2. REVIEW OF LITERATURE

This chapter summarizes the review of literature which helped in designing and executing the present piece of work.

2.1 MYOCARDIAL INFARCTION - OVERVIEW

Myocardial infarction is defined as myocardial cell death due to prolonged ischemia. After the onset of myocardial ischemia cell death is not immediate but takes a finite period to develop. It takes 6 hours before myocardial necrosis can be identified by standard macroscopic or microscopic postmortem examination. Myocardial necrosis results in and can be recognized by the appearance in the blood of different proteins, released into the circulation due to the damaged myocytes. Myocardial infarction can be experimentally induced in animals by invasive methods, which includes ISO induced myocardial infarction ( Arnaldo et al., 2004).

2.1.1 EXPERIMENTAL INDUCTION OF MYOCARDIAL INFARCTION BY ISOPROTERENOL

Isoproterenol[1-(3’,4’-dihydroxyphenyl)-2-isopropylaminoethanol hydrochloride] (ISO) a synthetic catecholamine (Fig 2.1), acts as a β-adrenergic agonist and has been found to cause severe stress in the myocardium resulting in the depletion of energy reserve of cardiac muscle cells and causes complex biochemical and structural changes leading to cell damage and necrosis (Rona, 1985). Catecholamines rapidly undergo auto-oxidation and it has been suggested that the oxidative products of catecholamines are responsible for changes in the myocardium (Yates and Dhalla, 1975). ISO induced myocardial necrosis also involves membrane permeability alterations that bring about loss of function and integrity of myocardial membranes (Todd et al., 1980).
ISO induced myocardial damage is considered as one of the most widely used experimental models to study the beneficial effects of many drugs and cardiac function (Grimm et al., 1998). Exposure of the heart to high concentration of ISO had been reported to result in the development of necrotic lesions in the myocardium of experimental animals. The pathophysiological changes due to ISO induced myocardial infarction in rats are comparable to those taking place in human myocardial infarction (Wexler and Greenberg, 1978).

![Fig 2.1 Structure of Isoproterenol](image)

**Fig 2.1 Structure of Isoproterenol**

where, $R$ is $\text{CH(OH)CH}_2\text{NHC}_3\text{H}_7$

### 2.1.2 ROS in the Pathogenesis of Isoproterenol Induced Myocardial Infarction

ISO, a synthetic catecholamine, develop cardiotoxicity when administered in higher concentrations (Jewett et al., 1989). ISO undergoes auto-oxidation which results in the generation of excess amount of electrons (Chen et al., 2000). These electrons can reduce oxygen molecules resulting in the generation of reactive oxygen species (ROS). ROS can initiate lipid peroxidation reactions and propagate cell membrane damage (Ferrari et al., 1991). Moreover, it is also suggested that ROS causes vascular damage by two mechanisms like, ROS can cause severe functional and metabolic disorders,
modification of proteins and membrane lipids, and depletion of glutathione in endothelial cells and also ROS can indirectly affect the function of endothelial cells by reacting with endothelial derived relaxation factor (EDRF or NO), thus, decreasing endothelium dependent vasorelaxation (Acworth et al., 1997). Oxidation of ISO produces the metabolite catechol-o-quinone, a potent reactant with the sulphhydryl groups and in turn, oxidation of protein sulphhydryl groups. Intra-cyclization of o-quinone produces aminochromes which can cause myocardial cell damage (Thompson and Hess, 1986).

**Fig 2.2 Auto-oxidation of ISO and Genesis of Heart Disease**

![Diagram of auto-oxidation of ISO and genesis of heart disease](image)

Isoproterenol

\[
\text{O}_2^- + \text{H}_2\text{O}_2 + \cdot \text{OH}^- \quad \text{Reactive Oxygen Species}
\]

Imbalance in oxidants and antioxidants

Oxidative stress, Tissue injury and cell death
An overview of the potential role of oxidative metabolites derived from ISO auto-oxidation in the genesis of heart disease is shown in Fig 2.2. The ISO induced oxidative stress is reported by various researchers. Sathish et al., (2003) have mentioned that ISO generate free radicals which in turn, stimulate lipid peroxidation a causative factor for irreversible damage to the myocardial membrane. ISO induced oxidative stress and cardiotoxicity is also reported by other researchers including Rajadurai and Prince (2006); Farvin et al., (2004); Ahmed et al., (2004).

2.1.3 LIPID ABNORMALITIES IN THE PATHOGENESIS OF ISOPROTERENOL INDUCED MYOCARDIAL INFARCTION

Reactive oxygen species formed from auto-oxidation of ISO results in the oxidation of LDL. Generation of ox-LDL takes place in micro domains in the arterial wall (Carmena et al., 1996). NADP(H) oxidase, myeloperoxidase, Cytochrome P450, mitochondrial electron transport chain, peroxynitrite, xanthine oxidase, ceruloplasmin, and lipoxigenase are involved in the oxidation of LDL. LDL is oxidized by different mechanisms like transition metal mediated oxidation, oxidation mediated by reactive oxygen species and myeloperoxidase (Heinecke, 1998). The mechanism of oxidation of LDL by ROS generated from ISO is very interesting.

ROS formed from ISO attacks the double bonds of polyunsaturated fatty acid (PUFA) present in LDL and thereby, initiate lipid peroxidation. This results in the removal of hydrogen atom from a methylene group. Molecular rearrangement of the resulting unstable carbon radical results in a more stable conjugated diene. The conjugated diene reacts with molecular oxygen and forms a peroxyl radical (Abuja and Esterbauer, 1995). This peroxyl radical derives a hydrogen atom from a neighboring
PUFA and results in the formation of hydroperoxide and other lipid radicals, thereby, initiating the chain propagation. Removal of hydrogen atoms by the peroxyl radical from other lipids including cholesterol eventually yields oxysterols. Lipid hydroperoxides fragment to shorter chain aldehydes, malondialdehyde and 4-hydroxynonenal. These reactive aldehydes in turn, may bind to amino groups of apo-B100 giving the protein an increased net negative charge. The classical LDL receptor recognizes a specific domain of the positive charges. Alteration of the domain results in failure of binding by the apoB/E receptor and an increase in the negative surface charge of apo-B100, resulting in increased recognition by the scavenger receptor present on the surface of macrophages (Xu et al., 1999).

Ox-LDL potentially promotes atherogenesis by binding with macrophages and forms foam cells. It is a chemo-attractant for monocytes (Parthasarathy et al., 1999) and T cells. It also stimulates the release of monocyte chemo-attractant Protein-1 along with macrophage colony stimulating factor from endothelial cells. Thereby, ox-LDL inhibits endothelial cells dependent arterial relaxation. Moreover, ox-LDL activates matrix digesting enzymes which play a role in plaque instability (Young and McEneny, 2001).

2.1.4 CHANGES IN LYSOSOME AND MITOCHONDRIA IN THE PATHOGENESIS OF ISOPROTERENOL INDUCED MYOCARDIAL INFARCTION

Oxidative stress, in the form of hydrogen peroxide, formed from auto-oxidation of ISO, rapidly ruptures lysosomes. Rupture of lysosome is followed by caspase dependent apoptosis or caspase independent apoptosis like cell death or necrosis (Brunk et al., 2001). The lysosomal membrane permeabilisation may occur due to lipid peroxidation of the membrane PUFA and the generated apparent ultra structural changes results in release
of a large number of acid hydrolases. Moreover, the lysosomal content of redox-active iron may induce intra-lysosomal Fenton-type reactions at oxidative stress situations with ensuing peroxidation of lysosomal limiting membranes and releases the damaging contents, such as cathepsins and redox-active iron to the cytosol (Tenopoulou et al., 2005). Consequences of decreased oxygen supply to myocardium are increased anaerobic glycolysis, and increased lactic acid associated with decrease in pH. In the acidic pH most of the acid hydrolases are activated resulting in further damage of the cell (Williamson et al., 1976).

The acid hydrolases induce mitochondrial permeability, through various proteins like phospholipases and the Bcl-2 family members like Bid, Bax and Bak. These Bcl-2 family members are cleaved by cathepsins. The end products are translocated to mitochondria resulting in Bcl-2 family members activation. Consequently, pro-apoptotic mitochondrial factors such as cytochrome C, enzymes involved in TCA cycle enzymes and proteins involved in the complexes of oxidative phosphorylation and apoptosis inducing factor are released into the cytosol, inducing cell death (Alexei et al., 2006).

2.1.5 OTHER PHYSIOLOGICAL CHANGES IN ISOPROTERENOL INDUCED MYOCARDIAL INFARCTION

Yogeeta et al., (2006) have mentioned that ISO administration induced morphological and functional alterations in the heart. ISO also increases calcium overload, leads to a deleterious high energy phosphate deficiency by the excessive activation of calcium dependent intracellular ATPases and by impairing the phosphorylating capacity of mitochondria, altered electrolyte level and phospholipase activation (Sathish et al., 2003). Farvin et al., (2004) have specified that ISO enhance
adenylate cyclase activity, increased cAMP formation and cardiac hypertrophy. Thus, it is evident that ISO induced myocardial infarction relates to myocardial necrosis as seen in humans with all related physiological and biochemical alterations.

2.2. **CYMBOPOGON CITRATUS**

*Cymbopogon citratus* (DC.) Stapf. (*C. citratus*) an ornamental grass (Fig 2.3) belonging to the family *Poaceae*, is global in distribution. The typical fresh taste of lemon grass made people of Srilanka, Vietnam, United States, Cambodia and other countries to use it as a spice and flavouring agent. It is a rich source of various phyto-constituents including terpenes, flavones, alkaloids, tannins, etc., This grass commonly known as fever grass is used in folk medicine for jaundice, to decrease the discharge from mucous membrane, antispasmodic and, analgesic agent and for the management of nervous disorders. It is commonly known as *lemon grass* in English, *Sera* in Hindi, *Karppurappul* in Tamil and *Kamanchi kasu* in Telugu.

2.2.1. **HABITAT AND MORPHOLOGICAL DESCRIPTION OF C. CITRATUS**

The herb is distributed in a large scale globally. Cymbopogon, the genus has about 55 species. *C. citratus*, a perrenial grass is universal in distribution. It is native to India and Malaysia. *C. citratus*, is now cultivated in India, West Indies, Africa, and tropical Asia. Among them India is considered as the main producer. Morphological characteristics of *C. citratus* is enumerated in Table 2.1.
FIG 2.3 *Cymbopogon citratus*

![Image of Cymbopogon citratus]

Division - Magnoliophyta  
Class - Liliopsida  
Subclass - Commelinidae  
Order - Poales  
Family - Poaceae  
Subfamily - Panicoideae  
Tribe - Androponeae  
Subtribe - Andropogoniniae
2.2.2. Ethnobotanical survey of *C. citratus*

*C. citratus* is a traditional herb used in Thai cooking. It is well known for its pungent lemony flavour. Tracking the rising popularity of Thai cooking in the United States, lemon grass is becoming a favourite of American gardeners. This fragrant grass is a versatile performer in the kitchen where it can be used in teas, beverages and herbal medicines (Giron *et al.*, 1991).

In traditional Chinese medicine, lemon grass is used to treat headaches, bloating, stomach aches, respiratory problems, cold, flu fever and rheumatic pains. It is also used as a blood purifier. It increases perspiration and can relieve muscle spasms. It is also used to treat muscle pain, infections, fever, colitis and indigestion (Lawless *et al.*, 1995; Blumenthal, 1998). In the Malay Peninsula, *C. citratus* is recommended in folk medicine for common colds, pneumonia and gastric problems (Carlini *et al.*, 1986). In Brazil, it is used in folk medicine for nervous conditions or gastrointestinal disturbances (Carlini *et al.*, 1986; Suzana *et al.*, 2001). A report of “Guatemalan use” lists *C. citratus* as a popular medicinal plant in Carib population (Lidia *et al.*, 1991).

In Thai, it is used in the treatment of fever, irregular menstruation, diarrhoea and digestive problems. It is also used in Central and South America for nervous conditions and helps to regulate blood pressure (Lawless *et al.*, 1995; Blumenthal, 1998).

In India, it is commonly used as an antitusive, antirheumatic and antiseptic agent. It is also used as an insecticide and food flavouring agent (Julia, 1992). Traditional Indian medicine employs lemon grass for fever, infection and sedation (Lawless, 1995).
### Table 2.1 Botanical Description of C. citratus

<table>
<thead>
<tr>
<th>S.no</th>
<th>Botanical description</th>
<th>Characteristics</th>
<th>Respective value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>General characteristics</td>
<td>Height</td>
<td>4 to 6 feet</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Spread</td>
<td>4 to 6 feet</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Plant habit</td>
<td>Round</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Plant density</td>
<td>dense</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Growth rate</td>
<td>Fast</td>
</tr>
<tr>
<td>2</td>
<td>Foliage</td>
<td>Leaf arrangement</td>
<td>most emerge from the soil, usually without a stem</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf type</td>
<td>Simple</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf margin</td>
<td>Entire</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf shape</td>
<td>Linear</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf venation</td>
<td>Parallel</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf type and persistence</td>
<td>Fragrant</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf blade length</td>
<td>90 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf blade width</td>
<td>16-18 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Leaf colour</td>
<td>Green</td>
</tr>
<tr>
<td>3</td>
<td>Flower</td>
<td>No flower</td>
<td>-</td>
</tr>
<tr>
<td>4</td>
<td>Fruit</td>
<td>No fruit</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>Trunk/bark/branches</td>
<td>Typically multi-trunked or clumping stems</td>
<td>-</td>
</tr>
</tbody>
</table>
2.2.3. Phyto-constituents of *C. citratus*

Plants are the rich source of various active constituents. Their isolation and purification is much important for evaluation of their pharmacological activity and molecular targeting. Various research work related with the isolation of phyto-constituents from *C. citraus* is reported earlier and some of them are compiled below.

*C. citratus* grass contains about 0.4% of volatile oil and that the oil contains 65% to 85% of citral and the concentration of citral depends on the geographical area grown (Carbajal *et al.*, 1989). Apart from citral, lemon grass oil also contains geraniol, myrcene, citronellal, limonene, linalool and dipentene (Torres, 1996).

The leaves also contain flavones like luteolin and its 7-O-β–Glucoside and 7-O–neohesperidoside, iso-orientin and 2-O-rhamnosyl iso-orientin, chlorogenic acid, caffeic acid p-coumaric acid, fructose, sucrose, octacosanol and triacontanol (De Matouschek, 1991).

Julia, (1992) has observed the presence of myrcene, linalool and geraniol in the oil. Sargenti *et al.*, (1997) have observed that GC analysis of essential oil from *C. citratus* is found to consist of geraniol and neral, geranic acid and nerolic acid. Other compounds found in the oil include Myrcene (12% to 25%), diterpenes, methylheptenone, citronellol, linalool, farmesol, other alcohols, aldehydes, linalool, terpineol and other minor fragrant components.

Various active constituents from *C. citratus* encompass stereoisomeric monterpene aldehydes like trans isomer geranial (40-62%), cis isomer neral (25-38%), nerol, limonene, linalool, β-caryophyllene etc., are reported by Sidibe *et al.*, (2001).
Composition of the oil separated by Cimanga et al., (2002) has revealed the presence of $\alpha$-Pinene – 1.1%, Camphene – 0.9%, Myrcene - 3.5%, Limonene - 3.1%, 1,8-Cineole - 0.2%, Citronellal - 0.2%, Linalool - 0.2%, Aromandendrene - 0.6%, Terpin-4-ol - 0.5%, Borneol - 3.7%, Geranial - 32.7%, Citronellol - 0.6%, Nerol 12.5%, Neryl acetate - 6.3%, Geranyl acetate - 0.1%, Eugenol - 0.3%.

Sharma et al., (2005) have mentioned the existence of various compounds like Cymbopogone, cymbopogonol, $\alpha$ and $\beta$ – citrals (72.29 %), myrcene (1.5 %), methylheptenone (0.2 %), linalool (1.2 %), linalyl acetae (0.1 %), 2- indecanone (0.3 %), geranyl acetate (0.1 %), citronellol (1.5 %), nerol, myrcene, limonene, methylheptenol, citral, geraniol, terpineol, Neral (29.0 %), Geranial (47.0 %), elemol (19.0 %), and unidentified sesquiterpene alcohol (19.0 %) in Saudi essential oil of $C.\textit{citratus}$. They have also brought up the presence of Geraniol (40.0 %), neral + geranial (13.0 %), $\alpha$ – oxobisabolene (12.0 %), menthone (0.2 %) and menthol (0.5 %) in Ethiopian essential oil.

The GC–MS spectra of the essential oil separated by Tognolini et al., (2006) from leaves of $C.\textit{bopogon citratus}$ by steam distillation shows the occurrence of various compounds at different percentages like Methyl-5-Hepten-2-one - 0.43 %, Myrcene - 15.48 %, Linalool - 1.28 %, Neral - 32.28 %, Geraniol - 3.35 %, Geranial - 41.28 %. They have also observed that percentage composition of non oxygenated monoterpenes as 15.48 %, oxygenated monoterpenes as 78.19 % and hydrocarbons as 0.43 %. The availability of flavonoids like luteolin and 6-C-glucoside has also been explained by Negrelle and Gomes, (2007). Structure of various phyto-constituents present in $C.\textit{citratus}$ are shown in Fig 2.4.
**FIG 2.4 STRUCTURE OF VARIOUS PHYTO-CONSTITUENTS OF *C. CITRATUS***

**CITRAL**

**GERANIOL**

**LINALOOL**

**LUTEOLIN 7-GLUCOSIDE**

**p-COUMARIC ACID**

**p-CAFFEIC ACID**

**CHLOROGENIC ACID**
2.2.4 Pharmacological activity of *C. citratus*

Recently many herbs have been discovered and biological activity of the crude extract and their active compounds have been partially studied. Various research works related with the hypolipidemic, antioxidant, cardio-protective, anti-diabetic, antibiotic, anti-malarial activity of various plant extracts are reported earlier. In this section, the pharmacological activity of *C. citratus*, crude extracts and isolated compounds are enumerated below.

2.2.4.1 Pharmacological activity of extract from *C. citratus*

Aqueous extract of *C. citratus* exhibits various activities such as antidepressant, antispasmodic, carminative activity (Flemming, 1965), fever reduction (Olaniyi *et al.*, 1975) and antihelmintic activity against ascariasis (Ross, 1984). Aqueous extract of *C. citratus* (commonly called as Abafado) displays the signs of various pharmacological activities like hypnotic, neuroleptic, anti-convulsant and anxiolytic effects along with decreasing body temperature and defecation scores (Carlini *et al.*, 1986). It also gives evidence of hypotensive activity (Carabajal *et al.*, 1989), analgesic activity against carrageenan or prostaglandin E2 induced hyperalgesic rats but not against hyperalgesia induced by dibutyryl cAMP (Berenice *et al.*, 1991; Lorenzetti *et al.*, 1991). Abafado demonstrate insecticidal, carminative, diuretic and pyretic activity (Arias *et al.*, 1992). Relaxation action on the mesenteric preparation of *C. citratus* extract has been described by Abeywardena *et al.*, (2002); Runnie *et al.*, (2004). Hypoglycaemic and hypolipidemic effect of *C. citratus* have been detailed by Adejuwon and Esther, (2007).

Methanolic extract of *C. citratus* causes the reduction of reactive oxygen species generation on the survival of the *Escherichia Coli* wild type AB 1157 strain (Suzana *et
al., 2001). The ferric reducing antioxidant potential, blood and intestinal cleansing activity of the same extract is observed by Runnie et al., (2004). Crude ethanolic extract inhibits enzymes like acetylcholinesterase, butyrylcholinesterase and lipoxygenase enzymes (Khattak et al., 2005).

Antimicrobial activity of crude ethanolic extract of C.citratus has been previously demonstrated by Onawunmi and Ogunlana, (1986); Mascolo et al., (1989); Syed et al., (1995); Tiziana Baratta et al., (1998): Cimanga et al., (2002). Antibacterial activity against Helicobacter pylori is reported earlier by Ohno et al, (2003). Synergistic effect of 13 antimicrobial drugs and 8 plant extracts has betrayed the better synergistic effect of C. citratus (Betoni et al., 2006). Similarly, C.citratus collected from India (Dikshit and Hussain, 1984; Prasad et al., 1986) have tried its antibacterial activity in In-vitro conditions against some human and animal pathogenic microorganisms.

Besides these bioactivities mentioned above, 80 % ethanol extract of C.citratus has given anti-mutagenic activity towards chemical induced mutations in Salmonella typhimurium strains TA 98 and TA 100 (Ross. 1984). Mutagenicity of Aflatoxin B1, 3-Amino-1,4-Dimethyl-5H-Pyrido (4,3-b)-indole, 3-Amino-1-methyl-5H-Pyrido(4,3-b)-indole, 2-amino-6-methylidipyrido-1,2-a:3’2’-d-imidazole, 2-aminodipyridol,2-a:3’2’-d-imidazole, 2-amino-3-methylimidazo (4,5-f) quinoline, N-methyl-N’-nitro-N-nitrosoguanidine and 2-(2-furyl)-3(5-nitro-2-furyl) acrylamide has inhibited by the extract of C. citratus in a dose dependent manner but no effect is found on the mutagenic activity of benzopyrene (Vinitketkumnuen et al., 1994). The extract also inhibits DNA adduct formation, protect abberant crypt foci formation, blocks faecal β- Glucuronidase activity and also antioxidant effect in rat colon (Ratchada et al., 1997; Puatanachokchai et
al., 2002). It also retards the growth of transplanted fibrosarcoma cells in mice in association with lung metastasis (Suaeyun et al., 1997).

2.2.4.2 Pharmacological activity of *C. citratus* oil

The essential oil extracted from fresh leaves of *C. citratus* has been found to have strong ascaricidal activity and anti-amoebic activity against *Entamoeba histolytica* (Ross, 1984). The oil has been used externally to treat skin eruptions, wound and bruises (Spring, 1989) and internally for alleviating colds and fever symptoms (Comerford, 1996). Oil is toxic against P388 mouse leukemia cells (Dubey et al., 1997). Further, the oil acts against filariasis causing microbes like *Culex quinquefasciatus* and dengue fever causing agent like *Aedes aegypti* (Tyagi et al., 1998). Oil also inhibits acetic acid induced writhing and controls the second phase of the response in the formalin test (Viana et al., 2000), mosquito repellency (Oyedele et al., 2002) and anti-malarial activity (Tchoumbougnang et al., 2005).

The oil is found to be the most active against human dermatophyte strains (Lima et al., 1993), *Trichophyton rubrum* and *Microsporum gypseum* (Kishore et al., 1993) and *Cryptococcus neoformans* (Violon and Chaumont, 1994). Moreover, the oil exhibits antibacterial activity against *Escherichia coli* (Ogunlana et al., 1987), *Escherichia coli, Staphylococcus aureus, Salmonella paratyphi, Shigella flexneri, Bacillus mycoides, Pseudomonas aeruginosa,* and *Bacillus subtilis* (Wannissorn et al., 1996), *Salmonella species,* *Escherichia coli* 0157, *Campylobacter jejuni* and *Clostridium perfringens* (Cimanga et al., 2002), *Listeria monocytogenes,* *Listeria innocua,* *Staphylococcus aureus* (Nguefack et al., 2004), *Klebsiella* and *Enterobacter species,* *proteus mirabilis,* *Morganella morganii* (Pereira et al., 2004) and *Salmonella typhimurium* TISTR 292,
Salmonella enteritis DMST 17368, Escherichia coli TISTR 292, Clostridium perfringens DMST 1519, Campylobacter jejuni DMST 15190 (Bhusita et al., 2005).

Wannissorn et al., (1996); Pattnaik et al., (1996) have found that the essential oil of C. citratus inhibit growth of Candida albicans. Some other organisms inhibited by C. citratus oil include Acinetobacter baumannii, Aeromonas veronii, Candida albicans, Enterococcus faecalis, Escherichia coli, Klebsiella pneumoniae, Psudomonas aeruginosa, Salmonella enterica, Serretia marcescens, Staphylococcus aureus and Proteus mirabilis (Hammer et al., 1999), Fusarium moniliforme, Aspergillus flavus and Aspergillus fumigatus (Nguefack et al., 2004) and Rhodotorula glutinis, Schizosaccharomyces pombe ATCC 60232, Saccharomyces cerevisiae ATCC 2365, Yarwwia lyolitica ATCC 16617 (Gianni et al., 2005). Thus, it is evident that the oil separated from C. citratus can be utilized as a good antimicrobial agent.

2.2.4.3 Pharmacological activity of phytoconstituents isolated from C. citratus

Monoterpene citral is the major constituent (75 %) of C. citratus. Citral isolated from C. citratus displayed expectorant, appetite stimulant, choleretic, carminative, spasmylytic, sedative action (Carlini et al., 1986), antiinflammatory, antiseptic, antirheumatism, deodorant, granulation-promoting effect (Carbajal et al., 1989), antiseptic, antimicrobial, anti-inflammatory, diuretic, central nervous system stimulating effects (Carbajal et al., 1989) and inhibition of the oxidation of β-carotene (Wang et al., 1992).

Citral possesses anticancer effect against prostate gland tumor in various strains of rats (Scolnik et al., 1994; Vinitketkumnuen et al., 1994). Citral is proved to be toxic
against P388 mouse leukemia cells (Dubey et al., 1997) and aided apoptosis in several hematopoietic cancer cell lines (Dudai et al., 2005).

Pattnaik et al., (1997) have established that citral acts against Cryptococcus and they have also suggested that citral is thought to be the component most likely to be antifungal than geranial and oil. Moreover, reports have suggested the anti-clastogenic effect of citral against irradiation in mice (Hosseinimehr et al., 2003; Paranagama et al., 2003; Abe et al., 2003).

Kauderer et al., (1991) have confirmed antimutagenic activity of myrcene, in mammalian tests in In-vitro assays. However, β-myrcene does not exhibit antinociceptive effect (Rao et al., 1990) and no protective effect on pentylenetetrazol induced seizures in mice (Da Silva et al., 1991). The peripheral analgesic effect of myrcene has confirmed the hyperalgesia induced by prostaglandin (Berenice et al., 1991; Lorenzetti et al., 1991). Myrcene has also reduced the toxic and mutagenic effect of cyclophosphamide in V79 cell line (Kauderer et al., 1991). Zheng et al., (1993) have explained that d-limonene and geraniol exhibit anti-carcinogenic effect. Limonene and Geraniol induce isoenzymes of CYP2B monooxygenase subfamily (De-Oliveira et al., 1997).

C.citratus and its phytoconstituents have been studied for various biological activities. However, there is scanty work in the cardioprotective effect of C.citratus. Hence the present study is carried out to identify the cardioprotective and hypolipidemic potential of C.citratus in ISO induced cardiotoxic rats.