Introduction
1. Epidemiology of Cardiovascular Disease:

Cardiovascular disease (CVD) is the number one cause of death worldwide (WHO, 2002b) and is the world’s largest killer, claiming 17.1 million lives a year (WHO, 2010). At the beginning of the 20th century, CVD was responsible for less than 10% of all deaths worldwide, but by 2001 that figure was 30%. CVD was the underlying cause for 36.3% of all deaths in 2004. About 80% of the global burden of CVD death occurs in low- and middle-income countries (WHO, 2010). Murray and Lopez predicted that CVD will be the leading cause of death and disability worldwide by 2020 mainly because it will increase in low- and middle-income countries. Nearly 50% of all deaths in high-income countries and about 28% of deaths in low and middle-income countries are the result of CVD (Mathers et al., 2006; WHO, 2002a). By 2030, almost 23.6 million people will die from CVD, mainly from heart disease and stroke. It is projected to remain the single leading cause of death (WHO, 2010). African Americans have a higher prevalence of CVD in comparison to Caucasian and Mexican Americans (American Heart Association & American Stroke Association, 2007; 2008). Even though 80% of CVD deaths occur in low- and middle-income countries, the death rates for most regions are still below the rate for high-income countries, which are 320 per 100,000 population annually. The marked exception is Europe and Central Asia, which has a rate of 690 CVD deaths per 100,000 population. The most common causes of CVD morbidity and mortality are ischemic heart disease (IHD), stroke and congestive heart failure (CHF) which accounts for at least 80% of the burden of CVD. In 2001 IHD was responsible for 7.3 million deaths and 58 million disability-adjusted life years (DALYs) lost worldwide (WHO, 2002b). In 2004, an estimated 7.2 million of these deaths were due to coronary heart disease and 5.7 million were due to stroke (WHO, 2010).

Seventy-five per cent of global deaths and 82% of the total DALYs resulting from IHD occurred in the low and middle-income countries. Some regions of India appear to be in the first phase, whereas others are in the second or even the third phase in experiencing an alarming increase in heart disease, which seems to be linked to changes in lifestyle and diet, rapid urbanization, and possibly an underlying genetic
Estimated Proportional Mortality (%), India, 2004

All NCDs 50.2%

Circulatory 26.1%

Injuries 12.0%

Other NCDs 8.4%

Diabetes 5.7%

Respiratory 2.7%

Other Causes 40.5%

Cancers 7.4%

Injuries 9.5%

Due to: A/C: Other major causes

Source: Author: Data: Indian National Cause of Death Register


5
component. In 2004, deaths due to non-communicable diseases in India were twice those from communicable diseases (WHO 2010). The WHO estimates that, by 2010, 60% of the world’s cardiac patients will be in India. About 50% of CVD-related deaths occur among people younger than 70, compared with about 22% in the West. Between 2000 and 2030, about 35% of all CVD deaths in India will occur among those age 35 to 64, compared with only 12% in the United States and 22% in China (Leeder et al., 2004). The CVD burden afflicts both men and women, with cardiovascular deaths accounting for 34% of all deaths in women and 28% in men in 1998 (WHO, 1999). CVD mortality per cent in India has reported as IHD 15% in men, 13.1% in women, rheumatic heart diseases 0.9% in men, 1.0% in women and other CVD 2.6% in men, 4% in women (Mathers et al., 2004). Between 1990 and 2020, the increase in IHD mortality (120% in women and a 137% in men) in the developing countries is expected to be much greater than among developed countries (29% and 48%, respectively) (Murray et al., 1996). It was suggested that endogenous hormones protect against CVD in women, and that estrogen deprivation after menopause increased their cardiovascular risk (Kannel et al., 1978). It was also evident that among the Asian Indian women, postmenopausal women were more susceptible to diabetes and cardiovascular diseases than premenopausal women (Ghosh and Bhagat, 2009).

In a survey conducted in 45 rural villages of Andhra pradesh in India, 32% of all deaths were due to CVD, outranking infectious diseases, which were responsible for 13% giving clear evidence that the epidemic has reached its advanced stage even in rural India (Joshi et al., 2006). CHD rates have ranged from 1.6% to 7.4% in rural populations and 1% to 13.2% in urban populations (Gupta, 2008). In 2000, there were an estimated 29.8 million people with CHD in India, out of a total estimated population of 1.03 billion people, or a nearly 3% overall prevalence (Gupta, 2008). In one study, Muslim men have been shown to have the highest CHD prevalence rates and Christian men have been shown to have the lowest CHD prevalence rates (Gopianth, 1995). However, another study demonstrated highest CHD prevalence
rates in Hindu men (Gupta, 2002), whereas two other studies have reported the highest rates in Gujarati men (Chadha, 1992; Gopianth, 1992).

Table 1. Estimate of Ischemic Heart Disease Mortality (Thousands) by Region and Sex and Projected Changes between 1990 and 2020

<table>
<thead>
<tr>
<th>Region</th>
<th>Women 1990</th>
<th>Women 2020</th>
<th>% Increase</th>
<th>Men 1990</th>
<th>Men 2020</th>
<th>% Increase</th>
</tr>
</thead>
<tbody>
<tr>
<td>EME</td>
<td>838</td>
<td>1107</td>
<td>32</td>
<td>829</td>
<td>1209</td>
<td>48</td>
</tr>
<tr>
<td>FSE</td>
<td>559</td>
<td>702</td>
<td>26</td>
<td>466</td>
<td>712</td>
<td>52</td>
</tr>
<tr>
<td>Total developed countries</td>
<td>1397</td>
<td>1809</td>
<td>29</td>
<td>1297</td>
<td>1921</td>
<td>48</td>
</tr>
<tr>
<td>India</td>
<td>556</td>
<td>1197</td>
<td>115</td>
<td>619</td>
<td>1405</td>
<td>127</td>
</tr>
<tr>
<td>China</td>
<td>377</td>
<td>684</td>
<td>81</td>
<td>386</td>
<td>811</td>
<td>110</td>
</tr>
<tr>
<td>OAI</td>
<td>227</td>
<td>552</td>
<td>143</td>
<td>233</td>
<td>581</td>
<td>149</td>
</tr>
<tr>
<td>SSA</td>
<td>117</td>
<td>263</td>
<td>125</td>
<td>92</td>
<td>222</td>
<td>141</td>
</tr>
<tr>
<td>Latin America</td>
<td>169</td>
<td>412</td>
<td>144</td>
<td>179</td>
<td>444</td>
<td>148</td>
</tr>
<tr>
<td>Middle East</td>
<td>291</td>
<td>717</td>
<td>146</td>
<td>319</td>
<td>874</td>
<td>174</td>
</tr>
<tr>
<td>Total developing countries</td>
<td>1717</td>
<td>3825</td>
<td>120</td>
<td>1828</td>
<td>4337</td>
<td>137</td>
</tr>
<tr>
<td>World</td>
<td>3134</td>
<td>5634</td>
<td>80</td>
<td>3125</td>
<td>6259</td>
<td>100</td>
</tr>
</tbody>
</table>

EME indicates established market economics; FSE, formerly socialist economies; OAI, other Asian and Pacific Island countries; SSA, Sub-Saharan Africa; LAC, Latin American/Caribbean; and MEC, Middle East Crescent.
2. Cardiovascular Physiology:

The cardiovascular system consists of the heart, blood vessels, and blood. Blood is propelled away from the heart in the arteries and returns to the heart in the veins.

2.1. Anatomic organization of the cardiovascular system

The heart has four chambers. The two atria serve as reservoirs for blood returning to the heart. The two ventricles are pumps that propel blood through the circulation. A septum divides the heart into right and left sides.

2.2. Cardiac cycle

The cardiac cycle consists of two phases, called diastole and systole. During diastole, oxygenated blood returns from the lungs to the left atrium, while deoxygenated blood from other parts of the body fills the right atrium. The sinus node generates an impulse that forces the two atria to contract. In this phase, the tricuspid and mitral valves are open, and blood is propelled from the atria into the relaxed ventricles. By the end of diastole, the electric impulse reaches the ventricles, causing them to contract. During systole, the contracting ventricles close the tricuspid and mitral valves. Shortly afterward, the pressure of the blood inside the ventricles rises sufficiently to force the pulmonary and aortic valves to open, and blood is ejected into the pulmonary artery and the aorta. Heart failure results from conditions that impair the ability of the heart to fill with, or to pump out, sufficient blood. The most common cause of heart failure is myocardial infarction (MI).

Heart rate is the number of heartbeats per unit of time, typically expressed as beats per minute (bpm). The typical healthy resting heart rate in adults is 60–80 bpm, with rates below 60 bpm referred to as bradycardia, and rates above 100 bpm referred to as tachycardia.

Blood pressure is the pressure exerted by circulating blood upon the walls of blood vessels. Normal blood pressure of a healthy person is 120/80 mm Hg, usually expressed in terms of the systolic pressure (120) over diastolic pressure (80). The risk of cardiovascular disease increases progressively in the abnormalities of normal blood pressure.
Fig: Heart anatomy

Heart and Valves

- Aorta
- Left Atrium
- Mitral Valve
- Mitral Valve Chordae
- Left Ventricle
- Pulmonary Valve
- Right Atrium
- Tricuspid Valve
- Right Ventricle
2.5. Cardiac muscle structure and function

In all types of muscle, the key event causing contraction is by the process of excitation-contraction coupling.

2.5.1. Excitation-Contraction coupling

The contraction of cardiac muscle in all animals is initiated by chemical impulses. The cells that create these rhythmical impulses are called pacemaker cells. Pacemaker cells spontaneously generate action potentials, which spread through gap junctions. A transverse tubule (T tubule) is a deep invagination of the sarcolemma, which is the plasma membrane, only found in skeletal and cardiac muscle cells. Action potentials conducted along T tubules are associated with opening of the voltage-gated Ca\(^{2+}\) channels and entry of extracellular Ca\(^{2+}\) into the cells. Ca\(^{2+}\)-induced Ca\(^{2+}\) release is triggered from internal sarcoplasmic reticulum stores. Almost all Ca\(^{2+}\) that interacts with troponin C to initiate contraction is derived from internal stores. Contraction occurs via the sliding filament mechanism. Relaxation of cardiac muscle depends on the removal of free cytosolic Ca\(^{2+}\). In cardiac muscle, Ca\(^{2+}\) is moved back into the extracellular fluid via the Na\(^+/-\)Ca\(^{2+}\) exchangers in the sarcolemma, which couple the inward transport of 3 Na\(^+\) from the extracellular fluid to the outward transport of one intracellular Ca\(^{2+}\). In addition, Ca\(^{2+}\) ATPase is used by all types of muscle and pumps Ca\(^{2+}\) back into the sarcoplasmic reticulum.

2.5.3. Sliding Filament Theory

The mechanism of active force generation in all muscle types is based on thin filaments being pulled over thick filaments. In a relaxed skeletal muscle, contraction is inhibited by the tropomyosin-troponin complexes, which obscure the active site on actin and prevent cross-bridge binding. When the muscle is stimulated, the intracellular Ca\(^{2+}\) concentration increases and Ca\(^{2+}\) binds to troponin C. The resulting conformational change exposes active sites on actin. A cycle of events now occurs in which myosin cross-bridges bind to actin, perform a powerstroke, detach, become cocked again, and then reattach. The cycle repeats continued in the presence of sufficient Ca\(^{2+}\) and ATP. As a result, thin filaments from each end of the sarcomere move toward the center by sliding over thick filaments, causing neighboring Z disks to
approach each other. In the example shown in, muscle shortening has occurred and the sarcomere length is reduced.

**Fig: Excitation-Contraction coupling**

**Fig: Sliding filament theory of muscle contraction.** Ca\(^{2+}\) binding to troponin C causes actin active sites to be exposed (compare panels 2 and 3). In the presence of ATP, myosin repeats a cycle of binding to actin, performance of a power stroke, detaching, becoming cocked, and reattaching further along the actin molecule (panel 4). Actin filaments are drawn over myosin. In this example, muscle shortening occurs (compare panels 1 and 5). ADP, adenosine diphosphate.
3. Cardiovascular disease types

CVD is a group of disorders or diseases of the heart and blood vessels, including heart attack and stroke.

Types of CVD include coronary heart disease, stroke, rheumatic heart disease, deep vein thrombosis and pulmonary embolism and congenital heart disease (WHF, 2011).

3.1. Coronary heart disease

Coronary heart disease (CHD) is a narrowing of the small blood vessels that supply blood and oxygen to the heart that can lead to a heart attack. CHD is also called coronary artery (CAD) or atherosclerotic heart disease. It is the end result of the accumulation of atheromatous plaques within the walls of the coronary arteries that supply the myocardium (the muscle of the heart) with oxygen and nutrients. Typically, coronary artery disease occurs when part of the smooth, elastic lining inside a coronary artery develops atherosclerosis.

3.1.1. Atherosclerosis

Atherosclerosis is an inflammatory disease. Because high plasma concentrations of cholesterol, in particular those of low-density lipoprotein (LDL) cholesterol, are one of the principal risk factors for atherosclerosis (National Cholesterol Education Program 1993). The process of atherogenesis initiate by the accumulation of lipids within the artery wall. The lesions of atherosclerosis occur principally in large and medium-sized arteries and can lead to ischemia of the heart, brain, or extremities, resulting in infarction. The most recent version of hypothesis emphasizes endothelial dysfunction response to injury of atherosclerosis. Possible causes of endothelial dysfunction leading to atherosclerosis include elevated and modified LDL-C; free radicals caused by cigarette smoking, hypertension, and diabetes mellitus; genetic alterations; elevated plasma homocysteine concentrations; infectious
microorganisms such as herpesviruses or *Chlamydia pneumonia*; and combinations of these or other factors (Russell Ross 1999).

Atherosclerotic lesions begin as fatty streaks underlying the endothelium of large arteries. Recruitment of macrophages and their subsequent uptake of LDL-derived cholesterol are the major cellular events contributing to fatty streak formation. The oxidative modifications in the lipid and apolipoprotein B components of LDL drive the initial formation of fatty streaks (Steinberg and Witztum, 1999). Although the recruitment of monocytes to the arterial wall and their subsequent differentiation into macrophages may initially serve a protective function by removing cytotoxic and proinflammatory oxLDL particles or apoptotic cells, progressive accumulation of macrophages and their uptake of oxLDL ultimately leads to development of atherosclerotic lesions. The recruitment of monocytes to lesion-prone sites of large arteries is regulated by cell adhesion molecules that are expressed on the surface of endothelial cells in response to inflammatory stimuli. The development of macrophage "foam cells" that contain massive amounts of cholesterol esters is a hallmark of both early and late atherosclerotic lesions. Cholesterol accumulation in these cells is thought to be mediated primarily by uptake of modified forms of LDL (Yamada et al., 1998). The transition from the relatively simple fatty streak to the more complex lesion is characterized by the immigration of smooth muscle cells from the medial layer of the artery wall past the internal elastic lamina and into the intimal, or subendothelial, space. Intimal smooth muscle cells may proliferate and take up modified lipoproteins, contributing to foam cell formation, and synthesize extracellular matrix proteins that lead to the development of the fibrous cap (Ross, 1999). Although advanced atherosclerotic lesions can lead to ischemic symptoms as a result of progressive narrowing of the vessel lumen, acute cardiovascular events that result in myocardial infarction and stroke are generally thought to result from plaque rupture and thrombosis. Plaque rupture exposes plaque lipids and tissue factor to blood components, initiating the coagulation cascade, platelet adherence, and thrombosis. Plaque ruptures associated with acute myocardial infarction generally occur at the shoulder regions of the plaque and are more likely to occur in lesions with thin fibrous
caps, a relatively high concentration of lipid-filled macrophages within the shoulder region, and large necrotic cores (Lee and Libby, 1997).

Fig: Initiating Events in the Development of a Fatty Streak Lesion
LDL is subject to oxidative modifications in the subendothelial space, progressing from minimally modified LDL (mmLDL) to extensively oxidized LDL (oxLDL). Monocytes attach to endothelial cells that have been induced to express cell adhesion molecules by mmLDL and inflammatory cytokines. Adherent monocytes migrate into the subendothelial space and differentiate into macrophages. Uptake of oxLDL via scavenger receptors leads to foam cell formation. OxLDL cholesterol taken up by scavenger receptors is subject to esterification and storage as lipid droplets, or is exported to extracellular HDL acceptors via cholesterol transporters, such as ABC-A1.

Fig: Lesion Progression
Interactions between macrophage foam cells, Th1 and Th2 cells establish a chronic inflammatory process. Cytokines secreted by lymphocytes and macrophages exert both pro- and antiatherogenic effects on each of the cellular elements of the vessel wall. Smooth muscle cells migrate from the medial portion of the arterial wall, proliferate and secrete extracellular matrix proteins that form a fibrous plaque.

Fig: Plaque Rupture and Thrombosis
Necrosis of macrophage and smooth muscle cell-derived foam cells leads to the formation of a necrotic core and accumulation of extracellular cholesterol. Macrophage secretion of matrix metalloproteinases and neovascularization contribute to weakening of the fibrous plaque. Plaque rupture exposes blood components to tissue factor, initiating coagulation, the recruitment of platelets, and the formation of a thrombus.
3.1.2. Myocardial infarction

MI or acute myocardial infarction (AMI), commonly known as a heart attack is the interruption of blood supply to a part of the heart, causing heart cells to die. This is most commonly due to occlusion or blockage of a coronary artery following the rupture of a vulnerable atherosclerotic plaque, which is an unstable collection of lipids 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 or death (infarction) of myocardium.

The death of myocardial cells first occurs in the area of myocardium most distal to the arterial blood supply: the endocardium. As the duration of the occlusion increases, the area of myocardial cell death enlarges, extending from the endocardium to the myocardium and ultimately to the epicardium. The area of myocardial cell death then spreads laterally to areas of watershed or collateral perfusion. Generally, after a 6 to 8 hour period of coronary occlusion, most of the distal myocardium has died. The extent of myocardial cell death defines the magnitude of the MI. If blood flow can be restored to at-risk myocardium, more heart muscle can be saved from irreversible damage or death.

3.1.3. Angina pectoris

Angina pectoris commonly known as angina, is severe chest pain (Merck Medicus, 2009) due to ischemia of the heart muscle, generally due to obstruction or spasm of the coronary arteries. The main cause of angina is due to atherosclerosis of the cardiac arteries. Angina pectoris is classified as stable angina, unstable angina and micro vascular angina.

Stable angina is also known as effort angina, typical presentations of stable angina is that of chest discomfort and associated symptoms precipitated by some activity like running and walking with minimal or non-existent symptoms at rest. Unstable angina (UA) also known as crescendo angina is defined as angina pectoris
that changes or worsens (Merck Medicus, 2009). UA may occur unpredictably at rest which may be a serious indicator of an impending heart attack. Micro vascular Angina or Angina Syndrome X is characterized by angina-like chest pain, but have different causes. The cause of Micro vascular Angina is unknown, but it appears to be the result of poor function in the tiny blood vessels of the heart, arms and legs (Guyton and Arthur, 2006). Symptoms in most patients with angina complain chest discomfort rather than actual pain.
3.2. Stroke

Stroke or cerebrovascular disease, previously known medically as a cerebrovascular accident (CVA), is the rapidly developing loss of brain functions due to disturbance in the blood supply to the brain. This can be due to ischemic lack of blood flow caused by blockage like thrombosis, arterial embolism, or a hemorrhage (Sims and Muyderman, 2009). As a result, the affected area of the brain is unable to function, which might result in an inability to move one or more limbs on one side of the body, inability to understand or formulate speech, or an inability to see one side of the visual field (Donnan et al., 2008).

The two major mechanisms causing brain damage in stroke are ischemia and hemorrhage. Each of these stroke subtypes is mechanistically quite different. Ischemic stroke is generally caused by embolism and/or thrombosis, hemorrhage by rupture of small penetrating vessels, and by rupture of intracranial aneurysms contained within the subarachnoid space surrounding the brain.

3.3. Rheumatic heart disease

Rheumatic fever (RF) is an autoimmune disease caused by the gram-positive bacteria Streptococcus pyogenes that follows a nontreated throat infection in susceptible children and teenagers. The disease manifests as polyarthritis, carditis, chorea, erythema marginatum, and/or subcutaneous nodules. Carditis, the most serious complication, occurs in 30% to 45% of RF patients and leads to chronic rheumatic heart disease (RHD), which is characterized by progressive and permanent valvular lesions. Mitral and aortic regurgitation are the most common events caused by repeated valvulitis (Luiza Guilherme and Jorge Kalil, 2010).

The mechanism to cause RHD is molecular mimicry. It is defined as a sharing of epitopes between antigens of the host and the infectious agent, which in the case of RF/RHD is S. pyogenes.
3.4. *Deep vein thrombosis and pulmonary embolism*

Deep vein thrombosis also known as deep venous thrombosis and usually abbreviated as DVT is the formation of a blood clot i.e. thrombus in a deep vein. It is a form of thrombophlebitis which is the inflammation of a vein with clot formation. Deep vein thrombosis commonly affects the leg veins (such as the femoral vein or the popliteal vein) or the deep veins of the pelvis. Occasionally the veins of the arm are affected (if spontaneous, this is known as Paget-Schrötter disease). A DVT can occur without symptoms, but in many cases the affected extremity will be painful, swollen, red and warm.

The most serious complication of a DVT is that the clot could dislodge and travel to the lungs, which is called a pulmonary embolism (PE). Obstruction of the blood flow through the lungs and the resultant pressure on the right ventricle of the heart leads to the symptoms and signs of PE. Symptoms of pulmonary embolism include difficulty breathing, chest pain on inspiration, and palpitations. Clinical signs include low blood oxygen saturation and cyanosis, rapid breathing, and a rapid heart rate. Severe cases of PE can lead to collapse, abnormally low blood pressure, and sudden death (Goldhaber, 2005).

According to Virchow's triad, venous thrombosis occurs via three mechanisms: decreased flow rate of the blood, damage to the blood vessel wall and an increased tendency of the blood to clot (hypercoagulability). Several medical conditions can lead to DVT, such as compression of the veins, physical trauma, cancer, infections, certain inflammatory diseases and specific conditions such as stroke, heart failure or nephrotic syndrome.

3.5. *Congenital heart disease*

CHD is a defect in the structure of the heart and great vessels which is present at birth. Many types of heart defects exist, most of which either obstruct blood flow in
the heart or vessels near it, or cause blood to flow through the heart in an abnormal pattern. Other defects, such as long QT syndrome, affect the heart's rhythm. Heart defects are among the most common birth defects and are the leading cause of birth defect-related deaths.

Signs and symptoms are related to the type and severity of the heart defect. Symptoms frequently present early in life, but it's possible for some CHDs to go undetected throughout life (Heart Defects: Birth Defects, 2010). Some children have no signs while others may exhibit shortness of breath, cyanosis, syncope, (NHLBI, 2010) heart murmur, under-developing of limbs and muscles, poor feeding or growth, or respiratory infections. Congenital heart defects cause abnormal heart structure resulting in production of certain sounds called heart murmur.

4. Cardiovascular disease risk factors

'Risk' is defined as a probability of an adverse health outcome, whereas 'risk factor' refers to an attribute or characteristic or exposure of an individual whose presence or absence raises the probability of an adverse outcome (WHO, 2002). In the context of 'CVD, a risk factor' can be defined as a characteristic that is associated with increased or decreased likelihood of subsequent development of CVD. This concept can be used for different purposes, each of which has its own strengths and limitations: to study the cause or pathophysiology of CVD; to estimate total cardiovascular (CV) risk; understanding the dynamics of the CVD epidemic within and between populations (Guy, 2008).

4.1. Age

Age is among the strongest risk factor and its relationship with CV mortality is exponential. It is an important factor to consider in total CV risk estimation, but its nonmodifiable nature limits its use in the management of risk. Given the nature of its association with CVD, it explains the paradox that if prevention is successful in a given generation, total CV mortality will increase: by preventing premature deaths, a larger proportion of the population will grow old and enter the age group (>85 years)
where death is attributed to CVD in a majority of cases. Other nonmodifiable risk factors are gender and a family history of premature CVD. Total risk levels in women tend to resemble those of men 10 years younger. Thus, risk is merely deferred by 10 years and ultimately more women than men die from CVD.

4.2. Sedentary lifestyle

Systematic reviews and meta-analyses of observational studies have evidenced reduced CV risk in physically active subjects (Batty, 2002; Oguma and Shinoda, 2004). All available evidence indicates that the association between physical activity pattern and CVD is causal. Physical activity has both a direct protective effect on the development of CV events and an indirect effect through its influence on risk factors.

4.3. Nutrition and CVD

Worldwide there is strong and consistent evidence of graded relationships between saturated fat intake and the occurrence of CVD at the community level. However, the development of CVD is also associated with other aspects of dietary imbalances related to the intake of hydrogenated fats, trans fatty acids, fiber, refined and processed sugars, salt, whole-grain products, or fruits and vegetables. Recently it was shown that a high dietary glycemic load and glycemic index increase the risk of CVD, particularly in overweight women (Beulens et al., 2007).

4.4. Tobacco smoking

In long-term smokers, smoking is responsible for 50% of all avoidable deaths and one half of these are due to CVD (Bartecchi, 1994). Male and female smoking patterns in recent decades have become increasingly similar. In prospective studies, the relative mortality from vascular disease has been found higher in female smokers than in male smokers (APCSC, 2005). The impact of smoking on atherosclerosis progression is greater in subjects with diabetes and hypertension. The risk of future cardiovascular disease is particularly high if smoking starts before the age of 15 years. Passive smoking has now been shown to increase the risk of coronary heart disease and other smoking related diseases (Law et al., 1997); the effects of passive smoke on the cardiovascular system may even be greater than expected; some of these effects appear rapidly and can precipitate acute manifestations of CVD. Although the exact
mechanisms by which tobacco smoking increases the risk of atherosclerotic disease are not yet fully understood, smoking enhances both the development of atherosclerosis and the occurrence of superimposed thrombotic phenomena. Smoking after a myocardial infarction is potentially the most effective of all preventive measures.

4.5. Diabetes and Impaired glucose tolerance

Impaired glucose tolerance is associated with an increased risk of developing CHD as well as other atherosclerotic diseases (Barr, 2007). In diabetes, the relative risk of CVD is of the order of 2 to 3 in men and of 3 to 5 in women, while in people with impaired glucose tolerance the relative risk is 1.5 compared with people with normal glucose tolerance. Subjects with type 1 diabetes have a 2- to 3-fold increase in the risk of developing CVD. This increased risk is almost entirely confined to patients developing diabetic renal disease. All type 2 diabetes patients are at increased risk of CVD, even in the absence of diabetic nephropathy. The risk of developing a MI in patients with type 2 diabetes was of the same order as for patients without diabetes who had already suffered a first MI (Haffner et al., 1998). Diabetes was labeled as a "CVD equivalent" in terms of risk assessment. The impact of type 2 diabetes on CVD risk is influenced by a number of factors, including duration of diabetes, age, and sex (Hu, 2005; Pajunen, et al., 2005; Whiteley et al., 2005; Juutilainen et al., 2005). Thus, the relative impact of type 2 diabetes on CVD risk is stronger in women than in men, suggesting that type 2 diabetes can be more convincingly considered as a CVD equivalent in women than in men (MacMahon et al., 1990; EUROASPIRE, 2001; Danesh et al., 2000; Meisinger et al., 2005; WHO, 1998). A substantial proportion of the excess risk of atherosclerotic disease in both type 1 and type 2 diabetes is caused by the diabetic state itself.

4.6. Blood pressure (BP)

Elevated blood pressure has been identified as a risk factor for CHD, heart failure, cerebrovascular disease, and renal failure in both men and women in a number of epidemiological studies (MacMahon, 1990). Both systolic blood pressure (SBP) and diastolic blood pressure (DBP) show a continuous and graded independent
relationship with the risk of stroke and coronary events. The death from both CHD and stroke increases progressively and linearly from BP levels as low as 115 mm Hg systolic and 75 mm Hg diastolic upward. Increased risks are present in all age groups ranging from 40 to 89 years old. For every 20-mm Hg systolic or 10-mm Hg diastolic increase in BP, there is a doubling of mortality from both CHD and stroke (Lewington et al., 2002). The apparently simple direct relationship between increasing SBP and DBP and CV risk is confounded by the fact that SBP rises throughout adult age in the vast majority of populations, whereas DBP peaks at about age 60 in men and 70 in women, and falls gradually thereafter. Both systolic and diastolic BP were independently predictive of stroke and CHD mortality and to a greater extent than pulse pressure. A high SBP may be associated with a lower risk for developing CVD than a low SBP.

4.7. **Total cholesterol**

Cholesterol is a fatty substance which is manufactured in the body, particularly in the liver and plays a vital role in the functioning of cell membranes. Cholesterol is found in several forms in the body and, when bound to proteins, forms lipoproteins. Cholesterol and other fatty blood components are often referred to collectively as ‘blood lipids’. Blood lipids can be divided into different fractions or components: LDL-C and HDL-C and triglycerides. High levels of LDL-C and low levels of HDL-C are associated with increased risk of CHD. The ratio of plasma total cholesterol (TC) or LDL-C to HDL-C is often used in risk calculations. The higher the ratio, the higher the CHD risk (Cholesterol and CHD: screening and treatment 1998).

Most of the blood cholesterol is normally carried on LDL-C particles. Over the entire range of TC and LDL-C concentrations there is a strong, continuous, graded, and independent positive association with risk of CVD. This association applies to women as well as men, and to old as well as younger people. The relationship is exponential, indicating that a given absolute difference in total or LDL-C from any point in the distribution is associated with a constant percentage difference in CHD risk. This association is considerably modified by other risk factors such as age, sex, smoking, blood pressure, diabetes, and low HDL-C cholesterol. CAD is rare in
populations with TC less than 3 to 4 mmol/L (115-155 mg/dL), even in the presence of other risk factors. Conversely, CAD is inevitable in untreated patients with the severest forms of familial hypercholesterolemia, even in the absence of other risk factors. The results of epidemiological studies, as well as trials with angiographic or clinical end points, confirm that the reduction of LDL-C must be of prime concern in the prevention of CVD.

4.8. **Triglycerides**

Hypertriglyceridemia is also associated with the risk of developing CVD, but the association is not as strong as it is for hypercholesterolemia. Although the risk of CVD does increase with hypertriglyceridemia, the risk is associated more strongly with moderate than with very severe hypertriglyceridemia, probably because the former is often due to accumulation in plasma of triglyceride-rich atherogenic intermediate-density lipoprotein cholesterol (IDL-C) and small very-low density lipoprotein cholesterol (VLDL-C), whereas the latter can be due to nonatherogenic large VLDL-C and chylomicrons.

A triglyceride value >1.7 mmol/l (150 mg/dl) is considered a marker of increased risk, but concentrations <1.7 mmol/l are not considered a goal of therapy.

4.9. **High-density lipoproteins**

HDL-C is one of the major carriers of cholesterol in the blood. HDL-C is small, dense, and spherical lipid-protein complex and is normally considered to consist of those plasma lipoprotein particles which fall into the density range of 1.063–1.210 g/ml. Low concentrations HDL-C, are clearly associated, not only with early development of atherosclerosis, but also with poor outcome in those who already have CVD. It has to be stressed that smoking, sedentary lifestyle, obesity, and type 2 diabetes cause lower HDL-C. The combination of moderately elevated triglycerides and low concentrations of HDL-C is very common in patients with type 2 diabetes. HDL-C <1 mmol/l (40 mg/dl) in men and <1.2 mmol/l (46 mg/dl) in women is considered a marker of increased risk.

HDL-C particles are composed of an outer layer containing free cholesterol, phospholipid, and various apolipoproteins (Apo), which covers a hydrophobic core
consisting primarily of triglycerides and cholesterol esters (Barter et al., 2003). The major proteins are Apo A-I (Mr 28,000) and Apo A-II (Mr 17,000). Apo A-I, the primary protein constituent of these particles, accounts for about 60% of the protein content of HDL-C. Apo A-I is synthesized in the intestines and liver and is thought to be largely responsible for the antiatherogenic effects of HDL-C. Some HDL-C particles carry only Apo A-I, whereas others contain both Apo A-I and Apo A-II (Shah et al., 2001). Other apolipoprotein species found in HDL-C particles include Apo A-IV, Apo C (C-I, C-II, and C-III), and Apo E. Several subtypes of HDL-C particles have been identified on the basis of density, electrophoretic mobility, particle size, and apolipoprotein composition (Albers et al., 1984). Numerous prospective cohort studies support a powerful inverse correlation between circulating HDL-C and coronary risk among patients with normal or elevated LDL-C (Drexel, 2006; Toth, 2005). Low HDL-C levels increase the risk of CHD and analysis substantiated that, on average, each one (1) mg/dL increase in HDL-C reduces CHD risk by 2% in men and 3% decrease in women (Gordon et al., 1989). The Framingham Heart Study have supported the role of low HDL-C as an independent risk factor for CAD and demonstrated that subjects with the highest HDL-C levels exhibit the lowest risk of developing CAD.

4.10. Other lipoproteins and lipoprotein components

LDL-C, a spherical lipoprotein particle, contains mostly cholesteryl ester in its non polar core; its surface apolipoprotein consists almost exclusively of apolipoprotein B-100 (apo B-100, or apo B), one apo B molecule per LDL-C particle. Apo B is the major protein component of LDL-C, IDL-C, VLDL-C, and, in truncated form, chylomicrons. Since chylomicrons are not normally present in plasma in the fasting state, almost all apolipoprotein B is in atherogenic lipoproteins. Concentrations of apolipoprotein B are therefore a direct measure of the concentration of atherogenic lipoproteins in plasma. The measurement is a useful indicator of risk of atherosclerosis, particularly in patients with hypertriglyceridemia and in people with normal concentrations of LDL-C (Yusuf et al., 2004). Values >150 mg/dL are clearly associated with increased risk.
Apo B-100 contains 4563 amino acids and is synthesized in the liver. In contrast, apo B-48, which is present on triglyceride-rich chylomicrons, is produced by the intestine as a truncated form of apo B-100. The liver employs apo B-100 in the formation of triglyceride-rich lipoproteins called VLDL-C. VLDL-C particles are responsible for transporting triglyceride fatty acids from the liver to peripheral tissues for utilization. When VLDLs-C enters capillaries, they come into contact with lipoprotein lipase (LPL), an enzyme that hydrolyzes the triglycerides and releases fatty acids for tissue utilization. VLDL-C carries two other groups of apolipoprotein, the apo Cs and apo Es. Apo CII is a cofactor for the activation of LPL. Apo E plays a role in the disposition of VLDL-C.
Body Mass Index (BMI) has been extensively used to define overweight or obesity. In adults, overweight is defined by an increased BMI ranging from 25 to 29.9 and obesity by BMI >30. Increasing BMI is highly associated with CVD. Other indicators apart from BMI have been proposed to assess body fat distribution. The waist-hip ratio (WHR) and waist circumference (WC) are now frequently used. WHO report on obesity (WHO, 1998) recommends the use of WC as an additional indicator of CV risk. In longitudinal studies in men and women, increased WHR or WC was associated with increased risk of ischemic heart disease mortality.

4.12. Psychosocial factors

There is increasing scientific evidence that psychosocial factors contribute independently to the risk of CHD, even after statistical control for the effects of standard risk factors. Low socioeconomic status, lack of social support and social isolation, stress at work and in family life, and negative emotions including depression and hostility, have been shown to influence both the risk of contracting CHD and the worsening of clinical course and prognosis in patients with CHD. Several behavioral and psycho physiological mediators and moderators of these effects have been identified.

5. Cardiac marker enzymes for myocardial infarction

The criteria for the diagnosis of myocardial infarction have been redefined recently, as reported in a consensus document of the European Society of Cardiology (ESC) and the American College of Cardiology (ACC), (Alpert et al., 2000) and require at least 2 of the 3 following characteristics: (1) typical symptoms (2) characteristic rise-and-fall pattern of a cardiac marker [eg: MB isoenzymes of creatine kinase (CK)] or, preferably, serum troponins (T or I) and (3) a typical electrocardiogram (ECG) pattern involving the development of Q waves. Current cardiac marker technologies, particularly the serum troponins, can now detect extremely small amounts of myocardial necrosis. The superior troponin’s clinical value comes from its higher sensitivity to smaller myocardial injury and its virtually
total specificity for cardiac damage (Jaffe et al., 2000). Despite the ability to detect quantitatively smaller degrees of myocardial necrosis, cardiac troponins need 4-10 h after symptom onset to appear in serum, at about the same time as CK-MB elevations become detectable, and peak at 12-48 h, remaining then abnormal for several days (Panteghini et al., 1999). This prolonged release pattern indeed makes it difficult to diagnose a re-infarction by the use of serial troponin measurements, suggesting a continuing role for CK-MB for this purpose (Panteghini, 2002).

The interesting part of the history of cardiac biomarkers is that the field intuitively and empirically found the appropriate markers. The first practical test utilized was serum glutamic oxaloacetic transaminase [SGOT], now called aspartate aminotransferase [AST]. Art Karmen used the transaminases for identifying M.I (LaDue 1954). In 1959 and 1960, creatine kinase was demonstrated to be a possible marker of skeletal muscle (Ebashi 1959) and cardiac muscle (Dreyfus 1960) damage and lactate dehydrogenase (LD) (Wróblewski 1956) and its isoenzymes (Wróblewski 1960) were described as possible markers around the same time.

5.1. Creatine Kinase and CK-MB

In the past, measurement of nonspecific markers such as aspartate transaminase (AST), lactate dehydrogenase (LDH), and total CK were performed. Mass assays for CK-MB became the standard of care for cardiac marker testing in the mid-1990s. Subsequently, newer markers of myocardial injury, including CK-MB isoforms, myoglobin, and cTnT and cTnI have become available on automated commercial instruments.

The measurement of CK and CK-MB levels has long been used for the diagnosis of AMI. CK, an enzyme present in many tissues, including the myocardium and skeletal muscle, has 3 isoenzymes: MM, MB, and BB. CK-MB is present in a relatively high concentration in the myocardium (roughly 20% of the total myocardial CK), whereas the concentration of CK-MM is highest in skeletal muscle (98% of total muscle CK) with only a small amount of CK-MB (usually about 2%). However, healthy skeletal muscle can have up to 5% CK-MB, and higher levels (up to 20%) of
CKMB can be found in patients with renal failure and chronic myopathic skeletal muscle injury (as occurs in polymyositis and dermatomyositis) or in the muscle tissue of trained athletes. Although CK-MB constitutes about 20% of the total CK in the myocardium, it should be noted that CK-MM is still the most abundant CK isoenzyme in myocardial tissue. Elevation of the total CK level is not cardiac specific and may be observed in patients with skeletal muscle injury and other disorders. Until recently, measurement of the CK-MB level was the traditional gold standard test for AMI. Following myocardial injury, the initial CK-MB rise occurs 4 to 9 hours after the onset of chest pain, peaks at 24 hours, and returns to baseline at 48 to 72 hours. One advantage of CK-MB over markers that remain elevated for longer periods is that it is easier to detect reinfarction using serial CK-MB measurement.

5.2. Lactate dehydrogenase

LDH activity has also been found reliably elevated in the presence of myocardial infarction. Human LDH consists of five distinct components having a characteristic distribution (Wroblewski, 1963). Serum or plasma of humans contains all five isoenzymes, the concentrations diminishing in the order: LDH-4, LDH-5, LDH-3, LDH-2, and LDH-1. Estimation of serum LDH isoenzymes in combination with CK is a well-established laboratory procedure for diagnosing AMI (Lott and Stang, 1980; Lee and Goldman, 1986). As the myocardium has a preponderance of LDH-1, with lesser amounts of LDH-2, necrosis of the myocardium results in release of relatively more LDH-1 than LDH-2 into the blood, reversing the normal ratio, usually between 12 h and 24 h, reaching a peak 48 h after the infarct (Lott and Stang, 1980; Lott, 1984). Warburton et al. (Warburton et al., 1967) described an increased proportion of LDH-3 (above 20%) in serum of 10 of 50 patients with AMI. According to their investigation, the source for the increased LDH-3 proportion was necrotic myocardium. MI is also characterized by elevations of LDH-4 and LDH-5.

5.3. Transferases and Phosphatase

Aminotransferases may be raised in cardiac diseases and hepatic diseases. Clinical and laboratory studies performed on human subjects have shown that the serum level of glutamic oxalacetic transaminase (SGOT) rises significantly following
The aminotransferases (formerly transaminases) are the most frequently utilized and specific indicators of hepatocellular necrosis. These enzymes—aspartate aminotransferase (AST, formerly SGOT) and alanine amino transferase (ALT, formerly serum glutamic pyruvate transaminase-SGPT)—catalyze the transfer of the α amino acids of aspartate and alanine respectively to the α keto group of ketoglutaric acid. ALT is primarily localized to the liver but the AST is present in a wide variety of tissues like the heart, skeletal muscle, kidney, brain and liver (Rosen and Keefe, 2000; Friedman et al., 2003). AST is elevated in the serum with hepatic cell involvement, skeletal muscle fiber inflammation, and myocardial cell injury.

γ-Glutamyl transpeptidase (GGT) is a unique biomarker for cardiac and metabolic risk evaluation. It is a membrane bound glycoprotein which catalyses the transfer of γ-glutamyl group to other peptides, amino acids and water. GGT levels were first associated with CVD and all-cause mortality in a British Regional Heart Study by Wannamethee (Wannamethee et al., 1995) reported in October of 1995. Large amounts are found in the kidneys, pancreas, liver, intestine and prostate. Non-hepatic causes of increased levels of the enzyme include anorexia nervosa, Gullian barre syndrome, hyperthyroidism, obesity and dystrophica myotonica.

Alkaline phosphatases (ALP) are a family of zinc metaloenzymes, with a serine at the active center; they release inorganic phosphate from various organic orthophosphates and are present in nearly all tissues. In liver, ALP is found histochemically in the microvilli of bile canaliculi and on the sinusoidal surface of hepatocytes. ALP from the liver, bone and kidney are thought to be from the same gene but that from intestine and placenta are derived from different genes (Rosalki and McIntyre, 1999). Highest levels of ALP occur in cholestatic disorders.

5.4. Myoglobin

Myoglobin is an oxygen-binding protein, found in a high concentration in both cardiac and skeletal muscle. Myoglobin is a relatively small protein molecule that is released into serum as early as 1 hour after AMI, reaches a peak in the range of 4 to 12 hours, and then is rapidly cleared. The major advantage of myoglobin as a cardiac
marker is that it is released earlier from damaged cells than other cardiac markers, permitting earlier detection of AMI (Vaidya, 1994; Gibler et al., 1987; Adams et al., 1993; Mair et al., 1995; Mercer, 1997). Rapid release of myoglobin probably reflects its low molecular weight and cytoplasmic location. Myoglobin as an early marker of AMI exhibits a high negative predictive value. The main reason that myoglobin has not been used by most hospitals for the evaluation of chest pain is its poor clinical specificity (60%-80%) owing to the presence of large quantities of myoglobin in skeletal muscle. Myoglobin, therefore, is potentially useful for ruling out but not for confirming the diagnosis of AMI. The use of myoglobin for the detection of reinfarction is complex.

5.5. Troponin

Cardiac troponin I (cTnI) and T (cTnT) are the most sensitive and specific biochemical markers of myocardial cell damage. A consensus document issued by the European and American College of Cardiology Committee for the redefinition of myocardial infarction promoted cardiac troponin I and T as the preferred markers for myocardial damage because of their nearly absolute myocardial tissue specificity, high sensitivity, and the ability of these marker to reflect microscopic zones of myocardial necrosis (Myocardial infarction redefined, 2000).

Both cTnT and cTnI are stored in a 2-compartment distribution in the myocyte, including a small cytosolic pool (4%-6%), with the majority of the remaining troponin found in the sarcomere. Thus, TnT and TnI have similar release kinetics from damaged myocardium. Both troponins increase in serum within 4 to 9 hours after AMI, peak at 12 to 24 hours, and remain elevated for up to 14 days (Bertinchant et al., 1996; Wu et al., 1999). An appropriate testing strategy is sampling for TnT or TnI at baseline, 8, and 16 hours, which has been demonstrated to be optimal for the diagnosis of myocardial necrosis (Newsby et al., 1998). With currently available assays, cTnT and cTnI have equal myocardial tissue specificity, as well as high sensitivity (Wu, 1999).

5.6. C-Reactive Protein
C-reactive protein (CRP) is an acute phase reactant that has been used as a marker of inflammation. Various studies have suggested that CRP levels measured also may be useful as an independent predictor of new coronary events, including AMI and death in patients with IHDS (Liuzzo et al., 2001). The prognostic information derived from troponin and CRP is additive in patients with unstable angina and non-Q-wave AMI; negative and low levels of CRP are associated with an approximately 1% risk of death at 14 days vs a 9% risk for patients with CRP concentrations greater than 15 mg/L.

In more recent trials, other investigators have confirmed the increased risk in Acute Coronary Syndrome (ACS) associated with higher CRP concentrations (Morrow et al., 1998; Heeschen et al., 2000; Lindahl et al., 2000; Mueller et al., 2002). In each of the above studies, the predictive value of CRP was independent of, and additive to, cardiac troponin.

5.7. Homocysteine

Homocysteine is a sulfur amino acid and a normal intermediate in methionine metabolism. It is the role of the liver and kidney to remove excess homocysteine from the blood. McCully postulated that moderately elevated homocysteine due to heterozygous mutations in homocysteine-related genes or poor vitamin status would also lead to increased risk of CVD in the general population (McCully, 1969). By the early 1990s, elevated homocysteine was being considered an independent risk factor for CVD A prospective study of in 1992 found that AMI or death due to coronary disease was statistically related to increased homocysteine levels (Stampfer et al., 1992). It is conclude that homocysteine was an independent risk factor for CVD and estimated that 10% of the population’s CVD risk is attributable to elevated homocysteine (Boushey et al., 1995). The lowering of plasma total homocysteine to 3 μmol/L would reduce the risk of IHD by 16%, deep vein thrombosis by 25% and stroke by 24% (Wald et al., 2002).

6. Antioxidants
To protect the cells and organ systems of the body against reactive oxygen species (ROS), humans have evolved a highly sophisticated and complex antioxidant protection system. It involves a variety of components which function interactively and synergistically to neutralize free radicals (Hennekens and Gaziano, 1993). Myocardial antioxidants are defined as substances which inhibit or delay the oxidative damage to subcellular proteins, carbohydrates, lipids and DNA. Although the exact mechanisms and interactions among various antioxidants are not fully understood, it is possible that one antioxidant may equilibrate with another to establish a cellular redox potential and thus all antioxidants may act in concert to protect against oxidative insult. Many substances have been suggested to act as endogenous antioxidants such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and vitamin E.

SOD catalyzes the dismutation of superoxide anion ($\text{O}_2^-$) to $\text{H}_2\text{O}_2$. Subsequently $\text{H}_2\text{O}_2$ is reduced to $\text{H}_2\text{O}$ and $\text{O}_2$ by peroxidases such as GPx or CAT. SOD is present in the cytoplasm as well as on the endothelial cell surface with either copper or zinc (CuSOD, ZnSOD) and in the mitochondria with manganese (MnSOD) (Ohta et al., 1994). GPx catalyzes the peroxidation of $\text{H}_2\text{O}_2$ in the presence of reduced glutathione (GSH) to form $\text{H}_2\text{O}$ and oxidized glutathione (GSSG). The GSSG recycles back to give GSH by glutathione reductase, which requires NADPH from the hexose monophosphate shunt. Thus, GPx plays a significant role as $\text{H}_2\text{O}_2$ scavenger in the heart since its activity is much higher than CAT. On the other hand, CAT is a membrane bound enzyme which is present in peroxisomes but its activity has also been observed in the mitochondrial matrix (Steare and Yellon, 1993). Other endogenous antioxidants including vitamin E (tocopherols), vitamin C (ascorbic acid), and vitamin A are also present in the myocardium (Keaney et al., 1999). Vitamin E is a fat soluble substance and is mainly associated with plasma lipoproteins. It acts as a potent peroxyl radical scavenger via breaking the lipid peroxidation chain reaction (Spencer et al., 1999).

Glutathione-S-transferase (GST) also known as glutathione transferase found mainly in the cytosol is thought to play a physiological role in initiating the
detoxication of potential alkylating agents (Habig et al., 1974), including pharmacologically active compounds. These enzyme catalyze the reaction of such compounds with the -SH group of glutathione, thereby neutralizing their electrophilic sites and rendering the products more water-soluble.

Paraoxonase (PON1) is synthesized in the liver and is closely associated with HDL-C. Several laboratories have reported that HDL-C protects against LDL-C oxidative modification, (Klimov et al., 1987; Mackness et al., 1993) the antioxidant activity of HDL-C may relate, at least in part, to the enzymes associated with HDL-C (Mackness and Durrington, 1995). PON1 has been suggested as the factor largely responsible for the antioxidant role of HDL-C (Shih et al., 1996). Further studies indicated that PON1 could prevent lipid-peroxide accumulation on LDL-C (Mackness et al., 1991). The other enzymes associated with HDL-C and playing role in its antioxidant activity are platelet activating factor acetyl hydrolases (Watson et al., 1995) and lecithin cholesterol acyl transferase (Klimov et al., 1989). Studies have shown that serum PON1 activity is reduced in diabetes and familial hypercholesterolaemia (Mackness et al., 1991; Abbott et al., 1995) diseases that are associated with accelerated atherogenesis. It was proposed that the serum PON1 activity decreased in survivors of myocardial infarction (McElveen et al., 1986).

7. Oxidative stress in cardiovascular disease

Oxygen radicals are continuously formed in all living organisms, with deleterious effects that lead to cell injury and death (Tappel, 1973). Production of oxidative species occurs under physiological conditions at a controlled rate, but it is dramatically increased in conditions of oxidative stress. Exposure of biological systems to xenobiotics, pollutants, ionizing radiation or U.V. light and development of certain pathological conditions lead to oxidative stress, consequently increase production of oxy radicals (Sies, 1992). ROS include superoxide anion radical (O2-) hydrogen peroxide (H2O2), hydroxyl radical (·OH), nitric oxide (NO), and peroxynitrite (ONOO-). Under physiological conditions, ROS are produced in low concentrations and act as a signaling molecule that regulate VSMC contraction and
relaxation, and participate in VSMC growth (Touyz and Schiffrin, 1999). Cell damage caused by free radicals appears to be a major contributor in aging and degenerative diseases of aging such as cancer, cardiovascular disease, cataracts, compromised immune system, rheumatoid arthritis and brain dysfunction. The principle sources of ROS in the vasculature include NAD(P)H oxidase, xanthine oxidase (XOD), uncoupled eNOS (endothelial nitric oxide synthase) and the mitochondrial respiratory chain (Landmesser and Harrison, 2001; Cai and Garrison, 2000).

XOD is from the molybendum iron sulfur flavin hydroxylase group of enzymes and is found predominantly in the liver and gastrointestinal tract but also in the kidney and brain. Interestingly, it is also found throughout the cardiovascular system (George and Struthers, 2008). Expression of these has been shown to increase in ischaemia and in response to increased levels of proinflammatory cytokines (Berry and Hare, 2004). While the major role of XOD is conversion of hypoxanthine and xanthine to uric acid, an interconvertible form, xanthine dehydrogenase, also exists and is responsible for conversion of NAD+ to NADH (Zhang et al., 1998). The action of these enzymes yields hydroxyl free radicals and hydrogen peroxide which can add to or initiate oxidative stress (Hille and Massey, 1981).

Lipid peroxidation (LPO) is a natural metabolic process under normal conditions. Polyunsaturated fatty acids (the main component of membrane lipids) are susceptible to peroxidation. LPO is one of the most investigated consequences of ROS actions on membrane structure and function. Malondialdehyde (MDA) is one of the most known secondary products of LPO, and it can be used as a marker of cell membrane injury (Esterbauer et al., 1991). It has observed that diabetic patients with CHD had higher levels of MDA than those diabetics without this disease. It shows that CVD have also been related to free radical-mediated mechanisms and to LPO (Kesavulu et al., 2001)
Some of the major endogenous antioxidants and their sites of action in cardiovascular system:

<table>
<thead>
<tr>
<th>Name</th>
<th>Site</th>
<th>Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>Superoxide dismutase (SOD)</td>
<td>Cytoplasm, cell, extracellular, mitochondrial</td>
<td>Catalyzes O₂ dismutation to H₂O, NO⁻ + 2H₂O → H₂O₂ + O₂⁻</td>
</tr>
<tr>
<td>Cu/Zn SOD</td>
<td>Cytoplasm</td>
<td>H₂O₂ → H₂O + O₂⁻</td>
</tr>
<tr>
<td>Mn SOD</td>
<td>Plasma membrane and mitochondrial</td>
<td></td>
</tr>
<tr>
<td>Catalase</td>
<td>Plasma membrane and mitochondrial</td>
<td></td>
</tr>
<tr>
<td>Glutathione peroxidase</td>
<td>Cytoplasm</td>
<td>Break lipid peroxidation chain and LDL reaction</td>
</tr>
<tr>
<td>Glutathione</td>
<td>Intercellular</td>
<td>Break lipid peroxidation chain and LDL reaction</td>
</tr>
<tr>
<td>Glutathione peroxidase</td>
<td>Intercellular</td>
<td>Break lipid peroxidation chain and LDL reaction</td>
</tr>
<tr>
<td>Vitamin E (tocopherol)</td>
<td>Cytoplasm and platelet</td>
<td>Break lipid peroxidation chain and LDL reaction</td>
</tr>
<tr>
<td>β-Carotene (precursor A)</td>
<td>Plasma</td>
<td>Inhibit oxidation of LDL</td>
</tr>
<tr>
<td>Vitamin C (ascorbic acid)</td>
<td>Cytoplasm and platelet</td>
<td>Directly as an antioxidant or as a cofactor for vitamin E</td>
</tr>
</tbody>
</table>

---

![Fig. Sources of Reactive Oxygen Species generation and their effect on signaling system in cardiovascular disease.](image_url)

NO is known as a vascular smooth muscle tone controller, it inhibits platelet activation, modulates apoptosis and inflammatory cell aggregation and activation at low concentrations. On the other hand, NO can react with superoxide anion (O₂⁻) to form ONOO⁻, which is highly cytotoxic. The damage of the vascular endothelium is always followed by vasoconstriction, platelet aggregation and inflammatory cell...
adhesion, which lead to an increased production of NO and, consequently, ONOO-
Associated with other factors, the overproduction of NO is one of the most important
issues involved in the development of lipid atherosclerotic plaques (Shaw et al., 2005).

NO is well known as an important mediator of many physiologic functions,
and its role in the pathogenesis of CVD is gaining recognition. NO is synthesized by
nitric oxide synthase (NOS). In mammals, 3 isoforms of NOS have been identified:
endothelial NOS (eNOS), neuronal NOS (nNOS) are constitutive and inducible NOS
(iNOS) (Bredt et al., 1991). All forms of NOS catalyze the conversion of L-arginine to
L-citrulline in an NADPH-dependent manner, producing NO from the terminal N-
guanidino group of L-arginine (Kwon et al., 1990). All isoforms have been detected in
cardiovascular tissues because neuronal and endothelial cells, as well as monocytes
and macrophages, are represented in this organ. NOS expression and its modulatory
role on cardiac function have been extensively studied under physiological conditions.
Inhibition of constitutive NOS was deleterious, but that selective inhibition of iNOS
might be beneficial. iNOS expression in the heart might account for myocardial
depression. iNOS expression was deleterious not only with respect to hypotension and
vascular hyporeactivity but also in relation to myocardial dysfunction (Wright et al.
8. Lipid metabolizing enzymes

LPL is a central enzyme in overall lipid metabolism and transport, being responsible for catalysing the hydrolysis of triglycerides transported in the bloodstream by chylomicrons and VLDL-C, thereby providing non-esterified fatty acids and 2-monoacylglycerols for tissue utilization. Mature LPL is secreted to the vascular endothelium from the parenchymal cells of the adipose and muscle tissues, its major sites of synthesis, and a variety of other tissues which have also been implicated as lesser, but significant, sources of the enzyme (Braun and Severson, 1992; Enerback and Gimble, 1993). The most recent evidence implicating that LPL has both proatherogenic and anti-atherogenic roles. The effects of LPL on atherosclerosis have been controversial. As atheromatous plaques contain substantial amounts of LPL in situ, Zilversmit proposed that local LPL is atherogenic. Furthermore, LPL mediates the lipolytic conversion of triglyceride-rich lipoproteins to atherogenic cholesterol-rich lipoproteins such as LDL-C and chylomicron remnants. Supporting this, LPL deficiency in humans, a common genetic cause of chylomicronemia syndrome, results in very low plasma levels of LDL-C, and is believed to cause resistance to premature
atherosclerosis (Brunzell, 1995). However, Benlian et al. have reported that several LPL-deficient patients have developed relatively advanced atherosclerosis. Furthermore, several clinical studies have shown that fibric acid derivatives induce LPL activity, lower plasma triglycerides, and suppress atherosclerosis (Frick et al., 1987; Manninen et al., 1992).

HMG-CoA reductase (or 3-hydroxy-3-methyl-glutaryl-CoA reductase) is the rate-controlling enzyme of the mevalonate pathway, the metabolic pathway that produces cholesterol and other isoprenoids. Normally in mammalian cells this enzyme is suppressed by cholesterol derived from the internalization and degradation of LDL-C via the LDL receptor as well as oxidized species of cholesterol. Competitive inhibitors of the reductase induce the expression of LDL receptors in the liver, which in turn increases the catabolism of plasma LDL-C and lowers the plasma concentration of cholesterol, an important determinant of atherosclerosis.

Lecithin: cholesterol acyltransferase (LCAT) is a 63KDa glycoprotein enzyme which is synthesized in the liver and secreted in the plasma (Francone and Fielding, 1991; Wang et al., 1997). LCAT binds to HDL-C to catalyze the transfer of a fatty acid residue from the sn-2 position of lecithin to cholesterol to form cholesterol ester and lysolecithin (Jimi et al., 1999). A lack of LCAT activity leads to increase in free cholesterol and phospholipids and decrease in esterified cholesterol. Cholesterol esterification would be a key step in transferring cholesterol from the tissues of the body to the liver (Glomset et al., 1966). This process, termed reverse cholesterol transport (Glomset, 1966), was proposed to facilitate the removal of cholesterol from the body. Thus reverse cholesterol transport is one of several proposed mechanisms by which HDL-C provides protection from CVD (Miller and Miller, 1975; Rhoads et al., 1976). Traditionally LCAT activity has been considered 'antiatherogenic' as the cholesterol esterification apparently creates a gradient necessary for the flow of unesterified cholesterol from tissues to plasma. However, newer data suggest that higher plasma cholesterol esterification rate is not necessarily 'protective' and that the 'protective' or conversely 'atherogenic' role of LCAT may depend on the
concentration and quality of plasma HDL-C and LDL-C particles and on availability of lipid transferring proteins such as cholesteryl ester transfer protein (Milada Dobiasova and Jiri, 1999).

9. Cardioprotective therapeutics

A cardioprotective drug reduces the risk of suffering or dying from a coronary event. The Survival and Ventricular Enlargement (SAVE) study (Pfeffer et al., 1992) suggested a major role for captopril in the long-term management of patients following myocardial infarction who had impaired myocardial function and the more recent Acute Infarction Remipril Efficacy (AIRE, 1993) study has shown that ramipril reduces mortality. At this point the concept of a cardioprotective drug requires consideration.

9.1. Thiazide diuretics

Thiazides tend to produce metabolic effects which would increase the coronary risk and they have not been shown to have cardioprotective actions in animal models. Thiazides have some impact on coronary events in the elderly but overall thiazides could not be considered cardioprotective.

9.2. Beta-adrenoceptor blocking drugs (beta blockers)

There is an extensive literature on the effects of beta blockers on the processes which led to death from IHD. Thus beta blockers reduce endothelial damage (Kaplan et al., 1987 a), atheroma formation (Kaplan et al., 1987 b) and tend to inhibit clot formation (Frishman et al., 1978). The cardioprotective role of beta blockers in post-infarct patients, in particular their capacity to reduce the risk of sudden death has been shown for three lipophilic beta blockers, timolol (Norwegian Study Group, 1981), propranolol (Beta Blocker Heart Attack Trial Research Group, 1982) and metoprolol (Hjalmarson et al., 1981), but not for the hydrophilic sotalol (Julian et al., 1982). Therefore, beta blockers are the drugs which come closest to meeting the criteria for being cardioprotective drugs.
9.3. Calcium channel blocking drugs (calcium antagonists)

Calcium antagonists have a reputation for being metabolically neutral and of having the potential to delay atheroma formation. Dihydropyridines have not reduced clinical events though the number of new atherosclerotic lesions per patient was significantly fewer in those on nifedipine (INTACT Study Group, 1990). Verapamil appeared to reduce sudden deaths and all cardiac deaths, they were not statistically significant. Diltiazem also may have a clinically relevant impact. However, diltiazem also did not significantly reduce cardiac events or total mortality.

In conclusion, among calcium antagonists dihydropyridines have not been shown to be effective, verapamil and diltiazem have a cardioprotective effect at best, modest.

9.4. Angiotensin converting enzyme inhibitors (ACE inhibitors)

ACE inhibitors have been shown to have some potential to reduce coronary events for their cardioprotective actions (Pfeffer et al., 1992). Enalapril and captopril reduced the incidence of sudden death in heart failure patients. Ramipril treatment after an acute infarct reduced overall mortality and was seen to have a beneficial effect.

9.5. Thromboembolism

Thrombus formation and plaque rupture are believed to be the final step in the development of a coronary occlusion. Aspirin (Acetylsalicylic acid) to reduce platelet aggregation, anticoagulants to prevent clotting and fibrinolytics to encourage thrombus breakdown have all been evaluated in clinical studies. Aspirin was reported to reduce coronary events, since the fatal and non-fatal MI were lower in those on aspirin. An anticoagulant warfarin reduced coronary mortality in postinfarct patients (Manson et al., 1991).
9.6. Antioxidant therapy

Antioxidants may prove to be effective cardioprotective drugs with the potential to reduce endothelial damage and atheroma formation. A variety of small molecules with powerful reducing (electron-donating) properties are present in the extracellular fluids to react with (scavenge) free radicals preferentially before they can damage more important molecules. These include vitamin C (ascorbic acid), vitamin E (alpha-tocopherol) and beta-carotene. Vitamin E and beta-carotene protect the core polyunsaturated fatty acids whose oxidation provides most of the toxic products. Antioxidant vitamins and drugs such as probucol and beta-hydroxytoluene in animal models of atherosclerosis yielded promising results suggesting that the atherogenic process could indeed be retarded (Maxwell, 1993).

9.7. Statins in cardiovascular risk reduction

The inhibitors of Hydroxy Methyl Glutaryl-Coenzyme A Reductase (I-HMG-CoA-R) or statins have become the cornerstone of drug therapy that aimed at reducing cardiovascular risk. Statins are the pharmacological group with the highest reduction power of the serum LDL-C concentration. Thus, statins have become the most important pharmacological weapon for cardiovascular risk reduction when associated to atherosclerosis. The Statins mechanism of action is they specifically compete with HMG-CoA for the catalytic site of its reductase (HMGCoA-R). This competition inhibits the metabolic pathway of HMG-CoA into mevalonate, a precursor molecule for the synthesis of cholesterol and other molecules such as the isoprenoids, Farnesyl and Geranyl Pyrophosphates (Endo, 1992; Brown and Goldstein, 2004). Thus, by inhibiting the HMG-CoA-R, the mechanisms allow statins to reduce concentrations of cholesterol in serum.

Pravastatin or Atorvastatin Evaluation and Infection Therapy trial (Cannon et al., 2002) demonstrated improved outcomes, with hospitalisation rates for heart failure significantly reduced after an acute coronary syndrome. The Reversal of Atherosclerosis with Aggressive Lipid Lowering trial (Nissen et al., 2004) demonstrated reduced rates of progression of atherosclerosis after intensive
atorvastatin treatment when compared with moderate pravastatin treatment. Statins are responsible for a wide range of adverse effects, ranging from mild gastro-intestinal disturbances to life-threatening conditions. Excluding Cerivastatin, current statins on the market have a very good safety profile and a proven reduction in mortality due to CVD (Wilt et al., 2004).

10. Cardioprotective Medicinal plants and Natural products

Modern drugs are effective in the control of CVD, but due to their side effects those drugs utilization is limited (Stollberger and Finsterer, 2005). So there is in a need to search for an effective medicine to treat CVD without any side effects. The prevention of CVD has been associated with the ingestion of fresh fruits, vegetables or plants rich in natural antioxidants (Argolo et al., 2004). The Indian herbal medicine is one of the prominent systems to treat diseases without any side effects. Recently, several plants of Indian origin have been found to possess medicinal properties with their beneficial effects in ailments like atherosclerosis, ischemia, cancer, diabetes and liver dysfunction (Vandana and Suresh, 2008).

The traditional systems of medicines - Ayurveda, Siddha and Unani are based on the experiences in the use of plant products in amelioration of common diseases. More than 2000 plants have been listed in the traditional (Herbal/Alternative) systems of medicine and some of these are providing comprehensive relief to the people suffering from cardio-vascular diseases, specially “hyperlipidemia” and “IHD”. WHO reports indicate that around eighty percent of the global population still relies on botanical drugs and several herbal medicines have advanced to clinical use in modern times (Mahmood et al., 2010). The use of Western medicinal drugs for the treatment of hypertension, congestive heart failure and post myocardial infarction are widely accepted. For CVD, herbal treatments have been used in patients with CHF, systolic hypertension, angina pectoris, atherosclerosis, cerebral insufficiency, venous insufficiency and arrhythmia (Mashour et al., 1998). Various phytoconstituents from plants were responsible for cardioprotective activity including carotenoids (Eugenia...
uniflora); triterpenes (Ganoderma lucidum); flavonoids (Anacardium occidentale, Nelumbo nucifera); cardiac glycosides (Digitalis purpurea, Antiaris toxicaria); alkaloids (Desmodium gangeticum, Erythroxylon coca Tinospora cordifolia); saponins (Asparagus racemosus, Vaccaria pyramidata); terpenoids (Ginkgo biloba); fatty acids (Elaeis guineensis) (Arya and Gupta, 2011) etc. Herbal medicines have been given a valuable status and readily available products for primary health care, and WHO has endorsed their safe and effective use (WHO Research Guidelines, 1993).

11. Terminalia pallida

Terminalia pallida (T. pallida) belongs to the family Combretaceae is commonly known as “Haritaki,” is a small evergreen endemic tree (Chada, 1976) mainly distributed in Tirumala hills, Rayalaseema region, Andhra Pradesh, India which lie geographically in the South-eastern ghats, are known for the rich heritage of plants with known and unknown medicinal values. The fruits of T. pallida were collected from Mogalipenta, Andhra Pradesh, identified by the taxonomist of the Herbarium, Department of Botany, S.K.University, Anantapur. A voucher specimen was deposited in the herbarium of the department of Botany, S.K.University, Anantapur [Authentication Code No. 38609(SKU)].

11.1. Traditional and medicinal use of Terminalia pallida

Many plants are in use for the treatment of heart related ailments (Mamtani and Mamtani, 2005; Sun et al., 2005). The search for an effective medicine to treat cardiovascular disorders without any side effects has lead to the use of traditional plant based medicine. The indigenous plants such as Inula racemosa (Ojha et al., 2011), Punica granatum (Mohan et al., 2010), Moringa oleifera (Nandave et al., 2009) and Embelia ribes Burm (Bhandari et al., 2008) have been proved for their cardioprotective activity against isoproterenol-induced myocardial infarction in rats.

In the ethnobotanical claims, the T. pallida fruits are used for the treatment of diabetes by the tribal people of Andhra Pradesh, India. Maceration of fruit powder is orally given for the treatment of diabetes (Thammanna et al., 1990). The fruit decoction cures diarrhoea. The dry fruit kernel of T. pallida along with roots of
Pimpinella tirupatiensis are given in the treatment of venereal disease and peptic ulcers (Nagaraju and Rao, 1989). T. pallida fruits have antidiabetic (Kameswara rao et al., 2003), antibacterial activity and diuretic property (Malaya Gupta et al., 2002). The ethanolic extract of T. pallida fruits (TpFE) has antibacterial and antifungal activity (Jeevan Ram et al., 2004), antiulcer activity (Gupta et al., 2005), antioxidant and hepatoprotective activity (Palani et al., 2009). Kameswara rao et al. (2003) and Malaya Gupta et al. (2002) screened the T. pallida fruits for the presence of major phytochemical constituents like flavonoids, saponins, triterpenes, anthraquinones, alkaloids, phenolic acids, tannins, steroids, polyphenolic flavonoids, anthocyanidins and terpenoids. T. pallida fruit consists of active principles like β-sitosterol, oleic acid, oleanolic acid, maslinic acid, ellagic acid and gallic acid (Gunasekar et al., 1993).

In Ayurveda T. pallida fruit is esteemed as a powerful cardiotonic and appropriately known as “Hridya”, as it possesses heart strengthening and cardiotonic properties. The other species of Terminalia such as T. arjuna and T. chebula also showed the cardio-protective effect against ISO-induced stress in rats (Parveen et al., 2011; Subramaniyan Suchalatha et al., 2005).
<table>
<thead>
<tr>
<th>Plant name</th>
<th>Family</th>
<th>Chemical Constituents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aichum sativum</td>
<td>Liliaceae</td>
<td>Alcin, sulfur compounds</td>
</tr>
<tr>
<td>Anacardium occidentale</td>
<td>Anacardiaceae</td>
<td>Flavonoids, carotenoids</td>
</tr>
<tr>
<td>Aronia borealis</td>
<td>Rosaceae</td>
<td>Cardiac glycosides</td>
</tr>
<tr>
<td>Asparagus racemosus</td>
<td>Asparagaceae</td>
<td>Saponins-Shatavariins I-IV</td>
</tr>
<tr>
<td>Cinnamomum tamala</td>
<td>Lauraceae</td>
<td>Cinnamaldehyde</td>
</tr>
<tr>
<td>Deinomon densatum</td>
<td>Ranunculaceae</td>
<td>Campesterol, stigmasterol, sitosterol, cholesterol, deltaaervasterol and alkaloids</td>
</tr>
<tr>
<td>Digitalis purpurea</td>
<td>Scrophulariaceae</td>
<td>Cardiac glycosides</td>
</tr>
<tr>
<td>Eugenia uniflora</td>
<td>Orchidaceae</td>
<td>Carotenoids, flavonoids</td>
</tr>
<tr>
<td>Ganoderma lucidum</td>
<td>Ganodermaeae</td>
<td>Terpenes</td>
</tr>
<tr>
<td>Ginkgo biloba</td>
<td>Ginkgoalaceae</td>
<td>Ginkgo flavone glycosides, terpenoids (ginkgolides and baicalein)</td>
</tr>
<tr>
<td>Hedera helix</td>
<td>Araliaeae</td>
<td>Coumarans-ignoids, hernadesine</td>
</tr>
<tr>
<td>Leonotis nycthemera</td>
<td>Araliaceae</td>
<td>Triterpenoids</td>
</tr>
<tr>
<td>Neolimba nudifera</td>
<td>Neolimbeae</td>
<td>Quercetin, luteolin, alkaloids</td>
</tr>
<tr>
<td>Onosma procumbens</td>
<td>Boraginaceae</td>
<td>Tannins, Glycosides, resins, alkaloids</td>
</tr>
<tr>
<td>Eleusine guineensis</td>
<td>Araceae</td>
<td>Fatty acids, omega-3 fatty acid</td>
</tr>
<tr>
<td>Quercus resinae</td>
<td>Fagaceae</td>
<td>Tannins</td>
</tr>
<tr>
<td>Rosa damascena</td>
<td>Rosaceae</td>
<td>Lycope, rutaxanthin, zeaxanthin, quercetin, kaempferol and cyanid</td>
</tr>
<tr>
<td>Trapaescor copsulifera</td>
<td>Menispermaeae</td>
<td>Alkaloidal constituents, including berberine, bitter principles, including columbin,</td>
</tr>
<tr>
<td>Euphorbia ceroe</td>
<td>Euphorbiaceae</td>
<td>chasmanthin, palmarin and tinosporon, tinosporic acid and tinosporol</td>
</tr>
<tr>
<td>Hordeum vulgare</td>
<td>Gramineae</td>
<td>Luteolin and fusicoccic acid, saponins</td>
</tr>
<tr>
<td>Croton aurantium</td>
<td>Rutaceae</td>
<td>Vitamin C, β-glucan-enriched fraction</td>
</tr>
<tr>
<td>Gnidia maxima</td>
<td>Passifloraceae</td>
<td>Vanilla oil, alkaloid</td>
</tr>
<tr>
<td>Asperulus hypoxystorum</td>
<td>Hippocastanaceae</td>
<td>Protein, lecithin, saponins</td>
</tr>
<tr>
<td>Hoppea rosmarinoidae</td>
<td>Rosmarinaceae</td>
<td>Hydrocinnamolin</td>
</tr>
<tr>
<td>Rapanea cornutus</td>
<td>Cruciferae</td>
<td>Polyspholol</td>
</tr>
<tr>
<td>Veratrum pyreoides</td>
<td>Ranunculaceae</td>
<td>Caffeic acid</td>
</tr>
<tr>
<td>Euphorbia lathyris</td>
<td>Nymphaeaceae</td>
<td>Saponins</td>
</tr>
<tr>
<td>Stachysphora amplexicorns</td>
<td>Verbenaceae</td>
<td>Protein</td>
</tr>
<tr>
<td>Desmodium gongylosum</td>
<td>Fabaceae</td>
<td>Friedelin, stigmasterol, usnic acid, hespidin, securilarin, choline, phenolic acids</td>
</tr>
</tbody>
</table>

Table: A BRIEF DESCRIPTION OF COMMON CARDIOPROTECTIVE PLANTS
Fig: VARIOUS CARDIOPROTECTIVE PHYTOCONSTITUENTS FROM PLANTS
Fig: VARIOUS CARDIOPROTECTIVE PHYTOCONSTITUENTS FROM PLANTS
Fig: *Terminalia pallida* plant and *Terminalia pallida* fruits

- **Gallic acid**
- **Ellagic acid**
- **Beta-sitosterol**
- **Oleic acid**
Maslinic acid

The prevention of CVD has been associated with the ingestion of fresh fruits, vegetables or plants rich in natural antioxidants. The protective effects of plants can be due to the presence of triterpenoids (Jingjing et al., 2007). A variety of physiological activities have been reported to the class of triterpene compounds, including anti-inflammatory, hypolipidaemic, hepatoprotective, gastroprotective, anticancer, anti-diabetic, anti-human immunodeficiency virus (HIV), anti-atherosclerotic, antiulcerogenic, antimicrobial and immunoregulatory effects (Dzubak et al., 2006). Maslinic acid (MA) is a pentacyclic triterpenoid compound that exists widely in fruits and leaves of medicinal plants like *Olea europaea* and *Terminalia pallida* (Montilla et al., 2003; Gunasekar et al., 1993). The compound has attracted much interest due to its proven pharmacological safety and its many biological activities such as anti-hyperlipidaemic (Jun et al., 2007), antioxidant (Montilla et al., 2003), anti-inflammatory (Aladedunye et al., 2008), anticancer (Fernando et al., 2011), anti-malarial (Carlos Moneriz et al., 2011), anti-hyperglycemic, anti-diabetic (Xu-Zhen Tang et al., 2008), antibacterial and anti-viral activities (Scalon Cunha et al., 2007). MA has a similar structure with Oleanolic acid (OA). OA has proved for its potent...
cardioprotective effect against ISO-induced MI in rats (Senthil et al., 2007). As MA is a triterpenoid, the structure also resembles with the cardioprotective compound OA and there is no literature on its cardioprotective activity, we attempt to evaluate the cardioprotective effect of MA.

13. Bioinformatics

Bioinformatics can be viewed as the use of computational methods to make biological discoveries (Baldi and Brunak, 1998). It is an interdisciplinary field involving biology, computer science, mathematics, and statistics to analyze biological sequence data, genome content and arrangement, and to predict the function and structure of macromolecules. The ultimate goal of the field is to enable the discovery of new biological insights as well as to create a global perspective from which unifying principles in biology can be derived (Altman et al., 2001). Different biological problems considered within the scope of bioinformatics involve the study of genes, proteins, nucleic acid structure prediction, and molecular design with docking.

13.1. Molecular Docking

When two molecules are in close proximity, it can be energetically favorable for them to bind together tightly. The molecular docking problem is the prediction of energy and physical configuration of binding between two molecules. A typical application is in drug design, in which one might dock a small molecule that is a described drug to an enzyme one wishes to target. For example, HMG Co-A reductase is an essential regulatory enzyme for cholesterol synthesis. The chemical action of the enzyme takes place at a localized active site on its surface. HMG Co-A reductase inhibitor drugs are small molecules that bind to the active site in HMG Co-A reductase and stay there, so that the normal functioning of the enzyme is prevented. Docking software allows us to evaluate a drug design by predicting whether it will be successful in binding tightly to the active site in the enzyme. Based on the success of docking, and the resulting docked configuration, designers can refine the drug molecule (Lesk, 2002).
Since the initial discovery of epinephrine, the principal active substance from the adrenal gland, the pharmacology and physiology of a large group of endogenous and synthetic catecholamines have been characterized (Barger and Dale, 1910). Catecholamines mediate their cardiovascular actions predominantly through α1, β1, β2, and dopaminergic receptors. β1-Adrenergic receptor stimulation results in enhanced myocardial contractility through Ca²⁺ mediated facilitation of the actin myosin complex binding with troponin C and enhanced chronicity through Ca²⁺ channel activation. β2-Adrenergic receptor stimulation on VSMC through a different intracellular mechanism results in increased Ca²⁺ uptake by the sarcoplasmic reticulum and vasodilation.

Dopamine, an endogenous central neurotransmitter, is the immediate precursor to norepinephrine in the catecholamine synthetic pathway. When administered therapeutically, it acts on dopaminergic and adrenergic receptors to elicit a multitude of clinical effects. Dopamine effects the stimulation of dopaminergic D1 postsynaptic receptors concentrated in the coronary, renal, mesenteric, and cerebral beds and D2 presynaptic receptors present in the vasculature and renal tissues promotes vasodilation and increased blood flow to these tissues.

Dobutamine is a synthetic catecholamine with a strong affinity for both β1- and β2-receptors. With its cardiac β1-stimulatory effects, dobutamine is a potent inotrope, with weaker chronotrophic activity. Despite its mild chronotropic effects, dobutamine significantly increases myocardial oxygen consumption.

Norepinephrine, the major endogenous neurotransmitter liberated by postganglionic adrenergic nerves, is a potent α1-adrenergic receptor agonist with modest β-agonist activity. Prolonged norepinephrine infusion can have a direct toxic effect on cardiac myocytes by inducing apoptosis via protein kinase A activation and increased cytosolic Ca²⁺ influx (Communal et al., 1998).

Epinephrine is an endogenous catecholamine with high affinity for β1, β2, and α1-receptors present in cardiac and vascular smooth muscle. High and prolonged
doses can cause direct cardiac toxicity through damage to arterial walls, which causes focal regions of myocardial contraction band necrosis, and through direct stimulation of myocyte apoptosis (Singh et al., 2001).

Phenylephrine with its potent synthetic α-adrenergic activity and virtually no affinity for β-adrenergic receptors, it is used primarily as a rapid bolus for immediate correction of sudden severe hypotension. It can be used to raise mean arterial pressure (MAP) in patients with severe hypotension and concomitant aortic stenosis. This agent has virtually no direct effect on heart rate.

Isoproterenol (ISO) is a potent, nonselective, synthetic β-adrenergic agonist with very low affinity for α-adrenergic receptors. It has powerful chronotropic and inotropic properties, with potent systemic and mild pulmonary vasodilatory effects.

14.1. Isoproterenol-induced myocardial infarction

ISO (4-[1-hydroxy-2-(1-methylethylamino)ethyl]benzene-1,2-diol), a synthetic catecholamine and β-adrenergic agonist by its positive inotropic and chronotropic actions, increases the myocardial oxygen demand that causes severe stress in the myocardium resulting in necrosis of the heart muscle (Bin Liu et al., 2008). A number of pathophysiological mechanisms have been proposed to explain the ISO-induced myocardial damage, including altered permeability, increased turnover of norepinephrine, and generation of cytotoxic free radicals on autooxidation of catecholamine. Oxidative stress increases cAMP levels by exhausting ATP and decreases sarcolemmal Ca2+ transport, resulting in intracellular calcium overload, which leads to ventricular dysfunction and contractile failure in rat heart (Bhagat et al., 1976, Tappia et al., 2001). ISO causes severe stress in the myocardium by enhancing the free radical formation which causes increased LPO. This increased LPO causes infarct-like necrosis of the heart muscle (Rajadurai and Prince, 2005; Biemond et al., 1986). It has also been suggested that heart failure subsequent to MI may be associated with antioxidant deficit as well as increased myocardial oxidative stress (Hill and Singal, 1996). ISO increases the level of myocardial lipids, mainly the LDL-C in the blood (Manjula and Shyamaladevi, 1993). The increased levels of cardiac marker enzymes are better markers in the detection of cardiac injury (Mair, 1997). The lesions
produced by ISO in rat heart are similar to those found in myofibrillar degeneration in human IHD (Milei et al, 1978). Hence, the study of ISO-induced myocardial necrosis and its underlying mechanisms might provide better insight and new leads on the pathogenesis of IHD.

Fig: Simplified schematic of postulated intracellular actions of β-adrenergic agonists. β-Receptor stimulation, through a stimulatory Gs-GTP unit, activates the adenyl cyclase system, which results in increased concentrations of cAMP. In cardiac myocytes, β1-receptor activation through increased cAMP concentration activates Ca\(^{2+}\) channels, which leads to Ca\(^{2+}\) mediated enhanced chronotropic responses and positive inotropy by increasing the contractility of the actin-myosin-troponin system. In vascular smooth muscle, β2-stimulation and increased cAMP results in stimulation of a cAMP-dependent protein kinase, phosphorylation of phospholamban, and augmented Ca\(^{2+}\) uptake by the sarcoplasmic reticulum (SR), which leads to vasodilation.
Dopamine

Norepinephrine

Epinephrine

Dobutamine

Isoproterenol

Phenylephrine
Fig: Chemical structures and names of common catecholamines.

Fig: Mechanism of induction of myocardial injury by ISO
AIM AND SCOPE

The search for an effective medicine to treat cardiovascular disorders without any side effects has lead to the use of medicinal plants and natural products. However, extensive literature survey has shown that there are no systematic scientific reports available in support of the cardioprotective activity of *T. pallida* fruit and its active constituent MA. Therefore, first time we have assessed their role against ISO-induced MI in rats.

Objectives

- To prepare ethanolic extract of *T. pallida* fruits (TpFE).
- To select the effective dose of TpFE by dose dependent study.
- To standardize the TpFE by HPLC.
- To measure the heart weight and body weight to assess heart coefficient in control and experimental rats.
- To estimate cardiac marker enzymes like CK, CK-MB, LDH, AST, ALT, ALP, and GGT in serum and heart tissues.
- To study lipid profile parameters like total cholesterol, triglycerides, HDL-C, LDL-C, and VLDL-C in serum and heart tissues.
To estimate the extent of LPO in serum and heart tissues.

To estimate the activity of XOD in heart tissues.

To estimate electrolytes such as sodium, potassium and calcium in serum.

To estimate membrane bound transport enzymes like Na⁺/K⁺ ATPases, Ca²⁺ ATPases and Mg²⁺ ATPases in heart tissues.

To estimate the content of antioxidant GSH, and the activity of antioxidant enzymes GPx, CAT, SOD and GST in heart tissues.

To estimate the enzymes LCAT and Paraoxonase in serum.

To estimate the lipid metabolizing enzymes LPL in liver, and HMG Co-A reductase in both heart and liver tissues.

To estimate the content of NO in serum.

To assess the expression of iNOS protein by SDS-PAGE and western blot analysis in heart tissues.

To study the effect of MA on cell lines by MTT assay.

To analyze the docking studies of the T. pallida fruit active constituents with regulatory enzyme HMG Co-A reductase.

To carry out histological studies of heart tissues by simple light microscopy.