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EVALUATION OF ANTICONVULSANT ACTIVITY OF *PAVETTA TOMENTOSA* LINN. IN MES AND PTZ INDUCED EPILEPSY

Bibhu Prasad Moharana[1*], Sujit Kumar Martha[1], Pratit Kanchan Sahu[1], Ansuman Sahu[1]

[1]Jeypore College of Pharmacy, Rondapalli, Jeypore (K)-764002.

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***Correspondence for Author**
Bibhu Prasad Moharana
Jeypore College of Pharmacy, Rondapalli, Jeypore (K)-764002.

ABSTRACT

Disease of central nervous system is appearing as a major threat in the future because of increasing mental stress, strain and work, which are essential in the developing world. Herbal drugs which are having diversified uses are always an alternative option to the synthetic drugs which are well known for their adverse effects in which the need is not met. Epileptic seizures are caused by parts of the brain eliciting abnormal electrical activity. In persons with epilepsy, there is an imbalance between excitatory and inhibitory neurotransmitters. Ethanolic extract of *Pavetta tomentosa* linn (200mg/kg & 400mg/kg p.o.) was studied for anticonvulsant activity on maximal electroshock-induced seizures and pentylenetetrazole induced seizures in mice. EEPT 200 mg/kg and 400 mg/kg have shown 65.20% and 70.29% protection respectively against MES induced seizures and 60.77% and 81.5% protection respectively against PTZ induced seizures. It is found that treatment with EEPT on mice significantly reduces tonic hind limb extensor stage in MES induced epilepsy & a significant increase in onset of clonic convulsions in PTZ induced epilepsy.

KEYWORDS: Anticonvulsant, *Pavetta tomentosa* linn, MES, PTZ, Seizure, Mice.

INTRODUCTION

Epilepsy is one of the major neurological disorders affecting approximately 0.8% of the population. There has been considerable progress in the pharmacotherapy of epilepsy over the last few decades, including the introduction of new antiepileptic drugs such as felbamate, lamotrigine etc.[1] Attempts to find out a common neurochemical basis for human or experimental epilepsy have been disappointing. An imbalance between the excitatory and

inhibitory neurotransmitters is responsible for seizures.[2,3] At neuronal level, seizure activity often occurs when glutamatergic excitatory neurotransmitters overrides gamma-amino butyric acid (GABA) mediated inhibition.[4] Several animal models of convulsions have been developed to evaluate anti-seizure activity.

However, current drug therapy of epilepsy is complicated by side-effects, teratogenic effects; long term toxicity and about one third of patients are refractory to pharmacotherapy. Furthermore, there is currently no drug available which prevents the development of epilepsy e.g. after head trauma and all currently available anti-epileptic drugs are synthetic molecules.[5] According to data from the World Health Organization [WHO] plants are sources of biologically active compounds used by about 80% of the world population, both in natural form as teas, and as manufactured drugs. In wealthy countries, growing numbers of patients rely on alternative medicine for preventive or palliative care.[6] Centuries before the advent of modern medicine, synthetic chemistry and the pharmaceutical industry, virtually all medicines came from plants. These medicinal plants have been an important source for the discovery of novel bioactive compounds which served and continue to serve as lead molecules for the development of new drugs.[7]

Pavetta tomentosa linn. is an erect, nearly smooth or somewhat hairy shrub 2 to 4 meters or more in height, belonging to family Rubiaceae. The plant is reported from India to China and through Malaya to tropical Australia. The plant is used for the treatment of visceral obstructions, hemorrhoidal pain, constipation, dropsy, arthritis, brain disorders & as febrifuge traditionally.[8] In the southern part of India, the decoction & extract of the bark of this plant is used as a remedy for control of symptoms of epilesy. However, no scientific data are available to validate the folklore claim. The aim of the present study was, therefore, to evaluate the anticonvulsant activity of P. tomentosa in experimental animal models, with a view to provide a pharmacological justification for the ethno-medicinal use of the plant's bark in the management of convulsions.

MATERIALS AND METHODS

Plant Material

The bark of *Pavetta tomentosa* linn was collected from Tirupati, Andhra Pradesh & it was authenticated by Dr. K. Madhava Chetty, Plant Taxonomist, Department of Botany, Sri Venkateswara University, Tirupati, Andhra Pradesh.

Preparation of Extract

Bark of the whole plant were dried in shade, separated and made to dry powder. It was then passed through the 40mesh sieve for extraction purpose.[9] A weighed quantity (70gm) of the powder was subjected to hot solvent extraction in a soxhlet apparatus using ethanol, at a temperature range of 60-70°C. Before and after every extraction the marc was completely dried and weighed. The extract was concentrated to dryness (percentage yield 17.5%) at 40°C under reduced pressure in a rotary vacuum evaporator. The extract was stored at 4°C until used as a suspension with Sodium CMC.

Experimental Animals

Swiss albino mice (20-30g) of either sex were procured from Asterace labs Hyderabad, A.P. The animals were maintained in a well-ventilated room with 12:12 hour light/dark cycle in polypropylene cages. Standard pellet feed (Hindustan Lever Limited., Bangalore) and drinking water was provided ad libitum through out experimentation period. Animals were acclimatized to laboratory conditions one week prior to initiation of experiments. Ethical committee clearance was obtained from IAEC (Institutional Animal Ethics Committee) (013/IAEC/StJCOP/2012) of CPCSEA (Committee for the Purpose of Control and Supervision of Experiments on Animals). (Reg no.1278/ac/09/CPCSEA).

Drugs and Chemicals

Phenytoin (PHNY; Sigma Chemical Co.), Diazepam (DZP; Calmpose, Ranbaxy Pharma, India) was dissolved in 1%w/w Sodium CMC (Qualigens Fine Chem., Mumbai) to be used in this study as standard duugs.

Acute Toxicity Study

The toxicity study was determined in mice by modified method of Lorke.[10] Mice fasted for 16 h were randomly divided into groups of 10 mice per group and were administered i.p. with the extract in doses ranging from 1000-2000mg/kg.

Evaluation of Anticonvulsant activity

Maximum electroshock-induced seizures

The animals were divided randomly into 4 groups containing 6 animals each. Seizures are induced to all the groups by using an Electro convulsiometer. Maximal electroshock seizures were elicited by a 60Hz alternating current of 150 mA intensity for 0.2 sec. The EEPT were administrated for 14 days before induction of seizures.[11] The duration of various phases

(Flexion, Extension, Clonus, Stuper, and Recovery) of epilepsy were observed. The percentage protection was estimated by observing the number of animals showing abolition and duration of Hind Limb Tonic Extension (HLTE).

Pentylenetetrazol-induced seizures

The animals were divided randomly into 4 groups containing 6 animals each. The test groups received two doses (200mg/kg, 400mg/kg) of EEPT orally for 14 days and test conducted for antiepileptic activity 1 h after the last doses of each extract. PTZ (80mg/kg/i.p) was used as the inducing agent. After the administration of each extract & PTZ, each animal was placed in an individual plastic cage for observation lasting 1h seizures and tonic clonic convulsions were recorded. The control group animals were received 1% SCMC while standard groups animals were received diazepam (4.0mg/kg/i,p.) and observed for onset of convulsions up to 1h after PTZ administration.[12]

Statistical analysis

The data were expressed as mean ± standard error of mean (S.E.M).The Significance of differences among the group was assessed using one way analysis of variance (ANOVA). The test followed by Dunnet's T-test, p value < 0.05 were considered as significant.

RESULTS

Acute toxicity study

There was no mortality amongst the graded dose groups of mice up to a dose of 2000mg/kg for duration of 72 h. The animals were observed for further 14 days period for all toxicity signs. There was no considerable change in body weight before and after treatment and no sign of toxicity were observed. This finding probably suggests that the ethanol extract is relatively safe or non-toxic in mice at the doses used for this study.

Assessment of anticonvulsant activity

Maximum electroshock test

Phenytoin (PHT) treated animals have shown 100% protection against MES induced seizures where as EEPT 200mg/kg and 400mg/kg have shown 65.20% and 70.29% protection respectively against MES induced seizures. The EEPT at both doses and standard treated animals had shown a significant change in duration at all stages of convulsions. EEPT 200 mg/kg and 400mg/kg had shown a significant decrease in the duration of tonic extensor phase and comparable significance ($p<0.01$) with the control. The results were shown in **Table 1**.

Table 1: Effect of ethanolic extract of *Pavetta tomentosa* linn (EEPT) on maximal electroshock (MES) induced convulsions.

Groups	% protection	Flexion	Extensor	Clonus	Stupor	Recovery
I .Control	0	8.500±0.22	13.00±0.36	19.83±0.30	40.00±0.73	176.6
II. PHT	100	4.17±0.30**	0**	9.167±0.40**	16.17±0.79**	92.5
III.EEPT (200mg)	51.35	6.667±0.49**	4.00±0.36**	15.00±0.68**	31.83±1.53**	142.2
IV.EEPT (400mg)	66.21	5.167±0.21**	2.667±0.33**	13.0±1.03**	17.00±0.44**	112.43

*n=6 in each group. Values are expressed as mean ± SEM of six observations. **P<0.01 when compared to the vehicle by Dunnet's T-test.*

Pentylenetetrazol-induced seizures

Diazepam treated animals have shown 100% protection against PTZ induced seizures where as EEPT 200mg/kg and 400 mg/kg have shown 60.77% and 81.5% protection respectively against PTZ induced seizures. EEPT 200mg/kg and 400mg/kg had shown a significant increase in onset of clonic convulsions and comparable ($p<0.01$) with the control. Mortality of EEPT 200mg/kg and 400mg/kg treated groups was reduced as compared to control group animals. The results were shown in **Table 2**.

Table 2: Effect of ethanolic extract of *Pavetta tomentosa* linn (EEPT) on pentylenetetrazole (PTZ) induced convulsions.

Group	Treatment	% of Protection	Duration of convulsion	Onset of clonic convulsion	% of Protection mortality
I	CONTROL	0	72.29 ± 8.83	177.70 ± 2.36	50
II	DIAZEPAM	100	10.58 ± 2.62**	765.33 ± 9.06**	100
III	EEPT(200mg)	69.66	28.21 ± 3.30**	496.83 ± 3.70**	84.44
IV	EEPT(400mg)	80.48	21.87 ± 1.33**	566.83 ± 5.79**	100

*n=6 in each group. Values are expressed as mean ± SEM of six observations. **P<0.01 when compared to the vehicle by Dunnet's T-test.*

DISCUSSION

In the present study for the screening of aqueous extract of *Pavetta tomentosa* for anticonvulsant activity, two standard methods namely MES and PTZ methods have been employed. It is found that ethanolic extract of *Pavetta tomentosa Linn.* markedly protects epilepsy induced by MES and PTZ in mice. GABA is the major inhibitory neurotransmitter in the brain while glutamic acid is an excitatory neurotransmitter in the brain. The inhibition of GABA neurotransmitter and the enhancement of the action of glutamic acid have been shown to be the underlying factors in epilepsy.[12,13] Our study shows that the ethanol extract

of the barks of P. tomentosa protected some of the animals against seizures induced by maximal electroshock and pentylenetetrazole and also delayed the latency of the seizures.

In the present study maximal electroshock produced seizures in all the animals used. Antiepileptic drugs that block MES-induced tonic extension are known to act by blocking seizure spread.[14] Moreover, drugs that inhibit voltage-dependent Na+ channels, such as phenytoin can prevent MES-induced tonic extension.[14,15]

Pentylenetetrazole induced seizures in all the mice used. Pentylenetetrazole may elicit seizures by inhibiting gabaergic mechanisms.[16] Standard antiepileptic drugs, diazepam and phenobarbitone, are believed to produce their effects by enhancing GABA-mediated inhibition in the brain.[14] It is, therefore, possible that the anticonvulsant effects shown in this study by the drugs against seizures produced by PTZ might be due to the activation of GABA neurotransmission. Since the extract similarly antagonized seizures elicited by pentylenetetrazole in mice, it is probable, therefore, that it may also be exerting its anticonvulsant effects by affecting gabaergic mechanisms.

CONCLUSION

From the present study, it was concluded that the ethanolic extract of *Pavetta tomentosa* linn at both 200mg and 400mg exhibited significantly antiepileptic activity in MES and PTZ induced epilepsy. Further studies are in progress to isolate the active constituent and to establish the effect on various types of epilepsy like grand mal, petit mal, absence and partial seizure.

REFERENCES

1. Elger CE. Future trends in epileptology. Curr Opin Neurol., 2001; 14: 185–186.
2. McNamara JO. Drugs effective in the treatment of the epilepsies. In: Hardman JG, Limbird JE, Molinoff PB, Ruddon RW, Gillman AG, editors. Goodman and Gillman's The Pharmacological Basis of Therapeutics. 9th ed. New York: McGraw Hill, 1996; 461-86.
3. Rang HP, Dale MM, Ritter JM, Moore PK. Pharmacology 5th ed. India: Churchill Livingstone, 2005; 456-473.
4. Smolders I, Belle KV, Ebinger G, Michotte Y. Hippocampal and cerebellar extracellular amino acids during pilocarpine-induced seizures in freely moving rats. Eur J Pharmacol, 1997; 319: 21-9.

5. Pitkanen A, Lukasiuk K. Molecular and cellular basis of epileptogenesis in symptomatic epilepsy. Epilepsy Behav, 2009; 16-25.
6. Akerele O. Medicinal plants and primary health care: an agenda for action. Fitoterapia, LIX, 1988; 355-363.
7. Agosta W. Medicines and Drugs from Plants. Journal of Chemical Education, 1997; 74: 857.
8. Kirtikar KR, Basu BD. Indian Medicinal Plants. Lalit Mohan Basu, Allahabad, 2003; III: 2149-2151.
9. Khandalwal KR. Practical Pharmocognosy Techniques and Experiments. 3rd ed., Pune; Nirali Prakashan, 1996.
10. Lorke D. A new approach to practical acute toxicity testing. Arch Toxicol, 1983; 54: 275-287.
11. Balakrishnan S, Pandhi P, Bhargava VK. Effects of nimodipine on the efficacy of commonly used antiepileptic drugs in mice. Ind J Exp Biol, 1998; 36: 51-55.
12. Vidya Rao, Anjali Rao, Sudhakar Karanth K. J. Ethnopharmocol, 2005; 102: 231-235.
13. Westmoreland BF, Benarroch EE, Dube JR, Regan TJ, Sandok BA. Medicinal neurosciences, Rochester: Margo Foundation, 1994; 307-312.
14. Rogawski MA, Porter RJ. Antiepileptic drugs: pharmacological mechanisms and clinical efficacy with consideration of promising development stage compounds, Pharmacol Rev., 1990; 42: 223-286.
15. MacDonald RL, Kelly KM. Antiepileptic drug mechanisms of action. Epilepsia, 1995; 36: S2-S12.
16. De Sarro A, Cecchetti V, Fravoloni V, Naccari F, Tabarrinia O, De Sarro G. Effects of novel6-desfluoroquirolones and classic quinolones on pentylenetetrazole-induced seizure in mice. Antimicrob Agents Chemother, 1999; 43: 1729-1736.