Research Article

Anti-epileptic activity of *Mentha cordifolia* against PTZ induced epilepsy in Swiss Albino mice

Mallappa Shalavadi*, Ashwini Hosoor, Devraj, Muttu Mugali, Raghavendra Kinnal, Savita Patil, Chandrashekar V. M., Lingaraj Anawal, Shubham Teli

Department of Pharmacology, B.V.V.S Hanagal Shri Kumareshwar College of Pharmacy, Bagalkote 587101, Karnataka, India

Received: 3 February 2024 Revised: 25 March 2024 Accepted: 30 March 2024

Abstract

Objective: The present study was designed to investigate the anti-epileptic activity of hydroalcohol extract of *Mentha cordifolia* against Pentylenetetrazole (PTZ) induced epilepsy in Swiss albino mice. **Materials and Methods:** The hydroalcohol extract of *Mentha cordifolia* was screened for its antiepileptic activity against Pentylenetetrazole (PTZ) induced epilepsy. The extract was given orally at the doses of 200 mg/kg and 400 mg/kg for 15 consecutive days prior to the induction of seizure. The protection action against PTZ induced seizure was evaluated by behavioral paradigm. **Results:** In Pentylenetetrazole induced epilepsy model, hydroalcohol extract of *Mentha cordifolia* at 200 mg/kg and 400 mg/kg treated groups showed significant increase in onset of action and decrease in death latency, number of convulsions, number of Straub's tail, duration of convulsions, jerking, tonic, clonic, duration of Stuper and Straub's tail was observed. **Conclusion:** Present study shows that the potent antiepileptic role of hydroalcohol extract of *Mentha cordifolia* on Swiss albino mice showed significant changes in behavioral paradigm. Further studies are required to elucidation the potent action of *Mentha cordifolia* on treatment of epilepsy.

***Keyword*:** *Mentha cordifolia*, pentylenetetrazole, antiepileptic, convulsions, seizures

Introduction

Epilepsy is chronic serious neurological disorder, characterized by transient occurrence of abnormal, excessive and synchronous neuronal activity in the brain, associated with various neurological, cognitive and psychological signs and symptoms (Trinka et al., 2015). Around more than 50 million people in the world suffering from epilepsy and approximately 5-8% of the general population experience at least one seizure in their lives (Newton and Garcia, 2012). The various chemical classes of drugs like barbiturates (Phenobarbitone), Succinimides (ethosuximide), Gamma amino butyric acid (GABA) analogs etc. are used in management of seizures (Goldenberg, 2010). Currently, antiepileptic drugs have limited efficacy and negative properties limit their use and cause difficulties in patient management (Dalic and Cook, 2016). World health organization reported that approximately 80% of people live with epilepsy in developing countries (Reddy, 2005). Many new drugs like vigabatrin, topiramate, zonisamide levetiracetam, lamotrigine, lacosamide, rufinamide and stiripentol were developed and considered comparatively safe and represents a real progress in the treatment (Aneja and Sharma, 2013). However, adverse effects also not been completely circumvented and nearly 30-40% of the people continue to have seizures with these new antiepileptic drugs (Ezekiel et al., 2010).

In traditional system of medicine various types of medicinal plants used for the treatment of epilepsy was scientifically proved to possess promising anticonvulsant activities in screening models of anticonvulsant activity (Wannang et al., 2008). The use of traditional or folk medicinal plants represents “leads” compounds which is a short cut for discovery of modern medicines with novel structure which is cheaper and less time consuming (Soejarto, 1996; Plotkin, 1988; Holland, 1994).

Address for Corresponding Author:
Dr. Mallappa Shalavadi
Associate Professor & Head
Department of Pharmacology,
BVVS Hanagal shri Kumareshwar College of Pharmacy, Bagalkote 587101, Karnataka, India
E-mail: mallu.sha007@gmail.com

DOI: https://doi.org/10.31024/ajpp.2024.10.2.2

Mentha cordifolia commonly known as spearmint, marsh mint, pepper mint or mint and is locally well known in the Philippines as "yerba buena' or 'herba bueba' and it was classified by Quisumbing as *M. arvensis* (Quisumbing, 1978; Escueta, 1987; Cantoria, 1977; Mahendran and Rahman, 2020). *Mentha cordifolia* has couple of synonyms for this species like *Mentha viridis* (L.), *Mentha cordifolia*, etc. It is aromatic plant belong to Lamiaceae family. *Mentha cordifolia* is grown in Thailand and Southeast Asian countries. It is a popular flavoring herb of Thai food and herbal tea. *Mentha cordifolia* shows some pharmacological properties like Antihypertensive activity (Poungrat et al., 2011). The leaves of *M. cordifolia* contain 0.8% volatile oil, consisting mainly of pulgenone, pitoeitone, and limonene (Tan, 1978), menthol, menthene, and menthenone (Quisumbing, 1978). Other constituents include cadinene, 1-carvomenthone, isomenthone, 4,8-epoxy-p menthan-3-one, 2-isopropylcyclo pentanone, 3,7-dimethyl-1,6-octadien-3-ol (linalool) (major component of oil), and p-menthan-2,5-diol. The chief constituents of the essential oil are carvone (55-75%) and limonene (upto 21.4%). The herb gave flavonoids, diosmin and diosmetin. Caffeic acid derivatives include rosmarinic acid in the volatile oil (Sharma, 2016).

Materials and methods

Chemicals

Pentylenetetrazole (PTZ), 70% Alcohol, Diazepam and All other chemicals were of analytical grade.

Instruments

Double beam automated UV-Visible Spectrophotometer UV-1601 (Shimadzu, Japan), Refrigerator, Centrifuge (MPW-350OR, Korea), Auto analyzer (Star-21), Centrifuge (REMI/R248/99) and Compound Microscope (Olympus Magnus) was used.

Animals

Young *Swiss albino* mice of male sex (22-35gm) were procured from the Central Animal House of H.S.K College of Pharmacy Bagalkote. Animals were acclimatized to laboratory condition at room temperature prior to experimentation. Animals were kept under standard conditions of a 12-hour light/ 12-hour dark cycle with food and water ad-*libitum* in groups of plastic cages with soft binding. The protocol was approved by the Institutional Animal Ethics Committee of H.S.K. College of Pharmacy, Bagalkote (Ref. No: IAEC/HSKCOP/Aug 2022/UG1) and carried out in accordance with the CPCSEA Guidelines for the use and care of laboratory animals.

Collection Plant material and extraction

The Plant material was collected from the Bagalkote district of Karnataka. The plant was identified and authenticated at Basaveshwar Science College Bagalkote. The authentication number is BVS/Bot/2022/UG2. The plants leaves were washed thoroughly with water and allowed to shed dried. The dried plant leaves were subjected to coarse powder. The collected coarse powder was allowed to macerate with hydroalcohol up to 3 days with occasional stirring. The Macerated solution is allowed to dry at room temperature and obtained solid mass was stored in refrigerator at 2-4°C.

Acute toxicity Study

Acute oral toxicity of *Mentha cordifolia* was studied by Naoual et al. (2022). According to him there was no signs and symptoms of toxicity and mortality was observed at the dose of 2000 mg/kg. Based on this we have selected $1/10^{th}$ (200 mg/kg) and $1/5^{th}$ (400 mg/kg) ratio of this dose for the present activity.

Experimental Design for pentylenetetrazole induced model

Swiss albino mice were divided into 4 groups of 6 animals.

Group I: Control group received PTZ on 15^{th} day, (n=6)

Group II: Standard group received diazepam 10mg/kg for 15 days, (n=6)

Group III: This group, received 200 mg/kg of *Mentha cordifolia* for 15 days and PTZ on 15^{th} day, (n=6)

Group IV: This group, received 400 mg/kg of *Mentha cordifolia* for 15 days and PTZ on 15^{th} day, (n=6)

Mice were administered with respective treatments for 15 days and on same day, PTZ 80mg/kg was injected intraperitoneally to mice 60 min after treatments and 30 min after the diazepam administration. Immediately after PTZ administration mice were observed for behavioral parameters such as (1) Onset of convulsions (elapsed time from PTZ injection until convulsion occurred), (2) Duration of seizure (Total time how much the animal is in convulsions), (3) Mortality for the duration of 30 minutes, (4) Tonic seizures, (5) Clonic seizures, (6) Straub's tail, (7) Stuper and (8) Jerky moments (Viswanatha et al., 2016).

Results

The Effect of hydroalcohol extract of *Mentha cordifolia* on PTZ induced epilepsy in mice

The effect of HAMC on onset of action and death latency against PTZ induced epilepsy, The HAMC treated group shows significant ($p<0.001$) increase in Onset of Action and significant ($p<0.01$) decrease in death latency was observed as compared to Control group (Table 1).

The effect of HAMC on No. of Convulsion and No. of Straub's tail against PTZ induced epilepsy, The HAMC treatment group shows significant ($p<0.01$ to $p<0.001$) decrease in No. of Convulsion and No. of Straub's tail as

Table 1. The Effect of hydro alcohol extract of *Mentha cordifolia* on onset of action and death latency against PTZ induced epilepsy in mice

Groups	Time in minutes		Number of animals survived	% protection
	Onset of Action (Convulsions)	Death latency		
Control	3.14 ± 0.19	5.84 ± 0.74	0/6	0
Diazepam (10 mg/kg)	00 ± 00***	00 ± 00**	6/6	100
HAMC (200mg/kg)	4.59 ± 0.07***	00 ± 00**	6/6	100
HAMC (400mg/kg)	4.34 ± 0.35***	00 ± 00**	6/6	100

All the values are expressed as mean ± SEM, n=6, One way Analysis of Variance (ANOVA) followed by multiple comparisons Dunnett's test. The value significant** $p<0.01$, *** $p<0.001$ as compared to control group.

Table 2. The Effect of hydro alcohol extract of *Mentha cordifolia* on No. of Convulsion and No. of Straub's tail against PTZ induced epilepsy in mice

Groups	No. of Convulsion	No. of Straub's tail
Control	1.87 ± 0.23	4.0 ± 0.4
Diazepam 10 mg/kg	00 ± 00***	00 ± 00***
HAMC (200mg/kg)	0.87 ± 0.12**	3.0 ± 0.0*
HAMC (400mg/kg)	0.87 ± 0.12***	2.0± 0.0***

All values are expressed as mean ± SEM, n=6, One way Analysis of Variance (ANOVA) followed by multiple comparisons Dunnett's test. The value significant* $p<0.05$, ** $p<0.01$, *** $p<0.001$ as compared to control group

Table 3. The Effect of hydro alcohol extract of *Mentha cordifolia* on Duration of Convulsion, Jerking, Tonic, Clonic, Stuper and Straub's tail against PTZ induced epilepsy in mice

Groups	Time in Seconds					
	Duration of Convulsion	Jerking	Tonic	Clonic	Stuper	Straub's tail
Control	217.0 ± 8.13	6.0 ± 0.57	15.50 ± 0.28	4.75 ± 0.14	152.0 ± 12.06	25.25 ± 1.70
Diazepam (10 mg/kg)	00 ± 00***	00 ± 00***	00 ± 00***	00 ± 00***	00 ± 00***	00 ± 00***
HAMC (200mg/kg)	129.8 ± 5.94***	1.83 ± 0.44***	00 ± 00***	00 ± 00***	112.50 ± 0.86**	8.25 ± 0.75***
HAMC (400mg/kg)	104.3 ± 2.28***	1.66 ± 0.33***	00 ± 00***	00 ± 00***	55.50 ± 6.39***	8.50 ± 1.32***

All values are expressed as mean ± SEM, n=6, One way Analysis of Variance (ANOVA) followed by multiple comparisons Dunnett's test. The value significant ** $p<0.01$, *** $p<0.001$ as compared to control group

compared to Control group (Table 2).

The effect of HAMC on duration of Convulsion, Jerking, Tonic, Clonic, Stuper and Straub's tail against PTZ induced epilepsy, The HAMC treated group shows significant ($p<0.001$) decrease in Duration of Convulsion, Jerking, Tonic, Clonic, Stuper and Straub's tail as compared to Control group (Table 3).

Discussion

Epilepsy is an important problem in developing countries. GABA potentiating drugs like diazepam, benzodiazepine, barbiturate, valproate etc. have been adopted to treat epilepsy. However, prolonged use of such drugs develops tolerance and dependence. Further the side effects like weight gain, insomnia, reflex tachycardia; sexual dysfunction etc. limits the usage of such drugs.[19] As a result of this people worldwide are looking at the alternative system of medicines like Ayurveda, Unani and Homeopathy etc. for remedies to cure the epilepsy (Ekstein and Schachter, 2010).

In continuation of such search the field survey was carried

out and whole plant of *Mentha cordifolia* contains chemical constituents like flavonoids, Rutin, Quercetin 3-O-glucoside, Quercetin, 3-O-glucoside-7-O-rhamnoside. There is report the rutin and quercetin possess CNS depressant activity. Since facilitation of excitatory transmission or reduction of inhibitory transmission is important mechanism leading to epilepsy it was hypothesized that the plant *Mentha cordifolia* containing there phytoconstituents is useful in ameliorating epilepsy. Hence, the present study was planned to demonstrate the anti-epileptic activity of hydroalcohol extract of *Mentha cordifolia* against PTZ induced seizures in rats. In the first phase of the present study the plant material was collected, authenticated and the hydroalcohol extract of *Mentha cordifolia* was prepared. The chemical constituents like rutin and quercetin derivatives required for the antiepileptic activity which is highly soluble in hydroalcohol. So, extraction was carried out in hydroalcohol.

The effect of HAMC produced anticonvulsant activity in PTZ induced seizure model resembles diazepam in onset and duration of convulsion in rats. The probable mechanism associated by the extract may be due to GABA mediated occupational theory with the receptors there by augmenting inhibitory neurotransmission there by depressing neuronal brain parts. This signifies that the potential of anti-oxidant property offered by plant extract. Hence it may help in scavenge free radical which causes cellular damage of the brain cell and there by relieve oxidative stress produced in epileptic region of PTZ induced seizure in rat model (Zhu et al., 2014; Govindu and Adikay, 2014). The possible mechanism for all the protective action may be due to the presence of essential phytoconstituent (phenolics, steroids and flavonoids) present in them. The HAMC was subjected for preclinical studies using rats there is scope for clinical studies to validate further therapeutic benefits in human volunteers to find out the more detailed mechanism of action of this plant extract and ascertaining its isolated phytoconstituents responsible for the activity.

Conclusion

In the present study, hydro alcohol extract of *Mentha cordifolia* showed anti-epileptic activity against Pentylenetetrazole induced seizures in mice. Evidenced by decrease in the various phases such as stupor of convulsion in PTZ model and significant increase in onset in duration of seizure, decrease in duration of the seizure and reduced mortality rate in PTZ model in a dose dependent manner. In the PTZ model the effect shown by HAMC is similar to standard drug because of reduction in the facilitation of Na^+ ions to the neuronal area of the brain. So, this may be the reason for shortening in time of both the phases and delay in firing of neurons. Further studies are required to understand the molecular mechanism of this drug.

Declaration of competing interest

The authors declare no conflicts of interest

Funding sources

This research did not receive any specific funding.

Acknowledgement

We are grateful to the Department of Pharmacology, Hanagal Shri Kumareshwar College of Pharmacy Bagalkote for providing all the needful to carry out this research work.

References

Aneja S, Sharma S. 2013. Newer anti-epileptic drugs. Indian Pediatrics. 50(11):1033-40. doi: 10.1007/s13312-013-0284-9.

Cantoria MC, Pharmacognosy in Action. National Research Council of the Philippines, Bicutan, Tagig, MM, 1977.

Dalic L, Cook MJ. 2016. Managing drug-resistant epilepsy: challenges and solutions. Neuropsychiatric Disease and Treatment, 12:2605-2616. doi: 10.2147/NDT.S84852.

Dravyaguna vijnyana Vol-2 by Acharya Priyavrat Sharma, Publisher. Chaukhambha Bharti Academy, pp 365, 2016

Ekstein D, Schachter SC. 2010. Natural Products in Epilepsy- the Present Situation and Perspectives for the Future Pharmaceuticals 3: 1426-1445.doi:10.3390/ph3051426

El Menyiy N, Mrabti HN, El Omari N, Bakili AE, Bakrim S, Mekkaoui M, Balahbib A, Amiri-Ardekani E, Ullah R, Alqahtani AS, Shahat AA. 2022. Medicinal Uses, Phytochemistry, Pharmacology, and Toxicology of *Mentha spicata.* Evidence-Based Complementary and Alternative Medicine, 12; 2022.doi.org/10.1155/2022/7990508

Escueta CH, Herba Buena. Department of Agriculture and Food, Bureau of Plant Industry, Philippines, 1987

Ezekiel I, Mabrouk MA, Ayo JO, Goji AD, Okpanachi AO, Mohammed A, Tanko Y 2010. Study of the effect of hydro-ethanolic extract of Commiphora africana (stem-bark) on sleeping time and convulsion in mice. Asian Journal of Medical Sciences 2(3):85-88.

Goldenberg MM. Overview of drugs used for epilepsy and seizures: Etiology, diagnosis, and treatment. P T 2010;35(7):392-415.

Govindu S, Adikay S. 2014. Evaluation of antiepileptic activity of chloroform extract of Acalypha fruticosa in mice. Pharmacognosy Research. 6(2): 108.doi: 10.4103/0974-8490.128970.

Holland BK. 1994. Prospecting for drugs in ancient texts. Nature 369(6483):702. doi.org/10.1038/369702a0.

Mahendran G, Rahman LU. 2020. Ethnomedicinal, phytochemical and pharmacological updates on Peppermint (Mentha × piperita L.)-A review. Phytotherapy Research, 34(9):2088-2139. doi: 10.1002/ptr.6664.

Newton CR, Garcia HH. 2012. Epilepsy in poor regions of the world. Lancet. 380(9848):1193-201. doi.org/10.1016/S0140-6736(12)61381-6

Plotkin MJ. 1988. Conservation, ethnobotany, and the search for new jungle medicines: Pharmacognosy comes of age. again. Pharmacotherapy 8:257-62.doi.org/10.1002/j.1875-9114.1988.tb04081.x

Poungrat P, Upa K, Warinee B, Parichat P, Veerapol K, Saowanee N. 2011. *Mentha cordifolia* extract inhibits the development of hypertension in L-NAME-induced hypertensive rats. Journal of Medicinal Plants, 5(7):1175-83

Quisumbing, Medicinal Plants of the Philippines, Katha Publishing Co. Inc., 1978

Reddy DS 2005. Pharmacotherapy of catamenial epilepsy. Indian Journal of Pharmacology, 37(5):288-93. doi :10.4103/0253-7613.1685.

Soejarto DD. 1996. Biodiversity prospecting and benefit-sharing: Perspectives from the field. Journal of Ethnopharmacology, 51:1-15. doi.org/10.1016/0378-8741(95)01345-8

Trinka E, Cock H, Hesdorffer D, Rossetti AO, Scheffer IE, Shinnar S, Shorvon S, Lowenstein DH. 2015. A definition and classification of status epilepticus--Report of the ILAE Task Force on Classification of Status Epilepticus. Epilepsia. 56(10):1515-23.

Viswanatha GL, Venkataranganna MV, Prasad NB, Ashok G. 2016. Evaluation of anti-epileptic activity of leaf extracts of *Punica granatum* on experimental models of epilepsy in mice. Journal of Intercultural Ethnopharmacology. 5(4):415-421. doi: 10.5455/jice.2016090410285

Wannang NN, Anuka JA, Kwanashie HO, Gyang SS, Auta A 2008. Anti-seizure activity of the aqueous leaf extract of *Solanum nigrum* L. (Solanaceae) in experimental animal. African Health Sciences 8(2): 74-79.

Zhu HL, Wan JB, Wang YT, Li BC, Xiang C, He J, Li P. 2014. Medicinal compounds with antiepileptic/anticonvulsant activities. Epilepsia. 55(1): 3-16.doi: org/10.1111/epi.12463