Evaluation of Adaptogenic and Antidepressant Activity of hydroalcoholic root extract of plumeria alba

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ABSTRACT

Plumeria alba is a species of plant belonging to the family Apocynaceae, which is common to moist ground in tropical areas across the world. Extracts of P. alba L. are reportedly used in traditional medicine in Africa, South America, and Asia to treat a wide variety of human diseases. In Mexico, aqueous extracts from dried leaves of P. alba L. are used to treat anxiety.

The present study was carried out to evaluate the antidepressant and adaptogenic potential of hydroalcoholic extracts of Plumeria alba root on various animal models. Tail suspension test and elevated plus maze was used to evaluate the antidepressant activity while swim endurance test and anoxia stress tolerance were used to evaluate the adaptogenic activity. The adaptogenic effect was assessed by swimming time and estimation of various biochemical parameters like glucose, cholesterol, triglycerides, cortisol and BUN levels. These activities were tested at dose of 500 mg/kg as high dose and 250mg/kg as low dose extracts of Plumeria alba root using Withania somnifera (100mg/kg, p.o.) as standard drug. It was found that extracts significantly (p<0.01) increases swimming time in rats. It also showed significant (P<0.05) decrease in blood glucose, cholesterol, triglyceride, plasma cortisol and BUN levels as compared to control group. The hydroalcoholic root extract of Plumeria alba was also able to increase the anoxia stress tolerance significantly(P<0.01). The obtained results revealed that Plumeria alba root has got significant anti stress activity. The hydroalcoholic root extract of Plumeria alba was also able to decrease the duration of immobility significantly (p<0.01) when compared to the control using standard as Imipramine (10mg/kg).

Key words: Plumeria alba; adaptogenic activity; anoxia stress tolerance ; antidepressant activity ; tail suspension test.

INTRODUCTION
Adaptogens are herbs that are nontoxic, produce a nonspecific defensive response to stress, and have a normalizing influence on the body. Adaptogens help the body adapt to stress, support its normal functions, and restore balance. They increase the body’s resistance to physical, biological, emotional, and environmental stressors. They are unique from other substances in their ability to balance endocrine hormones and the immune system, and allow the body to maintain optimal homeostasis [1]. Stress is a factor in many illness from headaches to heart disease, and immune deficiencies to digestive problems. A substantial contributor to stress-induced decline in health appears to be an increased production of stress hormones and subsequently decreased immune function [2]. Depression is the most prevalent mental disorder. The disorder was characterized by apathy, loss of energy, retardation of thinking and activity, as well as profound feelings of gloominess, despair and suicidal ideation. In spite of the availability of antidepressant drugs like tricyclic antidepressants, selective reversible inhibitors of monoamine oxidase-A (MAO-A), selective serotonin reuptake inhibitors (SSRIs) and selective noradrenaline reuptake inhibitors (SNRIs), depression continue to be a major medical problem [3].

**MATERIALS AND METHODS**

**Drugs and chemical used:**

*Withania somnifera* was procured from the Sapthagiri Pharma, Bangalore, India. Imipramine was procured from HCG pharma, Bangalore, India. All other reagents used were of analytical grade.

**Experimental animals:**

Albino Wistar rats weighing between 160-220g gm and Albino mice weighing 20-30 gm of either sex were used for this purpose. The animals were randomized into experimental, normal and control groups, housed individually in sanitized polypropylene cages containing sterile paddy husk as bedding. The experimental animals were maintained under 12:12 h light dark cycle, in an animal house with controlled temperature (20-25°C) and humidity. The animals were maintained under standard condition in an animal house approved by the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

The study protocol was approved by the Institutional Animal Ethics Committee (IAEC), Office of institutional animal ethical committee (IAEC) of Mallige College of pharmacy, Bangalore. Reg. no. 1432/PO/Re/S/11/CPCSEA

**Collection and authentication of plant material:**

The roots of Plumeria alba were collected from the surrounding gardens of Tarabanahalli. The sample was identified and authenticated by the Professor Channabasappa Botanist , Sri Siddaganaga science college , tumkur, karnataka. The data will be complied on the basis of animal experiment done at our institution laboratory.

**Extraction procedure:**
The roots are washed, dried under air conditioning and reduced to powder with electric mill. The powder is cold extracted with ethanol 95% and water mixture (80:20) for 72 hr. The crude extracts are filtered and evaporated [4].

**Preliminary Phytochemical Analysis**

Plumeria alba root extract was subjected to chemical tests for the identification of their active constituents. Test for the presence of carbohydrates, glycosides, resins, tannin, alkaloid, fixed oil, flavonoids, terpenoids, protein, saponins, anthraquinone and amino acid were conducted as per the standard procedure [5].

**Acute toxicity and dose selection:**

No signs of toxicity (behavioral changes or mortality) were observed after single oral administration of hydroalcoholic extract (5000 mg/Kg p.o) in rats during two weeks of observation. Therefore the LD50 of oral administration of P. alba extract is higher than 5000 mg/Kg in Sprague Dawley rats [6].

**Pharmacological Screening**

Evaluation of Adaptogenic activity of Plumeria alba by anoxia stress tolerance test: Albino mice of either sex weighing between 18-22 g are divided into four groups of six in each. Hermetic vessel of 500 ml air capacity is used for this test. Each animal is kept in the hermetic vessel and the time to show the first sign of convulsion is noted, it is immediately removed from the vessel and resuscitated if needed. After one week of drug treatment the animals are once again exposed to the anoxia stress. Similarly the animals are also observed at the end of 2nd and 3rd weeks with the same treatment and the time duration for anoxia stress tolerance is noted.

**Experimental protocol**

Animals were divided into four groups of six animals each. The groups were as follows:

**Group 1:** Normal control (mice administered with (normal saline 10ml/kg b.w.p.o.) daily for 21 days

**Group 2:** Standard Withania somnifera (animals were administered with single dose of Withania somnifera (10 mg/kg, p.o.) daily for 21 days

**Group 3:** Animals received single dose hydroalcoholic root extract of P.alba (250 mg/kg p.o.) daily for 21 days

**Group 4:** animals received single dose of hydroalcoholic root extract of P.alba (500 mg/kg p.o.) daily for 14 days

Evaluation of antidepressant activity of hydroalcoholic root extract of Plumeria alba by tail suspension test (TST) [8]

Albino mice weighing about 20-30g are used. Animals were moved from their housing colony to laboratory in their own cages and allowed to adapt to the laboratory conditions for 1-2 hrs. Each animal was individually suspended in the wooden box with the help of hanging clip
the animal was suspended 50 cm above the floor of that wooden box and the 1cm part of the tail was clipped. Each animal under test was both acoustically and visually isolated from other animals during test. The total period of immobility was recorded manually for 6 min. Each animal showed vigorous movement during initial 2 min period of the test. The duration of immobility was manually recorded during the next 4 min of the total 6 min testing period. Animal was considered to be immobile when it didn’t show any body movement, hung passively and completely motionless. The test was conducted in a dim lighted room and each mouse was used only once in the test.

After successive 14 days of treatment with control, standard and extract drugs, the immobility is calculated and recorded.

**Experimental Design**

Group - I Control treated with vehicle

Group - II Standard drug (Imipramine-10mg/kg)

Group –III hydroalcoholic root extract of Plumeria alba (250 mg/kg,p.o.) Group -IV hydroalcoholic root extract of Plumeria alba (500mg/kg,p.o.)

**RESULTS AND DISCUSSION**

**Extraction and Phytochemical Investigation**

Successive Soxhlet extractions of Plumeria alba root was performed. The extract powder was brownish white in colour and hygroscopic in nature. The Plumeria alba extract was subjected to different preliminary chemical tests to determine the chemical constituents present in the extract. The results has indicated the presence of flavonoids, and triterpenoids, alkaloids carbohydrates compounds as shown in Table 1.

**Table 1** Test for phytoconstituents of R. arboreum flower extract
Test for phytoconstituents of *Plumeria alba* root extract

<table>
<thead>
<tr>
<th>Phytoconstituents</th>
<th>Plumeria alba root extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>+</td>
</tr>
<tr>
<td>Anthraquinones</td>
<td>-</td>
</tr>
<tr>
<td>Tannins</td>
<td>+</td>
</tr>
<tr>
<td>Steroids</td>
<td>-</td>
</tr>
<tr>
<td>Glycosides</td>
<td>-</td>
</tr>
<tr>
<td>Reducing sugar</td>
<td>+</td>
</tr>
</tbody>
</table>

(+) - Present  
(-) - Absent

**Evaluation of Adaptogenic activity**

**Model I: Anoxia Stress Tolerance**

The Anoxia tolerance test was determined by taking the appearance of convulsion as end point. The hydroalcoholic root extract of *plumeria alba* at low dose (i.e. HAEPA-I 250 mg/kg/ b.w) and high dose (HAEPA-II 500 mg/kg/ b.w.) showed significant (p<0.001) increasing tolerance stress time (i.e. onset of convulsion time) in 7th 14th 21st day as compared with the control. Results were depicted Table 2 and graphically represented in Figure 11

**Effect of hydroalcoholic root extract of Plumeria alba on anoxia stress tolerance test**
<table>
<thead>
<tr>
<th>Treatment group</th>
<th>Dose (mg/kg, p.o.)</th>
<th>Duration of Anoxia Stress Tolerance (min)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1st Day</td>
</tr>
<tr>
<td>Control</td>
<td>–</td>
<td>21.8±0.583</td>
</tr>
<tr>
<td>Standard (Withania somnifera)</td>
<td>100mg/kg</td>
<td>21.8±0.374**</td>
</tr>
<tr>
<td>HAEPAM-I</td>
<td>250 mg/kg</td>
<td>23.4±0.51</td>
</tr>
<tr>
<td>HAEPAM-II</td>
<td>500 mg/kg</td>
<td>21.8±0.49*</td>
</tr>
</tbody>
</table>

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett’s. *P<0.05, **P<0.01, ***P<0.001
Fig. 1: Effect of hydroalcoholic root extract of *Plumeria alba* on Anoxia stress test

**TAIL SUSPENSION TEST**

Table 3 Effect of hydroalcoholic root extract of *Plumeria alba* on tail suspension test

<table>
<thead>
<tr>
<th>Treatment group</th>
<th>Dose in mg/kg (p.o.)</th>
<th>Duration of immobility (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1st day</td>
</tr>
<tr>
<td>Control</td>
<td>-</td>
<td>239±1.83</td>
</tr>
<tr>
<td>Standard (withania somnifera)</td>
<td>100mg/kg</td>
<td>232±1.3*</td>
</tr>
<tr>
<td>HAEP A-I</td>
<td>250mg/kg</td>
<td>336±01.53</td>
</tr>
<tr>
<td>HAEP A-II</td>
<td>500mg/kg</td>
<td>224±1.93</td>
</tr>
</tbody>
</table>

Results are expressed as mean ±SEM, data analyzed by using one way ANOVA followed by Dunnett’s. *P<0.05, **P<0.01, ***P<0.001
DISCUSSION AND CONCLUSION

The present study suggests that the hydroalcoholic root extract of Plumeria alba has significant adaptogenic and antidepressant activity. This adaptogenic activity in the plant might be due to the presence of flavanoids and Antidepressant activity might be due to terpenes. However, further investigation should be carried out to elucidate the exact mechanism of action.

The present goal of the study was to evaluate the adaptogenic and antidepressant activity of hydroalcoholic root extract of Plumeria alba in mice & rat.

- Title plant was investigated for Adaptogenic activity using following animal models:
  a. Anoxia stress test

- Title plant was investigated for Antidepressant activity using following animal models:
  a. Tail suspension test

- In Anoxia stress test, oral administration of HAPEPA at the dose of 250mg/kg and 500mg/kg significantly increase the stress tolerance time.
- In Tail suspension test, oral administration of HAPEPA at the dose of 250mg/kg and 500mg/kg significantly reduced duration of immobility.
- The hydroalcoholic root extract of Plumeria alba showed significant adaptogenic and antidepressant activity.

Declaration of Interests: None declared
REFERENCES


