INTRODUCTION

Diabetes mellitus is a major endocrine disorder and growing health problem in most countries. Decreased physical activity, increasing obesity, stress and changes in food consumption have been implicated in this increasing prevalence in the past two decades. Diabetes mellitus is categorized as a metabolic disease characterized by common features of chronic hyperglycemia with disturbance of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action or both. The cause of type 2 diabetes is a combination of resistance to insulin action and an inadequate compensatory insulin-secretory response. It was estimated that 2.8% of world population was diabetic in 2000 and this figure would climb to be as high as 4.4% of the world’s population by 2030 (most of which will be type 2 diabetes mellitus). Hyperlipidemia is a secondary metabolic dysregulation associated with diabetes. Besides the cause effect relationship with diabetes, elevated serum level of triglycerides, cholesterol and LDL are major risk factors for the premature development of cardiovascular disease like arteriosclerosis, hypertension, coronary heart disease etc. Increased plasma lipid levels mainly total cholesterol, triglycerides, LDL and VLDL along with decrease in HDL are known to cause hyperlipidemia which is the reason for initiation and progression of atherosclerosis impasse. Elevated lipid levels result from increased absorption through the gut or enhanced degradation. Though different types of drugs are available for the treatment of diabetes and hyperlipidemic, there is an increased demand by patients to use natural products for regulate these problems. Since time immemorial, patients with non-insulin dependent diabetes have been treated orally in folk medicine, with a variety of plant extracts.

A number of plants are mentioned in ancient Indian literature for the treatment of hyperglycemic and hyperlipidemic conditions. One such drug is Sesbania grandiflora of the family (Leguminosae: Papilionoidea), being used by some local tribal people, were selected for the present study. Sesbania grandiflora also called agati, an open branching tree up to 15 m tall and 30 cm in diameter. Sesbania grandiflora native range through Tropical Asia including, India, Indonesia, Malaysia, Myanmar and Philippines with possibly Indonesia as the center of the diversity and Southeast Asia is non-contiguous. The chemical constituents found are galactomannans, linoleic acid, β-Sitosterol and Carbohydrates. Traditionally the bark is used as astringent and utilized for the treatment of smallpox, ulcers in the mouth and alimentary canal, in bitter, in juvenile, infantile disorders of stomach, scabies, the juice of the leaves are utilized for the treatment of epileptic fits and clinical research supports the anticonvulsive activity of Agati leaves, astringent, bitter, termogenic, styptic, alexeteric, anti-helmintic, vulnary, demulcent, constipating, expectorants and antipyretic, bronchitis, cough, vomiting, wounds, ulcers, diarrhoea, dysentery, internal and external haemorrhages, dental caries, oral ulcers, proctoptosis, stomatitis and intermittent fevers.

MATERIALS AND METHODS

Plant material

Sesbania grandiflora plants were collected from local areas of Berhampur, Odisha. The taxonomical identification of the plant specimen was done by Dr. P. Lakshminarasimhan, scientist, Central National Herbarium, Botanical Survey of India, Howrah (Authenticated no. CNH/23/2011/Tech.II/483). Voucher specimen was preserved in the Department of Pharmacognosy of the Royal College of Pharmacy and Health Sciences, Berhampur for further verification. The plant materials were air dried under shade, coarsely powdered and kept in airtight container until further use.
Animals
Prior conducting the experiment, the ethical clearance for the study was granted by Institutional animal ethics committee (IAEC) of Royal College of Pharmacy and Health Sciences, Berhampur, (bearing registration number 1018/C/06/CPSEIA and date of registration 19th Dec 2006) in resolution number 04/ 11/IAEC held on 18/02/2011.
As per the OECD draft guidelines 423 received from CPCSEA, young female albino mice were used for acute toxicity study. Whereas other in vivo methods were carried out by using Sprague-Dawley (SD) rats of both sexes. All the animals for the in vivo studies, with no prior drug treatment, were procured from the animal house of R.C.P.H.S., Berhampur and housed in polypropylene cages with clean sterilized husk bedding (six mice or three rats/ cage). Bedding was changed every alternate day to maintain proper hygienic condition. Animals were maintained under controlled room temperature (22 ± 2°C) and humidity (55 ± 5°C) with a 12:12 hour light: dark cycle. The animals were fed with standard laboratory food diet made in-house recommended by National institute of nutrition (NIN), Hyderabad and pure drinking water *ad libitum*. The animals were acclimatized to laboratory hygienic conditions in the departmental laboratory for 7 days before commencing the experiment.

Chemicals
Glibenclamide and Simvastatin were obtained from Dr. Reddy’s Laboratories, Hyderabad. Experimental hyperlipidemic agent, Triton -WR 1339, was purchased from Sigma, USA. Blood glucose test-strips of Ascensia Entrust of Bayer Health Care and Diagnostic kits of Crest Biosystems, a division of Coral clinical systems, India were purchased. All other chemicals used for study were of analytical grade.

Preparation of extracts
Dried and powdered plant material (400 gm) was extracted by successive extraction process using soxhlet apparatus. Solvents were chosen depending upon their increase in polarity like Petroleum Ether (60-80° C), Chloroform, Methanol and Water. The extraction was carried out for 72 hours for each solvent. All the extracts were dried using rotary vacuum evaporator and freeze dryer. Their percentage yields were determined and stored in dessicator until further use.

Phytochemical screening
Different extracts obtained from the above extraction process were analyzed for presence of various phytoconstituents such as alkaloids, glycosides, flavones, tannins, terpenes, saponins, fats and sugars by the method of preliminary phytochemical study (color reactions)5, 10, 11.

Acute toxicity studies
The acute oral toxicity studies of extracts were carried out as per the OECD guidelines, draft guidelines 423 adopted on 17th December 2001 received from CPCSEA, Ministry of social justice and empowerment, Govt. of India. Administration of the stepwise doses of methanolic extracts of *Sesbania grandiflora* from 100 mg/kg up to the dose 2000 mg/kg to young female albino mice and observed the signs of toxicity up to 72 hr in the tested animals7. The female albino mice 25-30 gm were divided into different groups of six animals each. The control group received 10 ml/kg body weight of distilled water orally. The other groups received the extracts of *Sesbania grandiflora* at dose levels of 100, 500, 1000, 1500, 2000 mg/kg body weight through oral route. After administration of dose the animals were observed continuously for the first 4 hr and occasionally up to 24 hr and at the end of 72 hr13 for recording mortality, if any. Additional observations like behavioral changes, somato motor activity, tremors, convulsions, tonic extension, strub tail, muscle spasm, loss of righting reflex, ataxia, sedation, hypnosis, lacrimation, diarrhea, salivation, writhing, changes in skin, fur, eyes, mucous membranes etc were recorded. One tenth of upper limit dose and its half dose and double dose were selected as the levels for examination of therapeutic activity.

Oral Glucose Tolerance Test
After acclimation for 7 days, the oral glucose tolerance test was performed in overnight fasted normal rats14. All the rats were randomly divided into five groups (n=6). Group 1 was received only vehicle 1 ml/100 g and served as control group animals; group 2, treated with 5 mg/kg of glibenclamide; the remaining three groups were treated with 100, 200 and 400 mg/kg of methanolic extracts of *Sesbania grandiflora*. The rats were fasted for 12h (free access to water) and administered the above drugs to respective groups. Zero minute blood sugar level was determined from overnight fasted animals. After 30 minutes of the drug treatment (p.o.) the rats of all groups were orally fed with glucose 4 gm/kg. Blood glucose concentration was determined after 30, 60, 90 and 120 minutes of glucose loading. The blood samples were collected from the tail tip and measured by using glucometer and blood glucose test-strips.

Hypoglycemic activity
The hypoglycemic activity was performed in overnight fasted normal rats as per the method described by Jarald et al., 2008. All the rats were randomly divided into five groups of six rats each. Group 1 was kept as control, and was given a single dose of 1 ml/100 g of the vehicle; group 2 was treated with glibenclamide (5 mg/kg) as the hypoglycemic reference drug. Groups 3, 4 and 5 were treated with methanolic extract at three dose levels i.e. 100, 200 and 400 mg/kg (p.o.). The rats were fasted for 12h (free access to water) and administered the above drugs to respective groups. Zero minute blood sugar level was determined from overnight fasted animals i.e. before oral administration of drug. The blood glucose concentration was also measured after 30, 60 and 120 minutes of oral administration of drug. The blood samples were collected from the tail tip of the rats and measured the glucose concentration by using glucometer and blood glucose test-strips5.

Triton-induced hyperlipidemic model
Several studies showed that systemic administration of triton WR 1339 (ionic surfactant) in fasted rats causes elevation in plasma lipid level. Triton WR-1339 has been widely used to block clearance of triglyceride-rich lipoproteins to induce acute hyperlipidemia in several animals15, 16. This model is widely used for a number of different aims16 particularly, in rats it has been used for screening natural or chemical hypolipidemic drugs17.
After 7 days acclimation, hypolipidemic activity was studied in triton WR 1339 induced hyperlipidemic rats. All the rats were randomly divided into six groups of six rats each. Group 1 and 2 were received only vehicle 10 ml/kg and served as normal control and hyperlipidemic control groups respectively; group 3, treated with Simvastatin 10mg/kg and served as standard animals; the remaining three groups were treated with 100, 200 and 400 mg/kg of methanolic extracts of *Sesbania grandiflora*. The different drugs were administered to the respective group of animals for 5 consecutive days. Hyperlipidemia was induced on 6th day of the experiment by single injection of 400 mg/kg of triton WR
The effect of methanolic extract in fasted normal rats

The hypoglycemic effect in fasted normal rats were evaluated, after administration of the methanolic extract at the dose of 100, 200 and 400 mg/kg and glibenclamide 5 mg/kg to respective group and the results are given in table-2. After 30 min. of drug administration up to the end of 2 hours the blood glucose levels of the standard animals were declined. The extract show hypoglycemic activity at 400 mg/kg dose.

The effect of methanolic extract in Triton-induced hyperlipidemic model

As expected, administration of triton WR1339 led to elevation of serum lipid levels, which were maintained over a period of study in hyperlipidemic control group. The results were comparable with reference standard simvastatin. There was a significant elevation in serum lipids in triton-induced hyperlipidemic control rats when compared with normal control. The methanolic extract of Sesbania grandiflora at 200 and 400 mg/kg dose levels showed significant serum lipid lowering effects in hyperlipidemic rats and reduced serum lipids significantly (P<0.05) as compared to hyperlipidemic control statistically (shown in table-3).

DISCUSSION

The present study was undertaken to examine the hypoglycemic and hypolipidemic activity of methanolic extract of Sesbania grandiflora. Antihyperglycemic effect was studied on glucose loaded rats and hypoglycemic effect was studied on the normal rats. Hypolipidemic effect of the methanolic extract was evaluated by using triton induced hyperlipidemic model. Effective blood glucose control is the key for preventing or reversing diabetic complications and improving the quality of life in patients with diabetes. Thus, sustained reduction in hyperglycemia will decrease the risk of developing microvascular complications and most likely reduce the risk of macrovascular complications10. On the basis of this statement, we have selected the glucose induced hyperglycemic model to screen the antihyperglycemic activity of the plant extracts.

In the glucose loaded hyperglycemic model, the plant tested for antihyperglycemic activity exhibited significant antihyperglycemic activity at the dose level of 200 and 400 mg/kg. Excessive amount of glucose in the blood induces the insulin secretion. This secreted insulin will stimulate peripheral glucose consumption and control the production of glucose through different mechanisms20. However, from the study (glucose control), it was clear that the secreted insulin requires more than 2 h to bring back the glucose level to normal. In case of the methanolic extract and drug treated groups, the glucose levels did not exceed the control group, giving an indication regarding the supportive action of the extracts and drug in the glucose utilization. The methanolic extract of the plant, when tested for hypoglycemic activity, exhibited significant activity at the dose level of 400 mg/kg, suggesting its mechanism might be similar to sulfonlyureas. Sulfonlyureas increase insulin secretion by act on β-cells of islets of langerhens. This was marked increase in the level of serum total cholesterol, triglycerides, LDL, VLDL and decrease in the level of good cholesterol carrier HDL in the animals treated with triton. Elevated level of blood cholesterol especially LDL was the major risk factor for the coronary heart disease and HDL as cardio protective protein. Treatment with 200 and 400 mg/kg methanolic extract of Sesbania grandiflora significantly decreased the level of cholesterol, triglycerides,
VLDL and LDL as compared to hyperlipidemic control. There was significant increase in HDL as compared to control. This effect may be due to the increased activity of lecithin: cholesterol acetyl transferase which incorporates free cholesterol, free LDL into HDL and transferred back to VLDL and intermediate density lipoprotein. Decrease in the triglyceride level may be due to the increase in activity of the endothelium bound lipoprotein lipase which hydrolyzes the triglyceride into fatty acid or due to inhibition of lipolysis so that fatty acids do not get converted to triglyceride. Lipoproteinlipase releases fatty acids from chylomicrons and very low-density lipoproteins (VLDL) in the circulation. Half of those are taken up for storage. Catecholamines can suppress lipolysis via their action on β-adrenoceptors, but they mostly have the opposite effect of insulin and, by acting on β-adrenoceptors, increase cyclic AMP and the phosphorylation of hormone-sensitive lipase, stimulating lipolysis and the release of fatty acids in the circulation. Hepatic cholesterol synthesis is accelerated by triton WR 1339. Moreover, triton physically alters very low density lipoproteins rendering them refractive to the action of lipolytic enzymes of blood and tissues, preventing or delaying their removal from blood13. Hence the hypolipidemic effect of extracts could be due to an increased catabolism of cholesterol into bile acids.

**CONCLUSION**

The results obtained from the pharmacological screening conclude that the methanolic extract of *Sesbania grandiflora* plant has shown potential activity in decreasing the serum glucose level and have significant antihyperlipidemic activity. This research supports the inclusion of this plant in traditional antidiabetic preparations.

**ACKNOWLEDGEMENTS**

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**REFERENCES**


22. Ramirez A, Arancibia A, Calvo MA. Hypolipidemic effect of extracts could be due to an increased catabolism of cholesterol into bile acids.
Fig. 1: The effect of methanolic extract of *Sesbania grandiflora* in glucose loaded animals

**Table 2:** The effect of methanolic extract of *Sesbania grandiflora* in fasted normal rats

<table>
<thead>
<tr>
<th>Groups</th>
<th>Blood glucose concentration (mg/dl) at different time</th>
<th>0 min</th>
<th>30 min</th>
<th>60 min</th>
<th>90 min</th>
<th>120 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vehicle</td>
<td>85.50 ± 2.88</td>
<td>84.33 ± 3.24</td>
<td>87.67 ± 2.72</td>
<td>83.67 ± 2.14</td>
<td>84.83 ± 3.04</td>
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</tr>
<tr>
<td>Glibenclamide</td>
<td>84.17 ± 3.27</td>
<td>59.83 ± 3.75***</td>
<td>47.67 ± 2.74***</td>
<td>39.83 ± 1.82***</td>
<td>35.17 ± 2.21***</td>
<td></td>
</tr>
<tr>
<td>MESG-100</td>
<td>86.50 ± 3.45</td>
<td>85.83 ± 3.66</td>
<td>83.17 ± 3.27</td>
<td>87.17 ± 3.30</td>
<td>86.5 ± 2.43</td>
<td></td>
</tr>
<tr>
<td>MESG-200</td>
<td>85.17 ± 2.51</td>
<td>83.83 ± 2.64</td>
<td>78.67 ± 2.26</td>
<td>76.17 ± 2.89</td>
<td>73.83 ± 2.71</td>
<td></td>
</tr>
<tr>
<td>MESG-400</td>
<td>84.17 ± 4.45</td>
<td>77.17 ± 2.47</td>
<td>69.17 ± 2.75*</td>
<td>64.83 ± 2.82**</td>
<td>61.67 ± 2.72***</td>
<td></td>
</tr>
</tbody>
</table>

The results were expressed as mean ± SEM, n=6. The difference between groups was analyzed by one-way analysis of variance (ANOVA) followed by Dunnet’s test with 5% level of significance (P<0.05).

***P< 0.001, **P< 0.01 and *P< 0.05; compared Standard and Test groups vs Vehicle control.

Fig. 2: The effect of methanolic extract of *Sesbania grandiflora* in fasted normal rats

**Table 3:** The effect of methanolic extract of *Sesbania grandiflora* in Triton-induced hyperlipidemic model

<table>
<thead>
<tr>
<th>Groups</th>
<th>Lipid profiles (mg/dl)</th>
<th>Total Cholesterol</th>
<th>Triglycerides</th>
<th>HDL</th>
<th>LDL</th>
<th>VLDL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>61.83 ± 1.54</td>
<td>55.17 ± 1.64</td>
<td>32.67 ± 1.28</td>
<td>18.13 ± 2.16</td>
<td>11.03 ± 0.33</td>
<td></td>
</tr>
<tr>
<td>Hyperlipidemic</td>
<td>193.17 ± 3.58***</td>
<td>114.33 ± 1.93***</td>
<td>17.83 ± 1.11***</td>
<td>152.47 ± 4.42***</td>
<td>22.87 ± 0.39***</td>
<td></td>
</tr>
<tr>
<td>Standard</td>
<td>118.83 ± 2.97***</td>
<td>79.67 ± 2.33***</td>
<td>29.83 ± 1.14***</td>
<td>73.07 ± 2.67***</td>
<td>15.93 ± 0.47***</td>
<td></td>
</tr>
<tr>
<td>MEGO-100</td>
<td>196.33 ± 1.99</td>
<td>111.17 ± 1.14</td>
<td>19.50 ± 0.99</td>
<td>154.60 ± 2.62</td>
<td>22.23 ± 0.23</td>
<td></td>
</tr>
<tr>
<td>MEGO-200</td>
<td>150.17 ± 3.69***</td>
<td>95.33 ± 1.76***</td>
<td>24.17 ± 1.42*</td>
<td>106.93 ± 3.84***</td>
<td>19.07 ± 0.35***</td>
<td></td>
</tr>
<tr>
<td>MEGO-400</td>
<td>127.83 ± 2.57***</td>
<td>87.33 ± 2.67***</td>
<td>27.33 ± 1.36***</td>
<td>83.03 ± 3.52***</td>
<td>17.47 ± 0.53***</td>
<td></td>
</tr>
</tbody>
</table>

The results were expressed as mean ± SEM, n=6. The difference between groups was analyzed by one-way analysis of variance (ANOVA) followed by Dunnet’s test with 5% level of significance (P<0.05).

***P< 0.001, **P< 0.01 and *P< 0.05; compared Normal control vs Hyperlipidemic control.

***P< 0.001, **P< 0.01 and *P< 0.05; compared Standard and Test groups vs Hyperlipidemic control.
Fig. 3: The effect of methanolic extract of *Sesbania grandiflora* in Triton-induced hyperlipidemic model

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