

Investigating the toxicity of the leaf extracts of lotus

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ABSTRACT

Lotus croniculatus belongs to Fabaceae family and grows in the temperate regions and grasslands of the countries of Africa and America. The plant is commonly known as common bird's foot tree foil. The plant is a herb and appears to be clover plant. The flowers of the plant are used mainly to induce sleep and other effects that regard to brain and heart. The plant is used to reduce spasms in the digestive tract. The infusions that are diluted to certain extent are helpful to reduce the anxiety and insomnia and also to treat exhaustion. In spite of the possession of lot of pharmacological activities, the plant is to be studied in a detailed manner for the toxicity also. Medicinal plants are known to cause low adverse effects and high potency. In the present investigation, the acute and chronic toxicity levels of the plant Lotus croniculatus extract of leaves was studied in laboratory animals. It is indicated that the animals did not show any change in behavior in acute toxicity and no mortality too. The extract also showed no signs of toxicity and change of values in the blood cells, liver enzymes and also kidney function tests were also normal at the dose of 2g/kg which denotes the safety of the plant in rats.

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INTRODUCTION

Lotus croniculatus belongs to Fabaceae family and grows in the temperate regions and grasslands of the countries of Africa and America. The plant is commonly known as common bird's foot tree foil. The plant is a herb and appears to be clover plant [1, 2]. The leaf consists of 5 leaf lets. The plant has forage properties and is used as fodder for liver stock and exhibits non bloating properties in the livestock. The flowers of the plant are used mainly to induce sleep and other effects that regard to brain

and heart. The plant is used to reduce spasms in the digestive tract. The infusions that are diluted to certain extent are helpful to reduce the anxiety and insomnia and also to treat exhaustion.

The plant serves as the highest quality of fodder in agriculture and do not cause any bloating in the animals. This plant is often used as an alternative for the use as alfa alfa that grows in poor soil. The plant is generally grown like an ornamental shrub [3]. The plant also prevents soil erosion and also provides the habitat for the wildlife. The plant was investigated and reported containing glycosides of cyanogenetic type. They release trace amounts of hydrogen cyanide and other cyanogen derivatives when they are macerated with solvents. The plant is usually non-poisonous to the human beings at a very low dose. At this low dose the metabolism of the cyanide is really quick and excretes out in a short duration of time. The presence of condensed tannins was also confirmed in the plant. These compounds were known to increase the absorption of proteins in the intestine. The production of cyanides in the plant is prevented by preparing the infusions and used to induce sedations [4]. The herb

was investigated to prove pharmacological properties and documented to contain many flavonoids like Cyanidin, Diphenidin, Quercetin. These are responsible for afore mentioned pharmacological activities of the plant [5].

In spite of the possession of lot of pharmacological activities, the plant is to be studied in a detailed manner for the toxicity also. Medicinal plants are known to cause low adverse effects and high potency [6]. The potency of the plant may convert it to the toxicity if it is overdosed or abused. Proper care and measures are to be taken to utilize the plant for its full potential and for safer usage. Hence this work is carried out to investigate on the toxicity profile of the plant and to establish the toxic doses of the aqueous extract of the plant. The expected clinical outcome of the research is to determine the safest dose of the plant extract [7, 8].

METHODOLOGY

The plant is procured from the farm land in the locality and was duly authenticated by a botanist and the herbarium sample is deposited in the college library for further reference. The leaves of the plant were collected and dried in the shaded area where there is free flow of air that facilitates the ambient temperature and normal relative humidity. The dried leaves were powdered and the powder is passed through a sieve to get fine and even powder. The plant powder is weighed for 50gm and mixed in distilled water of 100ml in a beaker. This is topped off with 5ml of chloroform to prevent contamination and fungal growth. The beaker is closed with an aluminum foil and left aside for 7 days [9, 10]. Occasional stirring was done or every 24 hrs. the extract was filtered through a filter paper and was dried using a rotary vacuum evaporator. The thick paste of the extract was 19.25 %w/w. It was a brownish green colour paste with a normal odour and is used for further study directly.

Laboratory animals

Rats of albino wistar strain which weighed around 160-200g were selected for the study. The animals were bought from the supplier from Bengaluru city. Animals of both sexes were included in the investigation and are allowed to acclimatize in well ventilated room for about 10days. The temperature and relative humidity were kept normal as the outside conditions. The animals were allowed to take the feed freely and water ad libitum.

Acute toxicity studies

Acute toxicity studies were performed by adopting the OECD guidelines 423. Weights were noted

from the animals that were selected for the study. The rats were administered at a dose of 2000mg/kg body weight. The extract is dissolved in distilled water at a specified concentration and this was administered to the animals at the prescribed dose via oral route. The animals were allowed to rest in the cages and are kept devoid of the pellet feed and water. The rats were monitored for the signs of toxicity like irritation, itching, redness and other abnormal signs. On the basis of the toxicity and the mortality of the rats that is produced after the first stage further grouping was to be performed using the standard procedures. So the groups were divided to get the drug administration at the doses of 250 mg/kg, 500 mg/kg and 1000mg/kg. the ED50 was determined using the chronic toxicity study [2, 11].

Chronic toxicity studies

The study for the chronic toxicity was done for about 6 weeks with about 6 animals in each group. 1% of sodium CMC at 5mg/kg orally is given to vehicle group and 0.9% simple saline at 5ml/kg orally is given to control group. For extract groups 250mg, 500mg, 1g and 2g/kg of doses were given orally. The vehicle group and the extract groups were administered one dose in one week time. This was continued for 6 weeks duration. After the specified duration of time the animals were sacrificed to collect the blood from the retro orbital plexus to perform the analysis of enzymes and cell counts. The blood sugar levels, bilirubin, gluconate, oxaloacetate, transaminase, glutamate pyruvate transaminase, alkaline phosphate, cholesterol, urea in blood, uric acid, protein and creatinine, hematological analysis WBC, RBC, Clotting time, Hemoglobin were estimated by utilizing the standard [12, 13]. The results were subjected to the ANOVA using T-test values of $P < 0.05$ and were considered significant. The results data was represented as the means and the standard errors of mean [14].

RESULTS & DISCUSSION

The acute toxicity analysis were conducted to notice any changes in the weight variations, seizures and excess of salivation change in the fur and skin, mucosal changes. There were no abnormal changes that are noticed in the rats. A slight change was noted in the weight of the animal that might be because of the excessive eating of rat. The results of the chronic toxicity study were tabulated and the data suggests that there no clear toxicity even at the dose of 2g/kg of the rat. There was a normal trend of values for all the groups regarding the hematological, and biochemical parameters. There is no significant difference between the variations from

Table 1: Chronic toxicity analysis effect on renal profile

Treatment	Urea (mg/dL)	Uric acid (mg/dL)	Creatine (mg/dL)	Protein (g/dL)	Glucose (mg/dL)
Saline-5ml/kg	23.42±0.58	5.62± 0.53	0.98 ± 0.34	7.12±0.46	86.04±3.41
S-CMC-5mg/kg	24.65±0.71	6.83 ±0.92	2.32 ± 0.53	8.47±0.65	87.31±4.84
LCA-250mg/kg	24.23±0.97	6.41±0.65	2.54±0.70	8.29±0.57	87.24±4.72
LCA-500mg/kg	23.70±0.82	5.73±0.46	2.02±0.51	7.01±0.37	86.07±3.90
LCA-1000mg/kg	24.31±0.43	6.57±0.78	2.77±0.67	8.35±0.24	87.44±4.59
LCA-2000mg/kg	25.58±0.64	7.39±0.76	3.40±0.56	9.28±0.71	88.20±5.85

Table 2: Chronic toxicity analysis effect on hepatic profile

Treatment	Bilirubin (mg/dL)	SGOT (unit/L)	SGPT (unit/L)	ALP (unit/L)	Cholesterol (mg/100ml)
Saline-5ml/kg	0.73±0.08	53.02±1.99	31.40±0.62	9.71±0.34	63.82±3.26
S-CMC-5mg/kg	2.24±0.11	54.71±3.25	32.52±0.73	10.35±0.43	64.53±4.16
LCA-250mg/kg	2.56±0.35	54.93±2.87	32.81±0.93	10.46±0.66	64.74±4.71
LCA-500mg/kg	0.81±0.13	53.05±2.36	31.74±0.89	9.12±0.58	63.92±3.01
LCA-1000mg/kg	2.32±0.16	54.64±3.19	32.06±0.68	10.22±0.37	64.48±4.9
LCA-2000mg/kg	1.04±0.58	55.8±4.38	33.56±0.97	11.0±0.61	65.23±5.10

Table 3: Chronic toxicity analysis effect on hematological parametres

Group	RBC(cu.mm) x10 ⁹	Hemoglobin Gm %	Monocyte %	Lymphocyte %	Esoinophil %	Neutrophils %	Clotting time in sec
Saline-5ml/kg	9.24± 0.36	12.25 ± 0.93	4.07 ± 0.21	73.21 ± 4.38	3.08 ± 0.42	22.9 ±2.75	76.2 ± 3.1
S-CMC-5mg/kg	10.41± 0.59	12.76± 0.98	5.23± 0.42	74.52± 5.73	3.87±0.63	23.40± 3.82	77.6± 4.8
LCA-250mg/kg	10.12± 0.62	13.47± 0.96	5.52± 0.53	74.64± 5.12	4.98± 0.74	23.13± 3.04	76.9± 4.5
LCA-500mg/kg	9.36±0.73	12.01± 0.77	3.97± 0.34	73.45± 4.82	2.75±0.55	22.32± 2.01	76.7± 3.4
LCA-1000mg/kg	10.29± 0.45	13.56± 0.92	5.42± 0.37	74.36± 5.91	4.24±0.86	22.28± 3.58	77.14± 3.9
LCA-2000mg/kg	10.23± 0.94	14.32± 0.48	6.41±0.56	75.8±6.61	5.19±0.81	24.60± 4.11	78.3 ±5.2

the normal control group animals. There was no abnormality in the behavior of animals and also no mortality in the rats. Even at the dose of 2g/kg the rats were normal and this indicates that the extract was safer at the specified dose. The hematological readings were normal and indicated no signs of hematopoiesis or the destruction of WBC. The extracts raised the numbers slightly that indicates the activity of the extracts. The renal data was evaluated and it also shows no elevation or random reduction in the values in any group which indicates that the extract did not show any toxicity to the kidneys. This proves the extract had no function in lowering the renal efficiency.

The liver function parameters were normal and the enzymes were unchanged even after administration of the extract at 2g/kg. This indicates the safety of the extract on the liver function. Finalizing it, the extract can be claimed safer and relatively effective at a dose of 2g/kg dose also.

CONCLUSION

In the present investigation, the acute and chronic toxicity levels of the plant *Lotus croniculatus* extract of leaves was studied in laboratory animals. It is indicated that the animals did not show any change in behavior in acute toxicity and no mortality too. The extract also showed no signs of toxicity and change of values in the blood cells, liver enzymes and also kidney function tests were also normal at the dose of 2g/kg which denotes the safety of the plant in rats.

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Conflict of Interest

The authors declare that they have no conflict of interest for this study.

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