A PROSPECTIVE, RANDOMIZED, OPEN LABEL, COMPARATIVE STUDY OF POLICOSANOL AS AN ADD ON THERAPY TO ATORVASTATIN IN PATIENTS WITH HYPERLIPIDAEMIA"

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In partial fulfillment for the award of the degree of

DOCTOR OF MEDICINE
IN
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INSTITUTE OF PHARMACOLOGY
MADRAS MEDICAL COLLEGE
CHENNAI - 600 003

APRIL 2017

CERTIFICATE

This is to certify that the dissertation entitled,

"A PROSPECTIVE, RANDOMIZED, OPEN LABEL, COMPARATIVE STUDY OF POLICOSANOL AS AN ADD ON THERAPY TO ATORVASTATIN IN PATIENTS WITH HYPERLIPIDAEMIA" submitted by Dr. A. SUBA SHREE, in partial fulfillment for the award of the degree of Doctor of Medicine in Pharmacology by The Tamilnadu Dr.M.G.R.Medical University, Chennai is a Bonafide record of the work done by her in the Institute of Pharmacology, Madras Medical College during the academic year 2014-2017.

DEAN
Madras Medical College &
Rajiv Gandhi Govt. General Hospital
Chennai – 600 003.

DIRECTOR AND PROFESSOR,
Institute of Pharmacology,
Madras Medical College,
Chennai – 600 003.

CERTIFICATE OF THE GUIDE

This is to certify that the dissertation entitled,

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Place:
Date:

Dr. K. M. SUDHA, M.D.,
Professor of Pharmacology,
Institute of Pharmacology,
Madras Medical College,
Chennai- 3.

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TURNITIN ANTI-PLAGIARISM SOFTWARE

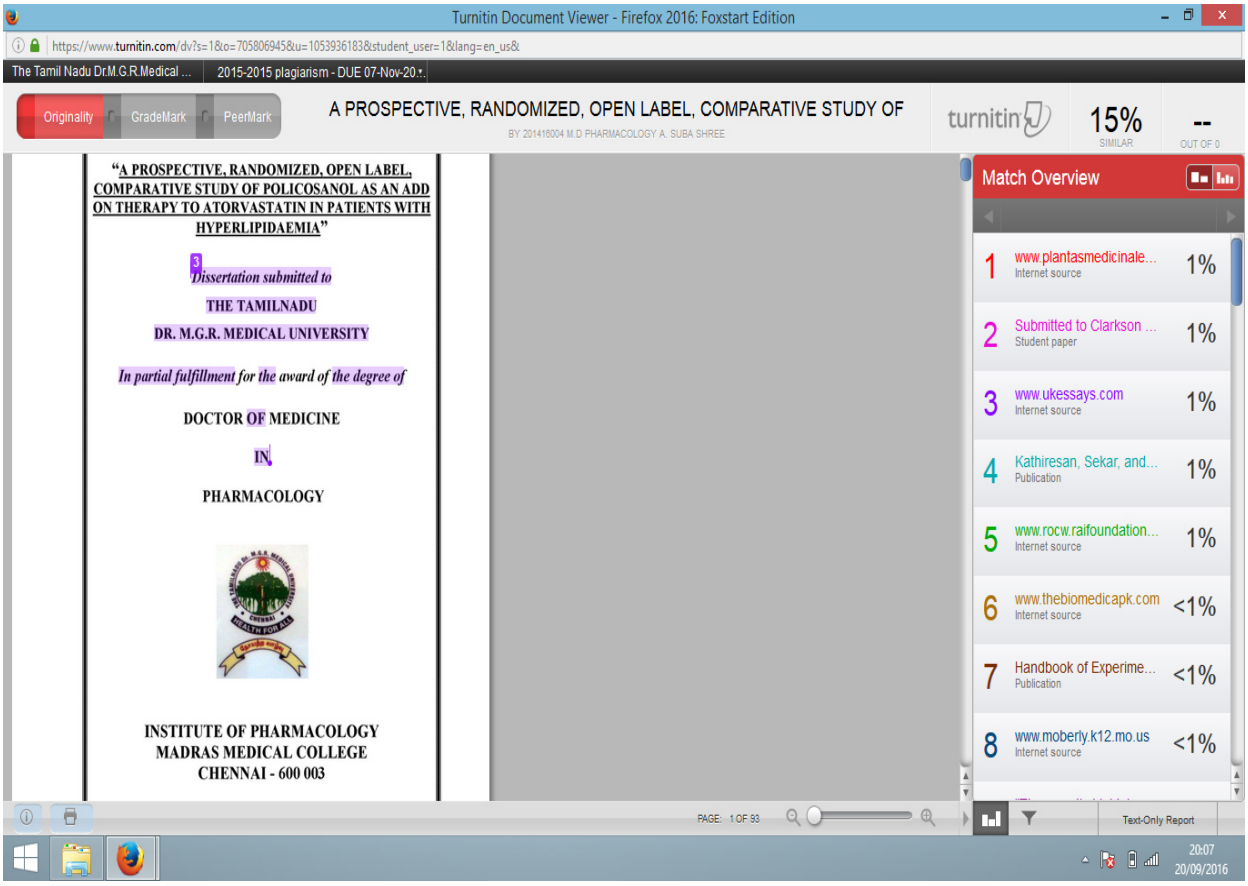


TABLE OF CONTENTS

INTRODUCTION

INTRODUCTION

Hyperlipidemia is one of the main cause of atherosclerosis and atherosclerosis-induced conditions such as coronary heart disease (CHD), ischemic cerebrovascular disease and peripheral vascular disease. In the majority of middle-aged and older adults, atherosclerosis accounts for the increase in morbidity and also for about one third of all the deaths[1].

A number of risk factors contribute to and are also associated with atherosclerosis. The risk factors which are modifiable will be of great value in the prevention of atherogenesis. The non modifiable risk factors include age and genetic factors mainly. The modifiable risk factors include diseases like hypertension, hypercholesterolemia, insulin resistance, and lifestyle factors such as smoking and diet. In these, hyperlipidemia is a potential risk factor for atherogenesis.

The treatment of Hyperlipidaemia[2] is either non-pharmacological or medical management which includes Statins, Fibric acid derivatives, Cholestyramine resins, Niacin & Ezetimibe. Statins are the first line therapy for lowering lipid levels.

Policosanol[3] is a mixture of eight primary long chain aliphatic alcohols, extracted from sugarcane (*Saccharum officinarum*) wax. They are of 24-34 carbons in length. Octacosanol is the predominant alcohol comprising approximately about 63% of the policosanol mixture. Other major constituents include triacontanol (13%) and hexacosanol (6%). The minor constituents include tetracosanol, heptacosanol, nonacosanol, dotriacontanol, and tetratriacontanol.

Policosanol reduces the levels of low-density-lipoprotein (LDL) cholesterol and total cholesterol while increasing levels of high-density-lipoprotein (HDL) cholesterol when given at higher concentrations[4].

Policosanol acts by inhibiting hepatic cholesterol synthesis prior to the formation of mevalonate. It also enhances the binding, receptor mediated uptake, and degradation of the LDL cholesterol in the endoplasmic reticulum, other than its inhibitory effect on cholesterol synthesis. It is unclear whether policosanol also inhibits hydroxylmethylglutaryl-coenzyme A (HMG-CoA) reductase enzyme[5-8].

Policosanol decreases levels of thromboxane A_2 and increases the levels of prostacyclin. In large doses, it can inhibit platelet aggregation induced by arachidonic acid and collagen[9-13].

A number of well-designed, short and long term studies on policosanol found that it significantly lowered both LDL-C and total cholesterol levels in patients with familial hypercholesterolemia[14-34] and also in patients with type 2 diabetes mellitus,[25,26] elderly patients[23,30] and in postmenopausal women[27]. Some long term trials have observed that policosanol significantly raised HDL cholesterol levels. In addition to its hypolipidemic effect, policosanol has shown promising results in treating patients with intermittent claudication[35,36].

In Indian population, no studies have been conducted to evaluate the lipid lowering effect of policosanol. Hence, the present study was planned to evaluate the efficacy of Policosanol as an add on therapy with Atorvastatin in patients with hyperlipidemia.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Rise in the blood cholesterol levels are the major cause for atherosclerosis and atherosclerosis related conditions such as coronary heart disease, cerebrovascular disease and peripheral vascular diseases. Atherosclerosis accounts for about one third of all the deaths and for the increase in morbidity and mortality of the middle aged and older adults worldwide.

Age, genetic factors and other factors like insulin resistance, life style modifications and diseases like hypertension are also associated with atherosclerosis. So there is a need to lower the increased lipid levels to normal levels to avoid atherosclerosis and other atherosclerosis induced conditions.

The high level of cholesterol in the blood is termed as Hypercholesterolemia. The cholesterol content is more in the Low density lipoprotein(LDL) and Very low density lipoprotein (VLDL) and less in the High density lipoprotein (HDL).

Higher the level of LDL-cholesterol is associated with a higher risk for atherogenesis. These increased levels of LDL-C are oxidized by reactive oxygen species in the endothelium of blood vessels. The oxidised LDL-C are scavenged by the macrophages and result in foam cell formation and accumulation of numerous adhesion factors, monocytes and neutrophils on the vascular endothelium. The atherosclerotic lesion so formed is the predisposing factor for various cardiovascular diseases like hypertension, coronary heart disease etc which affects the quality of life.

So, to lead a hale and a healthy life, there should be a balance in cholesterol homeostasis which can be achieved by a gene network involved in cholesterol synthesis, absorption, metabolism and elimination.

METABOLISM OF LIPIDS

LIPIDS:

Lipids are greasy substances occurring widely in nature. They are insoluble in water and soluble in fat solvents.

CLASSIFICATION[37,38]:

We classify lipids into 1) Simple lipids 2) Compound lipids 3) Derived lipids 4)Substances associated with lipids (Miscellaneous lipids).

SIMPLE LIPIDS:

These are the esters of fatty acids and alcohol. They are subclassified into a) fats and oils b) waxes.

COMPOUND LIPIDS:

In addition to alcohol and fatty acid, they contain groups like nitrogenous base, phosphate, carbohydrate, protein etc. They are subclassified into a)Phospholipids b) Glycolipids c) Sulpholipids d)Lipoproteins.

Lipoproteins – They are lipids attached to proteins present in plasma and tissue.

DERIVED LIPIDS:

These are hydrolysis derivatives of simple and compound lipids. They have fatty acid + alcohols other than glycerol + Glycerides + bases.

MISCELLANEOUS LIPIDS:

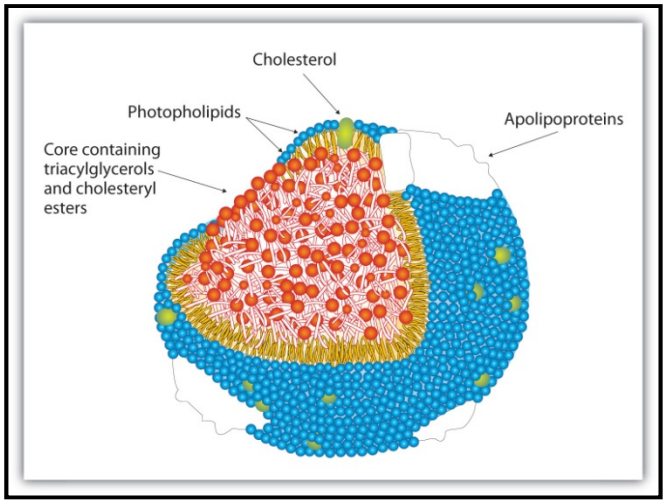
These are the substances which possess characteristics of lipids. Ex: Fat soluble vitamins, steroid hormones, carotenoids etc.

We do have another group called Neutral lipids. These lipids are uncharged and include mono, di and triacyl glycerol, cholesterol and cholesterol esters.

LIPOPROTEINS:

The major lipids in plasma such as triglycerides, phospholipids and cholesterol are combined with specific apoproteins to form molecular complexes called lipoproteins. They are circulated in blood and delivers the lipid to various tissues.

STRUCTURE OF A LIPOPROTEIN[39]:



The lipoprotein consists of an inner hydrophobic neutral lipid core with triacyl glycerol and /or cholesterol esters and an outer hydrophillic coat shell of phospholipids, apoprotein and cholesterol. The outer layer is polar in nature so that lipoprotein is soluble in aqueous solution.

The lipoproteins differ in their cholesterol, triglycerides, phospholipids and in the chemical composition of the protein carriers.

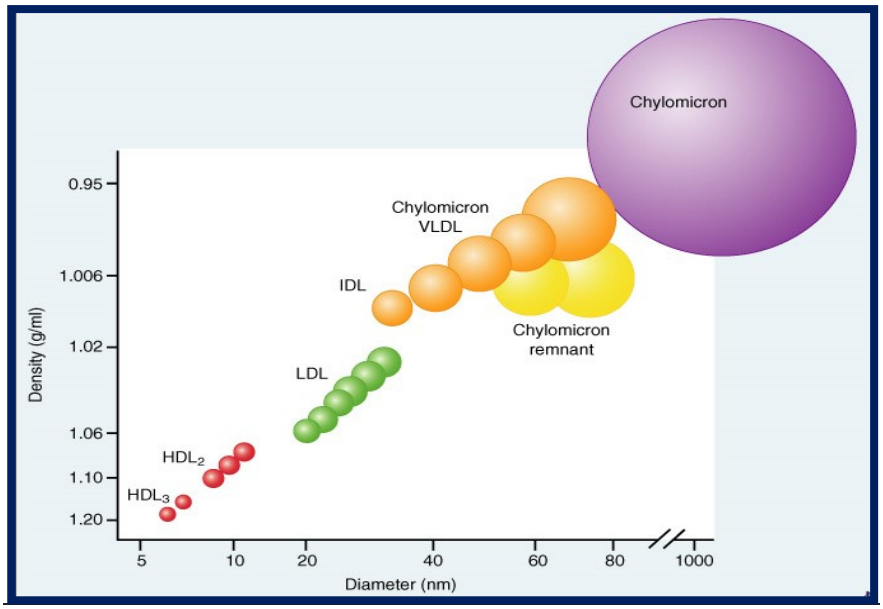
There are five major classes of lipoproteins in human plasma classified based on their densities depending on the proportion of lipids and proteins in them. 1) chylomicrons 2) Very low density lipoprotein (VLDL) 3)Low density lipoprotein (LDL) 4) High density lipoprotein (HDL) 5) Free fatty acids – albumin.

TYPES OF LIPOPROTEINS[39]:

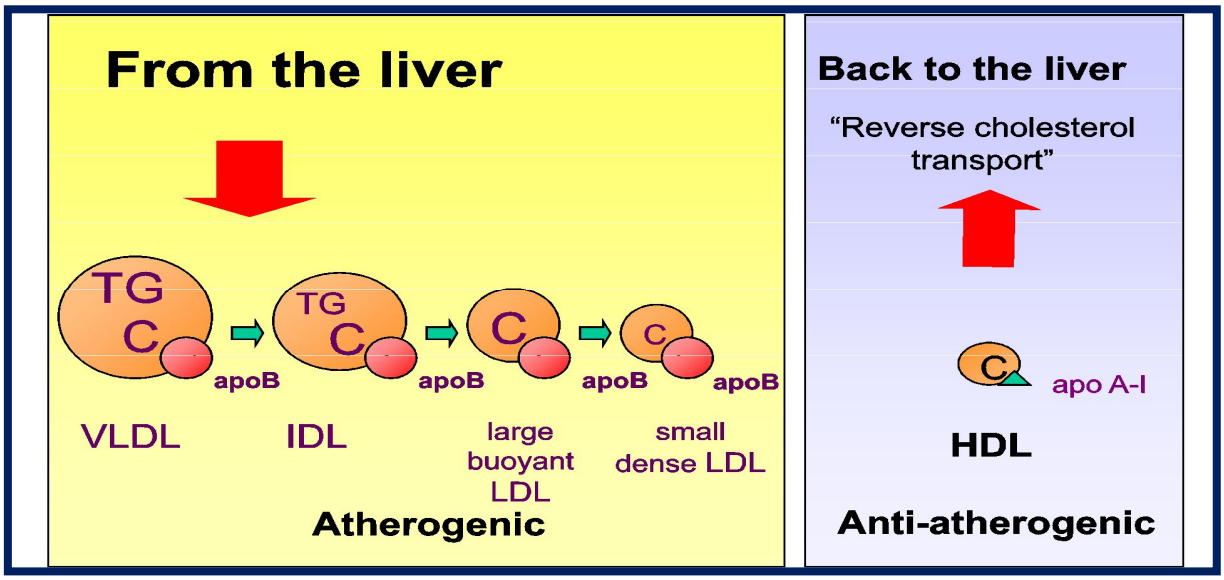
The lipoproteins are categorized into five types based on their diameter and relative density. It is described from larger and less dense molecules to smaller and denser molecules.

1. **Chylomicrons**: The lipoprotein with the greatest diameter and lowest density is known as **Chylomicrons,** which are derived from absorption of triacylglycerol and other lipids from the small intestine.
2. **Very low density lipoproteins** (VLDL, or pre-ß-lipoproteins): They have a diameter lesser than and density higher than chylomicrons. These are derived from the liver for the endogenous transport of newly synthesized triacylglycerol.
3. **Intermediate density lipoprotein (IDL):** This is a lipoprotein with a lower diameter and a little higher density than VLDL.
4. **Low-density lipoproteins** (LDL, or ß lipoproteins): These lipoproteins represent the end product in the catabolism of VLDL. They are also termed as “bad cholesterol” because of their association with the increased progression of atherosclerosis**.**

Major classes of Lipoprotein particles



5. **High-density lipoproteins** (HDL, or α-lipoproteins): These lipoproteins have the highest density and the lowest diameter which are involved in the reverse cholesterol transport (carrying fat molecules from the tissue back to the liver). They are also termed as "good cholesterol" because they prevent the formation of atherosclerotic plaques.



APOLIPOPROTEIN[37]

The protein part of lipoproteins are called apolipoproteins or apoproteins. we have eight different proteins with subgrouping. They are A I ,A II, B (B 100, B 48) , C I, C II, C III, D, E. One or more apoproteins will be present in each lipoprotein.

APOPROTEINS	LIPOPROTEINS
APO A I, APO A II	HDL, Chylomicrons
APO B 100	LDL, VLDL, IDL
APO B 48	Chylomicron and its remnants
APO C I, APO C II	VLDL, HDL, Chylomicrons
APO D	HDL
APO E	VLDL, HDL, CM and CM remnants

The main functions of apoproteins are

1) They form the structure of lipoproteins.
2) They act as enzyme inhibitors or enzyme cofactors.
3) They act as ligands for binding to the receptors.

EXOGENOUS PATHWAY OF LIPID TRANSPORT[40]

In the proximal part of the small intestine, fat-soluble vitamins, dietary cholesterol and fatty acids are all absorbed. Intestinal lipases hydrolyse the dietary triglycerides in the lumen and emulsify with bile acids to form micelles.

Within the enterocyte, the cholesterol gets esterified by the addition of a free fatty acid to form cholesteryl esters. The cholesteryl esters along with the triglycerides form the core of chylomicrons. The outer surface coat is composed of the phospholipids, Apo B48, Apo C, E and A. In this Apo B48 is unique to the chylomicrons. Apo C and E are transferred from HDL.

This newly secreted chylomicrons are called nascent chylomicrons and are absorbed into the intestinal lymph and carried directly through the thoracic duct into the blood stream. They are transported to the peripheral tissues before they enter the liver. These chylomicrons are responsible for the transport of all the dietary lipids into the circulation.

In the heart, skeletal muscle and adipose tissue, these nascent chylomicrons and lipoprotein lipase are attached and anchored by a protein called phosphatidyl inositol-anchored protein, GPIHBP1. These reactions occur on the endothelial surface of the capillaries. The triacylglycerol in chylomicrons are hydrolysed by these lipoprotein lipase and free fatty acids are released. HDL transfers the apo C-II to the chylomicron that acts as a cofactor for lipoprotein lipase.

The released free fatty acids are taken up by the heart and skeletal muscles where they are oxidized to produce energy. They can also be re-esterified and again stored as triglyceride. Some of the free fatty acids released will enter into the hepatocytes by binding with the plasma protein like albumin.

ENDOGENOUS PATHWAY OF LIPID TRANSPORT[41]

The endogenous transport of cholesterol involves two subsystems

- Atherogenic Apo B100 guided system for VLDL, IDL AND LDL.
- Anti atherogenic Apo A1 guided system for HDL.

Atherogenic Apo B100 guided system:

The liver secretes apo-B lipoproteins mainly VLDL and in the peripheral tissues, the triglycerides particles present in them are metabolized.

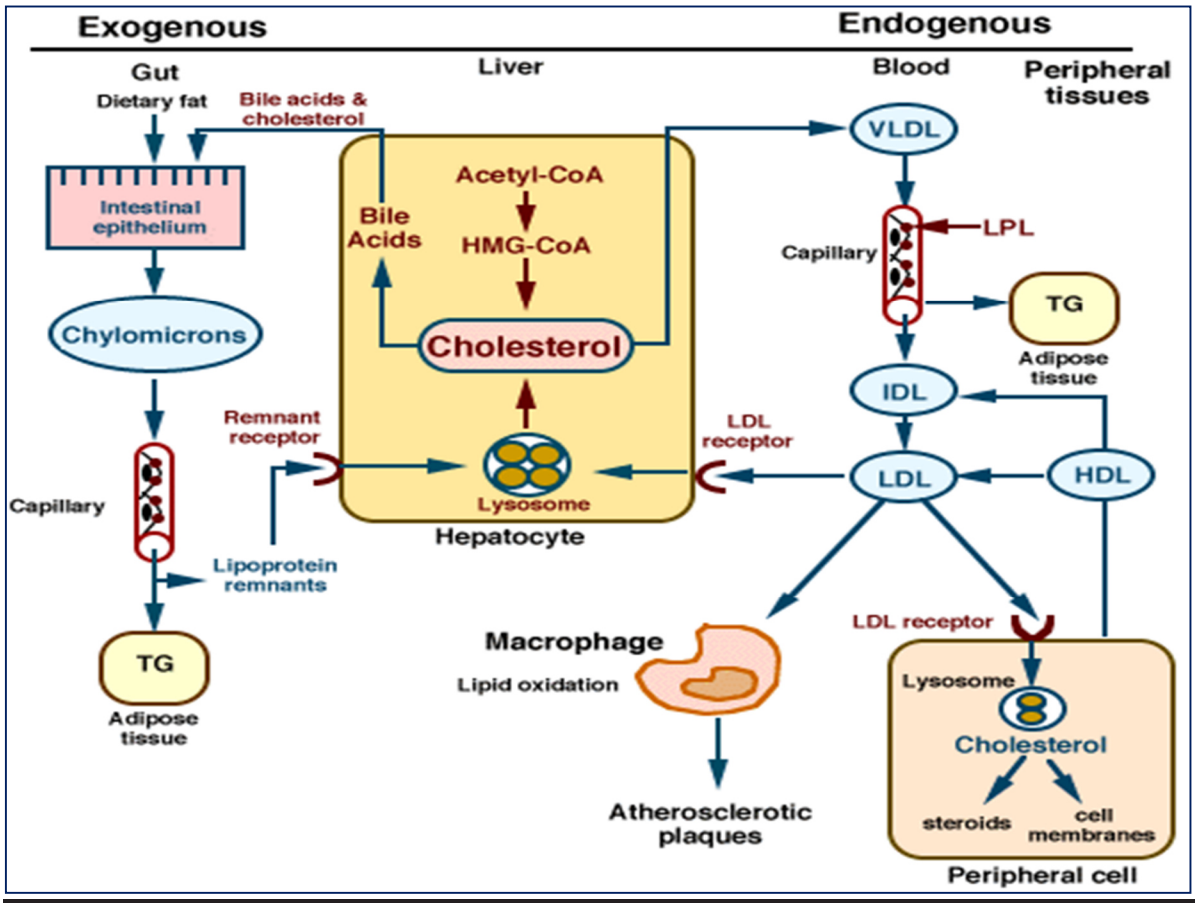
The VLDL particles resemble chylomicrons in their protein composition, but the difference is in VLDL, apo-B48 is replaced by apoB-100. They have the higher ratio of cholesterol and triglycerides.

In the liver, the triglycerides are derived mainly from the esterification of long-chain fatty acids and are incorporated in the hydrophobic core of VLDL. Microsomal triglyceride transfer protein (MTP) helps in the formation of the nascent VLDL particle by combining the hepatic triglycerides with the other major components like apoB-100, phospholipids and cholesteryl esters.

In the plasma, HDL transfers the apolipoproteins (Apo-E and the C series) to the VLDL particle. In the heart, skeletal muscle and adipose tissue, the lipoprotein lipase (LPL) enzyme helps in the hydrolysis of the triglycerides present in the VLDL similar to the process occurring in case of chylomicrons. This results in the formation of VLDL remnants which are now separated from the enzyme lipoprotein lipase. These remnants are called as IDL (intermediate density lipoprotein).

IDL contains almost the same amounts of triglyceride and cholesterol as VLDL. Only 40-60% of these IDL particles are removed by the liver through endocytosis by binding to apo-E and apoB-100 receptors. The remaining IDL forms LDL by hepatic lipase enzyme which hydrolyses its triglycerides.

Diagram shows both exogenous and endogenous pathway

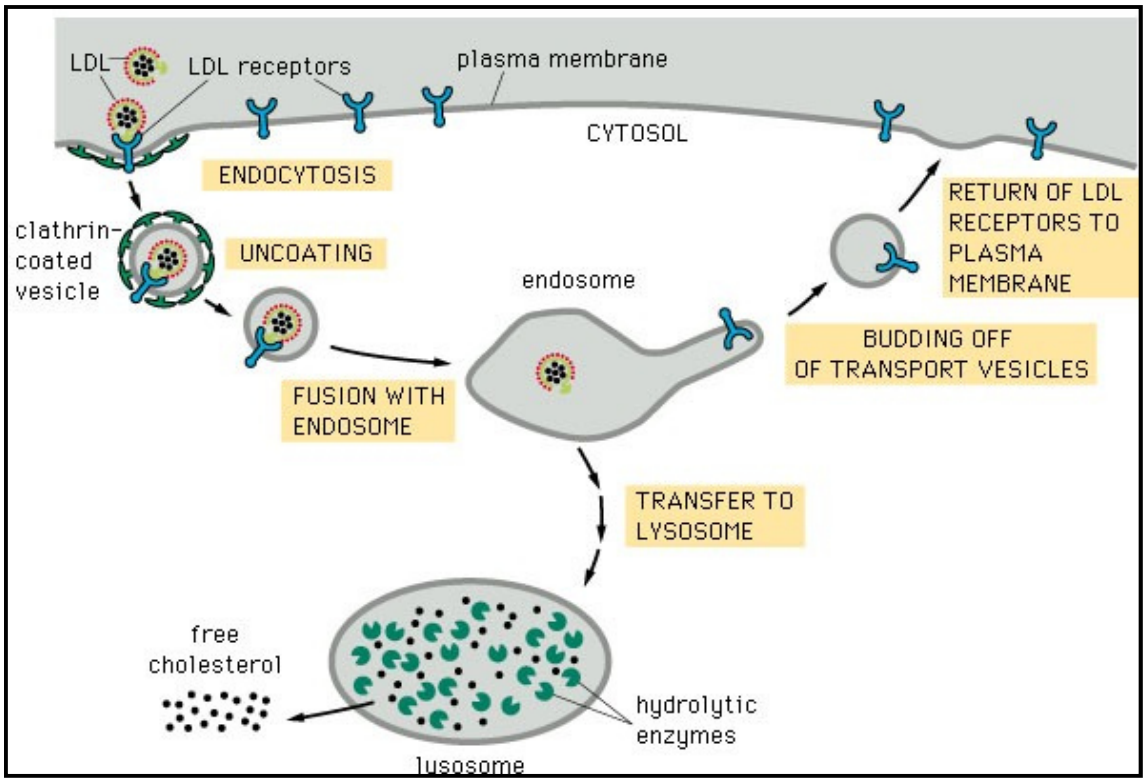


Now LDL carries only apoB-100. It is generally considered that the concentration of plasma cholesterol is equivalent to the amount of cholesterol present in the LDL particle. 70% of the circulating LDL cholesterol is cleared by LDL receptor-mediated endocytosis in the liver.

Receptor-mediated endocytosis of LDL – Cholesterol[42,43]:

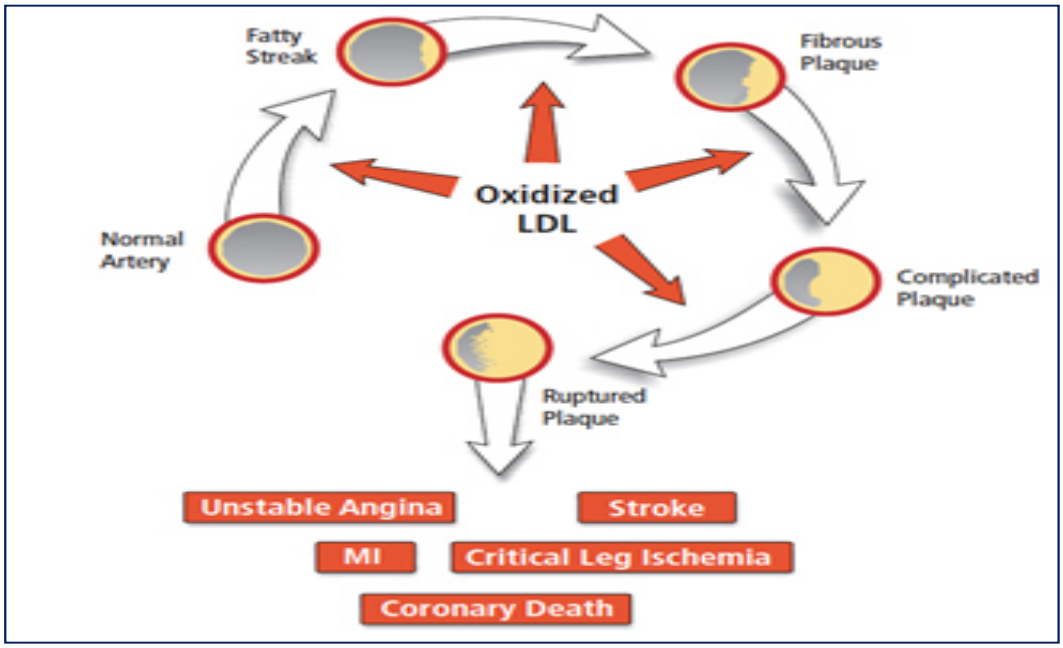
1. In the cells other than liver, the receptors for LDL are present in the plasma membrane in a specialized region called clathrin coated pits. Apo B-100 present on the surface of the LDL particle binds to this receptor protein clathrin and forms a LDL- receptor complex.
2. This LDL- receptor complex is engulfed by a process called endocytosis and taken up inside the cell to form an endocytic vesicle.
3. This endocytic vesicle later fuses with lysosomes which carries a number of degradative enzymes. The cholesteryl esters of low density lipoprotein are hydrolysed by lysosomal acidic enzyme, lipase to form unesterified cholesterol. The proteins are degraded and free amino acids are released. After this lysosomal degradation, the LDL receptor alone returns unmodified to the plasma membrane. The turn- around time for LDL receptor is usually 10 minutes. So in a lifetime of about a day, LDL receptor carries many LDL particles into the cell.

Receptor mediated endocytosis



1. The un-esterified free cholesterol is either used for cell membrane biosynthesis or re-esterified again and stored inside the cell. In fact, the free cholesterol activates acyl CoA: cholesterol acyltransferase (ACAT), the enzyme catalyzing the re-esterification reaction.
2. The re-esterified cholesterol contains Oleate and palmitoleate which are monounsaturated fatty acids but the cholesterol esters in LDL contain linoleate, a polyunsaturated fatty acid. High concentrations of un-esterified cholesterol disrupt the integrity of cell membranes.
3. The synthesis of LDL receptor is subjected to negative feedback regulation. When cholesterol is abundant inside the cell, new LDL receptors are not synthesized and plasma cholesterol are not taken up inside the cell.

ROLE OF LDL-C IN LIPID METABOLISM AND ATHEROSCLEROSIS[44]



In our body, there is a precise regulation in the metabolism of cholesterol to prevent the formation of atherosclerosis. The major carrier of cholesterol in the blood is low density lipoprotein.

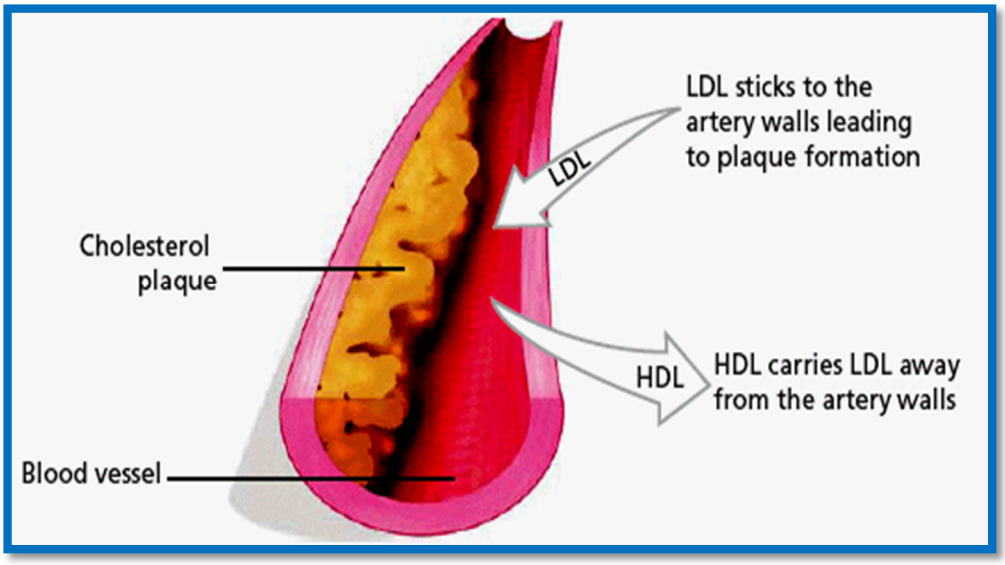
Higher the LDL levels in the blood, higher is the risk of atherogenesis. The main role of LDL-C is to transport the cholesterol from the liver to the peripheral tissues.

Check of cholesterol synthesis in hepatic cells

In the hepatic cells, low density lipoproteins control the synthesis of cholesterol by decreasing the effect of 3- hydroxy-3-methyl glutaryl CoA reductase (HMG CoA reductase) enzyme, the rate limiting enzyme in cholesterol synthesis.

Check of cholesterol synthesis in non-hepatic cells

The Low density lipoproteins are the prime source of cholesterol in the non-hepatic cells. In almost all the cases of familial hypercholesterolemia (Type II hypercholesterolemia), the major molecular defect is a deficiency or absence of LDL receptor. So LDL cannot be taken inside the cell for degradation by lysosomal enzymes resulting in increased level of LDL-C. The lipids in the LDL-C are then oxidised and they are deposited as atherosclerotic plaques in the vessel wall.

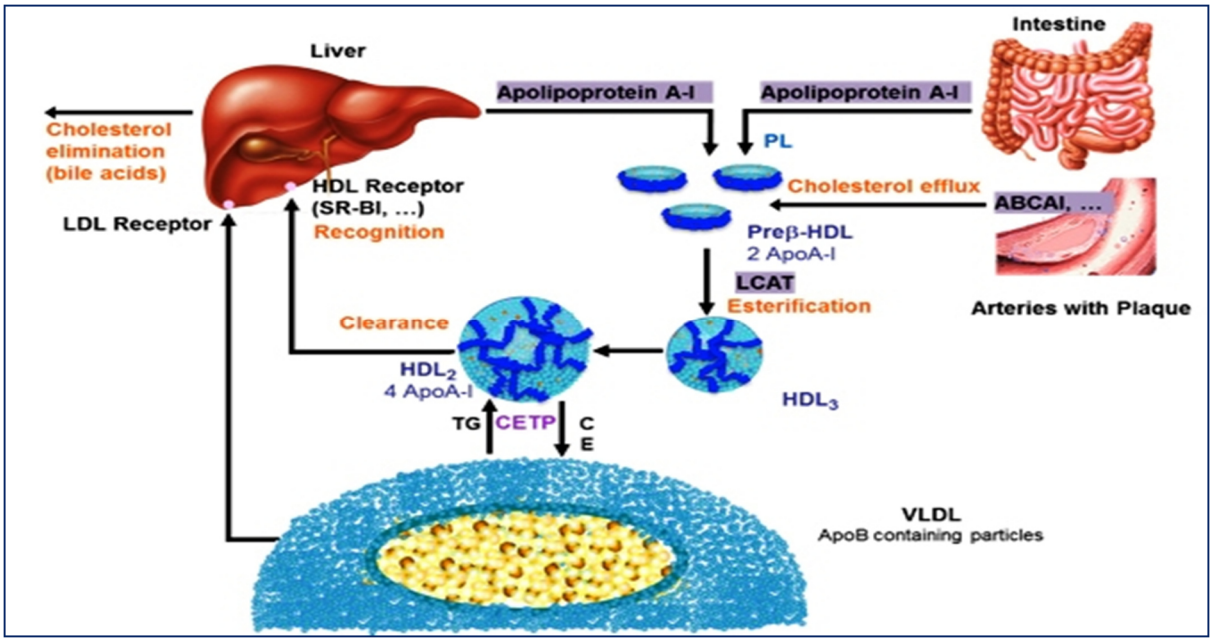


ANTIATHEROGENIC APO AI GUIDED LIPOPROTEIN SYSTEM

ROLE OF HDL IN LIPID METABOLISM

Apoproteins of HDL-C are synthesized from the liver and intestine. HDL gets its cholesterol from the surface monolayers of the chylomicrons and VLDL. It also acquires its cholesterol from the peripheral tissues and brings them to the liver thereby protecting the cells and maintaining cholesterol homeostasis.

REVERSE CHOLESTEROL TRANSPORT



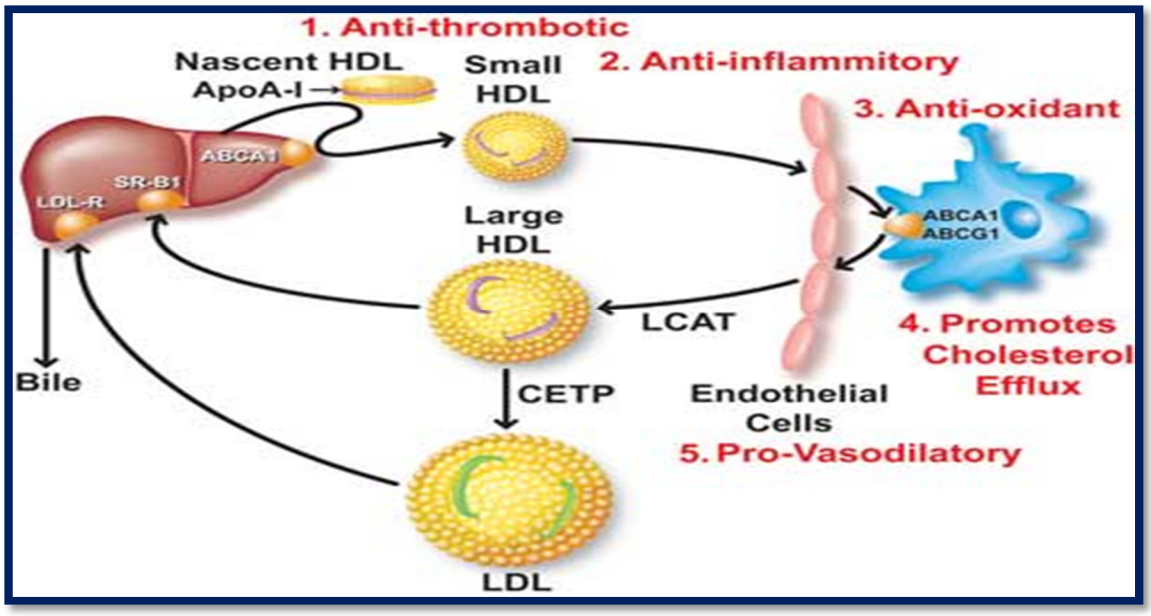
The free cholesterol is mainly transported by a transporter called ABC-1 (ATP binding cassette protein-1), which is acquired by a small particle termed prebeta-1 HDL. The mature HDL particle is formed by the esterification of its cholesterol by the enzyme lecithin:cholesterol acyl transferase (LCAT) in circulation. Apo-CI of HDL activates LCAT facilitating in this esterification.

The cholesteryl esters of the mature HDL particle are then transferred to chylomicrons remnants, VLDL, IDL and LDL, by a transfer protein called Cholesteryl ester transfer protein (CETP).

The transferred cholesteryl esters are finally taken up by the liver by means of two processes- Endocytosis & Docking receptor or scavenger receptor, SR-B1.

The process of HDL transporting the cholesterol from the periphery back to the liver is called reverse cholesterol transport mechanism.

PROTECTIVE ROLE OF HDL



1. HDL has an anti-inflammatory property.
2. It also acts as an anti-oxidant by improving endothelial function through stimulating the endothelial nitric oxide production.

   It also inhibits the expression of cell adhesion molecules like vascular cell adhesion molecule -1 (VCAM-1), E-Selectin, intercellular adhesion molecule-1, and also the accumulation of inflammatory infiltrates in the vessel wall.

DYSLIPIDEMIA

DEFINITION - DYSLIPIDEMIA

Dyslipidemia is the condition where there is alteration of one or many of the lipoproteins which may be an elevation of triglycerides or low density lipoproteins cholesterol, or decrease in high-density lipoprotein cholesterol. Hyperlipidemia refers to only an increase in lipid levels[45].

Various terminologies,

- Hyperlipidemia : increase in lipid levels,
- Hypercholesterolemia : increase in cholesterol levels,
- Hyperlipoproteinemia : increase in lipoprotein levels.

HYPERLIPOPROTEINEMIA - CAUSES[46]

1. **Primary causes**

Includes environmental factors, dietary factors and most commonly genetic defects which may be either single gene or multiple gene defects.

2. **Secondary causes[47]**

- Diabetes mellitus
- Hypothyroidism
- Lipodystrophy
- Stress
- Alcohol excess/ Acute hepatitis
- Anti-hypertensive drugs, diuretics
- Glucocorticoid treatment
- Nephritic syndrome
- Chronic renal failure
- Glycogen storage diseases
- Pregnancy
- Sepsis
- Acute intermittent porphyria
- Protease inhibitor therapy
- Obstructive liver disease/ Cholestasis
- Anorexia nervosa

CLASSIFICATION OF HYPERLIPOPROTEINEMIA [48]

Hyperlipoproteinemias are classified into several types based on their lipid levels and on the pattern observed in the electrophoresis of plasma lipoproteins.

Type I Hyperlipoproteinemia- Familial Hyperchylomicronaemia

- This type occurs due to a genetic deficiency of LPL enzyme resulting in increase in the chylomicron fraction leading to hyperchylomicronemia. There is increase in the serum triglyceride levels even after normal intake of fat in diet.
- These patients usually present with eruptive xanthomas.
- Type I usually does not cause an increased risk of coronary heart disease.
- No drug therapy is effective. Diet low in fat should be followed.

Type II a- Familial Hypercholesterolemia

- Defect is in the synthesis of LDL-C receptors. So degradation of LDL is stopped causing a rise in LDL-C with normal VLDL-C and triglyceride levels. Total serum cholesterol level is raised.
- In the electrophoresis, there is seen a prominent beta-lipoprotein band.
- Risk of Ischaemic heart disease is very much increased.

Type II b - Familial combined or mixed Hyperlipidemia

- In this type IIb, there is an increased VLDL-C resulting in elevated serum triglyceride, LDL and cholesterol levels. Overproduction of VLDL-C in the liver is the primary cause.
- In electrophoresis, there is a prominent beta and pre-beta lipoprotein bands.
- These patients have an increased risk of developing heart diseases.

- Treatment: Diet modulation and drugs.

Type III - Familial dysbetalipoproteinemia

- This type III is also known as broad beta disease as in the electrophoresis, there is a broad beta band between the beta and the pre beta band. This lipoprotein is also known as floating lipoprotein.
- There is apo E deficiency leading to decreased clearance of chylomicrons and VLDL remnants leading to an increase in the triglycerides and cholesterol.
- These patients also have increased risk of developing atherosclerosis and heart diseases and present with xanthomas usually.
- Treatment: Diet modification and drugs like niacin and fenofibrate or statin.

Type IV - Familial Hypertriglyceridaemia

- In type IV, there is an increased VLDL-C level, normal or decreased LDL-C level, normal to increased total cholesterol level with an increased circulating triglyceride level.
- The increase in VLDL cholesterol level is due to an overproduction and/or impaired removal of serum VLDL-C triglycerides.
- In electrophoresis, there is prominent pre-beta lipoprotein band.
- These patients present with a few clinical manifestations apart from accelerated risk of atherosclerosis and ischaemic heart diseases.
- Treatment is with dietary changes and drug therapy like niacin and/or fenofibrate.

Type V - Familial mixed Hypertriglyceridemia

- There is overproduction and reduced clearance of chylomicrons and VLDL-C due to a genetic defect causing a greatly increased triglyceride levels.
- There is normal or reduced LDL-C levels.
- In electrophoresis, there is prominent chylomicron and pre-beta band.
- These patients have no risk of atherogenesis.
- Treatment is with a change in the diet and drugs like niacin, and/or fenofibrate

RISK FACTOR FOR ATHEROSCLEROSIS

- **Major non modifiable** risk factors include elderly people, male sex, a positive family history and genetic abnormalities.
- **Lesser, uncertain modifiable** risk factors include physical inactivity, Stress (Type A personality), Obese persons, Post menopausal women, persons on high carbohydrate diet and foods rich in unsaturated fat , Increased Lipoprotein [LP(a)]
- **Potentially controllable hyperlipidemia** risk factors includes **hyperlipidemia,** diabetes, hypertension, cigarette smoking, Chlamydia pneumonia infection and C-reactive protein.

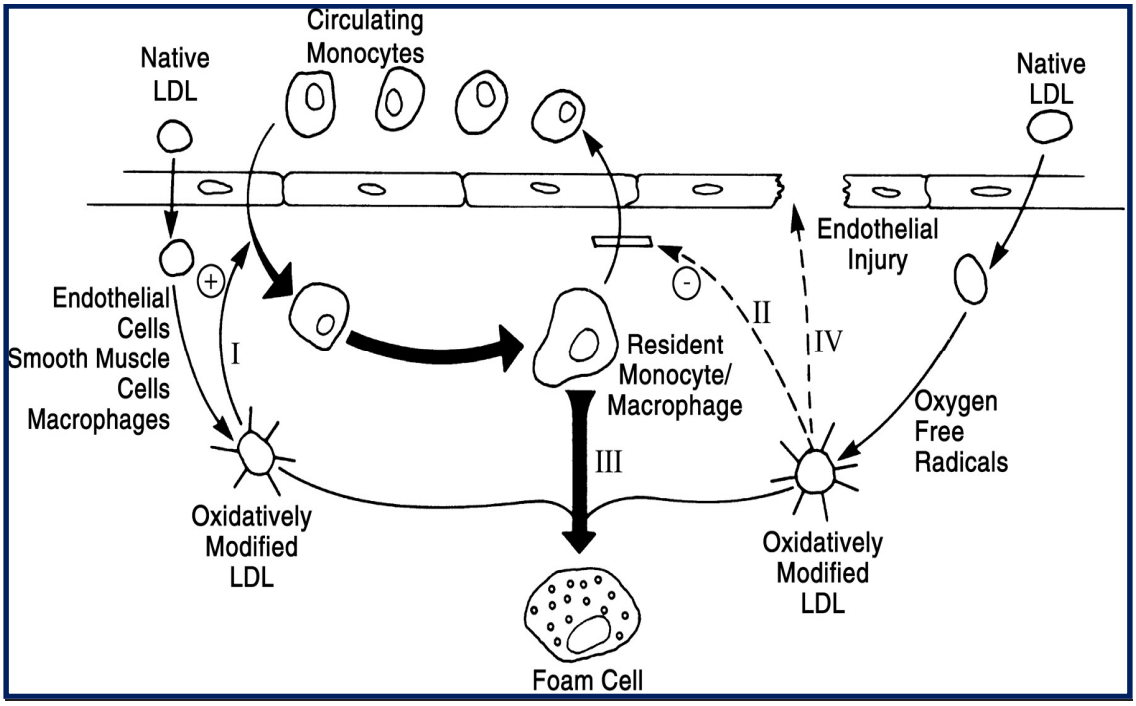
Hyperlipidemia comes under potentially controllable risk factor in causing atherosclerosis.

HYPERLIPIDEMIA – MAJOR RISK FACTOR FOR ATHEROSCLEROSIS.

Hyperlipidemia contributes to atherogenesis by

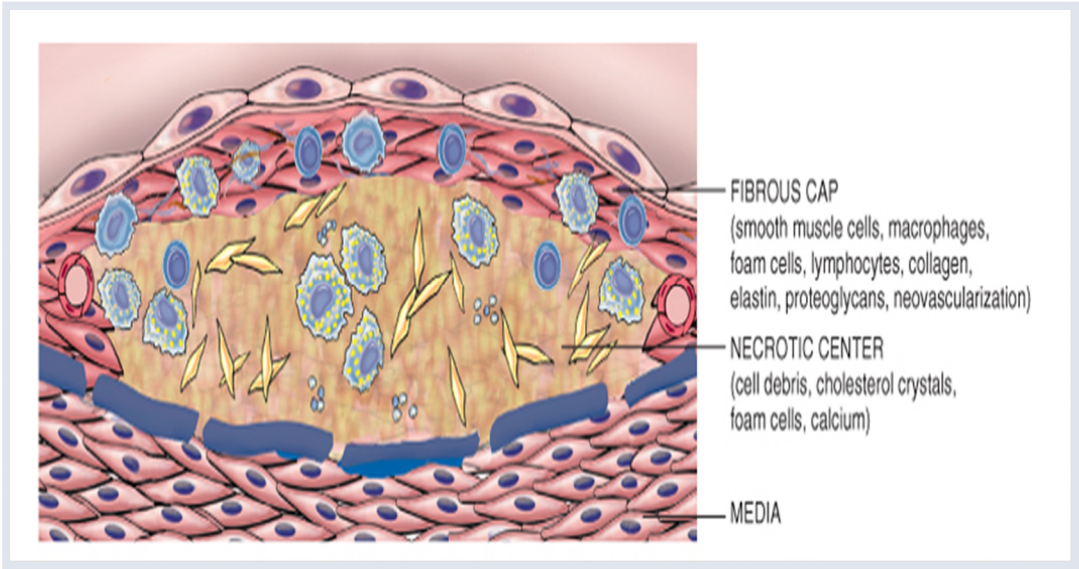
1. An increase in the blood cholesterol levels directly impairs the function of the endothelium by increasing the production of oxygen derived free radicals in the endothelium and decreasing the release of EDRF (Endothelium Derived Relaxing Factor), Nitric oxide.
2. There is increased accumulation of LDL lipoproteins at the site of tunica intima of the vessel wall and are oxidized by the generated free radicals formed.
3. The resultant oxidized LDL cholesterol is readily scavenged by the macrophages through scavenger receptor resulting in the formation of foam cells.

Role of Oxidized LDL-C in atherosclerosis



ATHEROSCLEROSIS[49]

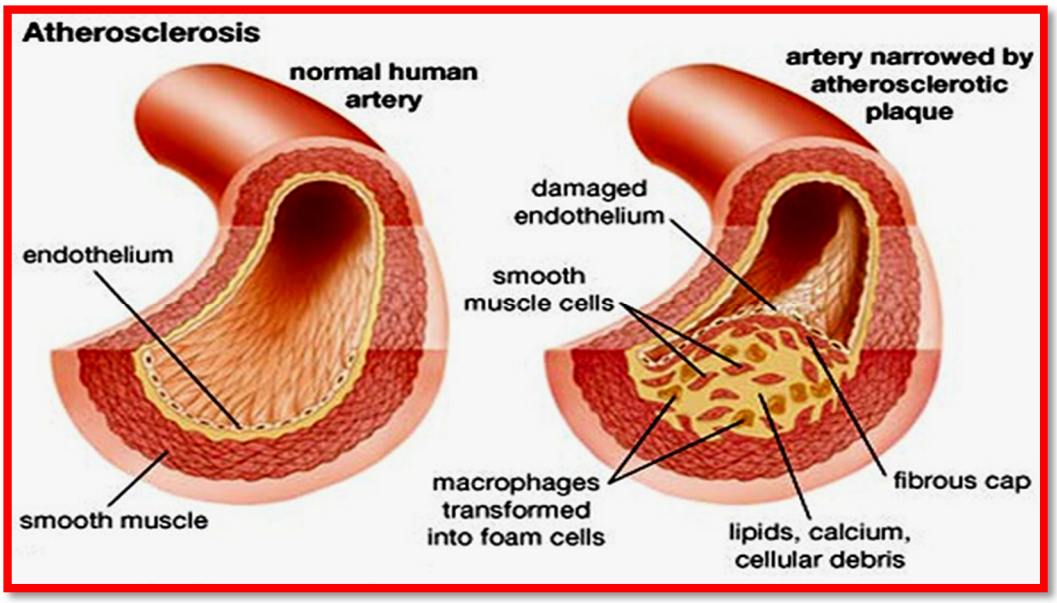
The pathogenesis of atherosclerosis primarily involves the intima of the large and medium sized muscular arteries. Atherosclerosis is composed of fibrofatty cap and a necrotic center. The fibrofatty cap is made up of smooth muscle cells, foam cell, lymphocytes, collagen, macrophages, neovascularisation, proteoglycans, elastin. The necrotic center consists of foam cells, cell debris, calcium and cholesterol crystals.



The Greek word “athera” means lump of gruel gave rise to the term atheroma.

Atheroma is a chronic inflammatory condition resulting from endothelial injury to the arterial vessel wall causing invasion of leukocytes(monocytes) and formation of oxidized LDL lipoproteins. It is usually meant as “hardening” or “furring” of arteries.

Atherosclerosis of vascular endothelium



Atherosclerosis and its associated vascular events involving arteries like coronary artery causing cardiovascular disease (CVD), cerebral artery causing stroke and periphery artery causing peripheral arterial disease (PAD) are the leading cause of morbidity, disability and mortality[50].

Pathogenesis of atherosclerosis

Pathogenesis of atherosclerotic changes in the vessel wall [51]

Chronic injury to vascular endothelium

Increased permeability, adhesion of leukocytes and platelets

LDL Lipoprotein and its oxidized forms accumulate in the intima of the vessel wall

Adhesion of monocytes and migration into the intima of vessel wall

Formation of macrophages from migrated monocytes and foam cells are formed by scavenging of oxidized LDL by the macrophages

Release of platelet adhesion factor from activated platelets, macrophages

Recruitment and proliferation of the smooth muscle cells occur

Lipid accumulates both extracellularly and intracellularly (macrophages and SMCs)

GUIDELINES FOR THE OPTIMAL PLASMA LIPID LEVELS[52]:

Total cholesterol:

Desirable	less than 200 mg/dl
Borderline high	200-239 mg/dl
High	more than equal to 240 mg/dl

HDL cholesterol:

Low	less than 40 mg/dl (50 in women)
High	More than 60 mg/dl

LDL cholesterol

Optimal for very high risk	Less than 70 mg/dl
Optimal	Less than100 mg/dl
Near optimal	100-129 mg/dl
Borderline high	130-159 mg/dl
High	160-189 mg/dl
Very High	≥190 mg/dl

Triglycerides

Normal	Less than 150 mg/dl
Borderline high	150-199 mg/dl
High	200-499 mg/dl
Very High	≥ 500 mg/dl

HYPERLIPIDEMIA - MANAGEMENT

The management of hyperlipidemia includes:

1. Diet modification
2. Physical exercise
3. Avoidance of risk factors
4. Medical management

1. DIET MODIFICATION

The first treatment of hyperlipidemia is dietary modification. Drugs are added only later to escalate the decrease in lipid levels.

The specific dietary interventions are

1. **Avoiding trans fatty acids:**

Foods rich in trans fatty acids have the ability to increase the LDL cholesterol levels and to decrease the HDL levels.

2. **Reduced intake of saturated fat**:

Reduced intake of saturated fat has the greatest ability in reducing LDL cholesterol levels in the blood. Saturated fatty acid rich foods are hydrogenated peanut butters, partially hydrogenated oils and fats, commercially available fried food, commercial bakery products and fat containing animal products

3. **Reduced intake of dietary cholesterol**:

The influence of dietary cholesterol on plasma cholesterol level is minimal. Dietary intake also varies from individual to individual. However, reduced intake of dietary cholesterol to < 300 mg/day helps in reduction of LDL-C levels.

4. **Consumption of MUFA and PUFA:**

Increased intake of monounsaturated and polyunsaturated fatty acids are good substitutes for saturated fats in lowering the LDL cholesterol levels in the blood. The PUFA rich oils are fish oils, soyabean oil, sunflower oil etc.

5. **Increased omega-3-fatty acid intake**:

Omega-3-fatty acids are rich in fish and its consumption helps in the reduction of Triglyceride levels which also contributes to the total cholesterol content of blood. Omega-3-fatty acids also have cardio protective effects.

6. **Increase dietary fiber intake**:

Soluble dietary fiber intake will reduce the LDL cholesterol levels. The good sources of soluble dietary fibre includes vegetables, fruits, whole grains, legumes etc

7. **Intake of antioxidant rich food sources**:

There is clearcut evidence of various antioxidants like vitamin C and E, playing an important role in decreasing the incidence of coronary heart disease. So, the recommended intake of antioxidant rich foods reduce the bad cardiovascular events better than other food supplements.

8. **Increased plant sterols intake**:

Plant-sterol containing foods lower the LDL-C by decreasing the absorption of cholesterol and fat soluble vitamins from food in the intestine.

So the AHA (American Heart Association) recommends the use of plant sterol foods, such as "cholesterol-lowering" margarine-type spreads and salad dressings, mainly for adults who are hyperlipidemic and usage for adolescents should be minimised[53].

2. INCREASED MODERATE PHYSICAL ACTIVITY

The AHA, American Heart Association recommends daily exercise programs like 30 minutes of moderate physical activity which burns about 210 kcal/day approximately consuming 4-7 kcal/min. These type of fitness therapies help in reducing the cardiovascular risk. This is actually a reasonable and feasible fitness therapy.

3. ELIMINATION OF RISK FACTORS ASSOCIATED WITH ATHEROSCLEROSIS:

Alcohol consumption is an important risk factor in increasing the cardiovascular risk. Red wine is usually beneficial in hypercholesterolemia since it has antioxidant effect. Also, patients who present with increased triglyceride levels are strictly recommended to stop or decrease the alcohol intake.

Smoking is another common risk factor for cardiovascular diseases. The smoking individuals are advised to quit smoking.. If individuals are unable to quit smoking, various smoking cessation therapies are available. These smoking cessation therapies may be either a drug therapy or nicotine replacement therapy[53].

4. DRUG THERAPY OF HYPERLIPIDEMIAS[55]:

- **HMG Co-A Reductase inhibitors**

  Atorvastatin, Rosuvastatin, Lovastatin,

  Simvastatin, Fluvastatin, Pravastatin

- **Lipoprotein lipase (LPL) activators**

  Bezafibrate, Clofibrate, Gemfibrozil, Fenofibrate

- **Bile acid sequestrating resins**

  Colestipol, Cholestyramine, Colesevelam

- **Cholesterol absorption inhibitor**

  Ezetimibe

- **Triglyceride synthesis inhibitor**

  Nicotinic acid

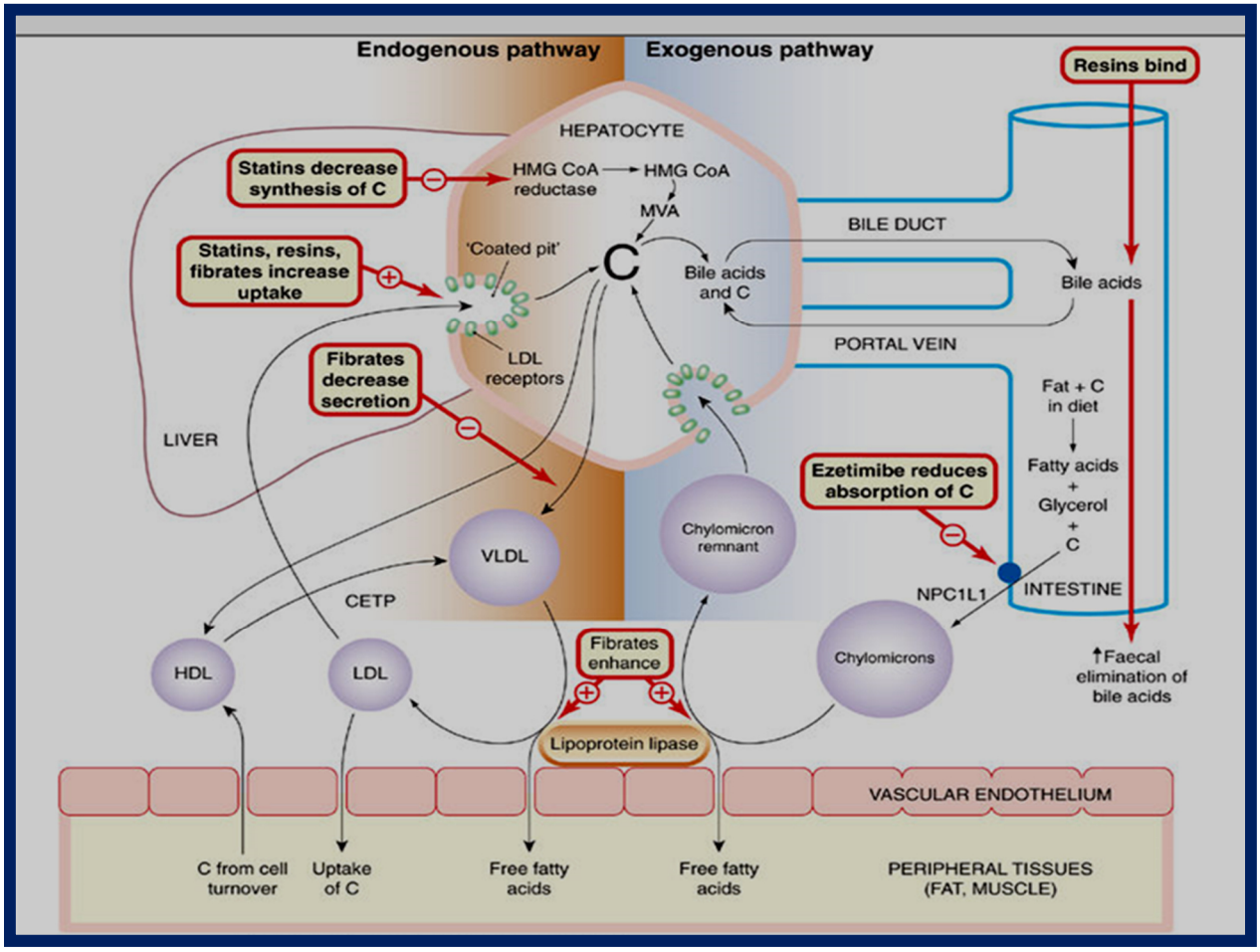
- **New drugs – CETP Inhibitors**

  Torcetrapib, Anacetrapib

- **Miscellaneous**

  Probucol[55], Gugulipid[40,] Fish oil derivatives[40]

Site of action of Statins, Fibrates, Ezetimibe and Resins used in Hypercholesterolemia



1. HMG-CO-A REDUCTASE INHIBITORS

ATORVASTATIN

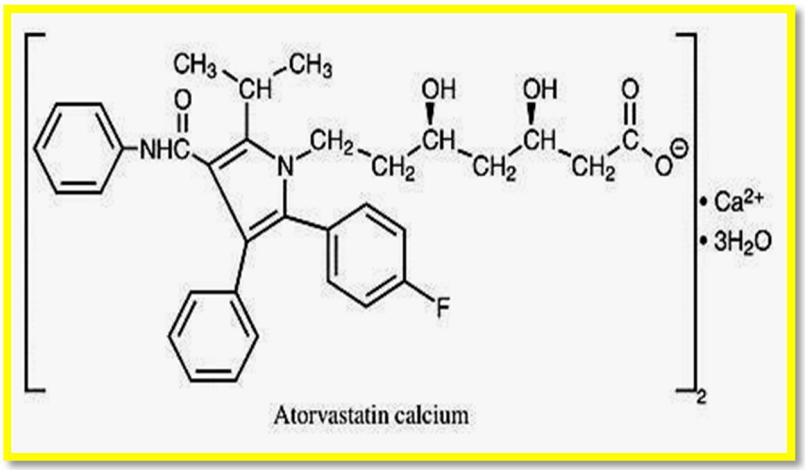
HISTORY:

In the year 1976, statins were first obtained from a mould called *Penicillium citrinum*, and it was identified as an inhibitor of cholesterol synthesis. Their mechanism of action is by inhibiting HMG-CoA reductase enzyme which is the rate limiting step in cholesterol synthesis.

The first statin to be studied in humans was Compactin, later renamed as mevastatin. The first developed statin and approved for use in humans was Lovastatin (formerly known as mevinolin). Lovastatin was isolated from the fungus, *Aspergillus terreus.* Six more statins are also available at present. Simvastatin and Pravastatin are modified chemical derivatives of lovastatin. Other statins like atorvastatin, rosuvastatin, fluvastatin, and pitavastatin are structurally distinct synthetic compounds.

CHEMISTRY

- The chemical structure of Atorvastatin is $(C_{33}H_{34}FN_2O_5)_2\,Ca_3H_2O$.
- The physical appearance of Atorvastatin calcium - white crystalline powder.
- It is insoluble in acidic pH.



Atorvastatin calcium

MECHANISM OF ACTION

The statins have a mevalonic acid like moiety which irreversibly inhibits the HMG-CoA reductase enzyme, the rate limiting enzyme in cholesterol synthesis.

So statins exert their action at an early step by inhibiting the conversion of HMG-CoA to mevalonate thereby reducing the LDL-C levels.

In the hepatic cells, the decrease in cholesterol synthesis results in transcription of genes responsible for the synthesis of LDL receptors. The membrane bound proteins called SREBPs undergo proteolysis and are translocated to the nucleus. This transcription factor binds with the sterol-response element, SRE of the LDL receptor gene resulting in increased transcription and synthesis of LDL receptors. So, the increased LDL receptors endocytose the LDL-C from the blood by receptor mediated endocytosis thereby clearing it from the blood and lowering its blood levels. There is also reduction in the degradation of LDL receptors. Statins also has the effect of reducing the LDL-C levels by enhancing the removal of precursors of LDL like VLDL and IDL and reduces the synthesis of hepatic VLDL.

PHARMACOLOGICAL ACTIONS OF ATORVASTATIN

1. **On Triglyceride levels**

Other than LDL-C, statins are also effective in reducing the triglyceride levels. The decrease in triglyceride levels are similar to the level achieved in case of LDL cholesterol levels. The observations in hypertriglyceridemic patients taking the higher dose of the most potent statins revealed a reduction of 35-45% of LDL-C levels and an equal amount of reduction in fasting triglyceride levels also.

2. On HDL-C Levels

Statins generally show a 5-15% modest rise in HDL cholesterol levels.

3. On LDL-C Levels

Statins can lower LDL-C levels upto 60%. This effect is augmented with the addition of nicotinic acid with statins.

4. Potential Cardio protective effects other than lowering LDL-C.

i. Action on endothelium

Vasoconstrictors and vasodilators both play a dynamic role in the vascular endothelium in the causation of coronary heart disease. Statins enhance the production of a potent vasodilator, Nitric oxide in the vascular endothelium. This vasodilating action of nitric oxide improves the endothelial function and results in protection against cardiovascular risk factors.

ii. On Plaque Stability

a) Statins inhibits the endothelial infiltration of monocytes into the arterial wall.
b) They also inhibit the secretion of matrix metalloproteinase (MMP) from the macrophages which normally destroys the extracellular matrix resulting in the weakening of the fibrous fatty cap.
c) This leads to the disruption of the atherosclerotic plaques. So by inhibiting MMP, statins prevent weakening of plaques that have formed.

d) Statins also inhibit smooth muscle cell proliferation and also enhance apoptosis thereby preventing atherosclerosis.

iii. Anti- Inflammatory action

C-reactive protein is an acute phase reactant increased in inflammatory conditions. Increase in C-reactive protein is a risk factor for coronary heart disease and atherosclerosis. The anti-inflammatory action noted in case of statins is their ability to decrease this C-reactive protein levels.

iv. On Lipoprotein Oxidation

Oxidation of LDL–C lipoproteins is the prime mediating factor in the formation of atherosclerotic foam cells which are nothing but oxidized LDL scavenged up by macrophages. The oxidized lipoproteins also promote cytotoxicity inside the atherosclerotic lesions. Statins reduce this oxidation of LDL-C lipoproteins and thereby decreasing the uptake of oxidized lipoproteins within the macrophages and also the formation of foam cells.

v. On Coagulation

Statins mainly reduce the platelet aggregation and also reduce its adhesion. Fibrinogenemia is associated with a greater incidence of cardiovascular risk.

❖ Atorvastatin has also an additional anti-oxidant property.

PHARMACOKINETICS

On oral administration, Atorvastatin is rapidly absorbed from the stomach. It has a longer plasma half life ($t_{½}$ = 18-24 hrs). Its plasma protein binding is more than 98%. It is metabolized in the liver. The HMG-Co-A reductase inhibiting action of atorvastatin is mediated through its metabolite. It is excreted primarily in bile.

USES

1. Statins are the first line drugs in the management of Hyperlipidemia with elevated LDL-C and total cholesterol levels.
2. It is also effective in most cases of secondary hypercholesterolemia eg: Diabetes, hypothyroidism and Nephrotic syndrome.
3. It also used in patients with risk factors for atherosclerosis for the primary prevention of arterial diseases.
4. It is used as a prophylactic therapy in the secondary prevention of myocardial and cerebral infarction.
5. **ADVERSE EFFECTS**

All statins are generally tolerated well.

Important side effects are:

1. Mild gastrointestinal complaints and headache.
2. Liver dysfunction noted by rise in serum transaminase levels occur occasionally.

3. Muscle aches (myopathy) are the most commonest side effect. Elevation in CPK levels occur infrequently.
4. Myopathy is more common when drugs like nicotinic acid / gemfibrozil or CYP3A4 inhibitors like ketoconazole/erythromycin/cyclosporine are used concurrently.

2. **LIPOPROTEIN LIPASE ACTIVATORS**

Gemfibrozil, Bezafibrate, Ciprofibrate and Fenofibrate cause reduction in the VLDL levels by degrading its triglycerides.

MECHANISM OF ACTION

Fibrates are derivatives of isobutyric acid. They activate the enzyme lipoprotein lipase, most important in the catabolism of VLDL by cleaving the triglycerides. This results in decrease in both VLDL and triglyceride levels. Fibrates activate peroxisomal proliferation activated receptor-alfa, a nuclear transcription gene regulating receptor. This PPARα activation enhances lipoprotein lipase synthesis and the expression of Apo A-I facilitating reverse cholesterol transport by HDL.

PPARα decreases the expression of Apo C-III, an LPL inhibitor. PPARα also enhances the expression of LDL receptor in the hepatocytes.

Fibrates lower the triglyceride levels by about 20-50% and lower LDL cholesterol levels by about 10-15%.

THERAPEUTIC USES

- Fibrates come under first line drugs for patients with increased triglyceride levels.
- To prevent acute pancreatitis in patients with severe hypertriglyceridemia and hyperchylomicronaemia.

ADVERSE EFFECTS

Side effects are rare which includes gastrointestinal symptoms, rashes, impotence, myopathy, blurred vision, hypokalemia and increased blood levels of aspartate and alanine aminotransferases and alkaline phosphatases.

3. **BILE ACID SEQUESTRATING RESINS**

Cholestyramine, Colestipol and Colesevelam are the bile acid sequestrants and are useful in patients with isolated increase in LDL-C levels.

MECHANISM OF ACTION

Bile acids are produced normally in the liver from cholesterol and secreted in the duodenum to help in the absorption of dietary fat. They are again taken up back to the liver through portal circulation. Bile acid sequestrants are the drugs which are anion exchange resins available in the chloride form. After oral administration these drugs can neither be digested nor be absorbed in the intestine.

It binds and complexes with the bile acids in the intestine and interrupts the entero-hepatic portal circulation of bile acids. So the faecal excretion of bile salts and cholesterol is increased. To this loss of bile acids, there is an indirect enhancement of cholesterol to bile acids in the liver.

ADVERSE EFFECTS

Constipation, flatulence and other gastrointestinal symptoms like accentuation of pre existing haemorrhoids can occur.

USES

a) Type IIa and Type IIb hyperlipoproteinemias along with niacin
b) To relieve pruritis in patients with cholestasis.
c) In digitalis toxicity, as these resins can bind to digitalis glycosides.

4. CHOLESTEROL ABSORPTION INHIBITORS[56]

Ezetimibe is a novel drug which not only inhibits the cholesterol absorption from the intestine but also inhibits the enterohepatic reabsorption of cholesterol excreted in bile.

PHARMACOKINETICS

It is absorbed rapidly from the gastro intestinal tract, undergoes conjugation with the glucuronide and then excreted. Its plasma half life is about 22 hours.

MECHANISM OF ACTION

The main mechanism of Ezetimibe is to inhibit a specific cholesterol transport protein called NPC1L1, a protein involved in the absorption of cholesterol from the intestinal mucosa. This results in decreased absorption of both dietary cholesterol and cholesterol excreted in the bile.

This loss of cholesterol accentuates the uptake of LDL via LDL receptors. Statins block the enhanced cholesterol synthesis and so the two drugs have a synergistic LDL-C lowering effect.

ADVERSE EFFECT

Reversible liver dysfunction and very rarely myositis have been noted with the use of this drug.

5. TRIGLYCERIDE SYNTHESIS INHIBITORS AND LIPOLYSIS

Nicotinic acid or Niacin comes under B group vitamin. In higher doses, it lowers the Triglycerides and VLDL-C very rapidly followed by moderate fall in LDL-C and Total Cholesterol levels. Its excretion is primarily in the urine.

MECHANISM OF ACTION

Niacin inhibits lipolysis in the adipose tissues and so of the circulating free fatty acids which are utilised by the liver for the synthesis of triglycerides in VLDL. So there is decreased release of VLDL from the liver and also of IDL and LDL indirectly. It could also increase the activity of lipoprotein lipase that hydrolyses triglycerides.

USES

Combined with bile acid sequestrants or statins, niacin reduces LDL cholesterol levels in Type II familial hypercholesterolemic patients.

ADVERSE EFFECTS

Nicotinic acid is a cutaneous vasodilator resulting in marked flushing and itching. This is due to the release of prostaglandin PGD_2 in the skin.

To minimise the symptom of flushing, Aspirin can be taken before niacin or Laropiprant, a specific antiflushing drug can be combined with niacin. The former acts by inhibiting PG synthesis. Laropiprant has no hypolipidemic action.

Gastro intestinal side effects like dyspepsia, vomiting, diarrhea can occur. Dryness and increased pigmentation of skin, liver dysfunction and jaundice, hyperglycemia and hyperuricaemia, atrial arrhythmia can also occur with niacin.

6. CHOLESTEROL ESTER TRANFER PROTEIN INHIBITORS[40]

Anacetrapib and Torcetrapib are the two drugs under this group. These are newly developed drugs in 2004 with the prime aim of increasing the HDL-C levels. They underwent clinical trials in 2007 and Anacetrapib proved to be effective in raising HDL-C levels by 129%. Cholesterol ester transfer protein (CETP) helps in the transport of cholesteryl esters from HDL-C to LDL-C and VLDL-C.

7. MISCELLANEOUS

PROBUCOL

It is also a hypolipidemic agent which inhibits the cholesterol synthesis and cholesterol absorption. It also increases the rate of degradation of LDL.

GUGULIPID

The drug is extracted from Guggul gum, consisting of Z and E Guggulsterones. This drug can reduce total cholesterol, LDL-C and reduce triglycerides to a moderate extent. It can increase HDL-C levels also. The only side effect noted is loose stools.

DERIVATIVES OF FISH OILS

The PUFA or the Omega-3 fatty acids, rich in fish oils like DHA and EPA are usually used as prophylaxis in hyperlipidemic patients. They have membrane stabilising and antioxidant action

POLICOSANOL[3]

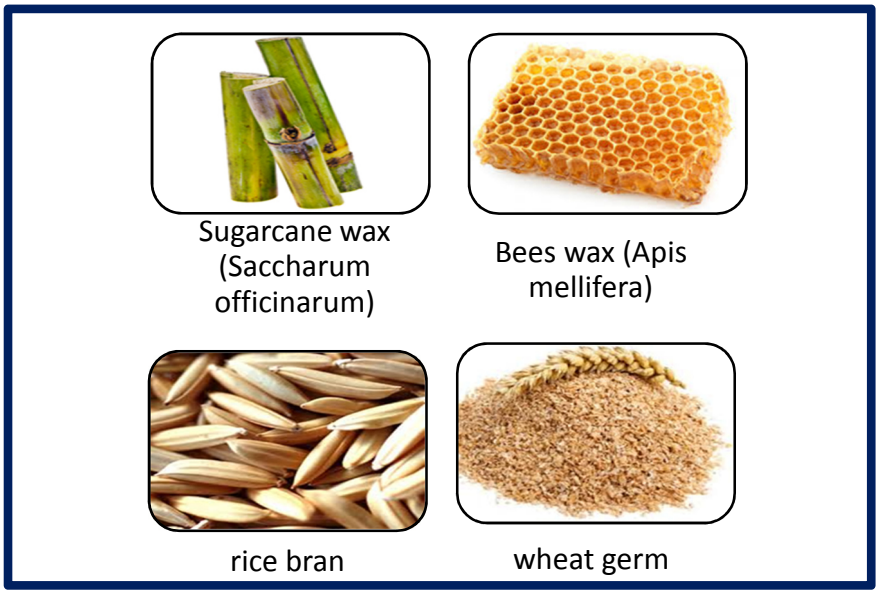
Policosanol is a natural mixture of eight primary long chain aliphatic alcohols of 24-34 carbon length.

Policosanol is isolated, extracted and purified usually from sugarcane wax. It is also obtained from bees wax, wheat germ or rice bran by solvent extraction and saponification .It is present in the plants epicuticular waxes which can be extracted and used as a dietary supplement.

COMPOSITION OF THE POLICOSANOL MIXTURE[57]:

The main constituent is octacosanol (60-70%).The next common constituents are Triacontanol (10-15%) and Hexacosanol (4-10%). The Minor components include Tetracosanol, Heptacosanol, Nonacosanol, Dotriacontanol and Tetratriacontanol.

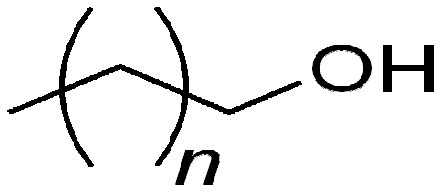
SOURCE[3]



STRUCTURE OF POLICOSANOL[57]

- The chemical structure of policosanol is

CH3-(CH2)n-CH2OH n=24-34



- The different components of policosanol differ in their n, number of carbon atoms.

For ex: Octacosanol has 28 carbon atoms with the molecular formula C_{28} H_{58} O.

BIOAVAILABILITY AND PHARMACOKINETICS[4]

- Policosanol is rapidly absorped from the small intestine
- Peak levels are achieved from 30 to 120 minutes after treatment with policosanol
- It is mainly distributed in the liver and extensively metabolized in the liver.
- It is excreted primarily in the faeces.

MAIN ACTIONS[3]

HYPOLIPIDEMIC ACTION:

Policosanol lowers LDL-C by 25% and increases HDL-C by 10% similar to that of statins. Policosanol has very minimal or no effect on triglyceride levels.

A number of studies in healthy volunteers, in type II hypercholesterolemia patients, Type II DM patients, post menopausal women have revealed that policosanol could lower LDL-C and Total cholesterol and increase HDL-C levels without any effect on triglycerides[58-60].

DECREASES THE OXIDATION OF LDL-CHOLESTEROL[61,62]:

This has been proved by an invitro study done by Menendez et al.

DECREASES PLATELET AGGREGATION[9-13]:

Clinical studies and animal models have confirmed that policosanol inhibits collagen induced platelet aggregation by decreasing the generation of thromboxane but not prostacyclin

ENDOTHELIAL PROTECTION:

It was found in studies that policosanol, in spontaneous hypertensive rats, caused a significant reduction in circulating endothelial cells.

ANTIHYPERTENSIVE EFFECTS[3]:

At very high doses in animals, policosanol enhances propranolol induced hypotensive effects. This higher dose is clinically irrelevant in humans.

EFFECT OF POLICOSANOL ON ATHEROGENESIS[3]:

In an animal study, it has been proved that policosanol reduced the thickness of fatty streaks and most policosanol pre-treated animals did not develop atherosclerotic lesions.

It also has performance enhancing action[63].

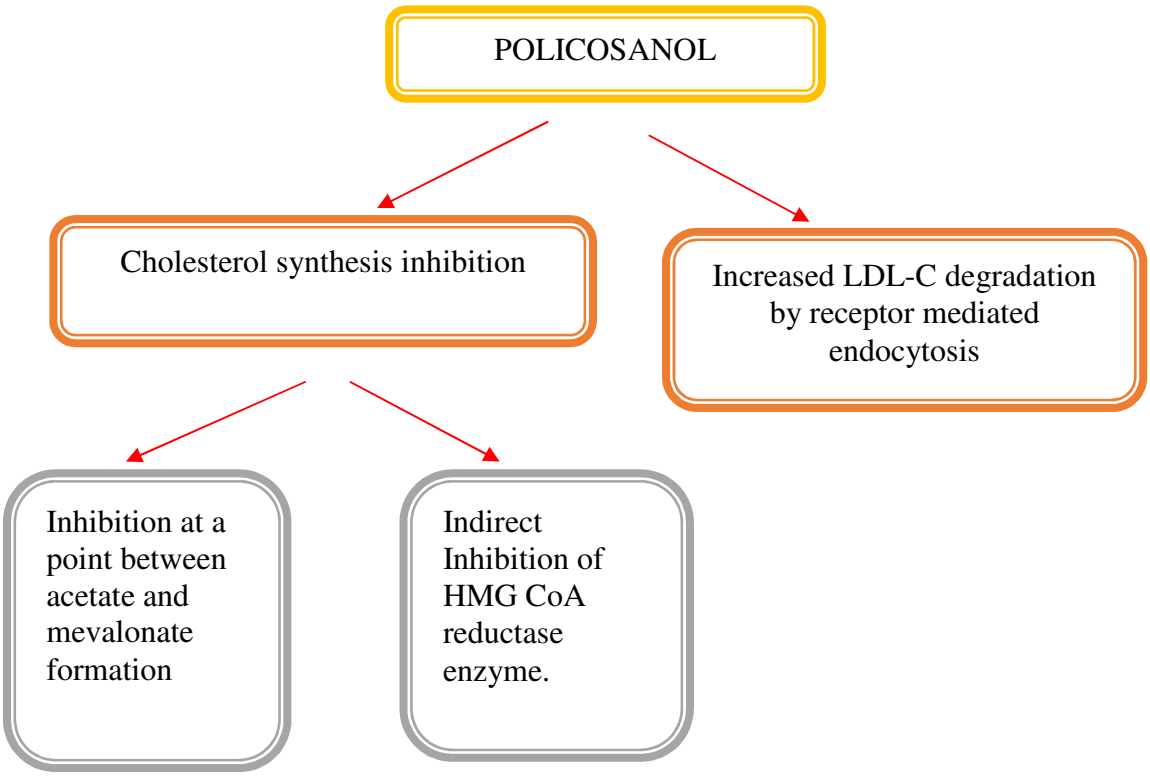
Some studies showed the evidence of antioxidant property in policosanol[61,62].

EFFECT OF POLICOSANOL ON LIPID METABOLISM

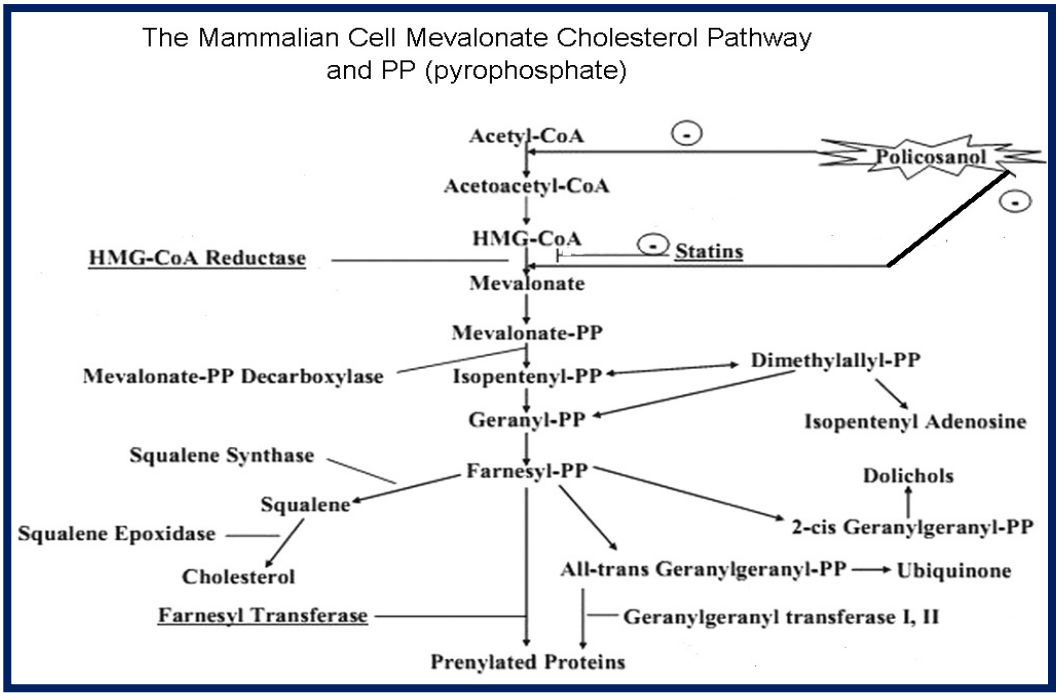
MECHANISM OF ACTION – HYPOLIPIDEMIC ACTION[57]

The mechanism of action for the hypolipidemic effect of policosanol i.e lowering total and LDL-C by

1. Hypocholesterolemic agent - inhibits the cholesterol synthesis at a point between the formation of acetate and mevalonate[6].
2. No direct inhibition of the enzyme HMG CoA reductase[7].
3. It significantly increases receptor mediated endocytosis of LDL-Cholesterol and stimulating its catabolism or degradation in the hepatocytes[8].



- Policosanol can effectively reduce serum cholesterol levels and also the cholesterol content in different tissues like liver, heart and fatty tissues.
- Policosanol has a persistent cholesterol lowering effect which does not change over time.
- Policosanol has an additional anti proliferative action on smooth muscle cells there by decreasing atheroma formation[64,65].



This picture depicts the site of action of policosanol in the cholesterol pathway i.e between acetate and mevalonate thereby inhibiting cholesterol synthesis.

EFFECT OF POLICOSANOL ON PLATELET AGGREGATION

Policosanol alters the prostaglandin synthesis thereby inhibiting platelet aggregation. Policosanol has the ability to lower the proaggregatory thromboxane A2 and increase the anti aggregatory prostaglandin, prostacyclin.

EFFECT OF POLICOSANOL ON INTERMITTENT CLAUDICATION[35,36]

In a study done by Castano et al in 2001, Policosanol has shown to significantly increase the initial claudication distance and the absolute claudication distance. Only very limited studies on the effect of policosanol in intermittent claudication are done.

Due to its antiplatelet action, policosanol is effective in reducing the symptoms of claudication.

EFFECT OF POLICOSANOL IN IMPROVING ANGINA[66]

Policosanol has been shown to improve the exercise ECG testing responses of Coronary heart disease patients with myocardial ischemia .

EFFECT OF POLICOSANOL ON PERFORMANCE ENHANCEMENT

There is only one evidence of octacosanol, the main constituent of policosanol as a performance enhancer which is a small double blinded trial[63].

COMMON USES

- Hyperlipidemia
- Intermittent claudication
- Performance enhancement

OTHER PROPOSED USES

- Antiplatelet action like aspirin
- In parkinsons disease[67]
- In Amyotrophic lateral sclerosis[68].

DRUG FORMULATIONS

It is available as

- Soft gel capsules - These are available in bottles containing 30, 60, 90 capsules per bottle and in a dosage of 5, 10 or 20 mg capsules. In our study we used capsules. Each capsule contains Policosanol (from Sugar Cane) 10 mg.
- Tablets – These are available as strips or in bottles containing 30, 60 or 90 tablets.

DOSAGE[3]

In clinical trials, the dose of policosanol usually tested, range from 5 to 20 mg/day.

SPECIFIC DOSE:

- For hypercholesterolemia - Dose is 5 – 20 mg/day. In this study, we used a dosage of 10 mg policosanol capsules for 8 weeks. Hepatic synthesis of cholesterol occurs in the evening. So policosanol is usually given in the evening once a day.
- For intermittent claudication - Dose is 10-20 mg/day for 3 months continuously.

ADVERSE EFFECTS[3,4]

Literally it does not produce any side effect[22]. Adverse effects occur in only less than 1% of patients taking Policosanol. It is safe upto a dosage of 5-80 mg daily for 3 years. Very rare side effects include Indigestion, Weight loss, Increased appetite, Headache, Insomnia or drowsiness, Skin rash and redness, Excessive urination.

In a pharmacovigilance study[69] conducted on 2252 subjects of age 60 years with coronary, cerebrovascular and peripheral arterial diseases treated with policosanol 5, 10 or 20 mg/day revealed that it does not produce any serious and also mild adverse events indicating its long term tolerability in elderly patients with high vascular risk.

Policosanol is relatively a non toxic drug, safe and well tolerated and proved by animal studies[70-74].

DRUG INTERACTIONS[3]

1. Being an antiplatelet aggregating agent, it will increase the risk of bleeding with aspirin, anticoagulants, heparin, anti-platelet drugs, NSAIDs.
2. It may potentiate the action of Levo dopa[67].
3. Policosanol may potentiate the action of sodium nitroprusside in hypertensive patients[75].
4. No drug interactions were studied between policosanol and antidiabetic and antihypertensive medications.

PRECAUTIONS[3]

1. Persons who are allergic or sensitive to Policosanol.
2. Pregnancy and lactation even though there is no evidence of teratogenicity or any embryonal toxicity.
3. Stop taking policosanol one week prior to any surgical procedure.

Policosanol by acting as a hypolipidemic agent, can prevent the progression of many atherosclerosis induced conditions at an early stage and could improve the quality of life.

Policosanol helps in lowering blood lipid levels and may be beneficial in patients with hyperlipidemia. Since few studies are available on lipid lowering effect of policosanol, this study has been undertaken to assess the hypolipidemic effect in comparison with Atorvastatin.

AIM & OBJECTIVES

AIM

To evaluate the efficacy and tolerability of Policosanol as an add on therapy to Atorvastatin in reducing the lipid levels in patients with hyperlipidaemia.

OBJECTIVES

Primary objective:

1. To assess the reduction in the total cholesterol, LDL and increase in the HDL levels.

Secondary objective:

1. To monitor any adverse effects with these drugs.
2. To assess any change in the triglyceride level.

METHODOLOGY

METHODOLOGY

STUDY DESIGN

This study was a randomized, open label, prospective and a comparative study.

STUDY CENTRE

Institute of Pharmacology, Madras Medical College (MMC) in collaboration with the Department of Internal Medicine, Rajiv Gandhi Government General hospital (RGGGH), Chennai.

STUDY PERIOD

The study was carried out from October 2015 to April 2016.

STUDY DURATION

8 weeks treatment period per patient.

STUDY POPULATION

Known hypertensive patients with Hyperlipidemia (Total cholesterol level 200-300 mg/dl) attending Hypertension OPD, RGGGH/MMC, Chennai.

SAMPLE SIZE

Totally 100 patients. 50 patients in each group (control and study groups)

ELIGIBILITY CRITERIA

INCLUSION CRITERIA:

1. Both genders.

2. Age- 25 – 70 yrs.

3. Hypertensive or type II diabetic patients with total cholesterol level between 200- 300 mg/dl and LDL-C $\geq$ 130 mg/dl.

4. Patients willing to give written informed consent.

EXCLUSION CRITERIA:

1. Pregnant and lactating women.
2. Subjects who have hypothyroidism and with the evidence of clinically significant gastrointestinal, renal, respiratory, hematological, neurological, psychiatric, cardiovascular dysfunctions or uncontrolled type II DM.
3. Triglycerides > 250 mg/dl.
4. Total cholesterol: HDL ratio > 4.5.
5. H/o allergy or intolerance to policosanol.
6. Those with the history of drug intake like thiazides and betablockers etc.
7. Patients unwilling or unable to comply with the study procedures.

STUDY PROCEDURE:

The study was conducted after obtaining approval from the Institutional Ethics Committee. Hypertensive patients with co-existing hyperlipidemia were explained about the details of the study.

An information sheet and informed consent form written in the regional language was provided to each patient and those willing to participate in this study were included.

SCREENING

The patients were screened with the detailed clinical history, physical examination and baseline investigations.

RECRUITMENT

Those who fulfilled the inclusion and exclusion criteria were recruited for the study.

RANDOMIZATION:

The enrolled patients were randomized to either control (Group A) or Study group (Group B) by simple randomization (odd numbers, control and the even numbers, study group).

TREATMENT PLAN

CONTROL GROUP

T. Atorvastatin 10mg/day for 8 weeks.

STUDY GROUP

T. Atorvastatin 10mg/day and Capsule Policosanol 10mg/day for 8 weeks.

The study medication was issued for 2 weeks. After assessing the compliance of the patient at the end of 2 weeks, study medication was issued for the subsequent 2 weeks. The same procedure was repeated till the completion of study.

ADVICE:

The subjects of both the groups are asked to continue their respective antihypertensive and antidiabetic medications and to avoid high fat diet.

INVESTIGATIONS:

BASELINE INVESTIGATIONS:

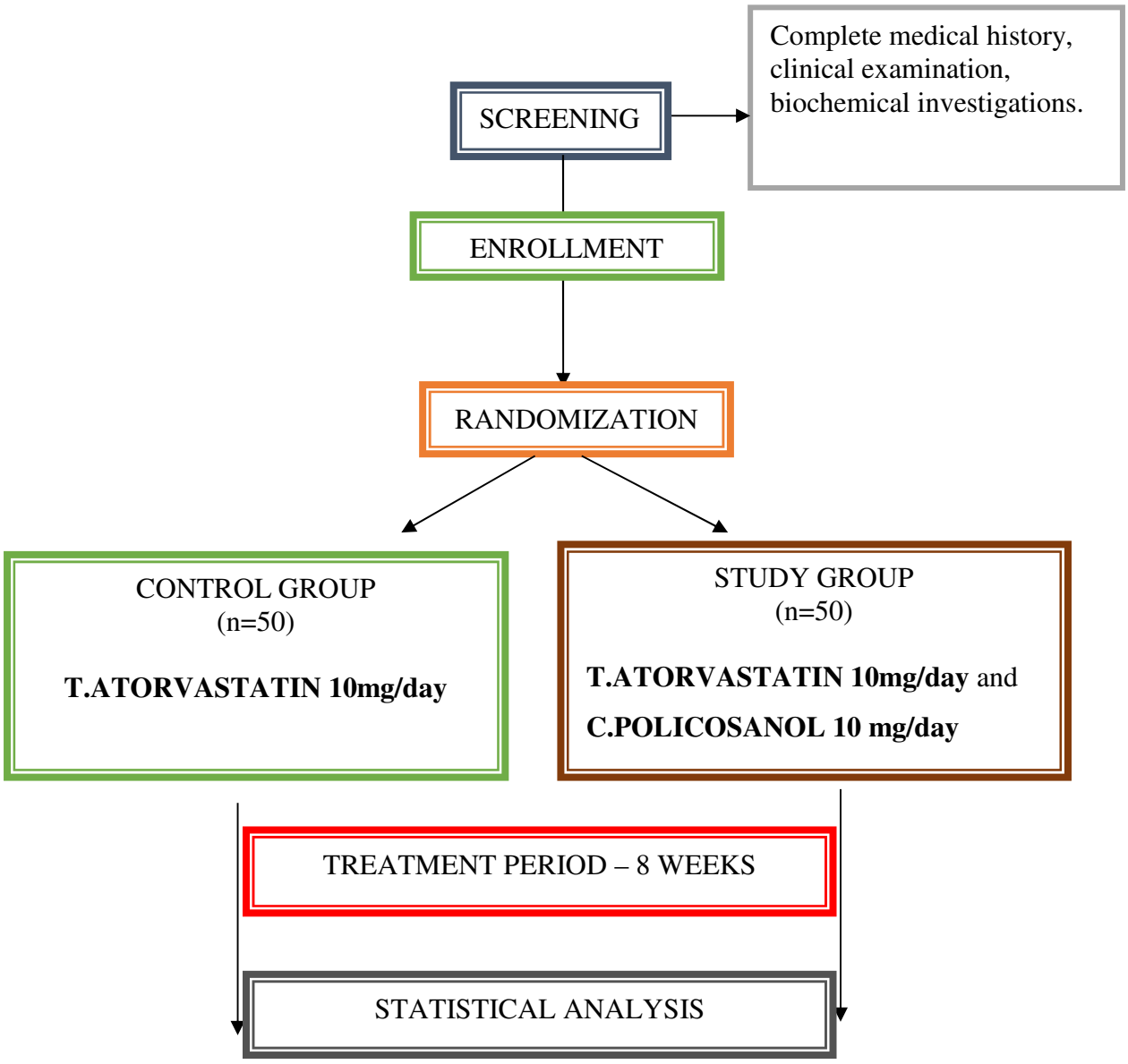
- Blood pressure and Body mass index
- Complete blood count (Hemoglobin, Total count, Differential count, ESR and Platelets)
- Fasting Blood sugar, Blood urea, Serum Creatinine.
- SGOT, SGPT

All the baseline investigations were done at the start of the study and at the end of the 8 weeks.

Fasting Lipid Profile: Serum (Total Cholesterol, LDL, VLDL, HDL & TGL) was done at baseline (0 week) and at 8th week of the study.

- Routine Urine Analysis
- ECG
- X ray Chest PA view

STUDY FLOW CHART



STUDY VISITS

Screening and Baseline

1. Demographic details obtained
2. Complete medical history recorded
3. Vitals recorded and clinical examination performed
4. Enrollment was done
5. Written informed consent obtained
6. Laboratory investigations were done

- Complete blood count
- Lipid profile
- Blood urea, Sugar
- Serum Creatinine
- SGOT, SGPT
- Urine analysis
- ECG
- X ray chest PA view

VISIT 1 (0 WEEKS)

1. Randomization of patient was done.
2. Physical & Clinical examination was done and BMI was calculated.
3. Vitals were recorded.
4. Drugs were issued for Control group and Study group patients.
5. Instructions given to return the empty bottle in the subsequent visit.
6. To report any adverse event if occurs.

VISIT 2 (2 WEEKS)

1. Received Empty bottle.
2. Clinical examination was done and BMI was calculated.
3. Vitals were recorded.
4. Drugs were issued for the subsequent 2 weeks.
5. Instructions were given to return the empty bottle in the subsequent visit.
6. Adverse events if any, were recorded.
7. Patients were advised to report any adverse event.

VISIT 3 (4 WEEKS)

1. Received Empty bottle.
2. Clinical examination was done and BMI was calculated.
3. Vitals were recorded.
4. Drugs issued for subsequent 2 weeks.
5. Instruction to return the empty bottle in the subsequent visit.
6. Adverse events if any, were recorded.
7. Patient advised to report any adverse event.

VISIT 4 (6 WEEKS)

1. Received Empty bottle
2. Clinical examination was done and BMI calculated.
3. Vitals recorded
4. Drugs issued for subsequent 2 weeks
5. Instruction to return the empty bottle in the subsequent visit.

6. Adverse events if any, were recorded.
7. Patient advised to report any adverse event.

VISIT 5 (8 WEEKS)

1. Received Empty bottle.
2. Clinical examination was done and BMI was calculated .
3. Vitals were recorded.
4. Adverse events if any, were recorded.
5. Laboratory investigation were done.

- Complete blood count
- Lipid profile
- Blood urea, Sugar ,Serum Creatinine
- SGOT, SGPT
- Urine analysis

Patients were advised to continue their routine medications at the end of 8 weeks.

INSTRUCTIONS TO PATIENTS

The patients were instructed clearly about the regular intake of the medicines. They were also given proper advice to report for assessment and collection of drugs. They were counseled to report any adverse reactions if occurs.

COMPLIANCE

Patients' compliance was monitored by the empty bottle returned at each visit.

ADVERSE EVENTS:

Any adverse event reported by the patient or observed by the physician during the study was recorded. The onset of adverse event, and its causal relationship to the study drug and the action taken for the adverse effect was recorded. Appropriate medical care was provided for the adverse event.

STATISTICAL ANALYSIS:

The obtained data were analyzed statistically using SPSS software version 21. Distribution of age was analysed using ANOVA.

The biochemical parameters were analyzed statistically in both the groups. The differences within the groups before and after treatment were analyzed using student's paired t-test whereas the difference between the Control and Study group were analyzed using students independent t-test.

p value of < 0.05 is considered as statistically significant.

RESULTS

RESULTS

This study was conducted to evaluate the effect of Policosanol in combination with Atorvastatin in reducing lipid levels.

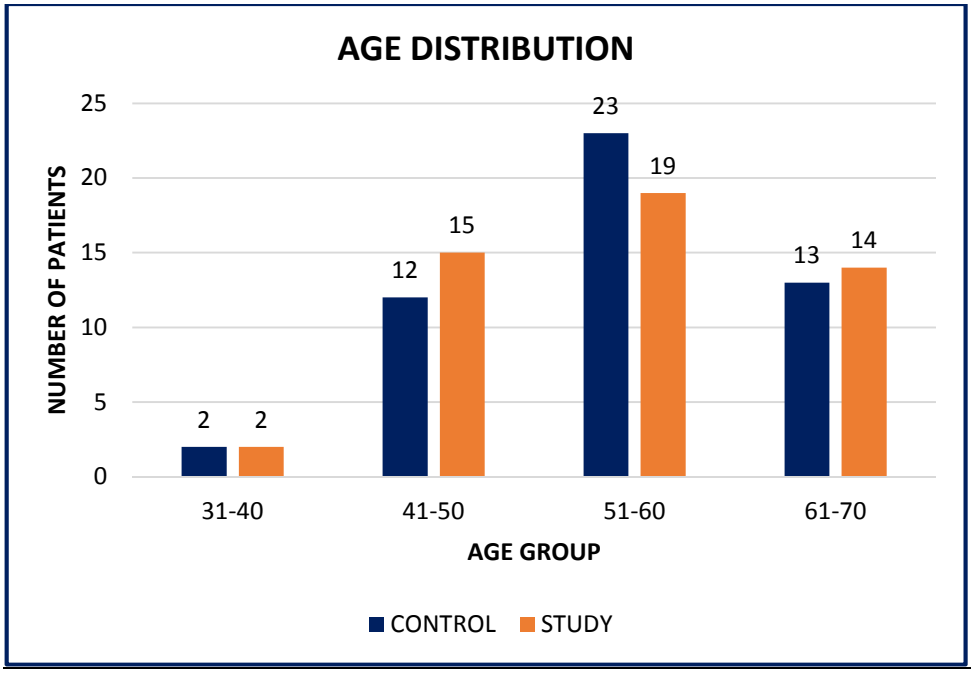
- 182 patients were screened of which 100 patients were enrolled and completed the study.
- There were no drop outs in the study.

TABLE 1: AGE DISTRIBUTION

AGE IN YEARS	CONTROL		STUDY	
	NO	PERCENTAGE	NO	PERCENTAGE
31-40	2	4%	2	4%
41-50	12	24%	15	30%
51-60	23	46%	19	38%
61-70	13	26%	14	28%
TOTAL	50	100%	50	100%

- Table 1 shows the age distribution of both the groups.
- Age group 51-60 years had more number of patients followed by age group 41-50 and 60-70 years.

FIGURE 1: AGE DISTRIBUTION



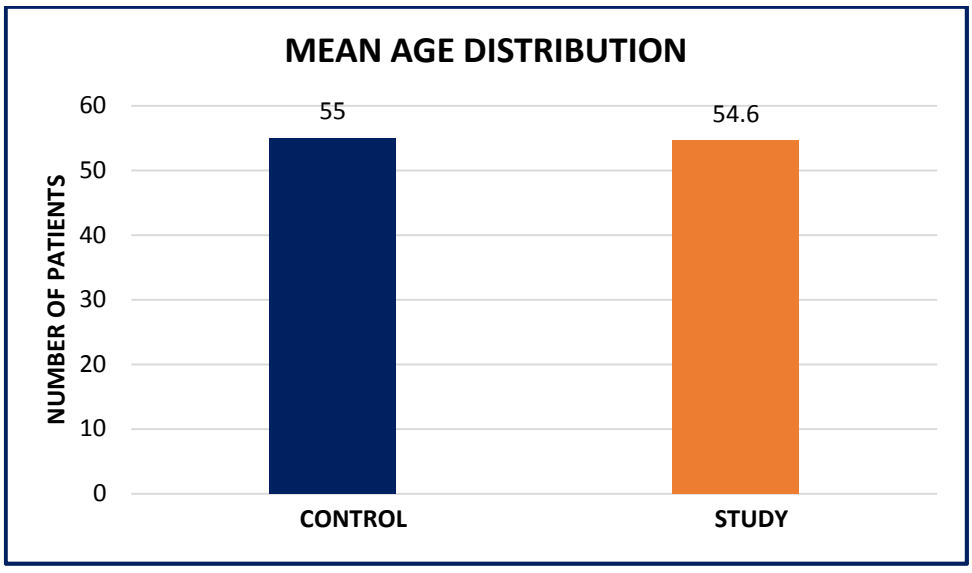
❖ Figure 1 depicts age distribution in both the groups.

TABLE 2: MEAN AGE DISTRIBUTION

GROUP	No of patients	MEAN AGE (in years)	SD
Control	50	55	7.095
Study	50	54.6	8.54
p value		**0.790**	

- Table 2 shows the mean age of both the groups.
- The mean age was similar in both the groups.
- There was no statistically significant difference between the groups.

FIGURE 2: MEAN AGE DISTRIBUTION



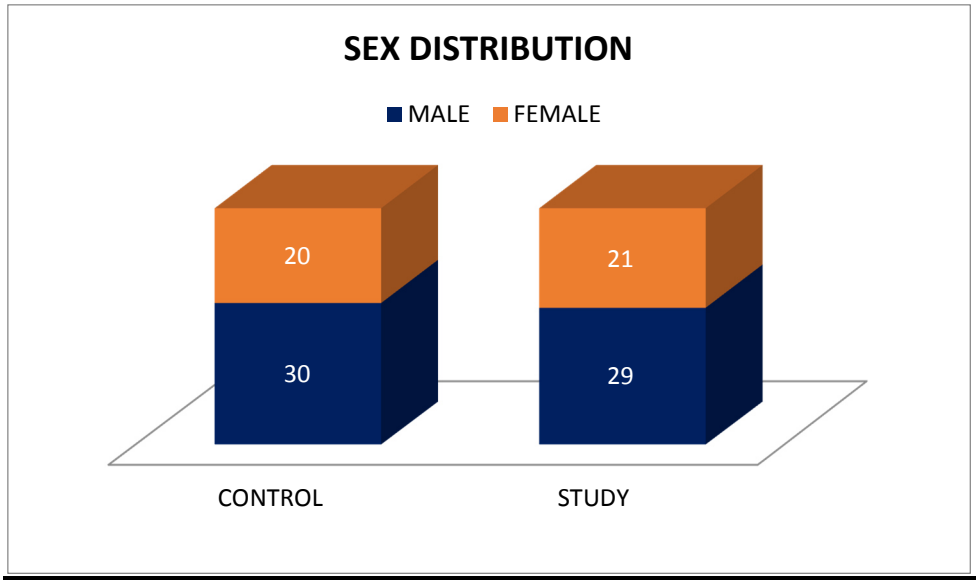
- Figure 2 is the graphical representation of Table 2.

TABLE 3: SEX DISTRIBUTION

SEX DISTRIBUTION	CONTROL		STUDY	
	NO. OF PATIENTS	%	NO. OF PATIENTS	%
MALE	30	60%	29	58%
FEMALE	20	40%	21	42%
TOTAL NO. OF PATIENTS	50	100%	50	100%

- Table 3 shows the sex distribution in both the groups.
- Males were more in number compared to females in both groups.

FIGURE 3: SEX DISTRIBUTION



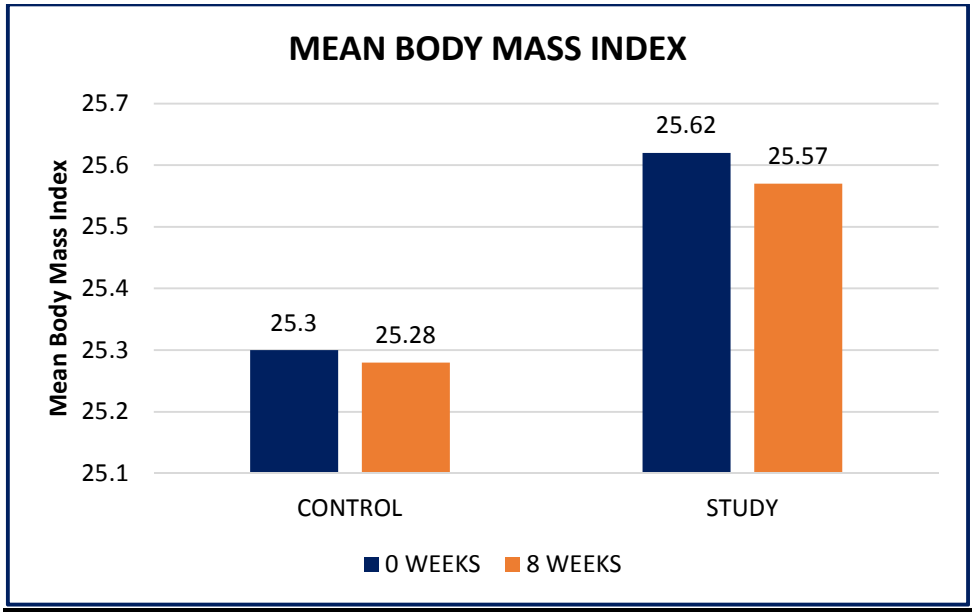
- Figure 3 depicts Table 3.

TABLE 4: BODY MASS INDEX

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	25.30	2.42	25.28	2.40	0.363
STUDY	25.62	3.05	25.57	2.99	0.070
p value	0.565		0.594		

- Table 4 shows mean body mass index in both the groups.
- Statistical analysis within the groups and between the groups did not show any difference in the body mass index at the end of 8 weeks.

FIGURE 4: BODY MASS INDEX



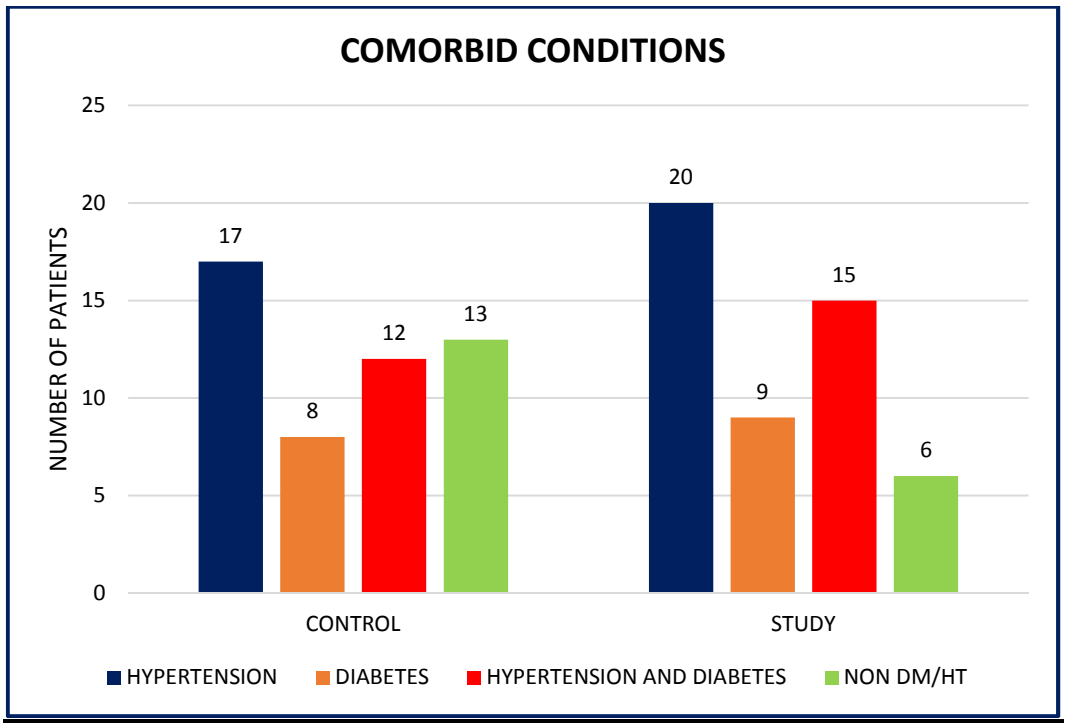
- Figure 4 depicts Table 4.

TABLE 5: COMORBID CONDITIONS

SEX DISTRIBUTION	CONTROL TOTAL=50		STUDY TOTAL=50	
	NO. OF PATIENTS	%	NO. OF PATIENTS	%
Hypertension	17	34%	20	40%
Diabetes	8	16%	9	18%
Hypertension and diabetes	12	24%	15	30%
TOTAL NO. OF PATIENTS	37	74%	44	88%

- Table 5 shows the associated comorbid conditions in patients of both the groups.
- 34% were hypertensives in control group and 40% in the study group.
- 16% were diabetics in the control group and 18% in the study group.
- In the control group, 24% were both hypertensives and diabetics.
- In the study group, 30% were both hypertensives and diabetics.

FIGURE 5: COMORBID CONDITIONS:



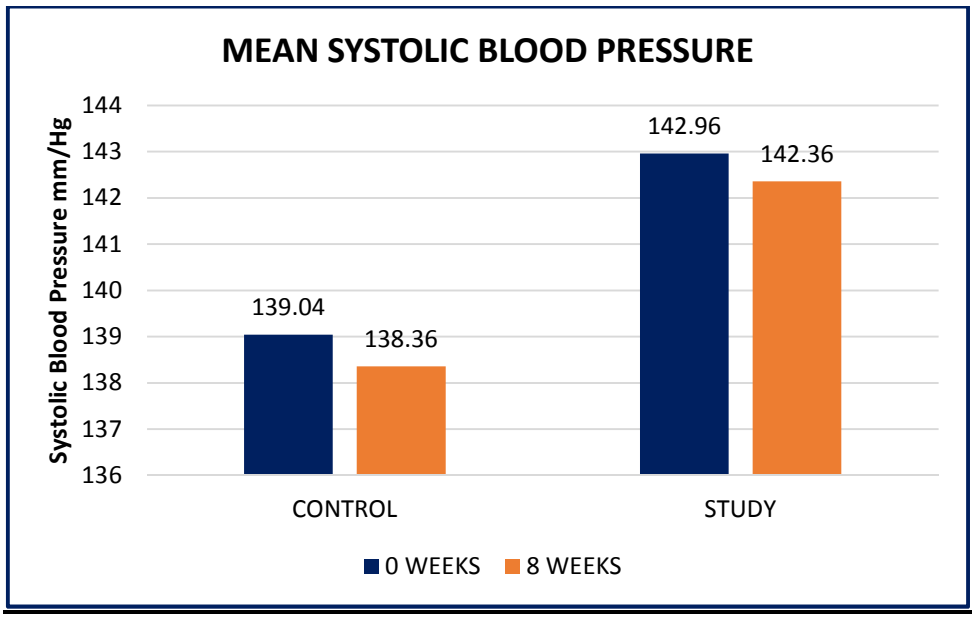
❖ Figure 5 is the diagrammatic representation of associated comorbid conditions in patients of both the groups.

TABLE 6: SYSTOLIC BLOOD PRESSURE

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	139.04	14.46	138.36	13.90	0.078
STUDY	142.96	12.68	142.36	12.61	0.046
p value	0.153		0.135		

- Table 6 shows mean systolic blood pressure in both the groups.
- Statistical analysis within the groups and between the groups did not show any significant difference in the systolic blood pressure at the end of 8 weeks

FIGURE 6: SYSTOLIC BLOOD PRESSURE



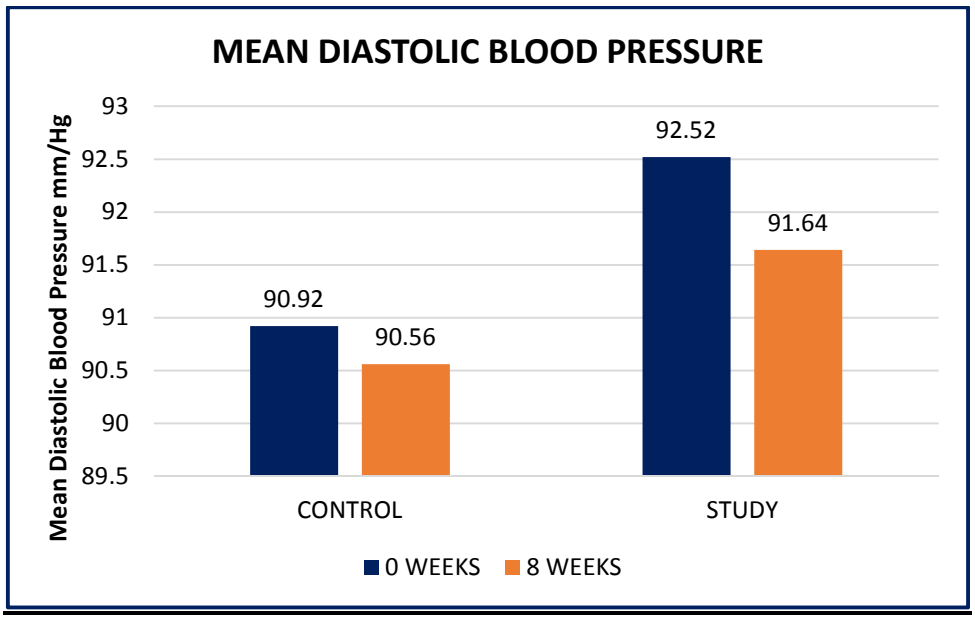
- Figure 6 depicts table 6.

TABLE 7: DIASTOLIC BLOOD PRESSURE

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	90.92	7.39	90.56	7.30	0.151
STUDY	92.52	6.39	91.64	6.96	0.094
p value	0.250		0.451		

- Table 7 shows mean diastolic blood pressure in both the groups.
- No statistically significant difference was observed within the groups and between the groups in the diastolic blood pressure at the end of 8 weeks.

FIGURE 7: DIASTOLIC BLOOD PRESSURE



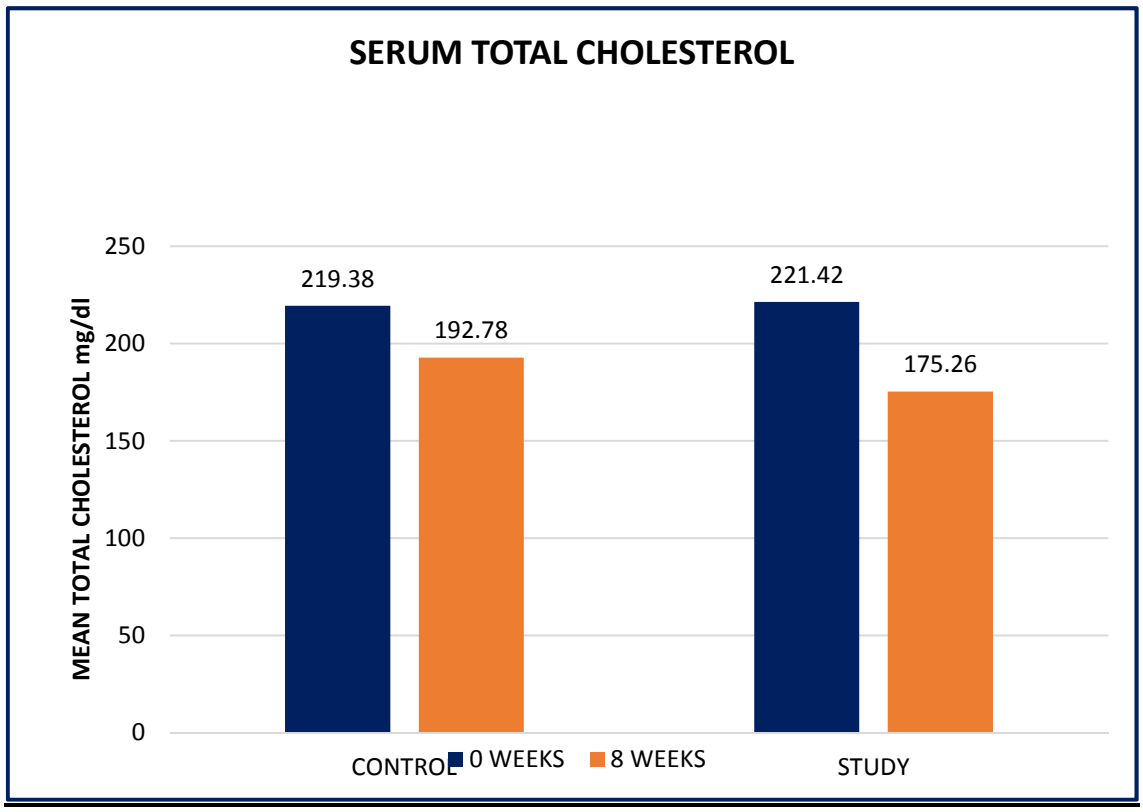
- Figure 7 depicts Table 7.

TABLE 8: SERUM TOTAL CHOLESTEROL

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	219.38	14.19	192.78	11.59	**<0.001**
STUDY	221.42	16.75	175.26	10.56	**<0.001**
p value	0.513		**0.01**		

- Table 8 shows the mean Cholesterol value of both the groups at baseline & end of 8 weeks.
- On comparing with baseline, both groups showed a decrease in mean Total cholesterol.
- Statistical analysis within the groups showed a significant decrease in the Total cholesterol at the end of 8weeks ($p < 0.001$).
- Between the groups, analysis showed a significant difference at the end of 8 weeks ($p = 0.01$).

FIGURE 8: SERUM TOTAL CHOLESTEROL



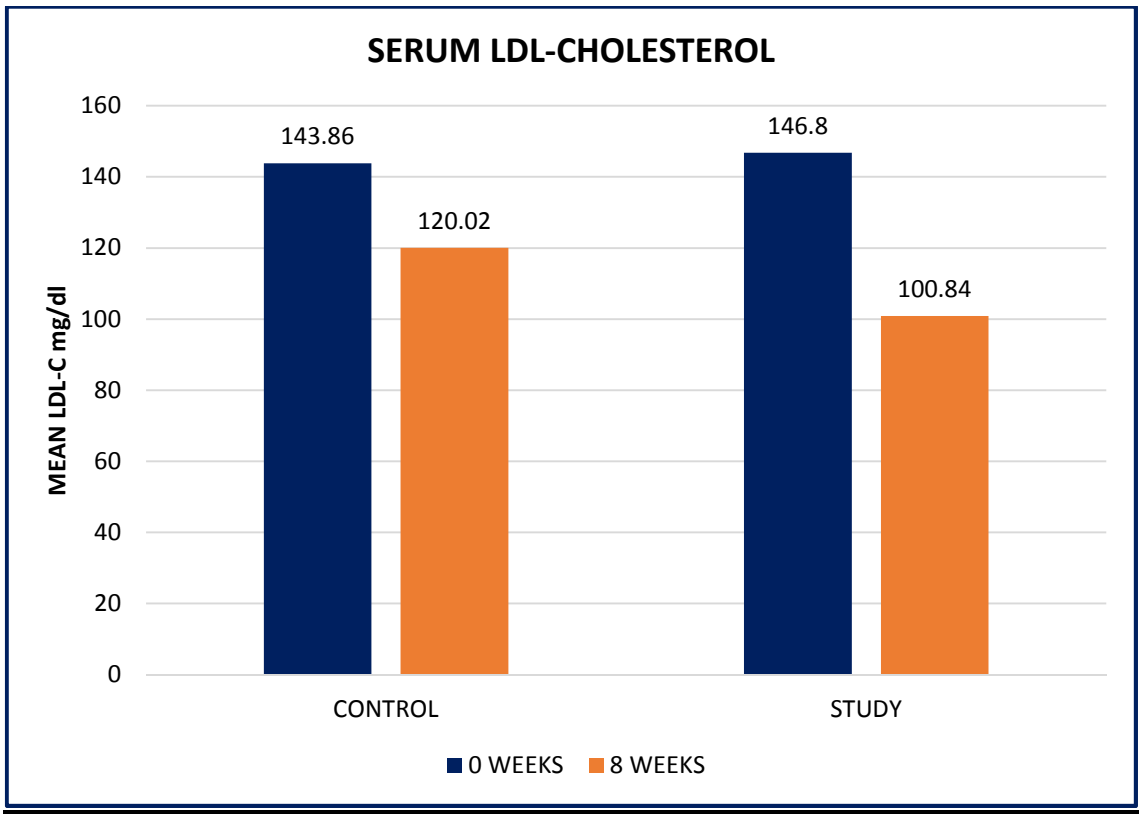
❖ Figure 8 is the graphical representation of Table 8.

TABLE 9: SERUM LDL CHOLESTEROL

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	143.86	14.38	120.02	11.87	**<0.001**
STUDY	146.80	17.18	100.84	10.76	**<0.001**
p value	0.356		**<0.001**		

- Table 9 shows mean serum LDL cholesterol levels of both the groups at baseline & end of 8 weeks.
- There was a significant reduction in the mean LDL cholesterol levels in the study group (100.84mg/dl) compared to the control group (120.02mg/dl) at the end of 8 weeks.
- Statistical analysis within the group showed a significant decrease in both the groups ($p < 0.001$)
- There was a statistically significant difference between the groups at the end of 8 weeks($p < 0.001$) .

FIGURE 9: SERUM LDL CHOLESTEROL



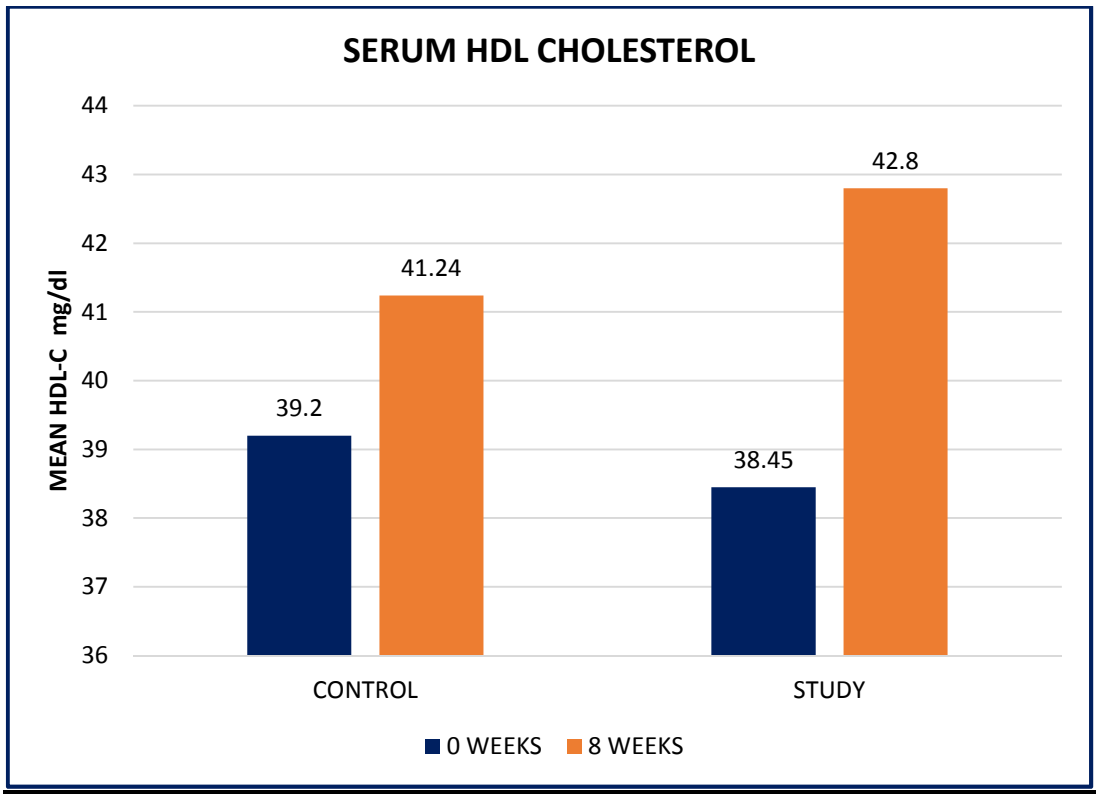
❖ Figure 9 depicts Table 9.

TABLE 10: SERUM HDL CHOLESTEROL

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	39.20	1.76	41.24	2.12	**<0.001**
STUDY	38.45	1.61	42.80	2.04	**<0.001**
p value	0.059		**<0.001**		

- Table 10 shows mean HDL cholesterol levels in both the groups at baseline & end of 8 weeks.
- Statistical analysis within the groups showed a significant decrease in the HDL level at the end of 8 weeks ($p < 0.001$).
- Analysis showed a significant difference between the groups ($p < 0.001$) at the end of 8 weeks.

FIGURE 10: SERUM HDL CHOLESTEROL



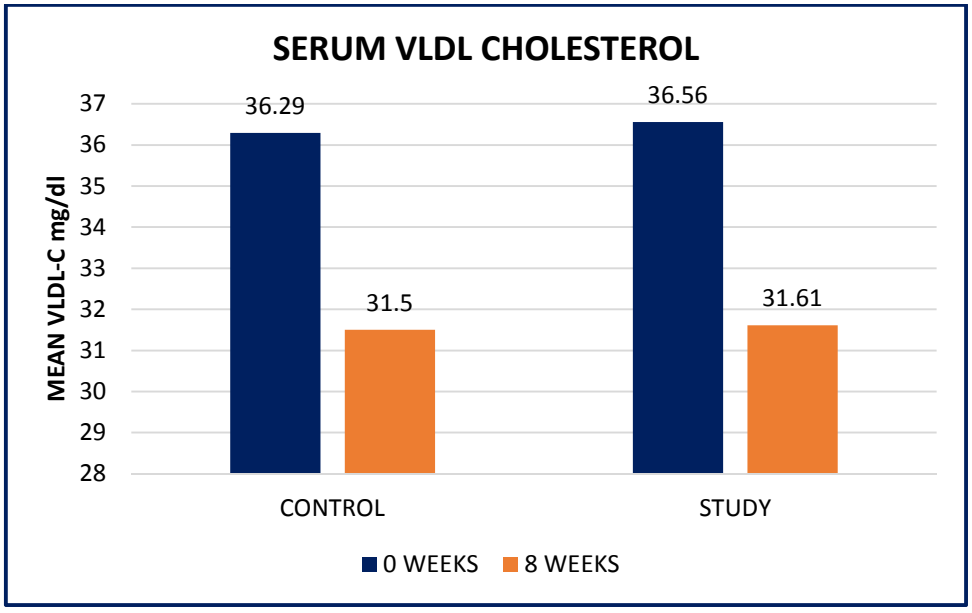
❖ Figure 10 is the diagrammatic representation of Table 10.

TABLE 11: SERUM VLDL CHOLESTEROL

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	36.29	3.19	31.50	1.59	**<0.001**
STUDY	36.56	3.64	31.61	1.69	**<0.001**
p value	0.692		**0.735**		

- Table 11 shows mean VLDL levels in both groups at Baseline and at the end of 8 weeks.
- Statistical analysis within the groups showed a significant decrease in the VLDL level at the end of 8 weeks ($p < 0.001$).
- There was no statistically significant difference between the groups at 8 weeks.

FIGURE 11: SERUM VLDL CHOLESTEROL



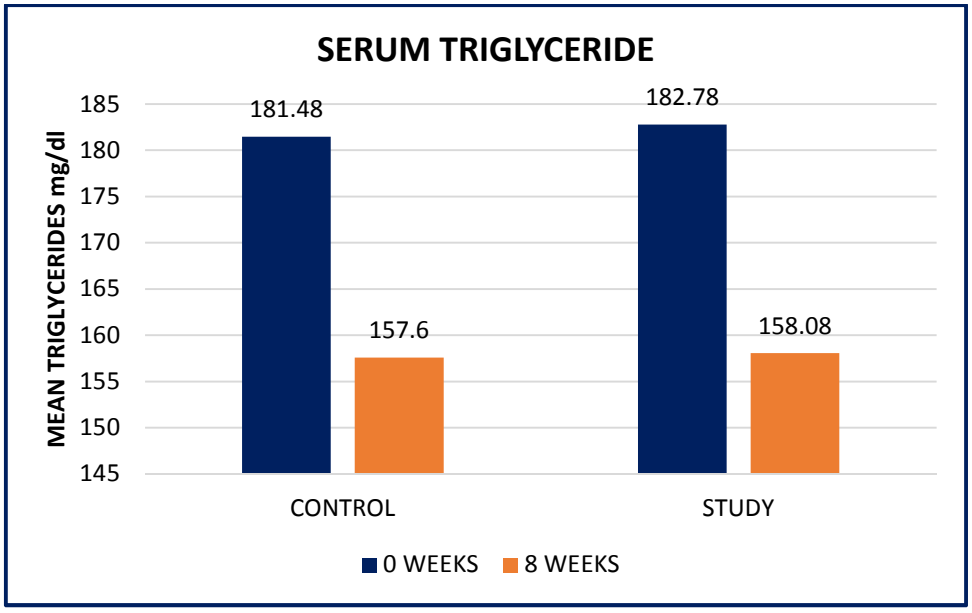
- Figure 11 depicts Table 11.

TABLE 12: SERUM TRIGLYCERIDE LEVELS

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	181.48	15.93	157.6	7.54	**<0.001**
STUDY	182.78	21.4	158.08	8.48	**<0.001**
p value	0.705		**0.771**		

- Table 12 shows mean Triglyceride level in both groups.
- Statistical analysis within the groups showed a significant decrease in the Triglyceride level at the end of 8 weeks (p <0.001).
- Statistical analysis between the groups did not show a significant difference at 8 weeks.

FIGURE 12: SERUM TRIGLYCERIDE



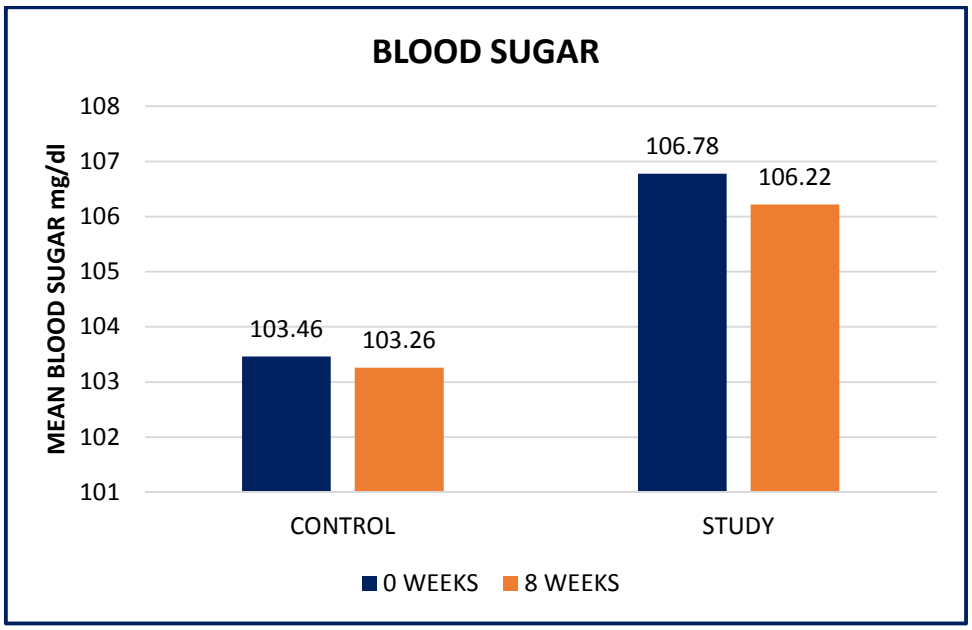
- Figure 12 is the graphical representation of Table 12.

TABLE 13: BLOOD SUGAR

GROUPS	0 WEEKS		8 WEEKS		p value
	MEAN	SD	MEAN	SD	
CONTROL	103.46	18.20654	103.26	18.16794	0.103
STUDY	106.78	18.20663	106.22	18.26594	0.098
p value	0.364		0.419		

- Table 13 shows mean blood sugar level in both the groups.
- No significant difference was observed in the blood sugar level within the group and between the groups in the statistical analysis done at the end of 8 weeks.

FIGURE 13: BLOOD SUGAR



- Figure 13 is the graphical representation of Table 13.

TABLE 14: BIOCHEMICAL INVESTIGATION (CONTROL GROUP)

Investigations	Control group		p value
	0 WEEKS	8 WEEKS	
Blood urea	24.88±3.37	24.46±3.24	0.528
Serum Creatinine	0.65±0.13	0.67±0.14	0.545
SGOT	28.26±4.66	27.62±4.54	0.561
SGPT	30.32±4.97	30.16±4.90	0.90
Total count	8188.8±1411.90	8159±1411.1	0.223
Hemoglobin	11.48±1.00	11.73±1.02	0.303

- Table 14 shows the Biochemical and Hematological parameters of the control group.
- Statistical analysis within the groups and between the groups did not show any significant difference at 8 weeks.

TABLE 15: BIOCHEMICAL INVESTIGATION (STUDY GROUP)

Investigations	Study group		p value
	0 WEEKS	8 WEEKS	
Blood urea	24.86±3.45	25.11±3.48	0.696
Serum Creatinine	0.68±0.14	0.67±0.14	0.744
SGOT	27.92±4.33	28.46±4.37	0.588
SGPT	30.30±4.72	30.04±4.71	0.770
Total count	8309.5±1438.7	8274.7±1407.6	0.514
Hemoglobin	11.68±1.09	11.75±1.24	0.222

- Table 15 shows the Biochemical and Hematological parameters of the study group.
- Statistical analysis within the groups and between the groups did not show any significant difference at 8 weeks.

TABLE 16: INCIDENCE OF ADRs

	CONTROL GROUP	STUDY GROUP
NUMBER OF ADRs	13	10

- Table 16 shows the incidence of ADRs presented by the patients in both the groups.
- In control group, 13 ADRs were reported and in Study group 10 ADRs were reported.

FIGURE 14: INCIDENCE OF ADRs

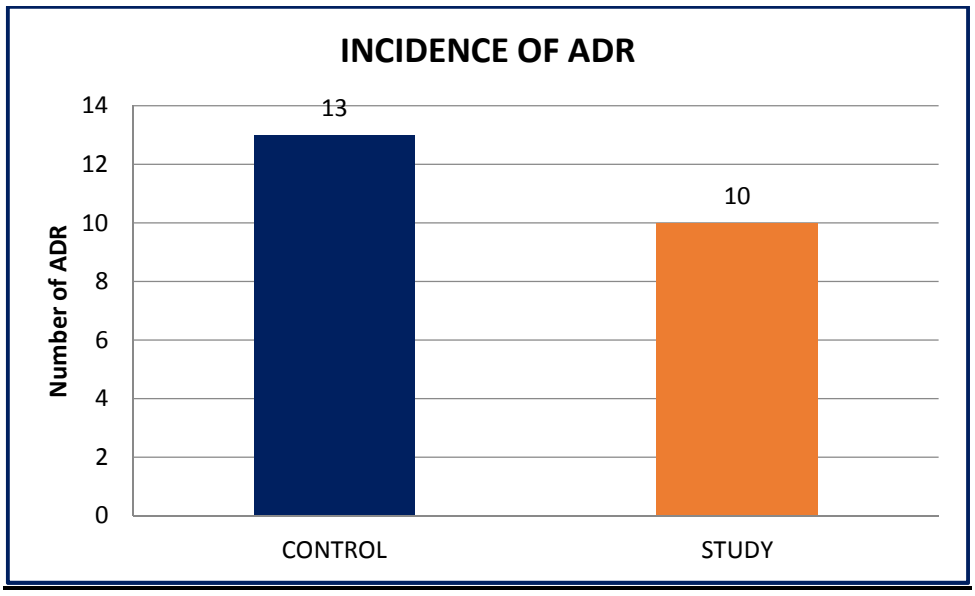


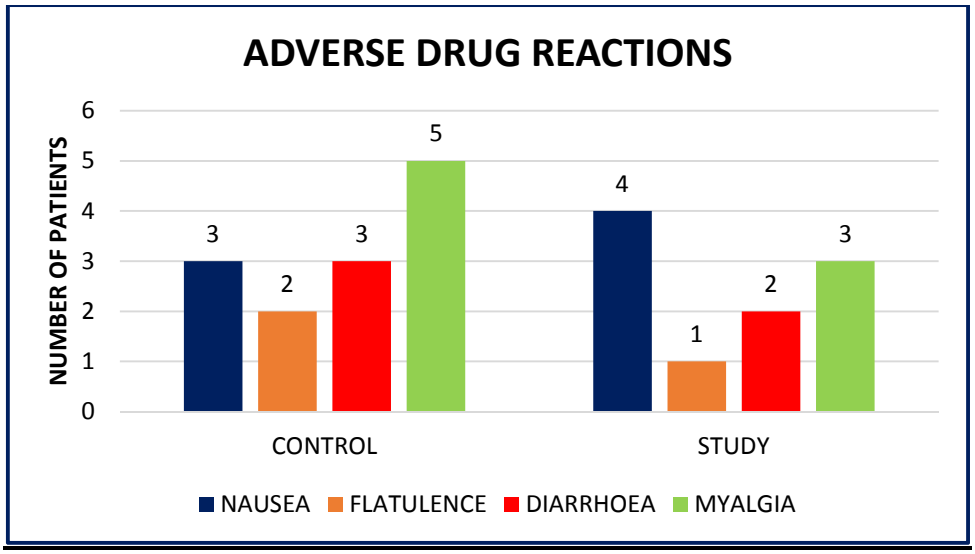
Figure 14 shows the graphical representation of incidence of ADRs.

TABLE 17: ADVERSE DRUG REACTION

ADVERSE DRUG REACTION	CONTROL GROUP	STUDY GROUP
Nausea	3 (6%)	4 (8%)
Flatulence	2 (4%)	1 (2%)
Diarrhoea	3 (6%)	2 (4%)
Myalgia	5 (10%)	3 (6%)

- Table 17 shows the adverse effect profile of both the groups.
- Gastrointestinal disturbances were reported more in both the groups.

FIGURE 15: ADVERSE DRUG REACTION



- Figure 15 is the graphical representation of adverse drug reaction.

TABLE 18: SEVERITY ASSESSMENT OF ADR

SEVERITY	CONTROL GROUP	STUDY GROUP
MILD	**13**	**10**
MODERATE	-	-
SEVERE	-	-

- Table 18 shows severity assessment of Adverse Drug Reactions.
- Severity assessment was done using Modified Hartwig and Siegel scale.
- All the Adverse Drug Reactions in control and study group were mild.

TABLE 19 : CAUSALITY ASSESSMENT OF INDIVIDUAL ADR IN CONTROL GROUP

ADRs	Certain	Probable	Possible	Un-likely	Un-classified	Un-classifiable	Total
Nausea	**-**	**-**	**3**	**-**	**-**	**-**	**3**
Flatulence	**-**	**-**	**2**	**-**	**-**	**-**	**2**
Diarrhoea	**-**	**-**	**3**	**-**	**-**	**-**	**3**
Myalgia	**-**	**-**	**5**	**-**	**-**	**-**	**5**
Total			**13**				**13**

- Table 19 shows causality assessment of individual ADR in control group.
- Causality assessment was done using WHO causality assessment scale
- All adverse drug reactions were categorized as possible.

TABLE 20 : CAUSALITY ASSESSMENT OF INDIVIDUAL ADR IN STUDY GROUP

ADRs	Certain	Probable	Possible	Un-likely	Un-classified	Un-classifiable	Total
Nausea	**-**	**-**	**4**	**-**	**-**	**-**	**4**
Flatulence	**-**	**-**	**1**	**-**	**-**	**-**	**1**
Diarrhoea	**-**	**-**	**2**	**-**	**-**	**-**	**2**
Myalgia	**-**	**-**	**3**	**-**	**-**	**-**	**3**
Total			**10**				**10**

- Table 20 shows causality assessment of individual ADR in study group.
- All ADRs were categorized as possible under WHO causality assessment scale.

DISCUSSION

DISCUSSION

Coronary heart disease remains the most common cause of death among middle aged and elderly people worldwide. Abnormalities in the metabolism of lipid and cholesterol constitute the major predisposing factor to atherosclerosis and in increasing the risk for CHD (Coronary heart disease).

Dyslipidemia which is characterized by increased levels of total cholesterol, LDL-C and triglycerides and a decrease in the HDL-C, is the major modifiable risk factor in the primary and secondary prevention of Coronary Heart Disease and atherosclerosis. Oxidative stress induced by highly reactive oxygen species (ROS) is considered to be an important factor in the pathogenesis of dyslipidemia i.e by oxidizing LDL-C.

As per the NCEP-ATP III guidelines, Statins are the first line and most frequently used drugs in the treatment of dyslipidemia. The dose of atorvastatin can be escalated to decrease the LDL-C by 20-60% if lower doses does not respond. The prolonged use of increased doses of atorvastatin can decrease only 6% of LDL-C levels additionally[40]. Besides the fact that statins are highly effective in controlling cholesterol levels, adverse effects including muscle pain, muscle weakness, and neuropathy are experienced by some patients taking it.

Policosanol is a natural mixture of long chain aliphatic alcohols, suppresses cholesterol synthesis and prevents the development of atherosclerosis.

The mechanism of action for the hypolipidemic effect of policosanol i.e lowering total and LDL-C is by

1. Inhibiting the cholesterol synthesis at a point between the formation of acetate and mevalonate[6].
2. No direct inhibition of the enzyme HMG CoA reductase[7].
3. It significantly increases receptor mediated endocytosis of LDL-Cholesterol and stimulating its catabolism or degradation in the hepatocytes[8].

So policosanol has possibly a potential additive and synergistic effect with atorvastatin since it inhibits the prior step in the cholesterol synthesis pathway. Also there wont be a need for dose escalation of atorvastatin if there is poor response to atorvastatin. Therefore this study was taken up to assess the efficacy of Policosanol in combination with Atorvastatin in patients with Hyperlipidemia without much side effects of Atorvastatin.

The patients for the study were screened by history, clinical examination and laboratory investigations.

Among 182 patients, 100 patients who fulfilled the eligibility criteria were enrolled and randomized into two groups, 50 patients in each group. Control Group received Tab.Atorvastatin and the Study Group received Tab.Atorvastatin plus Cap.Policosanol for 8 weeks duration. Post treatment patients were instructed to continue their respective anti-diabetic and anti-hypertensive medications.

Hypolipidemic effect was assessed by fasting lipid profile and the results were analyzed statistically.

The age and sex distribution did not show any statistically significant difference between the groups. This shows that all the patients belonged to the same population.

The percentage of hypertensives in the control group was 34% and 40% in the study group. They were found to be having coexisting diabetes in 24% in control group and 30% in the study group. The percentage of diabetics in control group was 16% and in the study group was 18%.

The body mass index and blood pressure did not show any significant difference between the groups. This shows that the addition of Policosanol did not affect these parameters.

Within the control group, the Total cholesterol, LDL-C, VLDL-C and Triglycerides decreased significantly ($p < 0.01$). The HDL-C was raised significantly at the end of 8 weeks .

Within the study group also who received Policosanol as an add on therapy with Atorvastatin , the Total cholesterol, LDL-C, VLDL-C and Triglycerides were decreased significantly and HDL-C was raised significantly ($p < 0.01$).

At the end of the study period of 8 weeks, the total cholesterol levels showed a reduction in patients who received Atorvastatin alone (12.12%) and also a significant reduction in patients who received Policosanol in addition to Atorvastatin (20.8%) ($p < 0.001$). On comparing both groups, there was a statistically significant reduction in Total cholesterol levels in patients with Policosanol as add on therapy to atorvastatin (Study Group) ($p < 0.001$).

The study group significantly reduced the LDL cholesterol levels by 31% and control group by 16.5% ($p < 0.01$). Comparing both the groups, the study group showed a statistically significant reduction in LDL levels ($p < 0.01$). Lowering of LDL-C by 30-40% is necessary to reduce the cardiovascular events[76]. In our study policosanol with atorvastatin could reduce the LDL-C levels by 31%

The HDL cholesterol levels showed a significant increase in the Control group (5.2%) and also in the Study group (11.2%) ($p < 0.001$). On comparing both groups, there was a statistically significant increase in HDL cholesterol levels in patients with Policosanol as add on therapy to atorvastatin (Study Group) ($p < 0.001$).

Both the groups showed a reduction in triglyceride and VLDL-C levels but there is no statistical significance between the groups.

This shows that addition of Policosanol contributes to the reduction of both LDL-C and Total cholesterol levels and to the elevation in HDL-C levels. The results of our study were similar to the meta-analysis review published in American Heart Association[4] in the year 2002, where policosanol 10-20 mg/day, lowered plasma total cholesterol levels by 17-21% and reduced LDL-C level by 21-29%. The review also showed a rise in HDL-C by 8-15% and no effect on triglycerides.

Our study results were also similar to the study conducted by Aneiros[77] et al, where policosanol supplementation for a period of 6 weeks reduced plasma LDL-C level by 21.5% and total cholesterol level by 16.2% in 45 patients with type II hypercholesterolemia.

In Castano[78] et al study, a twenty four weeks period of supplementation of policosanol in doses of 5 and 10 mg showed a significant reduction in total cholesterol by 16.2% and LDL by 24.4% and increased HDL by 29% suggesting an important role for policosanol in the prevention of atherosclerosis by its hypolipidemic action.

Comparative trials on type II hypercholesterolemic patients with statins and policosanol by Crespo[79] et al and Castano[80] et al showed statistically significant decrease in LDL and total cholesterol in the patients receiving both policosanol and lovastatin. Further adverse events were more in the lovastatin group compared to policosanol group indicating the safety profile of policosanol compared to statins.

The hematological parameters like complete blood count including hemoglobin, total count, differential count, ESR and platelets did not show any significant difference in both the groups.

There was no significant difference in biochemical parameters like blood sugar, urea, serum creatinine, SGOT, SGPT in both the groups at the end of the study period. This shows that addition of Policosanol did not alter the hematological and biochemical parameters.

The number of adverse events observed were less in patients receiving Policosanol as an add on therapy compared to patients receiving Atorvastatin alone. According to WHO causality assessment scale, all the Adverse Drug Reactions observed were categorized as **possible**. Based on the Modified Hartwig and Siegel severity assessment scale, all the reactions reported were **mild.** This explicits that Policosanol did not increase the occurrence of adverse events.

As evidenced by earlier studies, our study has also modestly observed that addition of Policosanol to Atorvastatin significantly reduced the total cholesterol and LDL cholesterol levels and increased the HDL cholesterol levels. Addition of policosanol to atorvastatin has not produced any significant changes in the hematological and biochemical parameters. Also the observed adverse drug effects were mild and resulted in significant improvement in the patients' quality of life.

The limitations of our study are that it was done for a shorter period and also Policosanol was given in combination with atorvastatin. The effects (therapeutic and adverse events) of policosanol in hyperlipidemia can be evaluated by administering policosanol as a monotherapy. Further studies are needed to be done in larger group of patients for longer duration to prove its effect in hyperlipidemia .

CONCLUSION

CONCLUSION

From our study, we conclude that in patients with hyperlipidemia

1. Policosanol as add on therapy to Atorvastatin is effective in reducing Total cholesterol, LDL-C and in increasing HDL-C levels.
2. Dose escalation of Atorvastatin was not required.
3. Policosanol was well tolerated.

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APPENDICES

APPENDIX - 1

LIST OF ABBREVIATIONS USED

CVD	-	Cardio Vascular Disease
CHD	-	Coronary Heart Disease
LDL-C	-	Low Density Lipoprotein Cholesterol
VLDL-C	-	Very Low Density Lipoprotein Cholesterol
IDL-C	-	Intermediate Density Lipoprotein Cholesterol
HDL-C	-	High Density Lipoprotein Cholesterol
LPL	-	Lipoprotein Lipase
ACAT	-	Acyl CoA Cholesterol Acyltransferase
SREBP	-	Sterol Regulatory Element Binding Protein
CETP	-	Cholesteryl ester transfer protein
LCAT	-	Lecithin Cholesterol Acyl Tranferase
PPAR	-	Peroxisome Proliferator Activated Receptor
AHA	-	American Heart Association
HMG-CoA	-	3-Hydroxyl-3-methylglutarylcoenzyme A
VCAM-1	-	Vascular cell adhesion molecule-1
PUFA	-	Poly unsaturated fatty acid
NCEP-ATP III	-	National Cholesterol Education Programme- Adult Treatment Panel III

APPENDIX – 2

CASE REPORT FORM

A PROSPECTIVE, RANDOMIZED, OPEN LABEL, COMPARATIVE STUDY OF ATORVASTATIN ALONE AND ATORVASTATIN WITH LYCOPENE IN PATIENTS WITH HYPERLIPIDAEMIA ATTENDING TERTIARY CARE HOSPITAL

PATIENT DEMOGRAPHY:

NAME :

AGE/SEX :

PLACE :

OP No :

OCCUPATION :

ADDRESS :

CONTACT NUMBER :

DIAGNOSIS :

S.No	Inclusion criteria	Yes	No	Exclusion criteria	Yes	No
1.	25-70 years			Triglycerides > 250		
2.	Total cholesterol level between 200-300 mg/dl and LDL-C≥ 130 mg/dl			Total cholesterol: HDL ratio > 4.5.		
3.	HT/DM			H/o allergy or intolerance to policosanol		
4.	Patients willing to give written informed consent			Pregnant and lactating women		
5.				Patient with chronic systemic disease		

SUBJECT: INCLUDED ☐ EXCLUDED ☐

RANDOMISATION: CONTROL GROUP ☐ STUDY GROUP ☐

REASON IF EXCLUDED:

SIGNATURE OF PRINCIPAL INVESTIGATOR:

Medical history:

VISIT 1 (DAY 1)

CHIEF COMPLAINTS:

PAST HISTORY:

ALLERGIC TO:

PERSONAL HISTORY:

CLINICAL EXAMINATION:

GENERAL

Pulse rate: BP: Ht: Wt: BMI:

LAB INVESTIGATIONS:

Complete Blood Count:

Hb: TC: DC: P L E M B ESR:

Fasting Blood sugar: mg/dl. Blood urea: mg/dl. Serum Creatinine: mg/dl.

Liver function tests: SGOT: IU/L SGPT: IU/L

Routine urine analysis: sugar albumin deposits

ECG:

Serum lipid profile: Total cholesterol: mg/dl.

LDL: mg/dl. HDL: mg/dl. Triglycerides: mg/dl.

TREATMENT:

VISIT 2 (2nd week)

CLINICAL EXAMINATION:

GENERAL

Pulse rate: BP: Ht: Wt:

BMI:

Adverse events:

VISIT 3 (4th week)

CLINICAL EXAMINATION:

GENERAL

Pulse rate: BP: Ht: Wt:

BMI:

Adverse events:

VISIT 4 (6th week)

CLINICAL EXAMINATION:

GENERAL

Pulse rate: BP: Ht: Wt:

BMI:

Adverse events:

VISIT 5 (8th week)

CLINICAL EXAMINATION:

GENERAL

Pulse rate: BP: Ht: Wt:

BMI:

Serum lipid profile: Total cholesterol: mg/dl.

LDL: mg/dl. HDL: mg/dl. Triglycerides: mg/dl.

Adverse events:

VITAL SIGNS:

VISITS	**VISIT 1 (Day 1)**	**VISIT 2 (2nd week)**	**VISIT 3 (4th week)**	**VISIT 4 (6th week)**	**VISIT 5 (8th week)**
Body Mass Index					
Pulse rate					
BP					
Temperature					
General/systemic examination					

INVESTIGATIONS:

Hematology Hb	**0 WEEKS**	**8 WEEKS**
TC		
DC		
ESR		
Platelet		

Biochemistry Blood sugar		
Blood urea		
Serum creatinine		
SGOT		
SGPT		

Lipid profile:	**0 WEEKS**	**8 WEEKS**
Total cholesterol		
HDL cholesterol		
Triglycerides		
LDL-C=[TC-HDL-VLDL]		
VLDL = TG/5		

ADVERSE EFFECTS:

S.No	**ADVERSE EVENTS**	**START DATE**	**STOP DATE**	**TREATMENT GIVEN**

TRIAL CHECK LIST

Particulars	**Screening & Baseline**	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**
Days		**0**	**2 weeks**	**4 weeks**	**6 weeks**	**8 weeks**
Informed Consent	✓					
Randomization	✓					
Patient Demography	✓					
Physical & Clinical examination	✓	✓	✓	✓	✓	✓
Blood pressure	✓	✓	✓	✓	✓	✓
BMI	✓	✓	✓	✓	✓	✓
TC,DC, ESR, Hb%	✓					✓
Lipid profile	✓					✓
SGOT, SGPT	✓					✓
Blood Sugar	✓					✓
Blood urea	✓					
Serum creatinine	✓					
Urine analysis	✓					✓
ECG	✓					
Dispense Study Medication		✓	✓	✓	✓	
Adverse Drug Event monitoring			✓	✓	✓	✓

APPENDIX – 3

INFORMED CONSENT FORM

Title: "A Prospective, Randomized, Open label, Comparative study of Policosanol as an add on therapy to Atorvastatin in patients with Hyperlipidemia"

Name of the Participant:

I _____ have read the information in this form (or it has been read to me). I was free to ask any questions and they have been answered. I am over 18 years of age and, exercising my free power of choice, hereby give my consent to be included as a participant in this study.

1. I have read and understood this consent form and the information provided to me.
2. I have had the consent document explained to me.
3. I have been explained about the nature of the study.
4. I have been explained about my rights and responsibilities by the investigator.
5. I am aware of the fact that I can opt out of the study at any time without having to give any reason and this will not affect my future treatment in this hospital.
6. I hereby give permission to the investigators to release the information obtained from me as a result of participation in this study to the sponsors, regulatory authorities, Govt. agencies, and IEC.

7.I understand that my identity will be kept confidential if my data are publicly presented.

8. I have had my questions answered to my satisfaction.

9.I have decided to be in the research study.

I am aware that if I have any question during this study, I should contact the investigator. By signing this consent form I attest that the information given in this document has been clearly explained to me and understood by me, I will be given a copy of this consent document.

1.Name and signature / thumb impression of the participant (or legal representative if participant incompetent)

Name _____Signature _____Date_____.

2.Name and signature of impartial witness (required for illiterate patients)

Name _____Signature _____Date_____.

Address and contact number of the impartial witness:

3.Name and signature of the investigator or his representative obtaining consent:

Name _____Signature _____Date_____.

சுய ஒப்புதல் படிவம்

ஆய்வு தலைப்பு

ஹைபர்லிபிடெமியா நோய் சிகிச்சையில் பாலிகோசனாலின் பங்கு வழக்கமான சிகிச்சை முறையுடன் ஓர் திறந்தநிலை ஒப்பிடுதல் ஆய்வு.

பெயர் : தேதி :
வயது : உள் நோயாளி எண் :

..... என்பவராகிய நான் இந்த ஆய்வின் விவரங்களும் அதன் நோக்கங்களும் முறையாக அறிந்து கொண்டேன். எனது சந்தேகங்கள் அனைத்திற்கும் தகுந்த விளக்கம் அளிக்கப்பட்டது. இந்த ஆய்வில் முழு சுதந்திரத்துடன் மற்றும் சுய நினைவுடன் பங்கு கொள்ள சம்மதிக்கிறேன்.

எனக்கு விளக்கப்பட்ட விஷயங்களை நான் நன்கு புரிந்தகொண்டு எனது சம்மதத்தைத் தெரிவிக்கிறேன். இச்சுய ஒப்புதல் படிவத்தைப் பற்றி எனக்கு விளக்கப்பட்டது.

இந்த ஆய்வினை பற்றிய அனைத்து தகவல்களும் எனக்கு தெரிவிக்கப்பட்டது. இந்த ஆய்வில் எனது உரிமை மற்றும் பங்கினைப் பற்றி அறிந்து கொண்டேன்.

இந்த ஆய்வில் பிறரின் நிர்பந்தமின்றி என் சொந்த விருப்பத்தின் பேரில்தான் பங்கு பெறுகிறேன் மற்றும் நான் இந்த ஆராய்ச்சியிலிருந்த எந்நேரமும் பின்வாங்கலாம் என்பதையும் அதனால் எந்த பாதிப்பும் ஏற்படாது என்பதையும் நான் புரிந்து கொண்டேன்.

இந்த ஆய்வில் கலந்து கொள்வதன் மூலம் என்னிடம் பெறப்படும் தகவலை ஆய்வாளர் இன்ஸ்டிட்யூசனல் எத்திக்ஸ் கமிட்டியினரிடமோ, அரசு நிறுவனத்தினடமோ தேவைப்பட்டால் பகிர்ந்து கொள்ளலாம் என சம்மதிக்கிறேன்.

இந்த ஆய்வின் முடிவுகளை வெளியிடும்போதே எனது பெயரோ, அடையாளமோ வெளியிடப்படாது என அறிந்து கொண்டேன். இந்த ஆய்வின் விவரங்களைக் கொண்ட தகவல் தாளைப் பெற்றுக் கொண்டேன். இந்த ஆய்விற்காக இரத்தப் பரிசோதனை செய்து கொள்ள சம்மதிக்கிறேன்.

இந்த ஆய்வில் பங்கேற்கும் பொழுது ஏதேனும் சந்தேகம் ஏற்பட்டால், உடனே ஆய்வாளரை தொடர்பு கொள்ள வேண்டும் என அறிந்து கொண்டேன்.

இச்சுய ஒப்புதல் படிவத்தில் கையெழுத்திடுவதன் மூலம் இதிலுள்ள அனைதது விஷயங்களும் எனக்கு தெளிவாக விளக்கப்பட்டது என்று தெரிவிக்கறேன். இச்சுய ஒப்புதல் படிவத்தின் ஒரு நகல் எனக்கு கொடுக்கப்படும் என்றும் தெரிந்து கொண்டேன்.

ஆய்வாளர் கையொப்பம் பங்கேற்பாளர் கையொப்பம்

நாள் :
இடம் :

APPENDIX – 4

INFORMATION TO PARTICIPANTS

Title: **"A Prospective, Randomized, Open label, Comparative study of Policosanol as an add on therapy to atorvastatin in patients with Hyperlipidaemia**"

Principal Investigator:

Name of Participant:

This study is being conducted in Hypertension OPD at Rajiv Gandhi Govt. General Hospital, Chennai. You are invited to take part in this study. The information in this document is meant to help you decide whether or not to take part. Please feel free to ask if you have any queries or concerns.

What is the purpose of this study?

Hyperlipidemia is a major cause of atherosclerosis and atherosclerosis-induced conditions such as coronary heart disease (CHD), ischemic cerebrovascular disease, and peripheral vascular disease. Atherosclerosis remains the major cause of increase in morbidity or mortality in a majority of middle-aged or older adults and account for about one-third of all deaths of persons in this age range. Policosanol is a dietary supplement made of long chain alcohols extracted from sugar cane wax. It is known to lower the blood cholesterol levels. Thus we want to test the efficacy and safety of treatment with Policosanol in reducing lipid levels.

We have obtained permission from the Institutional Ethics Committee.

The study design:

All patients in the study will be divided into 2 groups A & B. You will be assigned to either of the groups. Group A will receive standard treatment & Group B will receive standard treatment + Policosanol.

Study Procedures:

The study involves evaluation of decrease in lipid levels. The planned scheduled visits involve visits at 2^{nd}, 4^{th}, 6^{th}, 8^{th} week after your initial visit. You will be required to visit the hospital 5 times during the study. At each visit, the study physician will examine you. Blood tests will be carried out thrice during the study (at screening and at the end of study) and total of about 40 ml blood will be collected. These tests are essential to monitor your condition, and to assess the safety and efficacy of the treatment given to you.

In addition, if you notice any adverse events, you have to report it. You will be required to return unused study medicines when you report for your scheduled visits. This will enable correct assessment of the study results.

Possible benefits to you – Policosanol along with standard treatment will cause reduction in lipid levels.

Possible benefits to other people - The results of the research may provide benefits to the society in terms of advancement of medical knowledge and/or therapeutic benefit to future patients.

Confidentiality of the information obtained from you

You have the right to confidentiality regarding the privacy of your medical information (personal details, results of physical examinations, investigations, and your medical history). By signing this document, you will be allowing the research team investigators, other study personnel, and Institutional Ethics Committee to view your data, if required. The information from this study, if published in scientific journals or presented at scientific meetings, will not reveal your identity.

How will your decision to not participate in the study affect you?

Your decision not to participate in this research study will not affect your medical care or your relationship with the investigator or the institution. You will be taken care of and you will not lose any benefits to which you are entitled.

Can you decide to stop participating in the study once you start?

The participation in this research is purely voluntary and you have the right to withdraw from this study at any time during the course of the study without giving any reasons. However, it is advisable that you talk to the research team prior to stopping the treatment/discontinuing of procedures etc.

The expenditure for the treatment and investigation for this study will not be collected from you.

Signature of Investigator Signature of Participant

Date Date

ஆய்வு தகவல் தாள்

ஆய்வு தலைப்பு : ஹைபர்லிபிடெமியா நோய் சிகிச்சையில் பாலிகோசனாலின் பங்கு வழக்கமான சிகிச்சை முறையுடன் ஓர் திறந்தநிலை ஒப்பிடுதல் ஆய்வு.

ஆய்வாளர் :

பங்கேற்பாளர் :

இந்த ஆய்வு சென்னை இராஜீவ்காந்தி அரசு பொது மருத்துவமனையில் நடைபெற உள்ளது. நீங்களும் இந்த ஆய்வில் பங்கேற்க நாங்கள் விரும்புகிறோம். இதிலுள்ள தகவலின் அடிப்படையில் இந்த ஆய்வில் பங்கேற்பதா அல்லது வேண்டாமா என்று நீங்களே முடிவு செய்து கொள்ளலாம். உங்களது சந்தேகங்களை எங்களிடம் கேட்டு நிவர்த்தி செய்து கொள்ளலாம்.

இந்த ஆய்வின் நோக்கம் :

கரோனரி இதய நோய், இஸ்கிமிக் செரிபரோவாஸ்குலர் நோய் மற்றும் வாஸ்குலர் நோய் போன்ற நோய்களுக்கு ஹைபர்லிமிடெமியா தான் முக்கிய காரணியாக உள்ளது. ஹைபர்லிமிடெமியா இவை அனைத்து நோய்களையும் இரத்தக் குழாய்களில் தடிப்பு ஏற்படுத்துவதால் தான் உண்டாகின்றன. நடுத்தர மற்றும் முதியவர்களே பெரும்பான்மையாக இந்நோய்களால் பாதிக்கப்படுகின்றனர்.

பாலிகோசனால் என்பது ஒர இயற்கை மருந்து. இது கரும்பு, மெழுகிலிருந்து தயாரிக்கப்படுகிறது. பாலிகோசனாலுக்கு இரத்தத்திலுள்ள கொழுப்பினை கட்டுப்படுத்தும் தன்மை உள்ளது. இந்நோயின் கிசிக்சையில் பாலிகோசாலின் திறனை அறிவதே இந்த ஆய்வின் நோக்கம் ஆகும்.

இந்த ஆய்விற்கு இன்ஸ்டிட்யூசனல் எத்திக்ஸ் கமிட்டி சம்மதம் பெற்றிருக்கிறோம்.

இந்த ஆய்வில் கலந்து கொள்பவர்கள் A மற்றும் B என்று இரு குழுக்களாகப் பிரிக்கப்படுவர். A குழுவில் இருப்பவர்கள் வழக்கமான சிகிச்சையும், B குழுவில் இருப்பவர்கள் வழக்கமான சிக்சையுடன் பாலிகோசனால் மருந்தும் பெறுவர்.

இந்த ஆய்வில் நீங்கள் முதல்வாரத்தில் 2, 4, 6 மற்றும் 8வது வாரங்களில் பரிசோதிக்கப்படுவீர்கள். நோயின் தன்மையில் ஏற்படும் முன்னேற்றத்தினை அறிந்து கொள்வோம். இரண்டு முறை இரத்தப் பரிசோதனை செய்யப்படும். அதற்காக எடுக்கப்படும் இரத்தத்தின் மொத்த அளவு 40 மி.லி. மட்டுமே. இந்த ஆய்வினில் ஏதேனும் பக்கவிளைவுகள் ஏற்பட்டால் உடனடியாக எங்களிடம் தெரிவிக்க வேண்டும்.

இந்த ஆய்வில் கலந்த கொள்வதன் மூலம் நீங்கள் நோயின் தன்மையில் முன்னேற்றம் பெறலாம். மேலும் வருங்காலத்தில் பிற நோயாளிகளும் பயன்பெற இந்த ஆய்வு உதவியாக அமையும்.

முடிவுகளை அல்லது கருத்துக்களை வெளியிடும் போதோ அல்லது ஆய்வின் போதோ தங்களது பெயரையோ அல்லது அடையாளங்களையோ வெளியிடமாட்டோம் என்பதையும் தெரிவித்துக் கொள்கிறோம். இந்த தகவல்தாளில் கையெழுத்திடுவதின் மூலம் உங்களைப்பற்றிய குறிப்புக்களையோ எடுத்துக்கொண்ட சிகிச்சை முறையை பற்றியோ, ஆய்வாளரோ, அல்லது இன்ஸ்ட்டியூசனல் எத்திக்ஸ் கமிட்யைச் சார்ந்தவர்களோ தேவை ஏற்பட்டால் அறிந்து கொள்ளலாம் என்று சம்மதிக்கிறீர்கள்.

இந்த ஆய்வில் பங்கேற்காவிட்டாலும், நீங்கள் வழக்கமான சிகிச்சையை தொடர்ந்து பெறலாம்.

இந்த ஆய்வில் பங்கேற்பது தங்களுடைய விருப்பத்தின் பேரில்தான் இருக்கிறது. மேலும் நீங்கள் எந்நேரமும் இந்த ஆய்விலிருந்து பின்வாங்கலாம் என்பதையும் தெரிவித்துக் கொள்கிறோம்.

இந்த சிறப்பு சிகிச்சையின் முடிவுகளை ஆய்வின் போதோ அல்லது ஆய்வின் முடிவின் போதோ தங்களுக்கு அறிவிப்போம் என்பதையும் தெரிவித்துக் கொள்கிறோம்.

இந்த ஆய்வில் தாங்கள் கலந்து கொள்வதால் சிகிச்சைக்காகவோ, இரத்த பரிசோதனைகளுக்காகவோ தங்களிடமிருந்து எந்த கட்டணமும் வசூலிக்கப்படமாட்டாது.

ஆய்வாளர் கையொப்பம் **பங்கேற்பாளர் கையொப்பம்**

நாள் :
இடம் :

INSTITUTIONAL ETHICS COMMITTEE
MADRAS MEDICAL COLLEGE, CHENNAI-3

EC Reg No.ECR/270/Inst./TN/2013
Telephone No. 044 25305301
Fax : 044 25363970

CERTIFICATE OF APPROVAL

To
Dr. A. Suba Shree
Postgraduate in MD Pharmacology
Madras Medical College
Chennai 600 003

Dear Dr. A. Suba Shree,

The Institutional Ethics Committee has considered your request and approved your study titled **"A PROSPECTIVE, RANDOMIZED, OPEN LABEL, COMPARATIVE STUDY OF POLICOSANOL AS AN ADD ON THERAPY TO ATORVASTATIN IN PATIENTS WITH HYPERLIPIDEMIA" No. 18102015.**

The following members of Ethics Committee were present in the meeting held on 06.10.2015 conducted at Madras Medical College, Chennai-3.

1. Prof.C.Rajendran, M.D., : Chairperson
2. Prof.R.Vimala, M.D., Dean, MMC, Ch-3 :Deputy Chairperson
3. Prof.Sudha Seshayyan, M.D., Vice-Principal MMC, Ch-3 : Member Secretary
4. Prof.B.Vasanthi, M.D., Professor Pharmacology, MMC : Member
5. Prof.P.Ragumani, M.S., Professor, Inst.of Surgery, MMC : Member
6. Prof.Md.Ali, M.D., D.M., Prof. & HOD of Medl.G.E., MMC : Member
7. Prof.Baby Vasumathi, Director, Inst.of O&G, Ch-8 : Member
8. Prof.K.Ramadevi, Director, Inst.of Biochemistry, MMC : Member
9. Prof.Saraswathy, M.D., Director, Inst. Of Pathology, MMC: Member
10. Prof.Srinivasagalu, Director, Inst.of Inter Med. MMC : Member
11. Tmt. Rajalakshmi, Jr. Administrative Officer : Lay Person
12. Thiru S.Govindasamy, B.A., B.L., : Lawyer
13. Tmt.Arnold Saulina, M.A., MSW., : Social Scientist

We approve the proposal to be conducted in its presented form.

The Institutional Ethics Committee expects to be informed about the progress of the study and SAE occurring in the course of the study, any changes in the protocol and patients information/informed consent and asks to be provided a copy of the final report.

Member Secretary, Ethics Committee

MEMBER SECRETARY
INSTITUTIONAL ETHICS COMI
MADRAS MEDICAL COLLE
CHENNAI-600 003

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MADRAS MEDICAL COLLEGE
CHENNAI-3