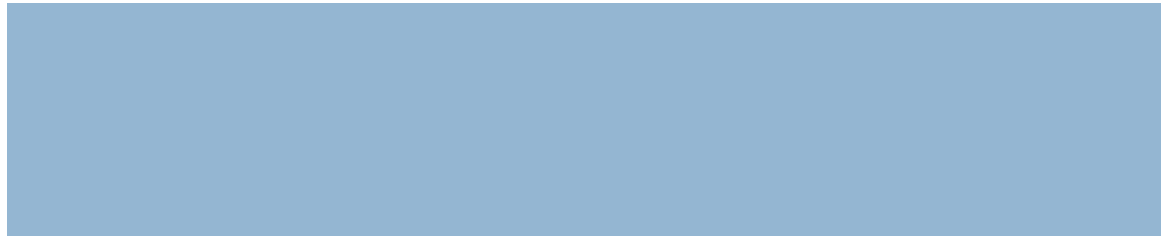


SUMMARY



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Cardiovascular diseases (CVDs) are a group of disorders of the heart and blood vessels which include coronary heart disease, cerebrovascular disease, peripheral artery disease, congenital heart disease. According to World Health Organisation (WHO), approximately 17 million people die of CVDs, particularly heart attacks and strokes, every year. Moreover, WHO has estimated that by 2020, CVDs will be the largest cause of death and disability in India. Sedentary life style and unhealthy diet are the major risk factors for development of CVDs. Further, the major fatal consequences of CVDs include heart failure and stroke, which are caused by the blockage that prevents blood from flowing to the heart or brain. Such blockage results from the build-up of fatty deposits in the inner walls of the major blood vessels.

Atherosclerosis is the accumulation of lipid-rich plaques within the walls of large arteries, caused by abnormalities in lipids and lipoprotein metabolism. It begins with endothelial dysfunction leading to generation of reactive oxygen species (ROS) that cause the oxidation of LDL in the subendothelial space progressing from minimally modified LDL (mmLDL) to extensively oxidized LDL (ox-LDL). Elevated levels of oxidized LDL have been considered to be a biochemical marker for coronary heart disease. Ox-LDL induces chemotaxis through chemokines (MCP-1 and CCR-2) and recruits monocytes to intima where they differentiate into macrophages with the help of M-CSF. Ox-LDL is taken up by macrophages via scavenger receptors and where it accumulates leading to development of foam cell formation, which is known as hall mark of both early and late atherosclerotic lesions. Acute myocardial Infarction (AMI) is the ischemic condition that develops due to Atherosclerosis which reduces

blood supply to the myocardium. The scarcity of oxygen and nutrients supply to cardiac cell causes damage to cardiac tissue and can compromise survival. AMI is commonly known as heart attack.

The treatment options for the CVDs are based on the treatment of underlying causes such as hypercholesterolemia and hypertension. These include drugs which reduce blood pressure (hypertension), lower cholesterol levels and prevent blood clots. However, they are generally associated with common side effects like dizziness, tiredness and headache and can interfere with heart and kidney functions. This has led researchers to develop medication based on herbal formulations for treatment of CVDs.

STUDY 1:

*Anethumgraveolens*L. (AG) belonging to *Apiaceae*(*Umbelliferae*) family, is an annual aromatic herb known for culinary and medicinal use since ancient times. This study was aimed to assess the toxicity profile of seed extract of AG by determining its effects after administration of acute and sub-chronic doses in swiss albino mice. Swiss albino mice were divided into four groups of six animals in each group. In acute toxicity, study the mice were administered single oral dose of 1000, 2000, 3000 and 5000mg/kg body weight and general behaviour, adverse effects and possible mortality were observed for 24 hours after administration. In the chronic dose study, the extract was administered orally at the doses of 1000, 2000 and 3000mg/kg body weight as treatment groups and Carboxy Methyl Cellulose (0.5%) as control group for 28 days. There was neither adverse effect nor death in treated groups. There were no alterations in organ weight, hematological profile and biochemical parameters. The overall finding of this study

indicates that AG extract is absolutely non-toxic and can be considered safe for human consumption as a therapeutic herb.

STUDY 2:

Isoproterenol (ISO) a synthetic catecholamine and β -adrenergic agonist, has been found to cause severe stress in the myocardium resulting in infarct like necrosis of the heart muscles. In the present study, we evaluated the preventive effect of AG on lipid peroxides, enzymatic, nonenzymatic antioxidants and histopathological findings in normal and ISO-induced myocardial infarction (MI) in male albino Wistar rats. AG pretreatment prevented the ISO induced increase in the serum levels of CK-MB, LDH, AST, ALT and uric acid levels in a dose dependent manner. AG pretreatment also improved lipid profile and prevented decrement in enzymatic (SOD, CAT, GPx and GST) and non-enzymatic (GSH and AA) antioxidants in cardiac tissue. AG extract decrease lipid peroxidation levels of cardiac tissues. Increment in the activities of Na^+/K^+ ATPase, Mg^{2+} ATPase and Ca^{+2} ATPase of cardiac tissues indicated the protective effects of AG extract on ISO treatment. Based on the results of present study it can be concluded that the presence of antioxidants in AG extract prevented myocardial damage.

Reactive oxygen species (ROS) plays pivotal role during myocardial infarction. The aim of the present study was to evaluate the protective effect of *Anethumgraveolens* L. (AG) against H_2O_2 induced oxidative stress in H9C2 cells. The H9C2 cells were incubated with 10 mM H_2O_2 and different concentrations of AG extract for 24 h. Cell viability and LDH release assays showed AG had successfully prevented H_2O_2 induced cardiomyoblasts death and prevention on LDH enzyme leakage from damage cells. Reduced levels of lipid peroxidation and intracellular ROS production indicated that the presence of antioxidants

in AG were capable to prevent H₂O₂ mediated ROS production and lipid peroxidation. AG extract also prevented H₂O₂ mediated reduction in mitochondrial membrane potential. Acridine Orange/Ethidium Bromide and Propidium iodide staining of cardiomyoblasts showed that AG prevented H₂O₂ induced cardiomyoblasts cell death. Based on the results of the present study, it can be concluded that AG extract has strong antioxidant property and it successfully prevents H₂O₂-induced cytotoxicity.

STUDY 3:

Present study was designed to evaluate the effect of *Anethumgraviolensis*L. extract (AG) on LDL oxidation and foam cell formation *in vitro*. We investigated the effects of AG extract on the oxidation of LDL and the uptake of lipid in RAW 264.7 macrophages. Initially we isolated LDL from blood donated healthy donors and subjected to oxidation using copper (II) sulfate (CuSO₄) as an oxidative inducer in presence and absence of AG extract. AG extract prevented LDL oxidation and it was observed by increased lag time and decreased Conjugate diene formation. AG successfully reduced levels of Malondialdehyde (MDA), Lipid hydroperoxide (LHP) and Protein carbonyls (PC). Oxidised LDL was incubated with RAW 264.7 macrophage cells in presence and absence of AG extract. AG extract successfully prevented lipid accumulation and reduced foam cell formations. AG extract also reduced levels of intracellular oxidative stress, mitochondrial membrane potential assay and cell viability. These results demonstrate the protective effect of AG on LDL oxidation and lipid accumulation in macrophages.

Atherosclerosis is caused by heightened levels of oxidative stress, cholesterol, inflammation induced endothelial dysfunction and formation of plaque beneath endothelial lining of aorta. The present study was design to evaluate the

antiatherosclerotic effect of AG extract using atherosclerosis animal model of rats. Twenty-four rats were assigned to the control, high cholesterol diet (ATH) and ATH + AG group that were fed with high cholesterol diet co-supplemented with AG. Serum levels of triglyceride (TG), total cholesterol (T-CHO), low density lipoprotein cholesterol (LDL-C), high density lipoprotein cholesterol (HDL-C) and malondialdehyde (MDA) were detected at the end of experiments. Vascular cell adhesion molecules (VCAM-1), and P-selectin were investigated by immunohistochemistry of thoracic aorta of rats. Serum levels of TG, TC, LDL-C and VLDL-C levels were significantly increased in ATH group as compared to Control group while AG co-supplementation showed significant decrement as compared to ATH group. HDL level also significantly increased in ATH + AG as compared to ATH group. Immunohistochemistry of thoracic aorta also suggested that the expression of VCAM-1 and P-selectin was significantly reduced as compared to ATH group. Based on the results of present study, it can be concluded that the AG extract prevented the atherosclerotic changes by lowering serum lipid levels, oxidation of lipids and suppressing the inflammatory response.