Pharmacological Screening of Anti-Asthmatic Activity of Ethanolic Extract of Calotropis Gigantea Leaves

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ABSTRACT
Calotropis gigantea Linn., (Asclepiadaceae) a widely growing plant has been reported to possess number of medicinal properties. It has been reported as a traditional folk medicine for a variety of ailments. The calotropis leaves also possess anti-diabetic activity. The present study deals with the effect of ethanolic extract of leaves of calotropis gigantea by using in vivo models. The study shows that the extract is effective against histamine induced bronchial muscle contractions in guinea pigs. Animal study involves the use of ethanolic extract of leaves that shows action against histamine induced broncho-constriction by using histamine chamber. These studies showed significant protection at lower doses while further increase in dose showed increased activity. The results of these studies indicated usefulness of ethanolic extract of Calotropis gigantea in asthma.

Keywords: Traditional plant, Anti-asthmatic, broncho constriction, Calotropis gigantea Leaves, ethanolic extract, Histamine chamber, chlorpheneramine maleate.

INTRODUCTION
Asthma is very commonly occurring condition that is most difficult to control in chronic stage. In the united state alone asthma affects almost 17 million people and this is a 75% increase in the last 20 years. This means that about 1 out of every 20 adults and close to 1 out of 13 children today have asthma. An alarming fact is that since 1980, asthma in children under age 5 years has risen remarkably. In school age children asthma has risen by 75%, India has alone an estimated 15-20 million asthmatics mortality data from developed countries, show that the rate varies from 0.1-0.8 per 10,000 person aged 5-34. For managing asthma attack symptomatic relief is foremost requirement. In India, in various traditional systems like Ayurveda, Unani and Siddha numerous herbs were mentioned for therapeutic use in asthma.

Calotropis gigantea (crown flower) is a species of Calotropis, commonly known as madar in Hindi, belonging to the family Asclepiadaceae, is a milky shrub up to 1-3 cm in height found throughout India. It is one of the important plants mentioned in Ayurveda and Unani system of medicine for asthma. According to Ayurveda dried whole plant is good tonic, expectorant, depurative, and anthelmintic. The leaves are useful in treatment of paralysis, arthralgia, swelling and intermittent fever. Leaves are bitter, astringent, stomachic, anthelmintic and tonic. Calotropis gigantea has been reported to contain proteases, 3′-methyl butanoates of amyrin, flavonol glycosides, calotropis stigmasterol and sitosterol, cardenolides, pregnanone etc.

MATERIALS AND METHODS
PLANT MATERIALS:
The plant of calotropis gigantea was collected from the road side location of Uppal (Telangana region) and was authenticated by Rana Kausar, Head of Department of Botany. Plant material was preserved in Department of Pharmacognosy GJCP, Uppal. The leaves from plant were seperated, dried and coarsely powdered.

PREPARATION OF PLANT EXTRACT:
The collected calotropis gigantea leaves were air dried under shade at room temperature and milled to coarse powder. The obtained dried powder was subjected to successive soxhlet extraction with ethanol as solvent. The powdered leaf material was packed in a thimble made of wattman’s filter paper. After complete extraction of powder the obtained extract is filtered through wattman’s filter paper, then the extract thus obtained was concentrated to
dryness in a flash evaporator under reduced pressure and controlled temperature. The obtained residues were yellowish brown colour to brown colour in form of thick and sticky paste. The extract was stored in refrigerator at 2-8°C and reconstituted uniformly by dissolving in suitable solvent before administration to animals orally using an intra gastric feeding tube.

EXPERIMENTAL ANIMALS:
Guinea pigs of either sex (350-450) were selected for present studies. 6 animals were taken in each group and maintained under standard laboratory conditions. They were allowed free access to standard dry pellet diet and water ad libitum during the experiment at standard conditions of temperature 22±1°C. All experimental procedures were followed in strict accordance with the guidelines prescribed by the committee for the purpose of control and supervision on experiment on animals (CPCSEA).

SCREENING OF ANTIASTHMATIC ACTIVITY IN VIVO STUDIES ON HISTAMINE INDUCED BRONCHOSPASM IN GUINEA PIGS:
Guinea pigs of either sex (350-450) were selected and randomly divided into four groups each containing six animals. The animals were kept on fasting overnight before treatment. The ethanolic extract and the standard drug i.e., chlorpheneramine maleate were administered orally. The single dose treatment was given one and half an hour before the study. Later the animals were exposed to an aerosol of 0.25% of histamine and time for preconvulsion states were observed for each animal and the experiment is repeated and observed for four groups of animals as described by (Sheth et al.,1972).

RESULTS AND DISCUSSIONS
The study dealt with screening of anti asthmatic activity of ethanolic extract of leaves of calotropis gigantea. Bronchial asthma is a chronic inflammatory disease characterised by both broncho constrictions and air way inflammation which leads to bronchial hyper-responsiveness to various stimuli, in which all types of cells play a role, more important being mast cells, eosinophils, and t-lymphocytes. Different agonist like ACh, histamine, 5HT and Bradykinin are responsible for contractile responses. Histamine is one of the major inflammatory mediators in the immediate phase of asthma causing airway hyper responsiveness and bronchial airway inflammation. This study regarding involvement of H₁ and H₂ receptors has been done in experimental asthma in guinea pigs using respiratory smooth muscle and it was confirmed that there is prominent involvement of H₁ receptors as compared to H₂ receptors in asthma.

ACUTE TOXICITY STUDY
From the acute toxicity studies lethal dose was found to be1000mg/kg and the dose selected is the 1/10th of the lethal dose i.e., 100mg/kg.
<table>
<thead>
<tr>
<th>Group (n=4)</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>Water</td>
</tr>
<tr>
<td>Standard</td>
<td>Chlorpheneramine maleate (2mg/kg)</td>
</tr>
<tr>
<td>Test leaves 1</td>
<td>Ethanolic extract (50mg/kg)</td>
</tr>
<tr>
<td>Test leaves 2</td>
<td>Ethanolic extract (100mg/kg)</td>
</tr>
</tbody>
</table>

Table No.1. Animals and Treatment for Anti-Asthmatic Activity of Leaves

<table>
<thead>
<tr>
<th>Control</th>
<th>Standard</th>
<th>Test-1</th>
<th>Test-2</th>
</tr>
</thead>
<tbody>
<tr>
<td>8.5±0.3</td>
<td>10.3±1.3**</td>
<td>9±0.9*</td>
<td>9.6±0.9**</td>
</tr>
</tbody>
</table>

Table No.2. Anti-Asthmatic Activity of Leaves
Comparison: standard v/s Test-1 and Test-2.
Statistical significance test for comparisons were done by One-way ANOVA.
Values are expressed in Mean±SEM; *p<0.01. **p<0.05 are extremely significant.

Figure No.2: Graph representing the Anti-Asthmatic Activity Results

DISCUSSION
The present study dealt with screening of anti-asthmatic activity of ethanol extract of leaves of *Calotropis gigantea*. Bronchial asthma is a chronic inflammatory disease, characterised by bronchoconstriction and airway inflammation which leads to bronchial hyper responsiveness to various stimuli, in which cell types play a role, more important being mast cells, eosinophils and T-lymphocytes. Different agonists like acetylcholine, histamine, 5-HT and bradykinin are responsible for contractile responses. Histamine is one of the major inflammatory mediators in the immediate phase of asthma, causing airway hyper responsiveness and bronchial airway inflammation. The study regarding the involvement of H1 and H2 receptors has been done in experimental asthma in guinea pig using respiratory smooth muscle and it was confirmed that there is prominent involvement of H1 receptors as compared to H2 receptors especially in asthma. The anti asthmatic activity is brought by inhibition of inflammatory mediators.

CONCLUSION
Investigation of anti-asthmatic activity of ethanolic extract of leaves of *Calotropis gigantea* was carried out. The broncho relaxant study comparable with that of standard chlorpheneramine maleate and statistical significance in post treated exposition time and mean exposition time also showed 100mg/kg as affected further increase in dose show decrease in activity. All the test extracts were screened for anti-asthmatic activity and they showed significant inhibition of inflammatory mediators.

REFERENCES
23. Rang and Dales pharmacology, 7th Ed.338 - 343.