Preliminary studies on cardio protective effect of *Hyoscyamus niger* linn in male albino rats

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ABSTRACT

In this present cardioprotective activity study the crude powder of the *Hyoscyamus niger* Linn is administered to the male albino rats at the dose of 100mg/100g b.wt. The markers of cardiac function test assess the normal functioning of heart. If there is a change in their normal values of these makers it indicates the dysfunction of heart. Biochemical parameters like TGL, Ck-MB and LPO were carried out to assess the cardioprotective effect of *Hyoscyamus niger* Linn crude powder in isoproterenol induced experimental animals. Thus, Plant based drugs have been used worldwide in traditional medicines especially Siddha medicines; suggest various herbal and herbomineral drugs for the treatment of various kinds of cardiovascular diseases. CVD will be the paramount cause of death in India which includes high blood pressure, chronic heart diseases, stroke and congestive heart failure. In the last few decades there has been an exponential growth in the field of Siddha herbal medicines or Tamil medicine.

Key words: *Hyoscyamus niger* Linn, Isoproterenol, TGL, Ck-MB, antioxidant

INTRODUCTION

Nature is the lifeline of our health as it provides all necessary things for survival. Medicinal plants are nature’s gift to human beings to make disease free healthy life and play a vital role to preserve our health [1]. Nowadays research has been focused on medicinal plants and food products derived from medicinal plants that have been found to have certain preventive measures in the treatment of cardiovascular disease (CVD) [2].

Although modern drugs are effective in preventing cardiovascular disorders, their use is often limited because of their side effects. Nowadays, it is being realized that herbs can protect the heart from heart diseases by their cardio protective action by providing an integrated structure of nutritional substances mainly phytochemicals which help in restoring and maintaining balanced body systems [3,4]. Cardiovascular disease (CVD) has become a universal cause of morbidity and a leading contributor to mortality in both developed and developing country. By 2020, CVD will be the paramount causes of death in India which include high blood pressure, coronary heart disease, stroke, congestive heart failure [5].

Myocardial infarction (MI) is defined as irreversible injury or subsequent necrosis of myocardial cells due to interruption of blood supply to a part of the heart [6,7]. Moreover, with advanced life style in developing countries, like India, particularly in metropolitan cities, MI is making increasingly important contribution to mortality statistics of such countries [8]. MI is a complex phenomenon affecting the mechanical, electrical and structural and biochemical properties of the cardiac system [9]. The use of complementary and alternative medicines is burgeoning globally for MI, especially in developed countries including US [10]. Although many studies identified the increasing prevalence of herbal use throughout the world, only a few reported on how patients perceived the efficacy of this healthcare modality in specific diseases including MI [11].
Herbal Medicine
Several medicinal plants have been found to possess antioxidant properties and have beneficial effects in pathological conditions like cancer, liver diseases, cataract and myocardial ischemia [4,12]. The use of herbal medicines has been steadily increasing over the past decade. A considerable number of these plants/plant based products have been widely used [13]. Therefore, interest in the examination of plants as potential sources of new drugs is increasing. The prophylactic and therapeutic effect of many plant foods and extracts in reducing cardiovascular disease has been reviewed as they are inexpensive, efficacious and safe [14]. As few systematic scientific studies are currently available, these medicinal plants need to be investigated scientifically. The study is an effort in the same direction thus the present investigation was undertaken to evaluate the cardioprotective effects of the crude powder the plant *Hyoscyamus niger* Linn in isoproterenol induced cardiovascular disease in albino male rats.

EXPERIMENTAL SECTION

Plant Material
Fresh plant sample *Hyoscyamus niger* Linn were collected from various parts of Thanjavur district. The seed were shade dried, powdered. Crude powder of *Hyoscyamus niger* Linn is given to the experimental rats at the dose of 100mg/100g body weight.

Henbane (*Hyoscyamus niger* L.), is a biennial herb indigenous to Europe, Western and Northern Asia, and Northern Africa. It has been introduced to Eastern Asia, North America, and Australia, and cultivated in several other countries [15, 16]. It is well-known for its traditional use as a hallucinogen. This species grows to a height of 1–2.5 feet (0.3–0.7 m) and has grayish-green leaves, white (or faintly yellow) flowers with purplish veins, and dark gray seeds. The plant has a distinctly unpleasant smell, hence its folk names were Stinking Nightshade and Stinking Roger [17].

Experimental Design
Male albino rats of 8 – 10 weeks of age weighing between 150-180g cardio-toxicity were used for the study. The animals were housed in polypropylene cages. Animals were divided into three groups of three animals. The animals were acclimatized for a week under laboratory conditions. All experiments were performed according to the norms of the local ethical committee.

Myocardial infarction was induced in rats by administration of isoproterenol at a dose of 70mg/ 100kg i.p of body weight intraperatonyally after drug standardization.

Experimental animals were distributed randomly, in three groups, containing three animals each.
- The animals in group I served as normal and received rat feed and normal water *ad libitum*.
- The group II rats served as test and were induced with isoproterenol at a dose of 70 mg/ 100g body weight intraperatonially.
- The animals in group III provided treated with normal rat feed and normal water *ad libitum* along with isoproterenol and crude powder of *Hyoscyamus niger* at the dose of 100mg/100g body weight the drug followed by it.

At the end of treatment, animals were fasted overnight, anaesthetized with ether, blood was drawn and the serum was separated for biochemical analysis like CK-Mb, TGL and LPO level were estimated and heart tissue also collected for the histopathological studies [18]. Mean values standard were calculated and percentage of inhibition for all the values carried out [19].

RESULTS AND DISCUSSION
Isoproterenol (ISO) [1-(3,4-dihydroxyphenyl)-2- isopropyl aminoethanol hydrochloride], a synthetic catecholamine and β-adrenergic agonist that causes severe stress in myocardium and infarct-like necrosis of the heart muscles [20]. ISO induced myocardial injury involves membrane permeability alterations, which brings about the loss of functions and integrity of myocardial membranes. ISO induced myocardial necrosis is a well known standard model to study the beneficial effect of many drugs on cardiac dysfunction [21].

The oral administration of crude powder of Siddha medicinal plant *Hyoscyamus niger* water suspension against isoproterenol for 30 days were found to protect the cardiac damage induced by lipid per oxidation and activation of antioxidant enzymes. The herbal drug administration was able to protect the cardiac necrosis as evidenced by the inhibitory activity of CK-Mb and TGL (Table: 1). The study also shows the significant efficacy of herbs in the treatment of cardiotoxicity was also evidenced by decrease in CK-Mb and TGL level in serum. Elevation of CK is
an indication of damage to muscle. It is therefore indicative of injury, rhabdomyolysis, myocardial infarction, myositis and myocarditis.

The observed increase in the body weight in isoprotenerol induced rats could be due to the accumulation of water content in the Oedematous intramuscular area in addition with necrosis of cardiac muscle fibres. Decreased activities of these cardiac marker enzymes in the cardiac tissue could be due to the leakage from damaged cardiac tissue into the circulation as a result of necrosis induced by ISO [22].

During myocardial injury cardiac membranes becomes leaky and results in higher serum level of creatine kinase (CK) enzyme. This enzyme enters into the blood stream and thus increasing their concentration in the serum, and serve as the diagnostic markers of myocardial tissue damage [23-25]. Isoprotenerol is well known cardiotoxic agent due to its ability to damage cell membrane. A significant elevated level of serum CK after isoprotenerol treatment was reported in rat earlier [26]. Lipid peroxidation and endogenous antioxidants were also measured to confirm the myocardial oxidative stress after administration of isoprotenerol. Isoprotenerol produce quinones which react with oxygen to generate superoxide anions $O_2^-$ and $H_2O_2$, which have damaging effects in cells [27]. Increased myocardial lipid peroxidation after isoprotenerol appears to be the initial event of oxidative damage.

| TABLE 1: Cardioprotective effect of *Hyoscyamus niger* on serum CK-MB, TGL and LPO |
|-------------------------------|------------------|-----------------|-----------------|
| **Groups**                   | **Dose**         | **Parameters**  |                  |
|                              |                  | **CK-MB** U/l   | **TGL** mg/dl   | **LPO** nM MDA/ ml |
| Normal                       | Isosaline        | 1.65±0.148      | 57.1±4.33       | 2.25±0.090         |
| Isoprotenerol induced        | 40mg/kg b.wt     | 13.2±0.792      | 133.3±9.31      | 2.71±0.130         |
| HN Treated                   | 100mg/kg b.wt    | 3.30±0.181      | 54.16±1.70      | 1.70±0.102         |

*Each values is the Mean ± SEM of three animals statistically significant from control*.

**Histopathology**

In histopathological examination of Heart tissues, normal group showed normal cardiac architecture and arrangement of myofibril, absence of interfibrillar necrosis, regular and normal multinuclear myofibrils arrangement, vacuolization and macrovesicular changes. In Isoprotenerol treated animals exhibited intense interfibrillar necrosis, vacuolization, macrovesicular changes and damage and irregular arrangement and morphological change of myofibrils associated with increased interfibrillar distance.

Photographs of heart sections were prepared (6 µm thickness) from different treatment groups stained with Haemotoxylin and Eosin, 40x.

**Plate A**: Normal group showed normal cardiac architecture and arrangement of myofibril, absence of interfibrillar necrosis, regular and normal multinuclear myofibrils arrangement, vacuolization and macrovesicular changes.

**Plate B**: control group animals(isoprotenerol induced rats) exhibited intense interfibrillar necrosis, vacuolization, macrovesicular changes and damage and irregular arrangement and morphological change of myofibrils associated with increased interfibrillar distance.

**Plate C**: *Hyoscyamus niger* treated group exhibited significant cardiac remodeling activity against isoprotenerol induced rat heart by normal cardiac architecture, arrangement of myofibrils, and absence of interfibrillar necrosis.
The crude powder of the plant *Hyoscyamus niger* has cardioprotective activity against the isoproterenol induced cardiotoxicity in male albino rats. Further studies, are needed to identify the chemical constituents of the plant *Hyoscyamus niger* Linn that may be responsible for the cardioprotective activity.

**REFERENCES**