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Research Article

PRELIMINARY PHYTOCHEMICAL ANALYSIS AND ACUTE ORAL TOXICITY STUDY OF *MUCUNA PRURIENS* LINN. IN ALBINO MICE

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ABSTRACT
Mucuna Pruriens Linn. is an annual, climbing shrub which has an important place among aphrodisiac herbs in India since the ancient times. The plant has been using traditionally for many medicinal purposes such as Infertility, Parkinson's disease, Loss of libido, Antioxidant, Anti venom, Anti microbial etc. The present study was carried out to investigate the preliminary phytochemical analysis and acute oral toxicity of the seeds of *M.pruriens* on albino mice. Matured seeds of *M.pruriens* were dried in shed and grinded in a mechanical grinder. The preliminary phytochemical analysis was done by following standard protocols. For acute oral toxicity study, methanolic extract of the seeds were used. The extract was prepared in a Soxlet apparatus. The preliminary phytochemical analysis showed the presence of protein, carbohydrates, glycosides, alkaloids, steroids, flavonoids, phenols and tannins. The acute oral toxicity study showed no mortality up to a dose of 4000 mg per kg body weight. The presence of plant chemicals revealed the medicinal values and the non toxic property of the plant indicated the value of the plant as medicine. Thus, we can conclude that, the seed of the plant can be used as a safe drug against many diseases.

Key Words: *Mucuna pruriens*, phytochemical analysis, acute oral toxicity, methanolic extract.

INTRODUCTION

In the ancient Indian Ayurvedic and Unani medicine systems, numerous plants and their products have been recommended for endurance against many diseases. *Mucuna pruriens* (Leguminoseae) is an annual herbaceous twining, climbing legume with long, thin branches and opposite, trifoliolate, lanceolate leaves 15 to 30 cm length. Its flowers are white to dark purple and hang in long clusters later form clusters of pods (Fig.1). The pods of the Mucuna have hair-like needles covering the outside which cause intense irritation to the skin. Traditionally the plant has been using for many diseases all over the world. In Brazil, it has been using as an aphrodisiac, diuretic, and nerve tonic, and for edema, intestinal worms. In Germany, it is used for diabetes, high blood pressure, high cholesterol, intestinal gas, muscle pain, rheumatism, worms etc. In India also, Mucuna has tremendous medicinal use. It has been using against abortions, cancer, catarrh, cholera, cough, debility, diabetes, diarrhea, diuretic, dysentery, edema, fertility, gout, impotency, kidney stones, menstrual disorders, nervousness, scorpion sting, snakebite, sterility, tuberculosis, worms, and as an aphrodisiac and uterine stimulant. The plant is used in other parts of the globe for asthma, burns, cancer, cholera, cough, cuts, diarrhea, diabetes, dog bite, edema, insanity, intestinal parasites, menstrual problems, mumps, nerves, pain, paralysis, pleurisy, ringworm, snakebite, sores, syphilis, tumors, wind-burns, worms, and as an aphrodisiac[1]. *Mucuna pruriens* has been found to have many beneficial properties some of which are Anti-parkinson activity [2], Anti-diabetic [3], Antioxidant, Metal chelating property [4], Green manure and fodder which has been successfully intercropped with maize in different parts of the world [5]. It was found to regulate Steroidogenesis and improve semen quality in infertile men [6]. The present study was carried out to investigate the preliminary phytochemical analysis and acute oral toxicity of the seeds in albino mice.

MATERIALS AND METHODS

Plant Sample Collection and Identification

Mature seeds of *M. pruriens* were collected from Barpeta district, Assam, India. The seeds were washed properly and dried in shade. For the identification of the specimen the plant was collected and prepared herbarium and authenticated in the department of Botany Gauhati University, Assam, India.

Extraction of Plant Materials

Dried seeds were grinded in a mechanical grinder to powdered form. 50 grams of the material were run with 200 ml of methanol in a soxhlet apparatus for 24 hours. The extract was collected and the methanol was evaporated in a rotary evaporator at 60° C. The dried extract was preserved in a refrigerator for future use.

Test Animals

All the animals used in the present study were obtained from the Animal House of the Department of Zoology, Gauhati University, Assam, India.

Preliminary Phytochemical Investigation

Preliminary phytochemical investigation was done using standard procedures [7,8,9]

- **Test for Proteins** Few drops of nitric acid were added by the sides of the test tube very gently to 1 ml methanol extract. Formation of yellow colour indicated the presence of protein in the sample.
- **Test for Carbohydrates** 1 ml each of Fehling A and Fehling B were added in diluted extract and heated for 30 minutes and observed for the formation of brick red colour.
- **Test for Resins** Five milliliter of distilled water was added to the methanol extract and observed for turbidity.
- **Test for Tannins** 5 ml of 45% ethanol was added to 2 g of the ground sample and boiled for 5 min. The mixture was cooled and filtered. Then 3 drops of lead sub acetate solution was added to 1 ml of the filtrate. A gelatinous precipitates were observed which indicates the presence

of Tannins. Another 1 ml of the filtrate was added 0.5 ml of bromine water. A pale brown precipitates were observed indicating the presence of Tannins.

- **Test for Saponins** 0.5 g of methanol extract was added to 5 ml of distilled water in a test tube. The solution was shaken vigorously and observed for a persistent froth. The frothing was mixed with 3 drops of Olive oil and shaken vigorously after which it was observed for the formation of an emulsion.
- **Test for Flavonoids** 0.5 g of the macerated sample of was introduced into 10 ml of ethyl acetate and heated in boiling water for 1 min. The mixture was then filtered. 4 ml of the filtrate was shaken with 1 ml of 1% aluminum chloride solution and kept. Formation of a yellow colour in the presence of 1 ml dilute Ammonia solution indicated the presence of flavonoids.
- **Test for Alkaloids** 5 gm of ground material was extracted with 10 ml Ammonical Chloroform and 5 ml chloroform. The mixture was filtered and the filtrate was shaken with 10 drops of 0.5 (M) H_2SO_4. Creemish white precipitate was observed for the presence of Alkaloids.
- **Tests for Steroids** 2 ml of acetic anhydride was added to 0.5 g of methanol extract and 2 ml of Sulphuric acid was added by the sides of the test tube and observed for the colour change from violet or blue-green.
- **Test for Phenols** Methanol extract was taken in a test tube and mixed with distilled water and warmed. To this 2 ml Ferric chloride solution was added and observed for the formation of green or blue colour.
- **Test for Glycosides** About 0.5 ml of methanol extract was taken in a test tube and added 1 ml glacial acetic acid containing traces of ferric chloride. To this solution 1 ml conc. Sulphuric acid was added and observe for the formation of reddish brown colour at the junction of the two layers and the upper layer turned bluish green in the presence of glycosides.

Acute Oral Toxicity Study

Acute Toxicity was studied by the method followed by Handa *et al*., 1990[10]. Albino mice of both sexes were taken for this experiment. Animals were divided in six groups (n=5) and were given different doses of plant extract via oral route (150, 300, 500,1000, 2000, 3000, 4000 mg/kg body weight) for four consecutive days and their mortality, loss of body wt. and general behaviour was recorded from the first dose up to 72 hours after the last administration of plant extract. One group was taken as control group and was administered with normal saline through the same route.

RESULTS

Preliminary Phytochemical Investigation

The preliminary phytochemical analysis of the seeds of the *M.pruriens* is shown in the Table1.

Acute Oral Toxicity Study

The investigation of acute toxicity is an initial step in the characterization of the biological effects of any substance which is necessary for conducting any biological experiment. The oral acute toxicity study revealed no mortality up to the dose 4000 mg/kg. The results are shown in the Table2.

DISCUSSION

Mucuna pruriens Linn. is the most popular drug in the Ayurvedic system of medicine[11] .The present study revealed the presence of phytochemicals like protein, glycosides, alkaloids, steroids, flavonoids, phenols, tannins and carbohydrates. All these phytochemicals help in preventing many diseases. It should be noted that steroidal compounds are of importance and interest in pharmacy due to their relationship with such compounds as sex hormones[12]. The presence of protein, glycosides and carbohydrate also indicate the palatability of the material. The medicinal values like anti-oxidant[13,14] , anti-venom[15],anti-tumour[16], anti-microbial[17] etc. have been evaluated scientifically. As already stated, the plant has many medicinal values, so it is an essential factor that, the plant should not have any toxicity. The acute oral toxicity study showed no toxicity up to a range of 4000 mg/kg body weight. Therefore, the oral administration of the root of the plant will not affect the animal in terms of its mortality. Hence the plant here can be seen as a potential source of useful drugs.

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Table1: Results of the preliminary phytochemical study.

Sl no	Phytochemicals	Result
1.	Protein	+ ve
2.	Carbohydrate	+ ve
3.	Resins	+ ve
4.	Tannins	+ ve
5.	Saponin	- ve
6.	Flavonoids	+ ve
7.	Alkaloids	+ ve
8.	Steroids	+ ve
9.	Phenols	+ ve
10.	Glycosides	+ ve

Table II: Results of Acute Oral Toxicity Study.

Group	Dosage mg/kg							Observation
Control	Normal saline							No Mortality No changes in Body weight No changes in General behaviour
Seed extract	150	300	500	1000	2000	3000	4000	

Fig.1 Photographs of the plant (A= plant; B= Flower; C= Pods; D=Seeds)

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