ANTIDIABETIC ACTIVITY OF MIMOSUPS ELENGI LEAVES ON ALLOXAN INDUCED DIABETIC RATS
MAMATHA M K¹, TINCY THOMAS², YOGESH H S¹, PRAMOD H J²
¹ R R College of Pharmacy, Chickbanavara, Bangalore- 560090, Karnataka, India.
²K.L.E.S’s College Of Pharmacy, Belgaum, Karnataka, India.
Corresponding author’s E-mail: mamthamkgowda@gmail.com

ABSTRACT
The polar and nonpolar solvent extracts of Mimosups elengi leaves were screened at dose of 200mg/kg body weight orally for antidiabetic activity using alloxan induced hypoglycemic rats on acute and prolonged treatment. Alcoholic and Aqueous extracts of leaves of Mimosups elengi, showed significant (p<0.01) antidiabetic results with both acute and prolonged treatment studies. The overall results were comparable with the standard drug glibenclamide.

Keywords: Mimosups elengi, Antidiabetic, Alloxan, glibenclamide.

INTRODUCTION:
In 2000, according to the World Health Organization, at least 171 million people worldwide suffer from diabetes. Its incidence is increasing rapidly, and it is estimated that by the year 2030, this number will double. Diabetes mellitus occurs throughout the world, but is more common (especially type 2) in the more developed countries. Diabetes is in the top 10, and perhaps the top 5, of the most significant diseases in the developed world, and is gaining in significance there and elsewhere.

Diabetes mellitus is a condition of hyperglycemic glycosuria due to the production, or utilization of insulin, or to a hyperactivity of those endocrine glands(pituitary, adrenals, thyroid) whose secretions normally balance, or counteract, the influence of insulin. There is also an associated disorder of fat metabolism.1 The characteristic symptoms are excessive urine production (polyuria), excessive thirst and increased fluid intake (polydipsia), and blurred vision. These symptoms are likely absent if the blood sugar is only mildly elevated. [1&2]

Insulin and oral hypoglycemic agents are the ways to treat diabetes but many drugs also have prominent side effects. Moreover, the treatment is costly and many time far away from the capacity of poor people resulting in the development of long term macrovascular and microvascular complications.3 Recently , the use of plant extracts in the treatment of diabetes mellitus is undergoing extensive investigation and many traditional plants are being screening throughout the world.[4,5] Mimusops elengi (Linn) a member of family sapotaceae it is commonly known as Bullet-wood tree in English and Bakul, Maulsiri in Hindi.6

This evergreen tree is 15mtrs, high and provides a thick shade. It is found in the Western Peninsula, southwards from the Khandala Ghat in the west and the N. Circars on the eastsides, Andaman, Burma, Ceylon, Martaban, Malay Peninsula and Archipelago. The leaves are given in snakebite. Powder of the leaves mixed in milk is administered twice a day for diabetes.7

MATERIALS AND METHODS
Collection of the plant material
The leaves of *Mimusops elengi* Linn were collected in the month of April-May from Dandeli, Uttara Kannada District. The leaves were authenticated by Dr. Harsha Hegde, Research Officer, Regional Medical Research centre, Indian Council of Medicinal Research, Belgaum.

**Preparation of plant extracts**
The air-dried leaves of *Mimusops elengi* Linn. Belonging to Family Sapotaceae were reduced to fine powder (40 size mesh) and around 100 gm of powder was subjected to successive hot continuous extraction (soxhlet) with petroleum ether, chloroform and ethanol. Another batch of powdered drug was macerated with chloroform-water I.P. (Each time before extracting with next solvent the powdered material was dried at room temperature). After the effective extraction, solvent were concentrated using rotary flash evaporator and water was removed by freeze drying.

**Experimental animals:**
Wistar albino mice of either sex weighing between 25 and 30 gm were selected for acute toxicity studies and Wistar albino rats of either sex weighing between 150 and 200 gm were selected for the antidiabetic studies. The animals were acclimatized to standard laboratory conditions of temperature (22±3°C) and maintained on 12:12 hr light: dark cycle. They were provided with regular rat chow (Lipton India Ltd., Mumbai) and distilled water *ad libitum*. The animal care and experimental protocols were in accordance with CPCSEA / IAEC.

**Acute Toxicity Study**
The acute oral toxicity study was carried out as per the guidelines set by organization for economic co-operation and development (OECD) revised draft guidelines 423 B (“Up and Down” method) received from committee for the purpose of control and supervision of experiments on animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India.

**Preparation and administration of doses**
Test solutions (2000 mg/kg) were prepared in distilled water using 2% tween 80 as suspending agent. The test solutions were administered in a single dose using a gastric intubation tube after fasting for 3 to 4 hr.

**Number of animals and dose levels**
In each steps three animals were used. Since there was no information on the substance to be tested (i.e. extracts), starting dose was selected to be 300 mg/kg b.w.

**Experimental design**
The acclimatized animals were kept fasting for 24 hrs with water *ad libitum* and injected intraperitoneally at a dose of 60mg/kg b.w. of alloxan monohydrate freshly prepared in normal saline solution. Before starting the experiment, animals were separated according to their body weight. After one hour of alloxan administration, animals were given feed *ad libitum* and 1 ml of (100 mg/ml) glucose i.p. to combat ensuring severe hypoglycaemia. After 72 hrs of the alloxan injection, the animals were tested for the evidence of diabetes by estimating their blood glucose level by using glucometer. The blood glucose level more than 150 mg/100 ml of blood was criteria. Experimental animals are divided into 7 groups of 6 animals each:

- **Group I**: Normal rats
- **Group II**: Diabetic control group
- **Group III**: Animals were receive a dose of 200mg/kg body weight of chloroform extract of *Mimosups elengi* leaves orally.
- **Group IV**: Animals were receive a dose of 200mg/kg body weight of ethanol extract of *Mimosups elengi* leaves orally.
- **Group V**: Animals were receive a dose of 200mg/kg body weight of aqueous extract of *Mimosups elengi* leaves orally.
- **Group VI**: Animals were receive a dose of 200mg/kg body weight of petroleum ether extract of *Mimosups elengi* leaves orally.
- **Group VII**: Animals were receive a dose of 10mg/kg body weight of Glibenclamide used as a reference standard.
The blood samples were obtained through the tail vein puncturing with Lancet. Blood was withdrawn at interval of initial (0 hr), 1, 3, 5 and 7th hrs of administration of single dose (for acute study) and at the end of 7th day (prolonged treatment). The animals were segregated into seven groups of six rats each, taking into consideration of diabetic blood sugar level.

**Estimation of Glucose**

The blood samples were obtained through the tail vein puncturing with lancet. A drop of blood so obtained was placed on the enzyme treated surface of the haemoglobinostrip, which was placed onto a glucometer (Medisense Glucometer, Abott Pharmaceuticals). The glucometer was kept on, then after 2 minutes glucomonitor reading was recorded.

The animals were continued with this treatment for 7 days and again blood glucose level was measured after prolonged treatment for 7 days. The statistical analysis was done by one way ANOVA followed by Dunnet’s ‘t’ test.

**RESULT AND DISCUSSION**

Chloroform, alcohol and aqueous extracts of *Mimusops elengi* Linn. Leaves were subjected for assessment of Acute oral toxicity study and Antidiabetic activity in alloxan induced albino rats in both acute and prolonged treatment.

Acute toxicity study was carried out according to OECD guidelines. Lethal dose of mice was calculated and was found to be as shown in Table no.1

Antidiabetic activity of Petroleum ether (40-60°C), Chloroform, Ethanol and Aqueous extracts were assessed for Antidiabetic activity. The Petroleum ether(40-60°C), Chloroform , alcoholic and aqueous extract were given orally at a dose 200mg/kg b.w with the help of a gastric tube to alloxan induced diabetic rats. The blood glucose level was analysed initially (0hr), 1th hr, 3rd hr, 5th hr and 7th hr. after single dose and at the end of the 7th day after prolonged treatment. Control animals received equal volume of the normal saline. Glibenclamide ( 10mg/kg b.w) served as the reference standard. Blood glucose level was estimated using glucometer(Medisense Glucometer, Abott Pharmaceuticals). Results obtained from alloxan induced diabetes indicated that chloroform,Alcoholic and Aqueous extract showed more significant (p<0.01) antidiabetic activity (192.5±0.67, 175.8±0.65 and 195.5±0.67 respectively) in acute as well as prolonged treatment (174.5±0.76, 162.3±0.66 and 180.5±0.76) compared to diabetic control (223.2±0.307, 217.3±1.5 respectively). The results were comparable with reference standard Glibenclamide (183.8±0.47, 163.±0.85). Petroleum ether extract did not show significant antidiabetic activity on prolonged treatment but showed significant (p<0.05) activity at 7th hour in acute study (204.5±0.42) compared to diabetic control.

The single dose of alcoholic extract has more significantly reduced the blood glucose level at 3rd hour (202.8±0.60 at 0 hr to 194.3±0.66 at 3rd hr) and significant hypoglycaemia was maintained for another four hours. Glibenclamide (10 mg/kg b.w.) has also significantly reduced blood glucose level at 3rd hr. (191.8±0.79 at 0 hr to 183.8±0.47 at 3rd hr) and significant hypoglycaemia maintained for another four hours. On prolonged treatment, the effect of alcoholic extract was (162.3±0.66) and nearly equal to that of the reference drug Glibenclamide (163.8±0.85). These findings clearly established that alcoholic exhibited better antidiabetic activity than other extracts.

The results of various extracts, control, diabetic control and standard are shown in table no.2.& 3, and graph no.1 & 2.

**Table No. 1: LD<sub>50</sub> doses of extracts**

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Name of Extract</th>
<th>LD&lt;sub&gt;50&lt;/sub&gt; Cut-Off mg/kg, b.w</th>
<th>Vehicle</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Petroleum ether extract</td>
<td>2000</td>
<td>Tween 80</td>
</tr>
<tr>
<td>2</td>
<td>Chloroform extract</td>
<td>2000</td>
<td>Tween 80</td>
</tr>
<tr>
<td>3</td>
<td>Ethanolic extract</td>
<td>2000</td>
<td>Tween 80</td>
</tr>
<tr>
<td>4</td>
<td>Aqueous extract</td>
<td>2000</td>
<td>Tween 80</td>
</tr>
</tbody>
</table>

1/10<sup>th</sup> of this lethal dose was taken as therapeutic dose for subsequent antidiabetic activity.
Table No.2: EFFECT OF MIMUSOPS ELENGI LEAVES ON BLOOD GLUCOSE LEVEL OF ALLOXAN INDUCED DIABETIC ALBINO RATS AFTER SINGLE DOSE

<table>
<thead>
<tr>
<th>Group (n)</th>
<th>Dose</th>
<th>Blood Glucose Level mg/100ml (Mean±SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Initial</td>
<td>1 hour</td>
</tr>
<tr>
<td>Normal Control (6)</td>
<td>2 ml saline</td>
<td>96.5±0.7</td>
</tr>
<tr>
<td>Diabetic Control (6)</td>
<td>2 ml Saline</td>
<td>203.5±0.76</td>
</tr>
<tr>
<td>Petroleum ether extract (6)</td>
<td>200mg/kg b. w.</td>
<td>211.7±0.88</td>
</tr>
<tr>
<td>Chloroform Extract (6)</td>
<td>200mg/kg b. w.</td>
<td>203 ±0.96</td>
</tr>
<tr>
<td>Alcohol extract (6)</td>
<td>200mg/kg b. w.</td>
<td>202.8±0.60</td>
</tr>
<tr>
<td>Water Extract (6)</td>
<td>200mg/kg b.w.</td>
<td>205.5±0.76</td>
</tr>
<tr>
<td>Glibenclamide (6)</td>
<td>10mg/kg b.w.</td>
<td>196.5±0.76</td>
</tr>
</tbody>
</table>

n=6,  *p<0.05 - Significant, **p<0.01 – More significant Vs. Diabetic Control  
SEM : Standard Error Mean, n = Number of animals.

Table No.03: EFFECT OF MIMUSOPS ELENGI LEAVES ON BLOOD GLUCOSE LEVEL OF ALLOXAN INDUCED DIABETIC ALBINO RATS AFTER PROLONGED TREATMENT

<table>
<thead>
<tr>
<th>Group (n)</th>
<th>Dose</th>
<th>Blood Glucose Level mg/100ml (Mean±SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Initial</td>
<td>7th day</td>
</tr>
<tr>
<td>Normal Control (6)</td>
<td>2 ml saline</td>
<td>96.5 ± 0.70</td>
</tr>
<tr>
<td>Diabetic Control (6)</td>
<td>2 ml Saline</td>
<td>203.5±0.76</td>
</tr>
<tr>
<td>Petroleum ether extract (6)</td>
<td>200mg/kg b. w.</td>
<td>211.7±0.88</td>
</tr>
<tr>
<td>Chloroform extract (6)</td>
<td>200mg/kg b. w.</td>
<td>203 ±0.96</td>
</tr>
<tr>
<td>Alcohol extract (6)</td>
<td>200mg/kg b. w.</td>
<td>202.8±0.60</td>
</tr>
<tr>
<td>Aqueous extract (6)</td>
<td>200mg/kg b.w.</td>
<td>205.5±0.76</td>
</tr>
<tr>
<td>Glibenclamide (6)</td>
<td>10mg/kg b.w.</td>
<td>196.5±0.76</td>
</tr>
</tbody>
</table>

n=6,  *p<0.05 – Significant, **p<0.01 – More significant Vs. Diabetic Control  
SEM : Standard Error Mean, n = Number of animals.

GRAPH NO.1: BLOOD GLUCOSE LEVEL OF ALLOXAN INDUCED DIABETIC ALBINO RATS AFTER SINGLE USE
GRAPH NO.2: BLOOD GLUCOSE LEVEL OF ALLOXAN INDUCED DIABETIC ALBINO RATS AFTER PROLONGED TREATMENT

REFERENCES
1) http://en.org/wiki/Diabetes_mellitus