Cardiovascular diseases (CVD) are important components of chronic diseases worldwide. CVD refer to the class of diseases that involve the heart and/or blood vessels. CVD remain the principal cause of death in both developed and developing countries accounting for roughly 20% of all deaths worldwide per year (Rajadurai and Prince, 2007). CVD incur a greater economical constraint than any other illness especially in the developing countries (Rekha et al., 2008). At present, the developing countries contribute a greater share to the global burden of CVD than the developed countries (Lopez, 1993; Whelton et al., 1995). According to World Health Organization (WHO) (2007) the impact of CVD worldwide is as follows: an estimated 17 million people die every year from CVD and there is one death every two seconds, one heart attack every five seconds and one stroke every six seconds. There were 58 million deaths in the year 2005 of which chronic diseases caused 35 million deaths and CVD caused 17.5 million deaths (Gupta, 2008). By 2020 it is predicted that the global deaths due to CVD reach approximately 25 million (Murray and Lopez, 2001). WHO estimates that by the year 2010, 60% of the world’s cardiac patients will be in India (Gaziano et al., 2006). Figure 1 depicts Global burden from CVD versus Disability Adjusted Life Years (DALY) for the period 1990-2020.

Pathophysiology of CVD

The cardiovascular system consists of three anatomical components: the autonomic nervous system, the heart and the vasculature. These three components interact in a complex manner to control blood flow to organs throughout the body. The autonomic nervous system controls a variety of bodily functions including blood pressure and heart rate. The heart is responsible for distributing blood to various tissues of the body (Lilly, 2003). The heart is a powerful muscle that pumps blood throughout the body by means of a coordinated contraction. The contraction is generated by an electrical activation which is spread by a wave of bioelectricity that propagates in a coordinated manner throughout the heart. The heart is a hollow, muscular organ made of four chambers with multiple arteries (arteries are the blood...
Fig 1. Global burden from CVD during the period 1990-2020.

Fig 2. CVD-Chain of events.
vessels that carry oxygenated blood) and veins (veins are the blood vessels that carry unoxygenated blood) connected to the heart. The top of the heart is called the base. The pointed tip of the heart is called the apex. The right, top chamber of the heart is called the right atrium. The right, lower chamber is called the right ventricle. The left top chamber of the heart is called the left atrium, and the left bottom chamber is called the left ventricle. The two sides of the heart are separated by the thick walled septum. In between the right atrium and right ventricle is the tricuspid valve. Between the left atrium and the left ventricle is the bicuspid valve which is also called the mitral valve (Jain, 2008; Tortora and Derrickson, 2006).

Unoxygenated blood from the upper and lower body enters into the heart through the inferior vena cava and the superior vena cava and enters into the right atrium. Once it enters into the right atrium it is pumped into the right ventricle and the tricuspid valve (also known as the right atrioventricular valve). Once the blood is in the right ventricle it is forcefully pumped out into the pulmonary artery through the pulmonary valve. The pulmonary valve prevents the blood from back flowing into the right ventricle. The blood moves through the pulmonary artery into the right and left lungs where it is oxygenated. The oxygenated blood returns to the heart through the right pulmonary veins and left pulmonary veins. The blood enters into the left atrium and then is pumped into the left ventricle through the mitral valve (bicuspid valve). This valve prevents blood from back flowing into the left atrium when the left ventricle contracts to push the blood through the aortic valve into the aorta. Once blood is in the aorta it travels from here to the rest of the body’s cells via arteries and eventually capillaries (Sembulingam and Sembulingam, 2006; Tortora and Derrickson, 2006).

There are many types of CVDs (Fig 3) which include arteriosclerosis, coronary insufficiency (atherosclerosis, myocardial infarction (MI), angina pectoris), heart valve disease, arrhythmia, heart failure, hypertension, orthostatic hypo tension, shock, endocarditis, diseases of the aorta and its branches, disorders of the peripheral vascular system (stroke, pulmonary embolism, diabetes mellitus (DM), thrombosis), and congenital heart disease. These conditions have similar causes, mechanisms, and treatment (Fig 2). The actively contracting heart muscle needs a steady supply of
Coronary heart disease
Disease of the blood vessels supplying the heart muscle
Risk factors: High blood pressure, high cholesterol, tobacco use, unhealthy diet, physical inactivity, diabetes, and advancing age.

Rheumatic heart disease
Damage to the heart muscle and heart valves from rheumatic fever, caused by streptococcal bacteria.

Congenital heart disease
Malformations of heart structure at birth, which may be caused by genetic factors or by adverse exposures during gestation. Examples are holes in the heart, abnormal valves, and abnormal heart chambers.

Other cardiovascular diseases
Tumours of the heart, vascular tumours of the brain, disorders of heart muscle, heart valve diseases, and disorders of the lining of the heart.

Other factors that can damage the heart and blood vessel system
Inflammation, drugs, high blood pressure, unhealthy diet, trauma, toxins, and alcohol.

Stroke
Strokes are caused by disruption of the blood supply to the brain. This may result from either blockage (ischaemic stroke) or rupture of a blood vessel (thrombotic stroke).

Risk factors: High blood pressure, atrial fibrillation (a heart rhythm disorder), high blood cholesterol, tobacco use, unhealthy diet, physical inactivity, diabetes, and advancing age.

Aortic aneurysm and dissection
Dilation and rupture of the aorta.

Peripheral arterial disease
Disease of the arteries supplying the arms and legs.

Deep venous thrombosis (DVT) and pulmonary embolism
Blood clot in the leg veins, which can dislodge and move to the heart and lungs.

Miriam syndrome
Congenital heart disorders, syphilis, and other infections and inflammatory disorders.

Fig 3. Different types of CVDs-Definitions.
oxygen and nutrients to function which are delivered by blood vessels known as coronary arteries. Their inner lining is normally smooth, and slowly become clogged with clumps of fats, cholesterol and other material called atherosclerotic plaques. Rupture of a plaque and the formation of a blood clot around the ruptured plaque occur suddenly. As a result, the supply of blood with its oxygen and nutrients going to the heart muscle is checked off (Ravi Kanth et al., 2008).

Some of the cardiovascular diseases can be defined as:

- **Angina pectoris:** This is often accompanied by symptoms of crushing, diffuse pain in the chest (directly over the heart), a shortness of breath that leads to gasping, weakness, anxiety, light-headedness, nausea and sweating. The coronary vessels narrow temporarily causing the heart muscle to suffer ischemia (lack of oxygen).

- **Hypertension:** Hypertension means that the pressure of blood exerts against the walls of the blood vessels, which is dangerously high. This excess pressure weakens artery walls and decreases their elasticity forcing the heart to pump harder. Arteries can also break due to this excess force and can cause hemorrhage (excessive bleeding).

- **Peripheral vascular disease:** It refers to a range of disorders that affect the blood vessels in the legs, feet, arms, or hands. The varieties of PVD are blood clots, atherosclerosis, and various veins.

- **Heart failure:** Heart failure, often called congestive heart failure, is a condition in which the heart can’t pump enough blood to body’s organs and tissues. It doesn’t mean heart has failed and can’t pump blood at all.

- **Atherosclerosis:** It refers to the formation and hardening of fatty plaques (atheromas) on the inner surface of the arteries (Fig 4).

Atherosclerosis develops from low density lipoprotein (LDL) becoming oxidized by free radicals, particularly oxygen free radicals (ROS) (Brown and Goldstein, 1990). Blood in arteries contains plenty of oxygen and is where atherosclerosis develops. Blood in veins contains little oxygen where atherosclerosis rarely develops. When oxidized LDL comes in contact with an artery wall, a series of
Fig 4. Progression of Atherosclerosis.
reactions occur to repair the damage to the artery wall caused by oxidized LDL. The body's immune system responds to the damage to the artery wall caused by oxidized LDL by sending specialized white blood cells (macrophages and T-lymphocytes) to absorb the oxidized-LDL forming specialized foam cells. Unfortunately, these white blood cells are not able to process the oxidized-LDL, and ultimately grow then rupture, depositing a greater amount of oxidized cholesterol into the artery wall. This triggers more white blood cells, continuing the cycle. Eventually, the artery becomes inflamed. The cholesterol plaque causes the muscle cells to enlarge and form a hard cover over the affected area. This hard cover causes a narrowing of the artery, reduces the blood flow and increases blood pressure (Mangin et al., 1993).

Mature lesions develop a fibrous cap composed of a dense extracellular matrix containing collagen and elastin. Underneath the fibrous cap, a lipid core forms that contains many macrophages, dead or dying macrophages, cellular debris including apoptotic bodies, and extracellular lipid accumulations. Proinflammatory mediators released from activated white cells and endothelial cells and smooth muscle cells (SMC) can potentiate cell death by apoptosis in the advancing lesion (Demer et al., 1994). As SMCs die within lesions, fewer remain to renew the extracellular matrix in the plaque's fibrous cap. In addition, the activated cells in the lesion, notably the macrophages, secrete proteinases that can degrade the macromolecules of the extracellular matrix (Galis et al., 1994). In particular, interstitial collagenases can attack the triple helical collagen fragments weakening the fibrous cap. Elastases can break down elastin required for migration of cells within the lesion, and arterial remodeling occurs during compensatory enlargement, and in the extreme, aneurysm formation. During this phase of atherogenesis neovessels form in the intima, often arising as extensions of vasa vasorum that originate in the adventitial layer.

> **Myocardial infarction:** In which a narrowed coronary artery becomes completely blocked, usually by a blood clot. The blocked artery dies if deprived of oxygen for over 40-60 minutes.

It occurs when the blood supply to part of the heart is interrupted causing some heart cells to die. This is most commonly due to occlusion (blockage) of a coronary artery following the rupture of a vulnerable atherosclerotic plaque, which is an
unstable collection of lipids (like cholesterol) and white blood cells (especially macrophages) in the wall of an artery. The resulting ischemia (restriction in blood supply) and oxygen shortage, if left untreated for a sufficient period of time, can cause damage and/or death (infarction) of heart muscle tissue (myocardium).

**Risk Factors of CVD**

The Framingham Heart Study (FHS) has contributed importance to understand the causes of coronary heart disease (CHD), stroke, and CVD. The FHS and other epidemiologic studies have shown that elevated low density lipoprotein cholesterol (LDL-C), elevated triglycerides (TG), and low high density lipoprotein cholesterol (HDL-C) are independent risk factors for CVD (Mehta and Bansal, 2008). Major risk factors of CVD in Indians are sedentary lifestyle, cigarette smoking, hypertension, high LDL-C, low HDL-C and diabetes mellitus (DM). Other factors that influence CHD risk are obesity, family history of premature CHD, small dense LDL particles, lipoprotein-a (Lp-a), hyperhomocysteinemia and abnormalities in several coagulation factors such as fibrinogen, plasminogen activation inhibitor-I (PAI-1) (Singh and Sen, 2003; Gupta et al., 2008). A higher cholesterol level is associated with higher risk of CHD and is continuous. Epidemiological studies in western population have revealed that there is a consistent, strong, positive, continuous and graded relationship between plasma total cholesterol and the incidence of Coronary Artery Disease (CAD) (Kennel, 1995). If the ratio of total cholesterol to HDL-cholesterol is greater than 4.5, it is considered as a powerful predictor of CAD (Rajmohan et al., 2000).

A meta-analysis of prospective population based case-control study has examined to study the relation between insulin and CVD (Ruige et al., 1998). Atherosclerotic vascular disease is the most common macrovascular complication of DM. "The Deadly Triangle" of coronary artery disease, cerebrovascular disease and peripheral vascular disease is the major clinical presentation of macrovascular disease in diabetes (Mohan et al., 1999). The Quebec cardiovascular study provides the strongest evidence that hyperinsulinemia is associated prospectively with the development of CAD.
The major constituents of tobacco smoke which are responsible for the cardiovascular effects are nicotine and carbon monoxide. Other chemicals that cause vascular injury include nitrogen oxides, hydrogen cyanide and tar, with cadmium, zinc and carbon disulphide being minor contributors. Smoking causes endothelial dysfunction (blood vessels cannot dilate normally), lipid alterations and platelet activation, leading to a prothrombotic state (increased tendency of the blood to clot). Tobacco use also increases the risk and severity of vascular disease by increasing the risk of diabetes, which itself damages the vessels by accelerating atherosclerosis (Fig 5).

Platelets are cellular components produced from bone marrow megakaryocytes that circulate in the peripheral blood for approximately 10 days. They interact with the endothelium to produce thrombi and thereby play a critical role in hemostasis. Once a break in the integrity of the vascular endothelium occurs (as during rupture of an atherosclerotic plaque) circulating platelets are exposed to thrombogenic collagen and avidly adhere to sites of intimal disruption. During activation, platelets lose their regular discoid shape and acquire an irregular morphology with multiple pseudopodia. The glycoprotein (GP) IIb/IIIa complex, the most abundant platelet adhesion receptor, is a receptor for fibrinogen, fibronectin, vitronectin, Von Willebrand factor, and thrombospondin. Platelet-bound fibrinogen can simultaneously bind more than one GP IIb/IIIa receptor, thereby promoting platelet aggregation, firm adhesion and spreading. Activated platelets release a number of substances, including thromboxane A2, fibronectin, fibrinogen, thrombospondin, adenosine-5-diphosphate (ADP), serotonin, and platelet-derived growth factor. These platelet-derived products recruit additional platelets, reinforce and stabilize aggregated platelets, and promote vasoconstriction. Platelets also play a role in the initiation and progression of atherosclerotic lesions by facilitating leukocyte recruitment and lipid accumulation in blood vessels.

Several epidemiological studies have reliably demonstrated that plasma fibrinogen is a strong and independent risk factor for cardiovascular disease that is at least as important as more traditional risk factors for the disease. The deleterious effects of this protein seem to be mediated through its role in hemorrhheology,
Fig 5. Effects of tobacco on the cardiovascular system.
hemostasis, and the atherogenic process itself. High plasma fibrinogen concentration
is associated with elevated risk of CHD and stroke. Prospective studies in healthy
men (Yarnell et al., 1991; Meade et al., 1993; Heinrich et al., 1994) and women
(Kannel et al., 1987) have shown that a single fibrinogen measurement predicts fatal
and non-fatal cardiovascular events as much as 16 years later (Meade et al., 1993).
Fibrinogen may promote, together with other haemostatic factors, atherosclerotic
changes and thrombosis through effects shown in vitro on platelet aggregability,
blood viscosity and foam cell formation. Such processes are compatible with a causal
role for fibrinogen. An alternative view is that the prospective fibrinogen-
cardiovascular disease association may be a consequence, rather than a cause, of the
disease process, perhaps due to an inflammatory response to progressive endothelial
damage.

Role of lipoproteins and apolipoproteins in CVD

Lipoproteins are complex aggregates of lipids and proteins that render the
lipids compatible with the aqueous environment of body fluids and enable their
transport throughout the body. The most abundant lipid constituents are
triacylglycerols, free cholesterol, cholesterol esters and phospholipids
(phosphatidylcholine and sphingomyelin especially). Apoproteins are required to
solubilize the non-polar lipids in the circulation. The physical properties of
apoproteins enable them to bind readily at the interface between water and
phospholipids, and specifically they bind to the phospholipids on the surface of the
lipoproteins. In addition to the apoproteins, lipoproteins contain a number of
important enzymes, including lipases, acyl transferases and transport proteins.

Lipoproteins are the macromolecular complexes containing lipids and at their
inner core surrounded by proteins, thereby act as carrier molecule for the insoluble
lipids in plasma. Physical properties and lipid compositions of various lipoprotein
classes have shown different proportions of lipid components (Table 1)
(Skipski, 1972). Apolipoproteins are the protein components of lipoproteins (Table 2)
(Jonas, 2002; Nelson and Cox, 2008). Apolipoprotein isolated from animal species
are generally similar to those of the humans in their physical and biological
significance and in their distribution among lipoproteins.
Table 1. Physical properties and lipid compositions of lipoprotein classes.

<table>
<thead>
<tr>
<th></th>
<th>CM</th>
<th>VLDL</th>
<th>LDL</th>
<th>HDL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Density (g/ml)</td>
<td>&lt; 0.94</td>
<td>0.94-1.006</td>
<td>1.006-1.063</td>
<td>1.063-1.210</td>
</tr>
<tr>
<td>Diameter (Å)</td>
<td>600-2000</td>
<td>600</td>
<td>250</td>
<td>70-120</td>
</tr>
<tr>
<td>Total lipid (wt%)</td>
<td>99</td>
<td>91</td>
<td>80</td>
<td>44</td>
</tr>
<tr>
<td>Triacylglycerols (wt%)</td>
<td>85</td>
<td>55</td>
<td>10</td>
<td>6</td>
</tr>
<tr>
<td>Cholesterol esters (wt%)</td>
<td>3</td>
<td>18</td>
<td>50</td>
<td>40</td>
</tr>
<tr>
<td>Cholesterol (wt%)</td>
<td>2</td>
<td>7</td>
<td>11</td>
<td>7</td>
</tr>
<tr>
<td>Phospholipids (wt%)</td>
<td>8</td>
<td>20</td>
<td>29</td>
<td>46</td>
</tr>
</tbody>
</table>

Table 2. Main properties of the apolipoproteins.

<table>
<thead>
<tr>
<th>Apolipoprotein</th>
<th>MW</th>
<th>Lipoprotein</th>
<th>Function</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apo A1</td>
<td>28,100</td>
<td>HDL</td>
<td>Lecithin cholesterol acyltransferase (LCAT) activation. Main structural protein.</td>
</tr>
<tr>
<td>Apo A2</td>
<td>17,400</td>
<td>HDL</td>
<td>Enhances hepatic lipase activity</td>
</tr>
<tr>
<td>Apo A4</td>
<td>46,000</td>
<td>CM</td>
<td></td>
</tr>
<tr>
<td>Apo B48</td>
<td>241,000</td>
<td>CM</td>
<td>Derived from Apo B100 – lacks the LDL receptor</td>
</tr>
<tr>
<td>Apo B100</td>
<td>512,000</td>
<td>LDL, VLDL</td>
<td>Binds to LDL receptor</td>
</tr>
<tr>
<td>Apo C1</td>
<td>7,600</td>
<td>VLDL, CM</td>
<td>Activates LCAT</td>
</tr>
<tr>
<td>Apo C2</td>
<td>8,900</td>
<td>VLDL, CM</td>
<td>Activates lipoprotein lipase</td>
</tr>
<tr>
<td>Apo C3</td>
<td>8,700</td>
<td>VLDL, CM</td>
<td>Inhibits lipoprotein lipase</td>
</tr>
<tr>
<td>Apo D</td>
<td>33,000</td>
<td>HDL</td>
<td>Associated with LCAT, progesterone binding</td>
</tr>
<tr>
<td>Apo E</td>
<td>34,000</td>
<td>HDL</td>
<td>At least 3 forms. Binds to LDL receptor</td>
</tr>
<tr>
<td>Apo(a)</td>
<td>300,000-800,000</td>
<td>LDL, Lp(a)</td>
<td>Linked by disulfide bond to apo-B100 and similar to plasminogen</td>
</tr>
</tbody>
</table>
Lp-a is a modified form of LDL that contains apoB-100 linked by a disulphide bridge to a highly polymorphic glycoprotein, apolipoprotein-B (Scanu et al., 1991). Elevated levels of Lp-a have been linked to an increased risk of CAD and other forms of vascular disease especially when associated with the elevated LDL or reduced HDL cholesterol level (Luthra et al., 1999). Increased concentration of Lp-a in plasma have been associated with an increased risk of the development of premature cardiovascular disease (Berg et al., 1974). Lp-a structure is similar to plasminogen and tPA and it competes with plasminogen for its binding site leading to reduced fibrinolysis. Lp-a stimulates secretion of PAI-1 leading to thrombogenesis. In addition, because of LDL cholesterol content, Lp-a contributes to atherosclerosis (Schreiner et al., 1993). Pathogenecity of Lp-a also includes inhibition of transforming growth factor-β, destabilization of plaque, increased smooth muscle cell proliferation and migration, formation of occlusive thrombus, impaired formation of collateral vessels, enhanced oxidation uptake and retention of LDL-C and upregulation of expression of the PAI-I (Rajasekhar et al., 2004).

The two major apolipoproteins of HDL are Apo-A1 and Apo-AII. Apo-A1 has cardioprotective role, whereas Apo-AII is associated with increased risk of atherosclerosis in animal models (Kawahiri et al., 2000). Apo-A1 activates the lecithin cholesterol acyl transferase (LCAT) and it is also involved in removal of free cholesterol from extra hepatic tissues. Apo-AII has also a role in catabolism of HDL perhaps by stimulating hepatic lipase (HL). Defects in the structure of Apo-B or in the assembly of Apo-B containing lipoproteins result in the failure of intestinal and hepatogogeneous triglyceride rich lipoproteins to be secreted. Apo-B-48 and Apo-B100 are the principal structural apolipoproteins of chylomicrons, and Apo-B100 is virtually the only apolipoprotein on LDL. Both of these lipoproteins are required as structural lipoprotein particles for the secretion of arginine rich lipoproteins from the intestine and liver (Brewer, 1977). Abnormalities of apolipoprotein synthesis including high apo-B100, high Lp-a and low apo-A1 levels have been reported in CHD cases (Gupta et al., 2008).
Role of lipid and lipoprotein metabolizing enzymes in CVD

Lipid and lipoprotein metabolizing enzymes play a role in the development of atherosclerotic cardiovascular disease, characterized by the presence of lesions due to accumulation of lipids in the walls of large and medium-sized arteries (Kannel, 1996). Fatty streaks are usually present in humans in several major vessels, namely aorta, coronary artery and cerebral artery during different stages of life. Subramanian \textit{et al.}, (2003) reported that variation in the activities and expression of lipoprotein lipase (LPL), HL, LCAT, cholesteryl ester transfer protein (CETP) can be lead to both pro- and antiatherogenic.

LPL and HL are the two major lipolytic enzymes responsible for the hydrolysis of triglycerides and phospholipids present in circulating plasma lipoproteins (Santamarina-Foja \textit{et al.}, 2004). LPL is synthesized predominantly in the skeletal muscle, cardiac muscle and adipose tissue, and to a lesser extent in the kidneys, adrenal glands, brain and macrophages. The importance of LPL in the atherosclerotic process was first proposed in 1973 (Zilversmit, 1973). It is a central enzyme in overall lipid metabolism and transport, being responsible for catalyzing the hydrolysis of triglycerides transported in the blood stream by chylomicrons and VLDL, thereby providing non-esterified fatty acids and 2-monoacylglycerols for tissue utilization. The physiological site of LPL action is at the luminal surface of blood vessels (Enerback and Gimble, 1993). It mediates the lipolysis of circulating VLDL and chylomicrons particles in the luminal surface of vascular endothelium thereby leading to both a decrease in their size and enrichment in their cholesteryl ester content (Zilversmith, 1973). Such remnants are readily taken up by macrophages in addition to free fatty acids as a consequence their transformation into foam cells (Mead and Ramji, 2002). In addition, LPL also stimulates the hepatic removal of lipolyzed lipoproteins and transfers the surface components of triglyceride-rich lipoproteins to HDL (Brunzell, 1995). Pathophysiological actions of LPL are concerned to promote foam cell formation and, ultimately, atherosclerosis.

HL is a form of lipase. It is expressed in the liver and adrenal gland (Dichek \textit{et al.}, 2006). HL involved in the hydrolysis of triglycerides and probably phospholipids in VLDL remnants which lead to a more efficient uptake of these particles and
Fig 6. Multiple roles of Hepatic lipase.
generation of LDL (Santamarina-Fojo et al., 2004). These multiple functions of HL (Fig. 6) facilitate not only plasma lipid metabolism but also cellular lipid uptake, and can be anticipated to have a major and complex impact on atherogenesis. In humans, low HL activity has been associated with increased risk of CAD. Like LPL, HL expression in the arterial wall may result in localized increased production of free fatty acid (FFA), increased cholesterol uptake, retention of LDL in the subendothelial wall, and macrophage recruitment, all of which would enhance lesion formation (Santamarina-Fojo, 2004). Accumulation of cholesterol by macrophages has been demonstrated to alter macrophage gene expression (Mikita et al., 2001) and promote atherosclerosis.

LCAT is responsible for the synthesis of cholesteryl esters in plasma (Jonas, 2000). It converts cholesterol to cholesteryl esters on the surface of lipoproteins, predominantly HDL (Sviridov and Nestel, 2000), after which the cholesteryl ester molecules migrate to the inner core of this lipoprotein. Through this action, LCAT plays a central role in the formation and maturation of HDL (Glomset, 1968), and in the intravascular stage of reverse cholesterol transport, the major mechanism by which HDL modulate the development and progression of atherosclerosis. A defect in LCAT function would be expected to enhance atherosclerosis by interfering with this process (Calabresi et al., 2005).

The relationship between CETP and lipoprotein metabolism is complex and may depend largely on the concentration of TG rich proteins (Harchaoui et al., 2007). CETP is a hydrophobic glycoprotein that is produced in the liver and adipose tissue. It regulates the exchange of TG and cholesteryl esters between the apo-B containing lipoproteins and HDL in plasma. Accordingly, the role that CETP plays in atherosclerosis is complex and may depend on several factors, including the plasma concentrations of CETP, the plasma levels and composition of lipoprotein donors as well as acceptors, and the overall metabolic conditions (Sikorski, 2006). In subjects with dyslipidemia, CETP concentration may increase up to two- to threefold (Le Goff et al., 2004).
Role of antioxidants in CVD

Antioxidants constitute the foremost defense system that limit the toxicity associated with free radicals. The equilibrium between antioxidant enzymes is an important process for the effective removal of oxygen stress in intracellular organelles (Andrew and Mathew, 1989). They are the first line of cellular defense against oxidative injury. There is evidence from recent epidemiological studies that antioxidants may reduce the risk of CVD (Gaziano and Hennekens, 1995). Low concentrations of antioxidant vitamins or enzymes and high concentrations of lipid peroxides in plasma will increase the risk of CHD (Singh et al., 1996). There are many experimental (Singal and Krishendaum, 1990; Dhalla et al., 1996) and clinical (Singh et al., 1996; Ghatak et al., 1996) studies documenting a close relationship between antioxidant deficit and cardiac dysfunction.

GSH functions as a free radical scavenger in the repair of radical induced cellular damage. Reduced availability of GSH in turn decreased the activity of GPx and GST. GPx plays an important role in protecting the heart from peroxidative attack (Doroshow et al., 1980). SOD and CAT are important antioxidant enzymes in mitigating free radical induced cell injury. A decrease in the activity of superoxide dismutase (SOD) and catalase (CAT) could result in the decreased removal of superoxide ion and hydrogen peroxide radicals which brings about a number of reactions which are harmful to the myocardium (Sumitra et al., 2001).

Role of nitric oxide (NO) in CVD

NO, a diffusible universal messenger mediates cell-cell communication throughout the body (Moncada et al., 1991; Pfeilschifter et al., 2003). NO, the major mediator of endothelium-dependent vasorelaxation is normally produced from L-arginine by endothelial nitric oxide synthase (eNOS) in the vessel wall (Cai and Harrison, 2000; Sachidanandam et al., 2005). There are three isoforms of the enzyme: neuronal NOS (nNOS), inducible NOS (iNOS), and endothelial NOS (eNOS). It is now known that each of these isoforms may be expressed in a variety of tissues and cell types. Only nNOS and eNOS are reported to be constitutive enzymes and are calcium-dependent and produce low levels of gas as a cell signaling molecule (Parvu et al., 2005). iNOS is calcium independent, activated by cytokines and
produces large amounts of gas which can be cytotoxic. NO is released from endothelial cells in response to stress produced by increasing blood flow and activation of a variety of receptors, hormones, neurotransmitters, and autacoids, and can be inhibited by superoxide anion (Ignarro et al., 1987; Koh et al., 2007).

NO exists as a nitrate form, and nitrite and nitrate are measured in serum, plasma, and urine as markers of NO generated (Koh et al., 2007). Physiologically the most relevant action of NO is to scavenge $\text{O}_2^-$ and other free radicals and also inhibits $\text{O}_2^-$-driven Fenton reaction and lipid peroxidation. The levels of NO will be increased under conditions of oxidant injury. The excess NO reacts rapidly with superoxide anion to form peroxynitrite, which may be cytotoxic by itself or easily decompose to the highly reactive and toxic hydroxyl radical and nitrogen dioxide ($\text{NO}_2$) (Ravi Kanth et al., 2008).

\begin{equation}
\text{NO} + \text{O}_2^- \rightarrow \text{ONOO}^-
\end{equation}
\begin{equation}
\text{ONOO}^- + \text{H}^+ \rightarrow \text{ONOOH}
\end{equation}
\begin{equation}
\text{ONOOH} \rightarrow \text{HO}^+ + \text{NO}_2^-
\end{equation}

It is well known that ONOO$^-$ is much more reactive than NO or superoxide, which will cause diverse chemical reactions in biological system including nitration of tyrosine residues of proteins, triggering of lipid peroxidation, inactivation of aconitase, inhibition of mitochondrial electron transport and oxidation of biological thiol compounds (Maeda and Akaike, 1998). NO could also be directly oxidized to $\text{NO}_2^-$, which induces DNA damage. In addition, the reaction of NO with $\text{H}_2\text{O}_2$ has been shown to produce potentially cytotoxic singlet oxygen (Oshima and Bartsch, 1994).

NO has been found to exert negative inotropic and negative chronotropic effect on cardiac muscle cells. There is also evidence for the release of NO along with other factors from the endocardium and may be involved in the beat-to-beat regulation of cardiac function (Shah, 1993). Basal level of NO regulates blood flow in the brain (Faraci and Breesse, 1993), heart (Max et al., 1993), lungs (Fineman et al., 1991), gastrointestinal tract (Iwata et al., 1992), and kidneys (Riabairo et al., 1992). Thus, nitric oxide is an endogenous autoregulator of blood flow. Endothelial
dysfunction may cause local deficiency of NO, which leads to platelet aggregation, and subsequent development of atherosclerosis.

Several studies revealed that NO interacts with various important biomolecules resulting in formation of nitrated lipids, nitrated proteins, nitrosamines, iron nitrosyls etc. Elfering et al., (2004) indicated that the nitration process may occur as a result of two conditions: i. a primary sequence that allows the nitration of tyrosine at the ortho position of the hydroxyl group and ii. the localization of the protein at a close to the membrane. Further, NO appears to be involved in DNA damage, regulation of metabolism and also in several membrane dependent processes. Membrane transport is a key process which is responsible for various specialized properties of individual cell types and ultimately responsible for the behavior of cells. Loss of NO signaling control results in excessive inhibition of respiratory chain leading to bioenergetic dysfunction (i.e. decreased ATP synthesis) and increased ROS production.

**Role of oxidative stress in CVD**

Imbalance between oxidant and antioxidant reaction either due to excess free radical formation or insufficient removal by antioxidant leads to oxidative stress (Kumar et al., 2008; Kaur et al., 2008). Recent studies have demonstrated that there is an association between increased oxidative stress and diabetes, hypertension and dyslipidemia (Ravi Kanth et al., 2008; D’Souza et al., 2008). Oxidation-reduction (redox) sensitive processes mediated by oxidative stress not only play an important role in the oxidation of LDL (Fig 7), which is a crucial step in the pathogenesis of atherosclerosis, but also cause vascular dysfunction by impairing the vascular relaxation response, promoting pathological growth of vascular smooth muscle cells (VSMC) and fibroblasts, triggering DNA breaks, causing apoptosis of endothelial cells, and increasing platelet adhesion, all of which contribute to the development of atherosclerotic disease (Taniyama and Griendling, 2003; Deng et al., 2004). Oxidative stress is a state in which excess formation of highly reactive molecules such as reactive oxygen species (ROS) and reactive nitrogen species (RNS) saturate the natural antioxidant defense mechanisms (Turko et al., 2001). Low levels of ROS are involved to some extent as signaling molecules under physiological conditions.
Fig 7. LDL oxidation – Atherosclerosis.

Fig 8. A number of Oxidative systems to limit free radicals.
ROS regulate the transcription of numerous genes that are relevant to CVD. The principle sources of ROS in the vasculature include NAD(P)H oxidase, xanthine oxidase, eNOs and the mitochondrial respiratory chain. A number of scavenging systems are in place to limit ROS levels (Fig 8). $O_2^-$ may be dismutated by a family of SODs to $H_2O_2$. $H_2O_2$ can be scavenged to water by catalase or by GPx in the presence of reduced glutathione (Hamilton et al., 2004).

The enhanced ROS production then dysregulates physiological processes. $O_2^-$ and other risk factors may react with NO causing endothelial dysfunction (Fig 9). In addition, ROS can stimulate vascular smooth muscle cell hypertrophy and hyperplasia. In general, increased production of ROS may affect four fundamental mechanisms that contribute to atherogenesis: oxidation of LDL, endothelial cell dysfunction, vascular smooth muscle cells growth, and monocytes migration (Berliner and Heinecke, 1996).

Oxygen is vital to life but as a diatomic molecule it is remarkably unreactive. However, oxygen is the substrate for the generation of a variety of ROS, some of which may be deleterious to cell function. ROS are generally speaking oxygen molecules in different states of oxidation or reduction, as well as compounds of oxygen with hydrogen and nitrogen. Although superoxide is produced directly from the reduction of oxygen, the biologically active species are hydrogen peroxide, hydroxyl radicals, hypochlorite ion and peroxynitrites. A number of sites within the cell participate in the production of superoxide including xanthine/xanthine oxidase reactions, autoxidation of catecholamines and arachidonic acid metabolism (Kukreja and Hess, 1992). ROS play an important role in modulating cell signalling pathways, and when produced at high levels inducing cell death. A number of cardiac ion channels respond to changes in oxygen partial pressure. Channel function can be modified by thiol reducing or oxidizing agents and thiol reducing agents mimic the effect of hypoxia. This would suggest that ion channel function is modified during hypoxia as a result of redox modification of the channel or a nearby protein (Hool, 2005).
Fig 9. Risk factors for endothelial dysfunction.

- Hypertension
- Hypercholesterolaemia
- Heart Failure
- Diabetes
- Smoking
- Homocysteine

\[ \text{O}_2 \uparrow \downarrow \text{NO} \]

- Endothelial Dysfunction

- Apoptosis
- Vascular smooth muscle
- Vasoconstriction
- Thrombosis
- Oxidation
- Lipids
- Proteins
- DNA
- Leucocyte adhesion

Fig 10. Reasons for hyperhomocysteinemia.

5-tetrahydrofolate

5,10 methylene-tetrahydrofolate

FADH2

MTHFR

N5-Methyl tetrahydrofolate

5-adenosylmethionine

S-adenosyl homocysteine

CBS

Cystathionine

Homoserine

Cysteine

Methylcobalamin

Adenosine

Homocysteine

CBS
Role of homocysteine in CVD

Several plasma biomarkers have been investigated to determine their use as tools for predicting the risk of CVD including homocysteine and C-reactive protein. Plasma homocysteine, a sulfhydryl-containing amino acid formed during the metabolism of methionine, is associated with endothelial damage and hypertension (Tayama et al., 2006). There is evidence that elevated plasma homocysteine is associated with CVD (Bhandari et al., 2008). A number of studies have indicated that hyperhomocysteinemia promotes atherosclerosis through increased oxidative stress, impaired endothelial function and induction of thrombosis (Bhandari et al., 2008). Homocysteine is present in plasma as protein bound or free forms. A majority of homocysteine is bound to albumin, while the free form exists as homocysteine disulfide, homocysteine-cysteine mixed disulfides, and trace amounts as reduced homocysteine (Nusier and El-Dwairi, 2007).

Homocysteine is formed from the demethylation of methionine and can subsequently follow one of two metabolic pathways: 1) remethylation to methionine by the vitamin B12-dependent enzyme 5-methyltetrahydrofolate-homocysteine methyltransferase, using 5-methyltetrahydrofolate as the methyl donor, a reaction catalyzed by 5,10-methylenetetrahydrofolate reductase (MTHFR); or 2) transsulfuration to cystathionine, catalyzed by the vitamin B-6-dependent enzyme cystathionine β-synthase. Folic acid, vitamin B-6, and vitamin B-12 are all cofactors in homocysteine metabolism (Miller et al., 1992; Panchanuniti et al., 1994; Woodside et al., 1998).

Auto-oxidation of homocysteine forms homocystine, mixed disulfides, and homocysteine thiolactone (Andersson et al., 1995). Potent cytotoxic ROS, including superoxide anion radical, hydroxyl radical, and hydrogen peroxide, are produced during this auto oxidation (Misra, 1974) and induce endothelial cell injury (de Groot et al., 1983), initiate lipid peroxidation, activate platelets and leukocytes (Harker et al., 1974), impairs oxidative phosphorylation and promotes the proliferation and fibrosis of smooth muscles by increasing calcium deposition. The known reasons (Fig 10) for an increased level of plasma homocysteine are genetic defects of cystathionine β-
synthase (CBS), methylenetetrahydrofolate reductase (MTHFR) and deficiency of folic acid, vitamins B₆ and B₁₂ (Domagala et al., 1997).

**Role of carnitine in CVD**

Carnitine plays an important role in the transmembrane transport of long chain fatty acids for its oxidation and has been found to offer protection against myocardial infarction induced by isoproterenol (IPL). Sushma Kumari et al. (1989) reported that carnitine offers protection to the myocardium in experimental myocardial infarction by preventing the lipid peroxidative system to be activated. The protective effects of carnitine are probably achieved by decreasing the levels of fatty acids and peroxides and activating the enzyme glutathione peroxidase. In addition carnitine decreases the level of taurine in turn decreases the uptake of calcium by the myocardium thereby preventing the myocardial cells to be overloaded by calcium. Taurine has also been found to increase the intracellular calcium level in the normal heart (Dolara et al., 1978). Myocardial ischemia has been found to be associated with massive loading of mitochondria with calcium, which interferes with mitochondrial functions and produces cellular injury (Shen and Jennings, 1972).

**Role of taurine in CVD**

Taurine, sulfur containing amino acid, is the most abundant free amino acid in excitable tissues and cells. Biosynthesis of taurine from cysteine and methionine occur by cysteine dioxygenase and cysteine sulfinate decarboxylase. Cysteine sulfinate decarboxylase is thought to be the rate-limiting enzyme in taurine biosynthesis (Reymond et al., 2000). Taurine makes up more than 50% of the total amino acid pool in the mammalian heart (Lombardini, 1996). It has extensive biological functions, such as stabilizing cell membrane, scavenging oxygen free radicals, regulating intracellular osmostasis, maintaining intracellular Ca²⁺ concentrations and neuromodulation (Shiny et al., 2005). Taurine is primarily utilized in biological system for the elimination of cholesterol from the body through the bile acid conjugation reactions (Sivakumar et al., 2007). Warskulat et al. (2004) have demonstrated that pathology develops in the myocardium if the animal is depleted of taurine stores either through a taurine deficient diet or taurine transport antagonists. There is a high gradient of taurine concentration between intra-and
extracellular space. This gradient is maintained by taurine transporter (TAUT) through the transportation of taurine on cellular plasma membrane. TAUT is usually involved in the maintenance of higher intracellular taurine concentrations and to play its physiological roles.

**Role of calcium in CVD**

Calcium ion is essential for vital body functions (Mitsuma et al., 1999). Calcium-dependent processes play a central role in several different cells of the cardiovascular system including vascular smooth muscle and endothelial cells and also in monocytes, macrophages and platelets. It is one of the essential factors for the excitation-contraction coupling in the cardiac muscle cell (Dhalla et al., 1981) as well as for the conduction of electrical impulses in certain regions of the heart particularly through AV node. Calcium is critical in the contractile process. Extracellular Ca\(^{2+}\) plays an important role in the contraction of cardiac muscle, which is rich in ion channels. The levels of free intracellular Ca\(^{2+}\) is restricted by the ion channels. In resting myocytes Ca\(^{2+}\)-Na\(^{+}\) exchanger maintain a low level of free intracellular Ca\(^{2+}\), which contributes to relaxation but may run in the reverse direction during excitation (Murry et al., 2000).

Ca\(^{2+}\) also plays a crucial role in regulating cardiomyocyte growth (Kane et al., 1981). Influx of extracellular calcium into the cell triggers the release of calcium from intracellular stores such as the cisternae of sarcoplasmic reticulum, mitochondria and various fixed and soluble intracellular proteins. Thus, there is increase in the intracellular free calcium concentrations from a resting level of 0.1 \(\mu\)m to an activating level of 10 \(\mu\)m (Marban et al., 1980; Shamkuwar et al., 2008). After increase in the level of intracellular calcium, it binds to sarcomeric protein, troponin-C resulting in a conformational changes in the tropomyosin protein, which causes myosin to interact with the active site on the actin filament. Sliding of the actin filaments over the myosin filaments produces the myocardial contraction (Huxely and Simmons, 1971).

 Accumulation of intracellular Ca\(^{2+}\) is a main event in the final stages of cell death. Ca\(^{2+}\) is thought to inhibit intracellular energy flow. When present in heart cells
it accumulates in mitochondria, causes uncoupling of oxidative phosphorylation and leads to decreased ATP production.

Calcium antagonist drugs principally act on L-type calcium channels to reduce the influx of calcium into the cells of the body. Calcium antagonist drugs are able to influence a wide range of cellular processes which have been implicated in atherosclerosis, glomerulosclerosis, left ventricular hypertrophy and insulin resistance.

**Role of C-Reactive Protein (CRP) in CVD:**

CRP is an acute-phase serum protein that is synthesized by hepatocytes. During infection, inflammation, and tissue injury, the serum level of CRP rises by a factor up to several thousand (Kitsis and Jialal, 2006). CRP values after acute myocardial infarction predict outcome including death and heart failure (Suleiman et al., 2006). CRP has numerous effects on endothelial cells that could support a pro-inflammatory and pro-thrombotic role (Fig. 11). Its procoagulant effects include inhibition of eNOS, prostacyclin, tPA and up-regulation of PAI-1, and the proinflammatory effects include up-regulation of IL-6, inter cellular adhesion molecule (ICAM) and vascular cell adhesion molecule (VCAM), and the chemokines, monocyte chemoattractant protein-1 (MCP-1) and IL-8.

In addition, CRP has been shown to promote the uptake of oxidized LDL which could be relevant to the genesis of the atherosclerotic lesion including plaque instability (Jialal et al., 2004; Jialal et al., 2006). CRP not only may be a marker of low grade chronic systemic inflammation but also may be directly involved in atherosclerosis. It can amplify the anti-inflammatory response through complement action, tissue damage, and activation of endothelial cells (Heald, 2007).

**Treatment of CVD**

The ideal goal of treatment/medicine for CVD is maximum reduction of morbidity and mortality through the maintenance/homeostasis of lipid and lipoprotein metabolism and other cardiac risk factors. Dieting, physical exercise and inclusion of dietary fiber have been used with limited success.
Fig 11. Effect of CRP on the vascular endothelium.
Antihyperlipoproteinemic Drugs

The hyperlipoproteinemias are conditions in which the concentration of cholesterol- or triglyceride-carrying lipoproteins in plasma exceeds an arbitrary normal limit. Administration of drugs that lower plasma concentrations of lipoproteins, either by diminishing the production of lipoproteins or by enhancing the efficiency of their removal from plasma. The combination of nicotinic acid and a bile acid binding resin lowers LDL concentrations (Kane et al., 1981). Clofibrate, Gemfibrozil, Probucol, Cholestyramine and Colestipol (Hashim and Van Itallie, 1965), Compactin and Mevinolin, are the drugs extremely effective in lowering plasma concentrations of LDL-cholesterol with minimal toxicity.

Beta (β)-Blockers

Congestive heart failure (CHF) is the only major CVD with increasing prevalence, incidence, and mortality. Studies proved that enhanced LPO and oxygen free radical damage persists in patients with CHF (Dave, 2000). Drugs like digoxin, vasodilators, and positive inotropic agents improve contractility, reverse haemodynamic abnormalities, and improve functional status, but they failed to confer a survival benefit. Their actions are mediated through activated cAMP and/or angiotension II and partially responsible for the pathological remodeling process that leads to progressive deterioration (Eichhom, 1998).

Swedish researchers in 1975 first gave evidence that β-blockers improve survival rate in some patients of dilated cardiomyopathy (Clelang et al., 1996). Metoprolol, Bisoprolol, Carvedilol, Alprenolol, Bucindolol and Labetolol (Mittal, 2000) have been used in different trials and all have shown survival benefit irrespective of the differences in their other pharmacological properties (Van Campen et al., 1998; Cleland et al., 1999). Sympathetic activation of patients with heart failure leads to increased circulating norepinephrine (NE) levels, which in turn stimulate α and β adrenergic receptors. Chronic elevation of NE may cause myocyte necrosis and progressive left ventricular dysfunction and is associated with poor prognosis in CHF patients. Treatment with β-blocker attenuates or inhibits progressive left ventricular dysfunction and remodeling.
Cardiac Resynchronization Therapy (CRT)

The main therapeutic mechanism of CRT is thought to be the correction of dyssynchronous myocardial contraction (Van Hemel and Scheffer, 2009). CRT is intended for patients with moderate to severe heart failure and also ventricular dyssynchrony, a condition in which the two ventricles are not beating synchronously. CRT was approved by the Food and Drug Administration (FDA) in 2001 (Panescu, 2005). A study sponsored by the National Institutes of Health (NIH) between 1997-2001 found that CRT devices increased the survival rate for heart failure patients.

CRT delivers electrical stimuli to the left and right ventricles simultaneously with the goal of synchronizing the activation of both ventricles. This is achieved by introducing a specially designed pacing lead into the left ventricle, usually implanted through an intravenous approach via the coronary sinus and into a lateral cardiac vein in addition to placement of standard right sided leads. The proposed mechanism of benefit by CRT is to correct the dyssynchrony between the right and left ventricles and the intraventricular dyssynchrony within the left ventricles by pacing the right ventricular apex and lateral or posterolateral wall of the left ventricle (Abraham et al., 2002; Bristow et al., 2004).

Cardiac Contractility Modulation (CCM)

CCM is a new form of electrical therapy. It is a potential device therapy for heart failure patients who are not CRT candidates. Only a small proportion of heart failure patients can benefit from CRT, because it is only applicable to patients with evidence of cardiac dyssynchrony. CCM has been proposed for enhancing ventricular contractile strength independent of the synchrony of myocardial contraction.

CCM involves implanting a pacing-type device, with a sensing lead in the right atrium and two right ventricular leads that deliver relatively large amplitude electrical stimuli during the absolute refractory period of the myocardium. The mechanism of effect is thought to be due to improved cardiac myocyte calcium handling without increasing myocardial oxygen demand (Borggrefe et al., 2008).
Hormonal approach

CVD is the leading cause of premature death of men and women in developed countries. Although death rates from CVD are similar, at 46% and 52% in men and women, the major difference is that the peak incidence in women occurs 10 years later than in men (Barter and Rye, 1996). In developing countries, half of all deaths of women over 50 are due to heart disease and stroke. Srinath and Yusuf (2001) reported that women will continue to experience disproportionately high mortality from CVD. By 2040, women in the study countries (Russia, Brazil, India, China, South Africa) will represent a higher proportion of CVD deaths than men.

The protection afforded to premenopausal women is in large part due to the beneficial effects of endogenous oestrogens. Cardiovascular benefits of oestrogen therapy particularly in postmenopausal women were highlighted by Colditz et al., (1987). Oestrogen deficiency, caused by surgical or natural menopause, increase the risk of CVD in women by threefold (Barrett-Conner, 1991; Barrett-Conner and Bush, 1991). Furthermore, epidemiological studies suggest that oestrogen replacement therapy in postmenopausal women reduces the risk of CVD by up to 50% (Bush, 1990). More studies on oestrogen use suggest a 35-50% reduction in the risk of CHD events with oestrogen therapy.

The precise mechanisms underlying the cardioprotective action of oestrogens are unknown. Although the mechanism is most probably multifactorial (Shewmon, 1994), a major reduction in CVD risk is attributed to the effect of oestrogen on the metabolism of apolipoprotein-A1 and HDL and LDL particles (Bush et al., 1987). Previous studies demonstrated that pre- or post-menopausal women treated with exogenous oestrogens have an increased abundance of apo-A1 and HDL (Walsh et al., 1991). Oestrogen is expected to alter transcription of the apo-A1 gene (Song et al., 1998).

Oestrogens modulation of coronary vasoreactivity includes both the promotion of coronary vasodilation via increased bioavailability of NO and increased prostacyclin production and the inhibition of endothelium-related vasoconstricting factors. Oestrogen interferes with the activity or release of the potent endothelial vasoconstrictor endothelin-1 and decrease the vascular responses to catecholamines.
Another possible mechanism by which oestrogen may be cardioprotective is by inhibiting the inflammatory response to atherosclerosis by decreasing myointimal proliferation, by inhibiting platelet aggregation and adhesion and foam cell formation and by decreasing the expression of circulating cellular adhesion molecules from the endothelium. Other postulated vascular benefits include attenuation of endothelial cell apoptosis, endothelium-dependent relaxation possibly via a Ca\(^{2+}\)-channel blocking effect and enhancement of neovascularization and collateral vessel formation.

**Antihypertensives**

Available literature indicates that the dietary management alone can’t control hypertension (Freis, 1971). Hypertension has been adversely associated with the risks of stroke, MI, CAD and peripheral vascular disease (PVD). The goal of treatment of hypertension is maximum reduction of morbidity and mortality in CVD and the primary goal is maintenance of optimal BP. Currently most commonly used drugs in the control of BP are:

- Diuretics (Thiazides (doxazosin, amlodipine), nondihydropyridines (Indapamide)
- β-blockers
- Calcium channel blockers (Dihydropyridines (nifedipine), nondihydropyridines (verampil and diltiazem))
- Angiotensin converting enzyme inhibitors (e.g. ramipril)
- Angiotensin receptor blockers (e.g. losartan)
- α-blockers

WHO- International Society of Hypertension (ISH) guidelines (Kaplan *et al.*, 2003) recommended that all the above drugs have equal efficacy in lowering BP in standard doses.

**Omega 3 fatty acids and CVD**

Both health professionals and the public are increasingly interested in omega 3 fatty acids role in the prevention and management of CVD. Omega 3 fatty acids from fish and fish oils can protect CHD. An inverse relation is observed between fish consumption and the risk of CHD (Kromhout *et al.*, 1985; Hu *et al.*, 2002). They reduce platelet aggregation at large doses, whereas at smaller amounts they have
modest platelet inhibitory effects (Mori et al., 1997). They may also influence the atherosclerotic process. These effects may be due to a reduction in lipids, inflammation, production of growth factor, or suppression of smooth muscle cell proliferation. They have direct effects on endothelial vasomotor function, and reduction in blood pressure (Din et al., 2004). However the optimal intake of omega 3 fatty acids is not firmly established and their mechanism of action is not fully understood. The ways that omega-3 fatty acids reduce CVD risk are still being studied. However, research to date suggests that they can: 1. decrease triglyceride and remnant lipoprotein levels and the rate of growth of atherosclerotic plaque 2. reduce inflammatory responses and the risk for arrhythmias and thrombosis 3. improve endothelial function and slightly lower blood pressure (Kris-Etherton et al., 2003).

**Coenzyme Q10 and CVD**

A large number of clinical trials demonstrate the relationship between coenzyme Q10 (CoQ10) deficiency and CVD, and the slowdown of CVD progression with CoQ10 supplementation. CoQ10, a fat soluble nutrient also known as ubiquinone (2,3-dimethoxy-5-methyl-6-decaprenyl-1,4-benzoquinone), is a major participant in electron transfer during oxidative phosphorylation in the mitochondria and is essential for the optimal functioning of an organism. It was discovered in 1957 as a component of beef heart mitochondria (Crane et al., 1975). CoQ10 is a potent antioxidant and free radical scavenger, and is a membrane stabilizer that preserves cellular integrity (Blizakov, 2002). Statins are prescribed to reduce CVD morbidity and mortality by interfere with biosynthesis of CoQ10 leading to low serum levels which cause muscle to atrophy.

**Bioinformatics approach – Docking studies**

Bioinformatics is the application of information technology to the field of molecular biology. Bioinformatics now entails the creation and advancement of databases, algorithms, computational and statistical techniques, and theory to solve formal and practical problems arising from the management and analysis of biological data. Over the past few decades rapid developments in genomic and other molecular research technologies and developments in information technologies have
combined to produce a tremendous amount of information related to molecular biology. The primary goal of bioinformatics is to increase our understanding of biological processes. What sets it apart from other approaches, however, is its focus on developing and applying computationally intensive techniques (e.g. data mining, and machine learning algorithms) to achieve this goal. Major research efforts in the field include sequence alignment, gene finding, genome assembly, protein structure alignment, protein structure prediction, prediction of gene expression and protein-protein interactions, genome-wide association studies and the modeling of evolution.

Bioinformatics was applied in the creation and maintenance of a database to store biological information at the beginning of the "genomic revolution", such as nucleotide and amino acid sequences. Development of this type of database involved not only design issues but the development of complex interfaces whereby researchers could both access existing data as well as submit new or revised data. In order to study how normal cellular activities are altered in different disease states, the biological data must be combined to form a comprehensive picture of these activities. Therefore, the field of bioinformatics has evolved such that the most pressing task now involves the analysis and interpretation of various types of data, including nucleotide and amino acid sequences, protein domains, and protein structures. The actual process of analyzing and interpreting data is referred to as computational biology.

Docking studies are computational techniques for the exploration of the possible binding modes of a substrate to a given receptor, enzyme or other binding site. Computational methods have developed as useful tools in facilitating new drug discovery (Kuntz, 1992). By the use of these methods, the biological activity of the candidate molecules can be estimated before experimental trials. Thus, they are simple, non-expensive and expedite to design molecules with desirable biological activity. Quantitative structure-activity relationship (QSAR) (Hemmateenejad et al., 2003 and 2005) and docking procedure (Wang et al., 1999) are two mostly used computational methods in drug design. In QSAR methodologies, a mathematical relationship, relating the biological activity to some molecular descriptors is obtained.
In docking studies, different search algorithms such as simulated annealing and genetic algorithm in combination with scoring function such as molecular mechanic calculations are used to study the binding of the candidate ligands to an enzyme with known structure (Hemmateenejad et al., 2007).

**Need for Alternative Medicine**

From the brief overview of CVD therapy, it is known that current methods for all types of CVD fails to achieve the maintenance of cardiac risk factors. A wide variety of therapeutic agents are available today with a range of action to fight against hyperlipidemia and CVD. These treatments are restricted by their limited action, pharmacokinetic properties and accompanying side effects. Moreover, the drugs and surgeries available today for the treatment of CVD are very expensive and the situation demands the need of alternative medicine.

**Importance of Medicinal Plants**

The history of plant chemicals began approximately 1.5 billion years ago, when algae first evolved in the primordial sea. About 435 to 500 million years ago the first known land plants began to appear in wet mud at the edge of bodies of fresh water. As millions of years passed, plants grew in size and differentiated.

Demand for medicinal plants is increasing in both developing and developed countries due to growing recognition of natural products, being non-narcotic, having no side effects, easily available at affordable price and sometime the only source of health care available to the poor. Among the entire flora, 35,000 to 70,000 species have been used for medicinal purposes. In India, of the 17,000 species of higher plants, 7500 are known for medicinal uses. This is the highest proportion of medicinal plants known for their medical purposes in any country of the world for the existing flora of that respective country. The Indian system of medicines, Viz Ayurveda, Siddha, Unani and Homeopathic system predominantly use plant based raw materials and most of their preparations and formulations. Ayurvedic and other indigenous systems of medicine are used commonly in India (Warier, 1995). According to WHO, 80% of the populations of developing countries relied on traditional medicines, mostly plant drugs for their primary health care needs.
natural products derived from medicinal plants have proven to be an abundant source of biologically active compounds, many of which have been the basis for the development of new lead chemicals for pharmaceuticals.

Plants show enormous flexibility in synthesizing complex material which have no immediate obvious growth and metabolic functions. These complex materials are referred to as secondary metabolites, which are also referred to as phytochemicals. The phytochemicals include alkaloids, saponins, tannins, anthraquinones, terpenoids, steroids, flavonoids, cardiac glycosides, etc. Phytochemicals are naturally occurring and biologically active compounds and have potential disease inhibiting capabilities. Hence the medicinal values of these plants lie on their component phytochemicals, which produce definite physiological actions on the human body. Constituents of herbal medicines are found in leaves, flowers, stems, seeds, roots, fruit and bark. Presence of several phytochemicals like alkaloids, terpenoids, tannins, glycosides, etc., herbs have been used to treat more than one hundred disorders in humans including atherosclerosis, arthritis, ischemia and reperfusion injury of many tissues, central nervous system injury, gastritis, cancer, diabetes and AIDS.

WHO has also recommended the evaluation of the plants effective in conditions where we lack safe modern drugs. An intact/native preparation may contain dote and antidote (Ponda and Anand, 1998) probably to mitigate side effect(s), if any, of the active (principle) components. Therefore, a majority (88%) of the global population has turned to plant derived medicines/therapeutic methods (Samy et al., 1999). Thus, it has become necessary to look for an economical as well as therapeutically effective treatment especially for usage in the developing and under-developed countries.

**Phytoremediation for cardiovascular disease**

Recent decades have seen a resurgent interest in traditional plant treatments for CVD. This has pervaded nutrition, the pharmaceutical industry and academic research, fuelled by a growing public interest and awareness of so called complementary and natural types of medicine. Many plants are reported which are
useful in the treatment of CVD (Varadharajan, 1985; Dwivedi and Somani, 1988; Eddouks et al., 2002). However few have received scientific or medical scrutiny.

**Herbal medicines - Common features**

- Herbal medicines are different from clinically defined medicines in their character as well as in their medicinal value.
- They are based mostly on herbal products.
- Usually, they are multi-drug formulations including animal and mineral products as essential components or additives.
- In herbal therapy, data on pre-clinical investigations are often incomplete although in majority cases the therapeutic experiences have been accumulated over centuries.
- Some of them follow practices based on, for example, mistaken beliefs, faulty experimentation, or inaccurate information that can be dangerous.
- They mostly include empirically defined doses and course of medication.
- The identity of plant species used is often controversial.
- Safety measures are poorly adopted, in most cases.
- Additives are frequently used; many of them also have therapeutic actions

**Cardiovascular active agents - mode of use**

According to available literature several plants used for the treatment of CVD and only a small number of these have received scientific and medical evaluation to assess their efficacy. A number of natural and synthetic products have been studied for their cardiovascular activities. The wide variety of chemical classes indicates that a variety of mechanisms must be involved in the lowering of the CVD risks. These drugs act in a variety of ways:

- As cardiotonic compounds
- By direct vasodilatory effect on blood vessels
- As CNS depressants
- As cAMP phosphodiesterase and platelet aggregation inhibitors
- As β-adrenergic blocking agents
- As antihyperlipidemic agent
- As antioxidant defense

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Plant drugs as cardiotonic compounds

The cardiac glycoside, peruvoside (Bisset et al., 1962) from *Thevetia pervulana* and asclepin from *Asclepias curassavica* act as cardiotonic agent for use in therapy (Patnaik and Kohler, 1978). A xanthone, Mangostin from *Garcinia mangostana* Linn. and its derivative, mangostin-3,6-di-o-glucoside have shown myocardial stimulation and rise in blood pressure which was partially blocked by propranolol.

Plant drugs as antihypertensives

The story of clinical testing of *Rauwolfia* in India is well known. *Rauwolfia* and related alkaloids have antihypertensive potential (Muller et al., 1952). The use of *Coleus spp.* in the treatment of heart diseases and other disorders has been reported in Ayurvedic material medica. Dubbey et al. (1981) reported that a diterpenoid, from *Coleus forskohlii* possesses hypotensive and positive inotropic effects, and exhibit nonspecific spasmolytic activity. The hypotensive activity of *Euphorbia maddeni* has been found to be due to flavonoid glycosides, kaempferol 4'-O-glucoside and hyperin.

Plant drugs as antihyperlipidaemics

Malhotra et al. (1970) reported the importance of guggulipid as antihyperlipidaemic drug. Salai guggal, the oleoresin of *Boswellia serrata* has been reported as an antiarthritic and antihyperlipidaemic agent (Atal et al., 1980). Their mechanisms of action is attributed to the presence of disulphides and sulphoxide substances (Reuter et al., 1990). The hypolipidemc effect of garlic (*Allium sativum* Linn) and onion (*Allium cepa* Linn) were demonstrated simultaneously by Mathew and Augusti (Augusti and Mathew, 1973) in India and Itokawa et al. (1973) in Japan. The hypocholesterolemic effect of garlic in humans was reported by Augusti (1977).

Plant drugs as antioxidants

Increased oxidative stress and antioxidant deficit have been suggested to play a major role in CVD. The antioxidant activities of the phenolic contents of *Centella asiatica* prevent CVD risks against adriamycin induced cardiomyopathy in rats (Gnanapragaam et al., 2004). Alkyl derivatives of cysteine sulphoxide, disulphides, polysulphides and their sulphoxides such as allicins and ajoene type compounds from *Allium sativum* Linn., *Allium cepa* Linn. are strong antioxidants (Reuter et al., 1990).
Flavonoids, gallotannins, ellagic acids, phyllembilic acids and related polyphenols, ascorbic acid present in *Emhlica officinalis* are strong antioxidants (Jeena and Kuttan, 1995).

The medicinal preparations in traditional medicines contain a variety of herbal and non-herbal ingredients that are thought to act on a variety of targets by various modes and mechanisms. Abana, a herbomineral preparation offers partial heart protection by protecting the membrane bound enzymes. Cardioprotective activity of Abana is comparable to other natural products such as gugulip, guggulsterone, colenol which are reported earlier (Tandon *et al.*, 1995). Polyherbal formulation and antioxidant compounds have shown protective effects in doxorubicin induced cardiotoxicity without reducing their therapeutic efficacy (Koti *et al.*, 2009).

Betaine from several microorganisms, plants, and animals has been reported to play a role in atherosclerosis and osteoporosis by reducing blood levels of homocysteine (Ganesan *et al.*, 2007). Oleanolic acid, a triterpenoid compound that exists widely in food and herbs, has a variety of biological effects, such as antioxidant, antihyperlipidemic (Senthil *et al.*, 2007), antiarrhythmic and cardiotonic effects (Somova *et al.*, 2004). Naringin, a flavonone found in grape fruit and related citrus species is reported to possess cholesterol lowering and antioxidant effects (Rajadurai and Prince, 2007).

**Aegle marmelos** — LITERATURE

*Aegle marmelos* (L.), popularly known as Maredu in Telugu, belongs to family Rutaceae. In English it is called as Bengal quince, golden apple, stone apple and bilwa in Sanskrit. It belongs to India, grown throughout the sub-continent as well as in Burma, Bangladesh and Pakistan. Its medicinal properties have been described in the ancient medical treatise in Sanskrit, Charka Sambha. It is used in several indigenous systems of medicine in India, China, Burma and Sri Lanka (Kamalakkannan and Prince, 2003).

Bael is considered to be an auspicious tree by the Hindus and has been called 'shivadruma' (the tree of Shiva). It is often planted near temples and the leaves and wood of the plant have been used for worshipping Lord Shiva and Parvati since time
Fig 12. Fruits of *Aegle marmelos*.
immemorial. It attain a height of 12 m, has unusual branches bearing straight spines. The bark is shallowly furrowed and corky, nearly 2 cm wide, white, fragrant, borne in clusters of 4-7. Its shallow calyx has 5 short broad teeth, pubescent outside. The flower usually has 5 petals of a pale greenish white colour, dotted with oil glands. Fruits are (Fig 12) globose, round, pyriform, oval or oblong, 5-20cm in diameter, with grayish yellow pericarp and sweet pulp, yellow to orange in colour. Seeds are numerous, compressed, closely packed and arranged in the cells surrounded by transparent mucilage, which on drying becomes hard. The whole plant and various parts (root, root bark, stem bark, leaf and fruit) are all used in traditional medicines (Sing and Malik, 2000).

**Phytochemical constituents**

A large number of individual phytochemical constituents have been identified in various parts of *A. marmelos*. These include alkaloids, terpenoids, coumarins, tannis, sugars and steroids.

**Phytochemical constituents present in root**

Auraptene, marmin, umbelliferone, and lupeol are reported to be present in the roots. The alcoholic extract of the root gave xanthotoxin, psoralen, dimethoxycoumarin, scopoletin, tembamide, umbelliferone, marmesin, marmin, skimmianine, and a glycoside skimmin (Varadarajan, 1985).

**Phytochemical constituents present in leaf**

Manandhar *et al.*, (1978) isolated four new alkaloids, O-(3,3-dimethylallyl)-halfordinol, N-2-ethoxy-2-(4-methoxyphenyl) ethylcinnamamide, N-2-[4-(3',3'-dimethylallyloxy)phenyl]ethylcinnamamide, and N-2-methoxy-2-(4-methoxyphenyl)-ethylcinnamamamide, besides aegelenine and aegeline from the leaves of bael. Condensed tannins, phlobatannins, flavan-3-ols, leucoanthocyanins, anthocyanins, flavonoid glycosides, skimmianine and r-sitosterol are also present.

On steam distillation, the leaves yield an essential oil, which contains: α,d-phellandrene (35%), cineol (3%), p-cymene (17%), citronellal (21%), citral (10%) and cumminaldehyde (5%). The essential oil from the twigs contains cineol (40-45%) and α,d-phellandrene only.
Phytochemical constituents present in gum

Graded hydrolysis of purified bael gum gives three neutral and two acidic oligosaccharides, together with monosaccharides. These sugars were identified through periodate oxidation, methylation, reduction with lithium aluminum hydride, co-chromatography, and preparation of crystalline derivatives. The neutral oligosaccharides were characterized as 3-0-(3-D-galactopyranosyl-L-arabonise, and 5-0-(3-D-galactopyranosyl-L-arabonise, and 3-0-(3-D-galactopyranosyl-D-galactose, and acidic oligosaccharides as 3-0-(β-D-galactopyranosyluronic acid)-3-0-(β-D-galactopyranosyl-D-galactose (Roy et al., 1975).

Phytochemical constituents present in seed

Linoleic acid, palmitic acid, stearic acid and linolenic acid are reported to be present in the seed (Sing and Malik, 2000).

Phytochemical constituents and nutritional value of fruit

Aegelin, marmelosin, marmesin, marmin, umbelliferone, alloimperatorin, imperatorin, marmelide, psoralen, rhamnose, scoparone, scopoletin, skimmin, xanthotoxol, tannic acid and β-sitosterol are reported to be present in the fruits of A. marmelos (Kamalakannan and Prince, 2003). Table 3 depicts the nutritional value of Bael fruit per 100 gms (Parichha, 2004).

Biological activity of A. marmelos - literature

The whole plant and various parts (root, root bark, stem bark, leaf and fruit) are used in traditional medicines (Kapoor, 1990; Singh and Malik, 2000). Nadkarini (1976) lists a number of uses for A. marmelos in the Ayurvedic, Siddha and Unani traditional systems of herbal medicine. A large number of putative biological actions have been claimed for A. marmelos in the literature includes:

- Hypoglycaemic, and antihyperglycemic activity (Kamalakannan and Prince, 2005; Kesari et al., 2006; Narendhirakannan et al., 2006; Narender et al., 2007).

- Antiparasitic (Haider et al., 1991), antimicrobial (Rani and Khullar, 2004; Saayi Krushna and Lakshmi Devi, 2005), antiviral (Badam et al., 2002) and antifungal (Rana et al., 1997) activity.
Table 3. Nutritional value of Bael fruit per 100 gms (Saswati Parichha, 2004).

<table>
<thead>
<tr>
<th>Nutritional constituent</th>
<th>Value</th>
</tr>
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<tbody>
<tr>
<td>Protein (g)</td>
<td>1.8</td>
</tr>
<tr>
<td>Fat (g)</td>
<td>0.3</td>
</tr>
<tr>
<td>Mineral (g)</td>
<td>1.7</td>
</tr>
<tr>
<td>Fibre (g)</td>
<td>2.9</td>
</tr>
<tr>
<td>Carbohydrate (g)</td>
<td>31.8</td>
</tr>
<tr>
<td>Calcium (mg)</td>
<td>85</td>
</tr>
<tr>
<td>Phosphorus (mg)</td>
<td>50</td>
</tr>
<tr>
<td>Vitamin-C (mg)</td>
<td>8</td>
</tr>
<tr>
<td>Potassium (mg)</td>
<td>600</td>
</tr>
<tr>
<td>Vitamin-B</td>
<td>Rich in $B_1$ and $B_2$</td>
</tr>
<tr>
<td>Edible portion</td>
<td>64%</td>
</tr>
<tr>
<td>Moisture (g)</td>
<td>61.5</td>
</tr>
<tr>
<td>Energy (K.cal)</td>
<td>137</td>
</tr>
</tbody>
</table>
Fig 13. 3’D-Structures of Phytochemicals of AMUFAEt

1, 5-dihydroxy-6-methoxy-2-methylantraquinone

Marmin

Umbelliferone
Impertonin

Marmesin
Free radical scavenging (Jagetia and Baliga, 2004), antioxidant (Kamalakannan and Prince, 2003) and radioprotective (Jagetia & Venkatesh, 2005) activity.

Anti-inflammatory, antipyretic and analgesic activity (Arul et al., 2005).

Anticancer (Costa Lotufo et al., 2005; Jagetia et al., 2005), antidiarrhoeal (Shoba and Thomas, 2001; Mazumder et al., 2006) activity.

Immunomodulatory (Citarasu et al., 2006), antileukoderma and antihyperthyroid (Kar et al., 2002; Panda & Kar, 2006) activity.

Cardiotonic, cardioprotective (Prince a Rajadurai, 2005), antihyperlipidemic (Rajadurai & Prince, 2005) and anticoagulant activity.

Induction of myocardial infarction/stress in experimental animals

Understanding CVD pathology would require serial studies of life time. The use of animal model largely circumvents this problem. The disease can also be induced in many animals, particularly laboratory rodents, using a variety of chemical agents and other experimental procedures. Both the spontaneous and the induced syndrome are under intensive study and they offer promise of new insight into CVD in man. The stable incidence of CVD risks in animal colonies affords a powerful tool for the study of preventive therapies for the disease. Isoproterenol (IPL) (Fig. ), clenbuterol, epinephrine and adriamycin (doxorubicin) are widely used to induce experimental cardiac infarction/stress in animals. The cytotoxic action of these agents are mediated by ROS, depletion in antioxidant levels, alterations in membrane permeability, loss of membrane functions and integrity and altered lipid metabolism (Todd et al., 1980; McCord, 1988). However, the source of their generation is different in the case of adriamycin (doxorubicin) (Koti et al., 2009), IPL, epinephrine (Miran et al., 2008) and clenbuterol (Valori et al., 1967; Sharma and Garg, 2003) (Fig. 14).
Fig 14. Chemical structure - Adriamycin, Clenbuterol, Isoproterenol and Epinephrine.
Alquist (1948) demonstrated that catecholamines act upon two receptors α and β. The α-receptor actions are through vasoconstriction where as β-receptor actions are through vasodilation. Valori et al. (1967), and Prakesh et al. (1972) found that association between catecholamine content in blood or urine and the severity of MI. Lands et al. (1967) reported that β-receptors found in adipose and cardiac tissues and have relatively high affinity to IPL compared to catecholamine. IPL is a synthetic β-adrenergic agonist that causes severe stress in the myocardium, resulting in infarct-like necrosis of heart muscle (Rona et al., 1959; Sushmakumari et al., 1989) with an increase in the level of myocardial lipids (Manjula and Shyamaladevi, 1993). It mainly increases LDL-cholesterol level in the blood which in turn leads to the build-up of harmful deposits in the arteries thus favoring coronary heart disease (Goldstein and Brown, 1984). The multitude of free radicals generated during oxidative stress associated with IPL-induced MI can damage every major cellular component, including carbohydrates, membrane lipids, protein and DNA (Downye, 1990). The patho-physiological consequences of such uncontrolled injury are widespread tissue damage and associated contractile dysfunction, arrhythmias, depletion of endogenous antioxidant network and enhanced lipid peroxidation resulting in increased myocardial malondialdehyde (MDA) content (Freeman and Caspo, 1982). IPL administration is known to produce electrocardiography and enzymatic changes in experimental animals. Moreover, IPL promotes lipolysis in the myocardium (Lech et al., 1977) and causes irreversible damage to the myocardial membrane (Chein et al., 1978).

AIM AND SCOPE

Many herbal extracts (Nandave et al., 2007; Rajadurai and Prince, 2007; Raju et al., 2008) and formulations (Bafna and Balaram, 2006; Suchalatha and Shyamaladevi, 2004; Sasikumar and Shyamaladevi, 2000) are reported to have cardioprotective activity. Fruit extract of A. marmelos is reported to possess antidiabetic, antioxidant, antimicrobial, antidiarrhoeal, anticoagulant, antihyperlipidaemic, radioprotective and antiulcer activities in experimental animals. The antidiabetic, antihyperlipidaemic and antioxidant effects of this plant suggest a possible cardioprotective effect. However, our exhaustive literature review could not
reveal any published data on the cardioprotective effect of *A. marmelos* unripe fruit aqueous extract (AMUFAEt) on MI in experimental animals.

The present study includes:

- To measure the body weight and heart weight to assess heart co-efficient in control and experimental rats.

- To assess the oxidative activity of AMUFAEt in IPL administered rats and to evaluate the protective effect of AMUFAEt and the extent of lipid peroxidation in the serum and heart tissue.

- To study the antioxidant activity of AMUFAEt in IPL administered rats by measuring heart tissue antioxidant enzymes i.e., glutathione (GSH), glutathione reductase (GR), glutathione-s-transferase (GST), glutathione peroxidase (GPx), catalase (CAT) and superoxide dismutase (SOD).

- To evaluate inotropic effect of AMUFAEt on the activities of heart tissue ATPases VIZ., Na\(^+\)-K\(^+\)-ATPase, Mg\(^{2+}\)-ATPase and Ca\(^{2+}\)-ATPase.

- To study the antioxidant activity of AMUFAEt in IPL administered rats by measuring heart tissue antioxidant enzymes i.e., glutathione (GSH), glutathione reductase (GR), glutathione-s-transferase (GST), glutathione peroxidase (GPx), catalase (CAT) and superoxide dismutase (SOD).

- To find out hypolipidemic effect of AMUFAEt on the plasma and heart tissue lipid profile (TC, HDL-C, LDL-C, VLDL-C, PL, FFA and TG) and phospholipids alterations in heart tissue.

- To find out cardiotonic property of AMUFAEt in IPL administered rats on the plasma electrolytes (sodium, potassium, calcium and iron) and transaminases (AST, ALT) and LDH, CK, ALP, taurine and troponin-T were measured in serum and heart tissues. Cardiac specific risk factors like CRP, Lp-a, Apo-A1, Apo-B and homocysteine in plasma were estimated.

- To understand the contribution of AMUFAEt protection in IPL-induced cardiac stress, by studying the electrocardiographic (ECG) and histopathological changes.

- To find out phytochemical efficacy of AMUFAEt, the total phenolic content and flavonoids were measured.

- To assess the extent of scavenging activity of AMUFAEt by in vitro method on hydroxyl radical scavenging, superoxide scavenging, DPPH radical scavenging, nitric oxide scavenging capacities and reducing power.


