Asian Journal of Pharmaceutical Technology & Innovation
ISSN: 2347-8810

Research Article

Received on: 29-07-2017
Accepted on: 05-08-2017
Published on: 15-08-2017

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Anticonvulsant Activity of Catharanthus Roseus Leaf

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ABSTRACT

Epilepsy is a neuropsychological disorder which causes seizures .The seizures are occurs due to imbalance between excitatory and inhibitory neurotransmitters. In the present study the Petroleum ether extract of Catharanthusroseus at the dose of 100,200 and 400 mg/kg was screened for anticonvulsant activity using pentylenetetrazole induced seizure test model. From present study it was observed that at the dose of 400mg/kg the petroleum ether extract showed better anticonvulsant activity.

Key-words: . Catharanthusroseus,anticonvulsant, pentylenetetrazole.

Cite this article as:
Manisha Shinde, Sanjay Chaudhari, Ritu Gilhotra, Anticonvulsant Activity of Catharanthus Roseus Leaf, Asian Journal of Pharmaceutical Technology & Innovation, 05 (25); 09-12, 2017. www.asianpharmtech.com

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Introduction

Central nervous system is complex, controlling various body functions through the balance of variety of stimulating and inhibitory neurotransmitters. Any drug that alters the action of any of the neurotransmitters may affect various neurobehavioral and neuroendocrinalfunctions. In India epilepsy is second most common neurological disorder.Epilepsy is a neuropsychological disorder which causes seizures.Seizures are occur due to over discharge of neuro transmitter substances result into an imbalance between excitatory and inhibitory neurotransmitters.[1]Inspite of tremendous advances in the epilepsy research the synthetic antiepileptic drugs are not affordable and posses many toxic adverse effects. Hence considering the above fact the development of safe and cheap anticonvulsant agent from plant sources is essential.

Catharanthusroseus commonly known as vincarosea,is an evergreen subherb or herbaceous plant growing to 1 m. tall. The leaves are oval to oblong, 2.5- 9.0 cm. long and 1- 3.5 cm. broad glossy green hairless with a pale midrib and a short petiole about 1- 1.8 cm. long and they are arranged in the opposite pairs. The flowers are white to dark pink with a dark red centre, with a basal tube about 2.5- 3 cm. long and a corolla about 2-5 cm. diameter with five petal like lobes. The fruit is a pair of follicles about 2-4 cm. long and 3 mm broad.*C. roseus*posses carbohydrate, flavinoid, saponin and alkaloids.[2]

From long time C.roseus has been used for the treatment of various diseases in different parts of world.In China it was used as diuretic,astringent and to treat cough.In India it was used as wasp stings,for nose bleeding,sore throat, mouth ulcer and for bleeding gums.The roots of Catharanthusroseuspossess hypotensive, sedative and tranquillising property[3].The plant has been reported to possess wide range of pharmacological activity like Anti cancer activity[4],Anti ulcer[5],Anti diabetic activity ,[6] Hypotensive,[7] Anti diarrheal[8],Anti microbial activity,[9]Anti oxidant [10], Wound healing [11,] Hypolipidimic[12] ,Memory enhancement activity.[13].

Materials and Methods

Collection of plant: The plant was collected from the Sangamner,Dist-Ahmednagar.The leaves were identified by Dr. P.G. Diwakar, Joint Director, Botanical Survey of India; Pune with a voucher specimen (MTS 01**)** . The herbarium has been kept in botanical survey of India, Pune.

Successive solvent extraction

The leaves of Catharanthusroseuswas collected and air dried under shade and then coarsely powdered with the help of mechanical grinder. 500gms of powdered materials were evenly packed in the soxhlet apparatus. It was then extracted with various solvents from nonpolar to polar such as petroleum ether, ethyl acetate, chloroform, and ethanol and aqueous successively. The dried concentrated extracts were used for phytochemical investigation, isolation, pharmacological activity.

Preliminary phytochemical evaluation

The preliminary phytochemical screening of extracts of the catharanthusroseuswas carried out for qualitative identification of type of phytoconstituentspresent.The presence of various phytoconstituents viz. steroids and terpenoids (LeibermannBurchardtest), alkaloids (Dragendroffs test), tannins and phenolics (Ferric chloride test), flavonoids(Shinoda test), Sugars (Fehling solution test), amino acids (Ninhydrin test), etc. was detected by usual methods prescribed in standard texts.[14]

Experimental study

Evaluation of anticonvulsant activity;

Pentylenetetrazole (PTZ) induced convulsion

Albino mice of either sex with a body weight 22-25g will be divided into five groups of 6 animals in each. Group I will be served as control treated with PTZ 80 mg/kg, intraperitonially, Group II with diazepam (4mg/kg i.p). Groups III, IV, Vreceive three different doses of petroleum ether extracts of C.roseusleaves(100,200 & 400 mg/kg) for seven consecutive days. On the eighth day one hr after oral administration of the std/extracts in respective groups, PTZ 80 mg/kg is administered intraperitoneally. Each animal was placed into individual plastic cage and were observed initially for 30min and later up to 24 hrs. The following parameters were recorded during test session of initial 30min and up to 24 hrs:

- ► Latency (onset of clonus)
- ► Onset of tonic-clonic convulsions
- ► Status of animal after 30 minutes
- ► Status of animal after 24 hrs

► Percent protection

The values were expressed as mean ± SEM from 6 animals. The results were subjected to statistical analysis by using ANOVA followed by Dunnett's- t -test to calculate the significance difference if any among the groups. P<0.05 was considered significant. Mice that did not convulse 30 min after pentylenetetrazole administration were considering protected[15].

Stastical analysis-

All data were expressed as Mean ± S.E.M. The results were analyzed statistically by one-way ANOVA followed by Dunnett's multiple comparisons test. The results obtained were compared with the standard drug.

Result

1. **Phytochemical analysis-**
   Phytochemical screening of petroleum ether extract of Catharanthusroseus leaves showed the presence of carbohydrates, phenols, saponins, tannins and alkaloids but devoid of steroids.
2. **Toxicity study-**
   A preliminary acute oral toxicity study, the plant extracts produced no adverse effects at dose 2000mg/kg and did not cause any death up to dose of 5000 mg/kg in mice.
3. **Effect of Catharanthusroseus leaf extract on PTZ induced convulsions-**
   It was observed that the petroleum ether extract of Catharanthusroseusat the dose 400 mg/kg produce a significant reduction in the duration of extensor,clonus and stupor phase as compared to control group.

Table no.1 Phytochemical Evaluation of Petroleum ether extract of Catharanthus roseus leaf .

Sr.No.	Phytochemical Constituents	Pet.ether extract
1	Steroids	+
2	Saponins	+
3	Tannins	+
4	Alkoloids	+
5	Carbohydrates	-
6	Proteins	+
7	Amino acids	-
8	Flavonoids	-
9	Diterpenes	-
10	Phenols	-

Table No: 2 Effect of Catharanthus roseus leaves extracts on PTZ induced convulsions.

Sr. No.	Groups	Treatment	Time spent in Various phases of convulsion (Sec)					
			Flexion	Extension	Clonus	Stupor	R/ D	% Protection
1.	Control	D/W 10ml/kg, p.o.	5.16±0.30	9.83±0.30	43.83±0.87	182.17±2.94	R	0
2.	Standard	Diazepam 4 mg/kg , i.p.	5.16±0.30ns	3.00±0.36**	16.50±0.76**	81.66±2.06**	R	69.48
3.	Pet.Ether Extract	100 mg/kg, p.o.	5.830.16ns	8.16±0.30**	39.83±1.22ns	136.50±2.40**	R	16.98
4.	Pet.Ether Extract	200 mg/kg, p.o.	6.50±0.22*	6.00±0.25**	38.66±1.78ns	131.50±1.72**	R	38.96
5.	Pet.Ether Extract	400 mg/kg, p.o.	5.50±0.34ns	4.66±0.21**	29.66±2.29**	119.17±1.22**	R	52.59

ns-nonsignificant ,* P< 0.05, **P<0.01 Values are Mean± SEM , n=6,when compared with control by using one way ANOVA followed by Dunnette's multiple comparison test

Discussion

PTZ is widely used acute experimental model in the preliminary screening of anticonvulsant drugs.The mechanism by which PTZ is believed to exert its action is by acting as antagonist at the $GABA_A$receptor complex[15].

Phytochemical screening of the extracts revealed the presence of tannin, alkaloids, steroids, tannins and saponins.Triterpine steroids and triterpenoidalsaponins are reported to posses anticonvulsant activity in some experimental seizure models such as MES and PTZ . Some alkaloids,monoterpenes and flavonoids also have protective effects against PTZ,picrotoxin and NMDLA induced convulsions [1]. As petroleum ether extract of Catharanthusroseus contains all the above constituents hence the anticonvulsant effect may be due to presence of one or more above chemical constituent.

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