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Evaluation of Anticonvulsant Activity of Ethanolic Extract of *Sebastiania chamaelea* on Wistar Albino Rats



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ABSTRACT

Objective: To evaluate the anticonvulsant activity of ethanolic extracts of areal parts of *Sebastiania chamaelea* (Euphorbiaceae) against Strychnine (STR) induced convulsions at different dose levels. **Methods:** The anticonvulsant effect of ethanolic extract of *Sebastiania chamaelea* by the Strychnine (STR)-induced seizure. **Results:** The extract at 200 and 400 mg/kg, produced a significant ($P < 0.01$) dose dependent increase in onset of convulsion compared to the control strychnine-induced seizures. **Conclusion:** The data obtained was shown that ethanolic extracts of *Sebastiania chamaelea* areal parts may help to control grand mal and petit mal epilepsy as dose dependent manner. Further study need to identify the exact phytochemical candidate showing anticonvulsant activity using different animal models.



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INTRODUCTION

Epilepsy is a disorder that is best viewed as a symptom of disturbed electrical activity in the brain, which may be caused by a wide variety of etiologies. It is a collection of many different types of seizures that vary widely in severity, appearance, cause, consequence, and management. Seizures that are prolonged or repetitive can be life-threatening. Each year, 120 per 100,000 people in the United States come to medical attention because of a newly recognized seizure.¹ At least 8% of the general population will have at least one seizure in a lifetime. The rate of recurrence of a first unprovoked seizure within 5 years ranges between 23% and 80%. 'Funny turns', black-outs or apparent seizures have many causes, including hypoglycaemia, vasovagal attacks, cardiac dysrhythmias, drug withdrawal, migraine and transient ischaemic attacks.

The plant *Sebastiania chamaelea* belonging to the family of *Euphorbiaceae*, the juice of plant astringent, Tonic It is used against diarrhoea, Syphilis. When cooked together with meat, and vegetables, whole plant is used for speedy recovery for women after giving birth. In Africa decoctions of the stems are used to relieve teething pain (as a bath), vertigo (applied on head) and taken with butter in the form of tonic^{ii, iii}.

Usually flowers and fruits as a herb but sometimes grows into a shrub about 1 m tall. Leaf blades about 20-50 x 3.5-8 mm, petioles 1-4 mm long, channeled on the upper surface. Leaf margin usually toothed but the teeth quite small, pale and visible only with a lens. Stipules caducous, small, short and triangular, about 0.5 mm long. Petioles and twigs emit a watery white to clear exudate. Flowers are small, male flowers about 0.5 mm diam., borne in slender spikes while the female flowers about 0.5-1 mm diam., tend to be solitary with petals have ciliate margins and two glands at the base. Ovary has three bi-lobed appendages on the outer surface. Cotyledons are obovate, about 13-14 x 7 mm, apex truncate. First pair of leaves are linear to obovate. At the tenth leaf stage: leaf blade linear to obovate, about 30-35 x 7 mm, apex retuse, base obtuse or cuneate, petiole about 1 mm long. Lateral veins about 8, forming loops inside the blade margin. Grows in open forest but also found in vine thickets and low closed forest on sand dunes close to the sea. Also occurs in Africa, Asia and Malaysia.

Ethanollic leaf extract shows the presence of Flavonoids, sterols, Tannins, Phenols (Phenolic compounds like caffeic acid, Melilotic acid, Aesculetin, P-hydroxy benzoic acid,

Coumarin, Cinnamic acid, Salicylic acid and Scopolamine. Aqueous leaf extract shows the presence of Flavonoids (myricetin, Quercetin, Kaempferol, and Luteolin) steroids, Tannins, glycosides, Terpenoids^{v,vi}.

MATERIALS AND METHODS

Animals

Albino rats of Wistar strain of either sex weighing between 150-200g were used. At room temperature (25±20C) they were housed in polyethylene cages in animal house, allowed to 4 days of acclimatization and maintained standard rat chow and standard laboratory conditions throughout the experiment. The study was conducted after obtaining approval by institutional ethical committee (IAEC) of Vijaya college of Pharmacy, Hyderabad 1292/PO/Re/S/09/CPCSEA.

Preparation of Extract

The powdered sample (200g) was extracted in 500 ml of ethanol for 72 hours. The extract was filtered using a vacuum pump and concentrated by removing the solvent completely using a water bath^{vii, viii}.

Preliminary phytochemical investigation

Preliminary phytochemical investigation was done as described in as per the Practical Pharmacognosy books.^{ix}

Toxicity Studies

Rats were kept overnight fasting prior to drug administration. A total of five animals were used which received a single oral dose (2000 mg/kg, b. wt.) of extract. Food was withheld for further 3-4 hrs and animals were observed individually at least once during the first 30 min after dosing, periodically during the first 24 h and daily thereafter for a period of 14 days. Once daily cage side observations included changes in skin and fur, eyes and mucous membrane (nasal) and also respiratory rate, circulatory (heart rate and blood pressure), autonomic (salivation, lacrimation, urinary incontinence and defecation) and central nervous system (tremors and convulsion)

changes and Mortality was determined over a period of 2 weeks. We did not find any changes including morbidity and mortality in rats^x.

ANTI EPILEPSY ACTIVITY

Strychnine (STR) induced seizure

Wistar albino rats of either sex weighing between 150-200gm were selected and divided into different groups required for the study; each group contains six animals^{xi-xiii}.

Group I - served as control (normal saline)

Group II -served as reference drug (Phenytoin) mg/kg body weight (i.p)

Group III - Hydro-alcoholic aerial part extract of *Sebastiania chamaelea* (200mg/kg, b.w, p.o)

Group IV - Hydro-alcoholic aerial part extract of *Sebastiania chamaelea* (400mg/kg, b.w, p.o)

Group II-IV Treated with one-hour later administration of test drugs, strychnine 5mg/kg i.p.
strychnine administered.

Statistical Analysis

Results were presented as statistically by using Graphpad prism version 5.0. All results were expressed as Mean±SD, analyzed by one-way analysis of variance followed by Tuckey's test.

RESULTS

The phytochemical screening showed the presence of alkaloids, terpenoids, tannins, sterol's, flavonoids and carbohydrates. The oral acute toxicity study showed that maximum dose of 2000mg/kg of extract is safe for 14 days of study. Strychnine was used to induce seizures and the test drugs were administered to assess effect on the seizures. The parameters assessed in animals were total number of convulsion, onset of first seizure; onset of clonic convulsions, duration of clonic convulsion, onset of tonic convulsion, duration of tonic convulsion, number of death within 30 min duration, and % age protection from death was also calculated and shown in table 1. The animals were divided into four groups of 6 animals each. Plain control was administered vehicle orally; standard control group was administered diazepam 5 mg/kg b. w. i. p.; test group A was administered 200 mg/kg orally, where test group B was administered 400 mg/kg orally.

After the administration of the above drugs, all animals were injected Strychnine after 30 min of the administration of diazepam i.p. In standard control and 60 min of administration of vehicle, test drug A and test drug B in plain control group, test group A and test group B respectively. Just after administration of Strychnine, animals were placed in an isolated cage and assessed for above mentioned parameters.

Mean total number of convulsion in plain control was 804.33 ± 103.74 ; in test group A was 686 ± 35.15 ; in test group B was 247 ± 126.96 , and no convulsion appears in animals of standard group. The mean score of each group was compared with each other using ANOVA one way with post Tuckey pair comparison test. Mean total number of convulsion in test A and test B were significantly ($p < 0.01$) reduced when compared with mean score of plain control, Test group B showed significant ($p < 0.01$) reduction in mean number of convulsion when compared with test group A.

Mean onset of first seizure in plain control was 33.83 ± 1.76 , in test group A was 57.5 ± 4.83 and in test group B was 64 ± 3.81 , test A and test B were significantly ($p < 0.01$) delayed the mean onset of first seizure when compared with plain control, but test A in comparison to test B there was no statistical significant ($p > 0.05$) difference.

The mean onset of clonic convulsion in plain control was 50.5 ± 5.59 , in test group A was 68.16 ± 3.63 and in test group B was 81 ± 4 , test drug A and test drug B showed significant ($p < 0.05$) increase in comparison to plain control, but there no significant difference was found when test group A was compared with test group B.

The mean duration of clonic seizures in plain control was 11.5 ± 1.39 , in test A was 9.84 ± 0.95 and in test B it was 9 ± 0.86 , there is no significant difference found between mean scores when compared with each other.

Table 1. Effect of SCHE on Strychnine induced seizures in rats

Parameters (Time in Sec.)	Groups			
	I Vehicle p.o.	II Diazepam 5mg/kg p.o.	III SCHE 200mg/kg p.o.	IV SCHE 200mg/kg p.o.
Number of convulsions	789.30±103.74	0.00	686±35.5**	247±127
Onset of 1 st convulsion	33.83±1.76	0.00	57.3±4.83*	64±3.80**
Onset of clonic convulsions	50.5±5.58	0.00	68.10±3.25*	82.5±3.45**
Duration of clonic convulsions	11.3±1.24	0.00	9.92±0.84	9.25±.46
Onset of tonic convulsions	108.12±8.24	0.00	348±62.10**	640.12±59.21***
Duration of tonic convulsions	17.98±1.25	0.00	16.20±1.02	5.89±8.10***
Number of death	6	0	2	0
% of Protection	0	100	66.66	100

Values are Mean±SD; n=6. P value *p<0.05, **p<0.01, ***p<0.001 vs. Control

DISCUSSION

Anticonvulsant activity of the hydroalcoholic extract of *Sebastiania chamelea* was screened by experimental model that was Strychnine induced seizure model. Strychnine-induced seizures test is considered as an experimental model for the "generalized absence seizures" and also a valid model for human generalized myoclonic seizures and generalized seizures of the petit mal type. In Strychnine induced seizure test parameters like onset of first seizure, onset of tonic convulsions, clonic convulsions and duration of tonic and clonic seizures, percent protection was observed. Test group A and test group B significantly (p< 0.01) increased the mean onset of first seizure when compared with plain control, but test group A in comparison to test group B there was no statistical significant (p>0.05) difference.

Test group A and test group B showed significant (p<0.05) increase in mean onset of clonic convulsion, in comparison to plain control, but there no significant difference was found when test group A was compared with test group B. In case of mean duration of clonic seizures there

was no significant difference found between mean scores when compared with each other; for the mean onset time of tonic convulsion, on the comparison of test A and test B with the plain control, test A group showed significant ($p < 0.05$) and test B also showed significant ($p < 0.001$) with respect to plain control, and in the comparison of test group A with test group B, test B showed significant ($p < 0.01$) increase in mean onset of tonic and clonic seizures with respect to Test A. For the mean duration of tonic convulsion on the comparison of control with test drugs, test A is not significant but test B showed significant reduction ($p < 0.001$) with respect to plain control and in comparison of test A with test B, test B showed significant reduction ($p < 0.01$) with respect to test A. After 30 min of interval protection from death was also assessed in the groups.

In control group all the animals died, whereas 66.66 % and 100% protection was observed in the animals of standard and test groups respectively. Thus, onset of first seizure, clonic and tonic seizure in the test groups was significantly increased ($p < 0.01$) when compared with plain control group and the effect was equivalent to standard group. The duration of tonic and clonic seizure was decreased in both the test groups and the reduction was significant when compared with the plain control group. **In conclusion**, the extract was shown significant antiepileptic effect at the dose of 200mg/kg and 400mg/kg.

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