	360	351-400	202	150	30	135	AWMI	0.093
4	Mr M	35	1	1	ACS	No	Yes	Yes	No	No	V	98	120/82	OW	N	No	N	200	310	301-350	246	146	35	133	AWMI	0.093
5	Mr J	60	3	1	CVA	No	No	Yes	Yes	Yes	NV	74	116/80	N	N	Yes	N	172	278	251-300	304	124	24	122	IWMI	0.096
6	Mr V	65	3	1	CVA	No	No	No	Yes	Yes	NV	72	120/80	N	N	No	N	180	298	251-300	252	136	38	128	Normal	0.113
7	Mrs K	50	2	2	PVD	Yes	No	No	No	No	V	74	130/70	N	N	No	H	170	364	351-400	158	82	16	88	Ishaemia	0.08
8	Mrs L	54	2	2	ACS	Yes	Yes	No	No	No	NV	74	160/100	OW	N	No	N	186	400	351-400	206	148	36	134	AWMI	0.093
9	Mr A	47	2	1	ACS	No	No	No	No	No	V	74	120/84	OW	N	No	N	164	307	301-350	352	118	24	119	AWMI	0.096
10	Mr R	65	3	1	CVA	No	No	Yes	Yes	Yes	NV	80	158/94	N	N	No	N	190	168	150-200	168	166	26	98	Ishaemia	0.078
11	Mr G	37	1	1	ACS	Yes	No	No	No	Yes	NV	74	170/100	OW	N	No	H	208	304	301-350	306	126	25	123	HLWMI	0.096
12	Mr N	52	2	1	PVD	Yes	No	Yes	No	Yes	NV	78	160/94	N	N	No	N	275	314	301-350	217	115	75	147	Normal	0.096
13	Mr A	55	2	1	CVA	Yes	Yes	No	No	No	V	82	160/100	N	N	No	N	180	274	251-300	252	136	38	128	Ishaemia	0.113
14	Mrs D	56	2	2	ACS	No	No	No	No	No	V	86	170/100	OW	N	No	N	140	248	201-250	204	184	46	153	AWMI	0.078
15	Mr S	56	2	1	ACS	No	Yes	Yes	Yes	Yes	NV	84	120/80	OW	N	No	N	198	209	201-250	246	160	38	141	AWMI	0.094
16	Mrs R	61	3	2	ACS	Yes	Yes	No	No	No	NV	80	160/90	OW	N	No	N	160	306	301-350	204	188	37	155	IWMI	0.1
17	Mr V	42	2	1	CVA	No	No	No	No	Yes	NV	64	170/100	OW	N	Yes	N	206	360	351-400	302	128	24	124	Ishaemia	0.116
18	Mrs J	55	2	2	CVA	Yes	Yes	Yes	No	No	V	76	160/90	OW	N	No	N	190	398	351-400	302	130	30	125	Ishaemia	0.083
19	Mr S	63	3	1	PVD	No	No	No	No	No	V	74	150/90	N	N	No	H	200	364	351-400	352	114	28	117	Normal	0.09
20	Mr N	65	3	1	PVD	No	No	No	Yes	Yes	NV	76	150/100	OW	N	No	H	210	307	301-350	246	160	38	114	Ishaemia	0.113
21	Mr L	78	3	1	CVA	No	No	Yes	Yes	No	V	78	110/70	N	N	No	N	180	368	351-400	354	116	28	118	Normal	0.096
22	Mrs M	40	1	2	CVA	No	No	No	No	No	NV	84	120/80	OW	N	No	N	188	216	201-250	240	162	37	142	Normal	0.106
23	Mr T	57	2	1	ACS	Yes	No	Yes	No	No	V	86	160/100	OW	N	No	N	162	304	301-350	304	124	24	122	AWMI	0.1
24	Mr R	38	1	1	ACS	No	Yes	No	Yes	Yes	NV	76	130/80	N	N	No	N	168	288	251-300	240	134	35	127	AWMI	0.1
25	Mr S	60	3	1	CVA	No	No	No	No	Yes	NV	78	130/80	N	N	No	N	170	306	301-350	248	142	36	131	Normal	0.112
26	Mr R	57	2	1	CVA	Yes	Yes	Yes	No	No	V	74	180/120	N	N	No	N	180	318	301-350	252	136	38	128	Hge	0.108
27	Mrs V	67	3	2	CVA	No	Yes	Yes	No	No	V	78	120/80	N	N	No	N	210	352	351-400	181	120	39	111	Ishaemia	0.106
28	Mr S	52	2	1	ACS	No	Yes	Yes	Yes	Yes	NV	74	120/80	N	N	No	N	212	320	301-350	179	122	37	109	AWMI	0.106
29	Mr B	76	3	1	ACS	Yes	Yes	No	Yes	No	NV	78	120/70	N	N	Yes	N	260	398	351-400	246	146	35	133	IWMI	0.112
30	Mr A	59	2	1	CVA	No	Yes	No	No	No	V	74	120/64	N	N	No	N	302	318	301-350	160	168	18	90	Ishaemia	0.113
31	Mrs S	51	2	2	CVA	No	Yes	No	No	No	V	82	140/80	OW	N	Yes	N	202	360	351-400	246	160	38	141	Normal	0.116
32	Mr K	47	2	1	ACS	No	No	Yes	No	No	V	76	130/74	OW	N	No	N	218	306	301-350	218	140	32	112	HLWMI	0.09

33	Mr R	62	3	1	PVD	No	Yes	No	Yes	Yes	NV	80	140/80	OW	N	No	N	310	280	251-300	240	332	30	126	Normal	0.096
34	Mr P	73	3	1	CVA	No	No	No	Yes	No	V	92	170/90	N	N	Yes	N	218	324	301-350	182	118	40	112	Ishaemia	0.113
35	Mr S	65	3	1	PVD	Yes	Yes	Yes	Yes	No	V	74	130/90	N	N	No	N	268	330	301-350	204	184	46	153	Ishaemia	0.096
36	Mrs R	60	3	2	PVD	No	Yes	No	No	No	V	80	120/70	OW	N	No	N	230	360	351-400	242	164	41	143	Normal	0.113
37	Mr P	50	2	1	CVA	No	Yes	No	No	Yes	NV	74	130/70	OW	N	No	N	219	310	301-350	208	186	37	154	Ishaemia	0.116
38	Mr P	58	2	1	ACS	Yes	Yes	Yes	No	Yes	V	78	160/110	OW	N	Yes	H	236	318	301-350	248	139	35	151	AWMI	0.11
39	Mr S	52	2	1	ACS	No	No	No	No	No	V	72	160/90	N	N	No	N	220	316	301-350	240	154	36	138	ASMI	0.096
40	Mr R	58	2	1	ACS	No	No	No	No	Yes	NV	74	120/80	OW	N	No	N	184	384	351-400	209	109	37	139	IWMI	0.113
41	Mrs S	46	2	2	CVA	No	No	Yes	No	No	V	70	120/74	OW	N	No	N	164	280	251-300	200	96	28	130	Ishaemia	0.09
42	Mr M	54	2	1	CVA	No	No	No	No	Yes	NV	74	130/80	N	N	No	N	156	270	251-300	195	91	23	132	Ishaemia	0.096
43	Mrs M	51	2	2	ACS	No	No	No	No	No	V	72	120/80	OW	N	No	N	170	263	251-300	260	109	36	139	AWMI	0.113
44	Mr A	62	3	1	ACS	Yes	Yes	No	Yes	No	V	74	110/70	N	N	No	N	160	218	201-250	306	190	40	157	IWMI	0.116
45	Mr RR	55	2	1	PVD	No	Yes	Yes	Yes	Yes	NV	70	130/90	N	N	No	H	168	302	301-350	318	200	36	162	Normal	0.093
46	Mr R	52	2	1	PVD	Yes	No	Yes	No	Yes	NV	78	120/80	OW	N	No	N	210	316	301-350	280	186	32	181	Normal	0.09
47	Mrs C	60	3	2	ACS	Yes	No	No	No	No	V	75	130/100	OW	N	Yes	N	172	312	301-350	218	162	18	100	AWMI	0.116
48	Mr A	58	2	1	ACS	Yes	No	No	No	No	NV	78	130/80	OW	N	No	N	166	308	301-350	171	160	29	101	HLWMI	0.096
49	Mr M	42	2	1	ACS	No	Yes	No	No	No	V	74	130/100	N	N	No	N	180	210	201-250	202	150	40	135	IWMI	0.11
50	Mr J	72	3	1	ACS	No	Yes	Yes	Yes	Yes	NV	70	120/80	N	N	No	N	190	290	251-300	174	158	32	106	IWMI	0.116

CONTROL GROUP DATA																								
						Past History			Personal History			Clinical examination					Investigations								
S No	Name	Age	M	Sex	Clinical status	HT	DM	CAD	Smoking	Alcohol	Diet	Pulse	BP	BMI	JVP	Carotid Bruit	CBC	FBS	PPBS	Cholestrol	TC	HDL	LDL	ECG	IMT
1	Mr D	45	2	M	T2DM	No	No	No	Yes	Yes	NV	74	110/80	OW	N	No	N	160	206	154	164	48	121	N	0.05
2	Mrs S	52	2	F	T2DM	No	No	No	No	No	V	76	130/90	OW	N	No	N	156	260	170	168	28	100	N	0.056
3	Mrs M	67	3	F	T2DM/SHT	Yes	Yes	No	No	No	NV	76	160/100	N	N	No	N	217	208	192	116	32	122	N	0.096
4	Mrs K	64	3	F	T2DM	No	Yes	No	No	No	NV	70	110/80	N	N	No	N	162	306	240	119	42	146	N	0.062
5	Mr A	60	3	M	T2DM/SHT	Yes	No	No	No	Yes	NV	74	120/80	N	N	No	N	200	320	250	111	40	145	N	0.083
6	Mr V	70	3	M	T2DM	No	No	No	No	Yes	NV	68	130/80	N	N	No	N	210	360	198	90	32	125	N	0.09
7	Mr M	36	1	M	T2DM	No	No	No	No	No	NV	70	130/70	N	N	No	N	172	280	180	157	26	118	N	0.056
8	Mrs R	50	2	F	T2DM	No	Yes	No	No	No	V	80	140/90	OW	N	No	N	226	234	194	120	46	143	N	0.062
9	Mr G	42	2	M	T2DM	No	No	No	Yes	No	NV	76	120/70	N	N	No	N	280	320	240	163	29	132	N	0.07
10	Mrs E	52	2	F	T2DM	No	Yes	No	No	No	NV	74	130/80	N	N	No	H	190	206	180	142	43	87	N	0.076
11	Mr M	56	2	M	T2DM	No	Yes	No	Yes	Yes	NV	72	150/80	N	N	No	N	146	190	186	116	44	116	N	0.09
12	Mr K	54	2	M	T2DM	No	Yes	No	No	No	NV	84	160/90	N	N	No	N	195	265	187	115	39	121	N	0.08
13	Mr F	60	3	M	T2DM	No	No	No	No	No	NV	80	120/70	N	N	No	N	184	260	180	139	46	112	N	0.08
14	Mrs P	50	2	F	T2DM	No	No	No	No	No	NV	70	130/70	N	N	No	N	260	402	159	85	34	89	N	0.078
15	Mr J	65	3	M	T2DM	No	No	No	No	No	V	82	120/70	N	N	No	N	230	216	192	128	24	96	N	0.09
16	Mr K	50	2	M	T2DM	No	No	No	Yes	Yes	NV	74	160/80	N	N	No	N	242	307	180	132	42	134	N	0.06
17	Mr N	56	2	M	T2DM/SHT	Yes	Yes	No	No	No	NV	76	140/100	N	N	No	N	290	306	167	115	26	119	N	0.083
18	Mrs T	55	2	F	T2DM	No	No	No	No	No	NV	70	110/70	N	N	No	N	202	362	153	98	24	113	N	0.076
19	Mrs A	60	3	F	T2DM	No	No	No	No	No	NV	78	120/80	OW	N	No	N	210	350	203	160	24	136	N	0.068
20	Mr V	66	3	M	T2DM	No	No	No	Yes	Yes	V	68	130/80	N	N	No	N	294	360	220	134	42	151	N	0.076
21	Mr P	60	3	M	T2DM	No	Yes	No	No	No	NV	70	140/80	N	N	No	N	216	320	196	145	39	124	N	0.083
22	Mr R	55	2	M	T2DM	No	Yes	No	Yes	No	NV	72	130/60	N	N	No	N	240	394	205	16	25	131	N	0.082

																				2					
23	Mrs K	63	3	F	T2DM	No	No	No	No	No	NV	76	110/74	N	N	No	N	220	296	182	124	29	142	N	0.078
24	Mr M	52	2	M	T2DM	No	No	No	Yes	Yes	V	70	100/80	N	N	No	N	212	304	195	116	32	98	N	0.068
25	Mrs S	60	3	F	T2DM/SHT	Yes	Yes	No	No	No	V	74	118/70	N	N	No	N	230	310	240	168	34	140	N	0.09
26	Mr A	60	3	M	T2DM	No	No	No	Yes	No	NV	80	140/80	OW	N	No	N	210	356	192	113	38	114	N	0.082
27	Mr P	63	3	M	T2DM/SHT	Yes	Yes	No	No	No	NV	82	130/100	N	N	No	N	202	341	254	190	24	130	N	0.11
28	Mr K	56	2	M	T2DM	No	No	No	Yes	Yes	NV	86	118/90	OW	N	No	N	198	240	215	148	32	100	N	0.083
29	Mrs R	43	2	F	T2DM	No	No	No	No	No	NV	70	130/60	N	N	No	N	164	300	218	162	26	119	N	0.054
30	Mr A	50	2	M	T2DM	No	No	No	Yes	No	NV	72	110/70	N	N	No	H	190	260	205	165	30	124	N	0.068
31	Mr I	55	2	M	T2DM	No	No	No	No	No	NV	78	116/80	N	N	No	N	162	204	195	152	32	112	N	0.078
32	Mr P	56	2	M	T2DM	No	No	No	No	No	NV	76	130/70	OW	N	No	N	169	226	208	160	28	96	N	0.068
33	Mr S	60	3	M	T2DM/SHT	Yes	Yes	No	Yes	No	NV	70	130/80	N	N	No	N	170	195	205	142	21	134	N	0.093
34	Mrs P	47	2	F	T2DM	No	No	No	No	No	NV	74	140/80	N	N	No	N	148	254	182	129	30	112	N	0.072
35	Mr L	50	2	M	T2DM	No	No	No	Yes	No	V	80	110/70	N	N	No	N	160	320	220	164	25	132	N	0.078
36	Mr V	50	2	M	T2DM	No	No	No	No	No	NV	82	120/70	N	N	No	N	224	360	200	132	36	98	N	0.056
37	Mr C	66	3	M	T2DM	No	No	No	No	Yes	NV	74	130/70	OW	N	No	N	215	342	216	147	23	120	N	0.083
38	Mrs S	55	2	F	T2DM	No	Yes	No	No	No	V	68	118/84	N	N	No	N	200	264	255	186	40	132	N	0.086
39	Mrs A	53	2	F	T2DM	No	No	No	No	No	NV	65	116/80	N	N	No	N	212	220	240	194	32	97	N	0.09
40	Mr B	60	3	M	T2DM	No	Yes	No	Yes	Yes	NV	70	120/70	N	N	No	N	205	280	242	168	28	120	N	0.078
41	Mrs L	50	2	F	T2DM	No	Yes	No	No	No	NV	72	110/80	N	N	No	N	168	226	194	132	40	89	N	0.056
42	Mr G	45	2	M	T2DM	No	Yes	No	No	Yes	NV	78	120/70	N	N	No	N	196	234	248	168	32	140	N	0.053
43	Mr P	55	2	M	T2DM/SHT	Yes	No	No	Yes	Yes	NV	76	130/90	N	N	No	N	204	234	216	144	30	124	N	0.086
44	Mrs K	62	3	F	T2DM	No	No	No	No	No	NV	72	140/90	OW	N	No	N	261	402	160	98	27	94	N	0.058

45	Mr S	60	3	M	T2DM	No	No	No	Yes	Yes	V	78	130/80	N	N	No	N	240	264	190	112	32	98	N	0.086
46	Mrs R	59	2	F	T2DM	No	No	No	No	No	V	70	110/70	N	N	No	N	230	194	215	164	48	148	N	0.068
47	Mrs C	60	3	F	T2DM	No	Yes	No	No	No	V	72	120/70	N	N	No	N	210	220	185	116	30	100	N	0.058
48	Mr K	62	3	M	T2DM	No	No	No	Yes	Yes	NV	70	130/84	N	N	No	N	200	232	245	168	32	114	N	0.09
49	Mr L	60	3	M	T2DM	No	No	No	No	No	V	76	110/70	N	N	No	N	212	200	140	100	22	96	N	0.082
50	Mr P	60	3	M	T2DM	No	No	No	No	No	NV	80	100/80	N	N	No	N	164	225	190	146	24	99	N	0.078