Research Article
AMERICAN JOURNAL OF PHARMACY AND HEALTH RESEARCH
www.ajphr.com
2014, Volume 2, Issue 10
ISSN: 2321–3647(online)

Experimental Evaluation of Anticonvulsant Activity of Hydrocotyle Asiatica linn (Centella asiatica) in Albino Mice

Praveen Kumar Uppala[1]*, Murali Krishna B[2]
1. Assistant Professor, KVK College of Pharmacy, Affiliated to JNTUH, Hyderabad.
2. Assistant Professor, Bhaskara College of Pharmacy, Affiliated to Andhra University, Visakhapatnam.

ABSTRACT

To evaluate the antiepileptic activity of Hydrocotyle asiatica linn (aqueous extract) in preventing maximal electroshock (MES) and pentylenetetrazole (PTZ) induced convulsions. To compare its efficacy with standard drugs- phenytoin for MES method and sodium valproate for PTZ method. 48 male albino mice weighing 18-30g are selected and divided into 2 groups of 24 mice each – one group for MES and other group for PTZ method. In MES method, seizures were induced via ear clip electrodes with a current of 50 mA for 0.2 seconds. Each mouse is pretreated with drugs (p.o.) one hour before MES test. The different groups include – Group C1 administered distilled water (0.25ml), Group S1 administered phenytoin (50 mg/kg), Group T1 administered aqueous extract of Hydrocotyle asiatica linn (100 mg/kg) and Group T2 administered aqueous extract of Hydrocotyle asiatica linn (300mg/kg). In PTZ method, seizures were induced by giving PTZ 80 mg/kg s.c. Each mouse is pretreated with drugs one hour before giving PTZ. The different groups include – Group C2 administered distilled water (0.25ml p.o.), Group S2 administered sodium valproate (300mg/kg i.p.), Group T3 administered aqueous extract of Hydrocotyle asiatica linn (100 mg/kg) and Group T4 administered aqueous extract of Hydrocotyle asiatica linn (300mg/kg). The aqueous extract of Hydrocotyle asiatica at a dose of 300mg/kg has shown statistically significant anticonvulsant activity against both MES and PTZ convulsions and its anticonvulsant activity is similar to that of standard sodium valproate (300mg/kg).

Keywords: Hydrocotyle, asiatica,, Maximal, electroshock, seizures, Pentylene tetrazole, Sodium valproate

*Corresponding Author Email Praveen.chintu32@gmail.com
Received 10 August 2014, Accepted 29 August 2014

Please cite this article as: Uppala PK *et al.*, Experimental Evaluation of Anticonvulsant Activity of Hydrocotyle Asiatica linn (Centella asiatica) in Albino Mice . American Journal of Pharmacy & Health Research 2014.

INTRODUCTION

Epilepsy has been recognized since antiquity. The word epilepsy is derived from the Greek word meaning "to seize" or "take hold of", indicating that the person having a seizure is "possessed" or at least out of control.[1] More than a century ago, John Hughlings Jackson, the father of modern concepts of epilepsy, proposed that seizures were caused by "occasional, sudden, excessive, rapid and local discharges of gray matter," and that a generalized convulsion resulted when normal brain tissue was invaded by the seizure activity initiated in the abnormal focus[2] . Epilepsy is a relatively common neurological disorder. Approximately 5-10% of the population will have at least one seizure, with the highest incidence occurring in early childhood and late adulthood.[3] Early treatment ranged from exorcism to blood letting.[1] Bromide was introduced in 1857 for treating epilepsy and Phenobarbital in 1912. Subsequently many other anticonvulsants were discovered.[4] Antiepileptic therapy has many drawbacks such as long duration of therapy, adverse effects of drugs, need for therapeutic drug monitoring etc.[2,5,6] There is clearly a need for more specific and effective drugs [5]. Medicinal plants have been an important source of new drugs. Hydrocotyle asiatica linn has been extensively used in Ayurvedic medicinal practice for the treatment of epilepsy, various skin diseases, leprosy and malaria.[7,] Experimentally; drugs with a potential antiepileptic activity are assessed by injecting medullary stimulants or by applying a maximal electrical shock. Drugs which antagonize chemically induced seizures are effective in petit mal epilepsy and which protect against electrically induced seizures in grand mal epilepsy.[8]

MATERIALS AND METHODS

Male albino mice weighing 18 – 30 g are obtained from Central animal house, KIMS, Hubli. All test animals are allowed free access to food and water ad libitum, both being withdrawn just prior to experimentation. They are divided into 8 groups, each group consisting of 6 animals.

Chemicals and solutions

Phenytoin sodium (Ciron pharmaceuticals) – standard drug for MES method

Valproic acid (sun pharmaceuticals) – standard drug for PTZ method

Pentylene tetrazole (PTZ) [Himedia labs] - chemoconvulsant

Aqueous extract of Hydrocotyle asiatica linn – test compound

Distilled water – vehicle

Equipment

Electroconvulsometer with accessories

This instrument provides an alternating current stimulus of 50 cycles per second. The electronic timing circuit contained in the apparatus automatically passes stimulus current for a preset period, which may be varied from 0.1 to 1 second in steps of 0.1 second. The current, variable from 0.25 to 350 milliamperes (mA), is suitable for producing minimal and supramaximal seizures required in the assay of anticonvulsant drugs.

1 ml syringes, Measuring jars, Chemical weighing balance, Animal weighing balance, Stop watch, Animal cages

Methods

Maximal electroshock (MES) method, Pentylenetetrazole (PTZ) method

Preparation of solutions of standard and test drugs:

Phenytoin sodium:

The standard solution of phenytoin sodium was prepared by dissolving 400 mg of phenytoin sodium powder in 100 ml of distilled water at room temperature. The solution was freshly prepared every time. It was protected from light. This solution had a concentration of 4 mg/ml.

Sodium valproate:

The standard solution of sodium valproate was prepared by diluting 2g of sodium valproate in 100ml of distilled water at room temperature. The solution was freshly prepared everytime. This solution had a concentration of 20 mg/ml.

Pentylenetetrazole (PTZ):

The standard solution of pentylenetetrazole was prepared by dissolving 100 mg of pentylenetetrazole powder in 20 ml of distilled water at room temperature. This solution had a concentration of 5 mg/ml.

Aqueous extract of Hydrocotyle asiatica linn:

Preparation of aqueous extract of Hydrocotyle asiatica linn:

The dried powder of whole plant was obtained from head of the department of Rasayanashastra, Ayurvedic maha vidyalaya, Hubli. The aqueous extract was obtained by cold maceration method. 30 g of the powder was soaked in 200 ml of cold water for ~ 18 hr at room temperature. The extract was first filtered through Whatman no. 1 filter paper to clarify and then through a 0.45 µm membrane filter. The filtrate was evaporated to dryness at room temperature in a steady air current and the yield recorded as a percentage of the quantity of

initial plant material used[9, 10]. The test solution of Hydrocotyle asiatica linn was prepared by dissolving 2g of aqueous extract of Hydrocotyle asiatica linn in 100 ml of distilled water at room temperature. This solution had a concentration of 20 mg/ml.

Figure 1:Hydrocotyle asiatica

Figure 2: Hydrocotyle asiatica

Experimental methods

Male albino mice weighing 18 – 30 g are obtained from Central animal house, KIMS, Hubli. All test animals were allowed food and water ad libitum, both being withdrawn just prior to experimentation.[11] All the test animals which were tested for standard convulsive responses with MES and PTZ stimuli were subjected to further experiments of this study after 24 hours. The test animals were divided into 2 groups – one each for MES and PTZ method.

Maximal electroshock (MES) method:

The mice were subjected to maximal electroshock convulsions with a current of 50 mA for 0.2 second via ear electrodes. The electrodes were moistened with saline solution before application. The resultant seizure passes through various phases: phase of tonic limb flexion, tonic limb extension, clonus, post-ictal depression followed by recovery or death. [12] The mouse was considered as protected if the drug prevented the appearance of hindlimb tonic extensor component of the seizure [13]. The animals were divided into 4 groups, each consisting of 6 animals.

Figure 3: Electroconvulsometer

Figure 4: Tonic hind limb extension in mice

Control group-1 (C1)

In group C1 for MES method, mice were administered 0.25 ml of distilled water orally. After one hour[14, 15], they are subjected to maximal electroshock with an alternating current of intensity 50 mA for 0.2 second through ear clip electrodes. The duration of various parameters like tonic hind limb flexion, tonic hind limb extension, clonus, postictal depression and the incidence of recovery or death were noted.

Standard group-1 (S1)

In group S1, all mice received 50 mg/kg of phenytoin orally[16]. After one hour, they are subjected to maximal electroshock with an alternating current of intensity 50 mA for 0.2 second through ear clip electrodes. The results obtained were recorded in a similar way as for group C1.

Test group-1 (T1)

In group T1, all mice received 100 mg/kg of aqueous extract of Hydrocotyle asiatica linn orally[17]. After one hour, they are subjected to maximal electroshock with an alternating current of intensity 50 mA for 0.2 second through ear clip electrodes. The results obtained were recorded in a similar way as for group C1.

Test group-2 (T2)

In group T2, all mice received 300 mg/kg of aqueous extract of Hydrocotyle asiatica linn orally[18]. After one hour, they are subjected to maximal electroshock with an alternating current of intensity 50 mA for 0.2 second through ear clip electrodes. The results obtained were recorded in a similar way as for group C1. The observations are presented in tables 1-6.

Pentylenetetrazole (PTZ) method

In this method, the mice received 80 mg/kg of PTZ subcutaneously.[19] Only those animals that exhibited convulsive response in the form of clonus, tonic fore and hind limb flexion, tonic limb extension, post-ictal depression followed by recovery were used for experiment. In this method

abolition of tonic hind limb extension phase was considered as protection conferred by the drug. The mice were divided into 4 groups, each consisting of 6 animals.

Control group-2 (C2)

This is the control group for PTZ method. In group C2, all mice received 0.25 ml of distilled water orally. After one hour, PTZ (80 mg/kg) was administered subcutaneously. The duration of various phases of ensuing convulsions were noted and subsequent mortality recorded.

Standard group-2 (S2)

In group S2, all mice received 300 mg/kg of sodium valproate intraperitoneally[20]. After one hour, PTZ (80 mg/kg) was administered subcutaneously. Results were recorded in a similar way as for group C2.

Test group-3 (T3)

In group T3, all mice received 100 mg/kg of aqueous extract of Hydrocotyle asiatica linn orally. After one hour, PTZ (80 mg/kg) was administered subcutaneously. Results were recorded in a similar way as for group C2.

Test group-4 (T4)

In group T4, all mice received 300 mg/kg of aqueous extract of Hydrocotyle asiatica linn orally. After one hour, PTZ (80 mg/kg) was administered subcutaneously. Results were recorded in a similar way as for group C2.

Statistical analysis

The results of this study are expressed as mean± standard error of mean (mean± SE). Results are analyzed by student 't' test. Significance is established when probability value (p value) is less than 0.05. P values are denoted as * $P < 0.05$ as significant, ** $P < 0.01$ as highly significant and *** $P < 0.001$ as very highly significant.

RESULTS AND DISCUSSION

Table-1: Mean duration of various parameters of group C1 (Control, 0.25 ml of distilled water)

Parameters (Duration in seconds)	Serial no. of animals						
	1	**2**	**3**	**4**	**5**	**6**	**Mean**
Tonic hind limb flexion	1	4	3	3	4	2	2.83
Tonic hind limb extension	13	16	15	10	11	12	12.83
Clonus	16	14	20	13	16	14	15.5
Post-ictal depression	178	180	212	170	120	182	173.66
Recovery (R)/ Death (D)	R	R	R	R	R	R	

Table-2: Mean duration of various parameters of group S1 (Standard, 50 mg/kg of Phenytoin sodium)

Parameters (Duration in seconds)	Serial no. of animals						
	1	2	3	4	5	6	Mean
Tonic hind limb	-	-	-	-	-	-	
Tonic hind limb	-	-	-	-	-	-	
Clonus	16	14	18	14	13	14	14.83
Post-ictal depression	-	-	-	-	-	-	
Recovery (R)/	R	R	R	R	R	R	

Table-3 Mean duration of various parameters of group T1 (Test, 100 mg/kg of Aqueous extract of Hydrocotyle asiatica linn)

Parameters (Duration in seconds)	Serial no. of animals						
	1	2	3	4	5	6	Mean
Tonic hind limb flexion	2	2	-	2	-	-	1
Tonic hind limb extension	10	12	-	11	-	-	5.5
Clonus	-	18	19	20	17	16	15
Post-ictal depression	-	90	-	63	-	-	25.5
Recovery (R)/ Death (D)	D	R	R	R	R	R	

Table-4 Mean duration of various parameters of group T2 (Test, 300 mg/kg of Aqueous extract of Hydrocotyle asiatica linn)

Parameters (Duration in seconds)	GROUP- C1	GROUP- S1	GROUP- T1	GROUP- T2
Tonic hind limb flexion	2.83±0.5	0	1^{NS}	0.33^{NS}
Tonic hind limb extension	12.83±0.95	0	5.5±0.4***	1.66^{NS}
Clonus	15.5±1.04	14.83±0.75	$15±0.66^{NS}$	$15.66±0.57^{NS}$
Post-ictal depression	173.66±12.45	0	$25.5±7.95^{NS}$	15^{NS}

Table-5 Comparison of mean duration (in seconds) of different parameters in MES method (with control)

Parameters (Duration in seconds)	GROUP- C1	GROUP- S1	GROUP- T1	GROUP- T2
Tonic hind limb flexion	2.83±0.5	0***	1**	0.33**
Tonic hind limb extension	12.83±0.95	0***	5.5±0.4***	1.66***
Clonus	15.5±1.04	$14.83±0.75^{NS}$	$15±0.66^{NS}$	$15.66±0.57^{NS}$
Post-ictal	173.66±12.45	0***	25.5±7.95**	15***

Table-6 Comparison of mean duration (in seconds) of different parameters in MES method (with standard)

Parameters (Duration in seconds)	GROUP- C1	GROUP- S1	GROUP- T1	GROUP- T2
Tonic hind limb flexion	2.83±0.5	0	1^{NS}	0.33^{NS}
Tonic hind limb extension	12.83±0.95	0	5.5±0.4***	1.66^{NS}
Clonus	15.5±1.04	14.83±0.75	$15±0.66^{NS}$	$15.66±0.57^{NS}$
Post-ictal depression	173.66±12.45	0	$25.5±7.95^{NS}$	15^{NS}

Data expressed as mean ± SE
n = 6; *p < 0.05, **p<0.01, ***p<0.001 (compared with control), NS- not significant

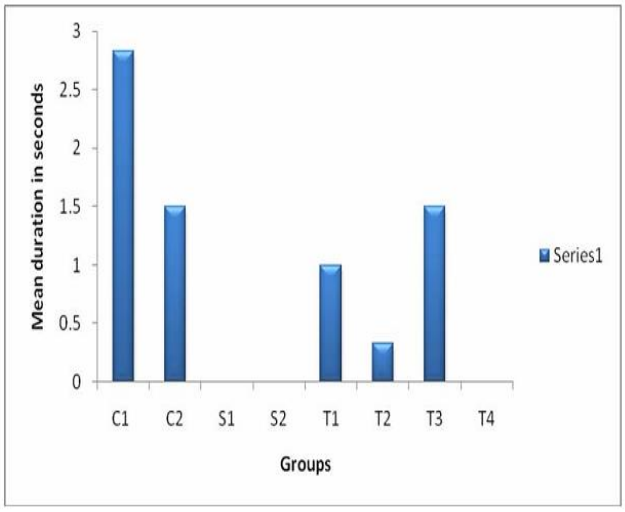

Figure 5: Mean durations of tonic hind limb flexion in MES and PTZ methods

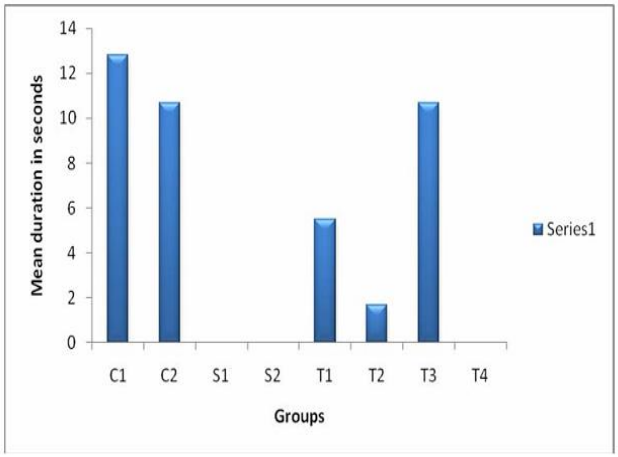

Figure 6: Mean durations of tonic hind limb extension in MES and PTZ methods

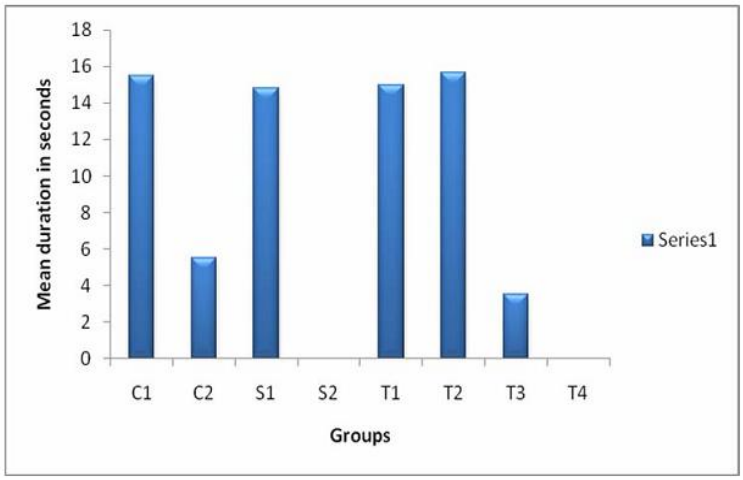

Figure 7: Mean duration of clonus in MES and PTZ methods

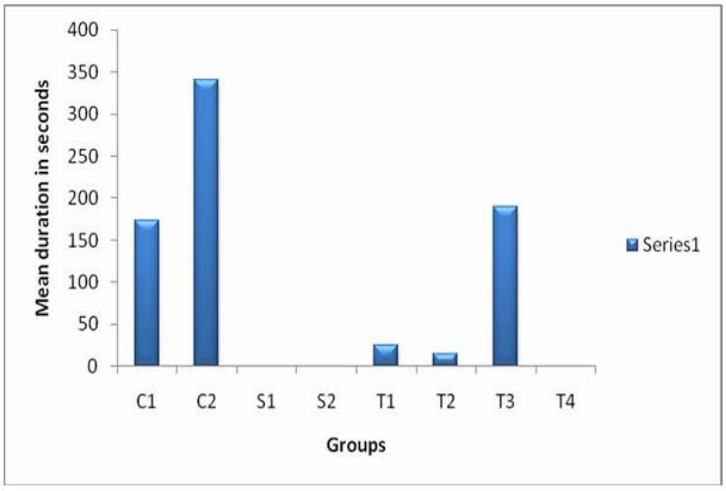

Figure 8: Mean duration of post ictal depression in MES and PTZ methods

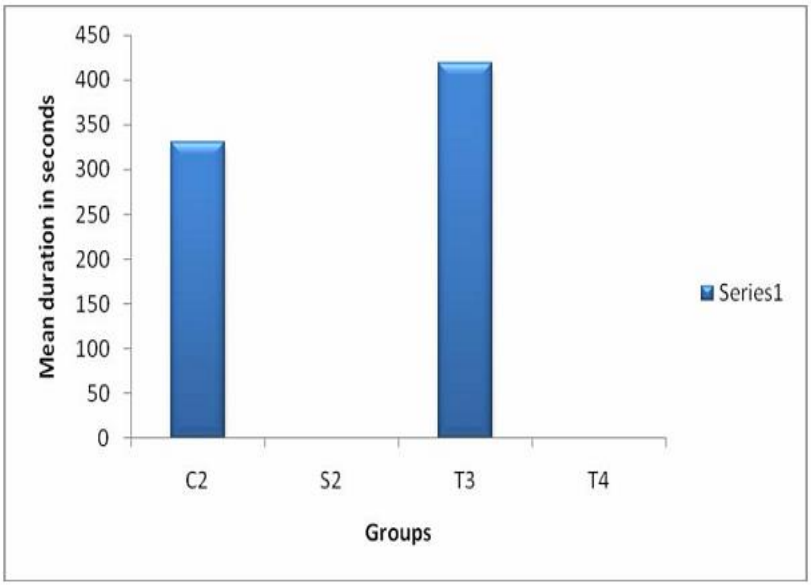

Figure 9: Mean duration of seizure latency in PTZ methods

Master chart showing variations in duration of different parameters in MES and PTZ methods

MES method										PTZ method									
Groups	Parameters	Serial number of animals								Groups	Parameters	Serial number of animals							
		1	2	3	4	5	6	Mean	SE			1	2	3	4	5	6	Mean	SE
C1	SL	-	-	-	-	-	-	-	-	C2	SL	360	320	290	353	342	319	330.66	10.8
	THLF	1	4	3	3	4	2	2.83	0.5		THLF	1	1	2	2	2	1	1.5	0.23
	THLE	13	16	15	10	11	12	12.83	0.95		THLE	11	12	10	13	9	10	10.66	0.61
	Cl	16	14	20	13	16	14	15.5	1.04		Cl	5	6	7	4	6	5	5.5	0.44
	PID	178	180	212	170	120	182	173.66	12.45		PID	337	350	343	351	328	332	340.16	3.78
S1	SL	-	-	-	-	-	-	-	-	S2	SL	-	-	-	-	-	-	-	-
	THLF	-	-	-	-	-	-	-	-		THLF	-	-	-	-	-	-	-	-
	THLE	-	-	-	-	-	-	-	-		THLE	-	-	-	-	-	-	-	-
	Cl	16	14	18	14	13	14	14.83	0.75		Cl	-	-	-	-	-	-	-	-
	PID	-	-	-	-	-	-	-	-		PID	-	-	-	-	-	-	-	-
T1	SL	-	-	-	-	-	-	-	-	T3	SL	410	460	412	400	417	415	419	8.73
	THLF	2	2	-	2	-	-	1	1		THLF	1	2	1	1	2	2	1.5	0.23
	THLE	10	12	-	11	-	-	5.5	0.4		THLE	10	11	9	11	11	12	10.66	0.43
	Cl	-	18	19	20	17	16	15	0.66		Cl	4	3	4	3	5	2	3.5	0.44
	PID	-	90	-	63	-	-	25.5	7.95		PID	285	-	287	-	281	282	189.16	1.15
	THLF	-	-	2	-	-	-	0.33	-		THLF	-	-	-	-	-	-	-	-
	THLE	-	-	10	-	-	-	1.66	-		THLE	-	-	-	-	-	-	-	-
	Cl	14	16	18	15	16	15	15.66	0.57		Cl	-	-	-	-	-	-	-	-
	PID	-	-	90	-	-	-	15	-		PID	-	-	-	-	-	-	-	-

In the present study, for the screening of aqueous extract of Hydrocotyle asiatica for anticonvulsant activity, two standard methods namely maximal electroshock (MES) and pentylenetetrazole (PTZ) methods have been used. Male albino mice weighing between 18-30 g were used as experimental animals. They were divided into two groups. One group of mice were subjected to maximal electroshock with an alternating current of 50 mA intensity for 0.2 seconds through ear clip electrodes. This immediately produced typical different phases of convulsions like tonic flexion of both fore and hind limbs, tonic extension of fore and hind limbs, clonus and post-ictal depression in 95% of animals. Only those animals showing above phases of convulsion were selected for this study. The parameters observed were the duration of tonic hind limb flexion, tonic hind limb extension, clonus, post-ictal depression and incidence of recovery and death. MES method has a high degree of predictivity for drugs useful in the management of tonic clonic seizures in man[9]. The second group of mice received an intraperitoneal injection of PTZ (80 mg/kg).[19] only those animals showing convulsive response were used for the experiment. The parameters studied were seizure latency, clonus, tonic hind limb flexion, tonic hind limb extension, post-ictal depression and recovery or death of the animal; duration of all parameters were recorded. In both MES and PTZ methods, the mouse was considered protected if the drug abolished the tonic hind limb extension.[11]

MES method

Tonic hind limb flexion:

A comparison of mean duration of tonic hind limb flexion of control group with other groups indicates that there is a decrease in mean time of tonic hind limb flexion in group T1 (100 mg/kg of aqueous extract of Hydrocotyle asiatica), group T2 (300 mg/kg of aqueous extract of Hydrocotyle asiatica) and S1 (Phenytoin 50 mg/kg). The test compound has shown statistically significant protection ($p < 0.01$) in both group T1 and T2. In group S1, there is complete abolition of flexor phase, which is statistically significant ($p < 0.001$). A comparison of mean duration of tonic hind limb flexion of test groups (T1 and T2) with standard (S1) indicates that there is no significant difference between S1, T1 and T2. Although the criterion of abolition of only tonic extensor phase has been recommended as a sufficient protective end point[49].

Tonic hind limb extension

A comparison of mean duration of tonic hind limb extension of control group with test groups indicate that there is a decrease in mean duration of tonic hind limb extension in both groups T1 and T2 and it is statistically significant ($p < 0.001$). In group S1, there is

complete abolition of tonic hind limb extension which is statistically significant ($p < 0.001$). A comparison of test groups (T1 and T2) with group S1 indicate that there is significant difference between S1 and T1 ($p < 0.001$) while no significant difference between S1 and T2. The abolition of tonic hind limb extension has occurred in 3 out of 6 mice in T1 and 5 out of 6 mice in T2. Since abolition of tonic hind limb extension is considered suggestive of protection against MES convulsions, the aqueous extract of Hydrocotyle asiatica has anticonvulsant effect against MES convulsions at a dose of 300 mg/kg. This effect is comparable to that of phenytoin in this study.

Clonus

Analysis of results when compared with control suggest that there is a decrease in mean duration of clonus in groups S1 and T1 while a slight increase in group T2. However these values are not statistically significant. Analysis of results when compared with standard suggests that there is no significant difference between groups S1 and test groups T1 and T2.

Post-ictal depression

A comparison of mean duration of post-ictal depression with control (Table 5, figure 8) indicates that there is a decrease in the mean duration in groups T1 and T2 which is statistically significant ($p < 0.001$). Group S1 has shown no post-ictal depression at all which is statistically significant ($p < 0.001$). A comparison of mean duration of post-ictal depression with standard (Table 6, figure 8) indicates that there is no significant difference between the groups S1, T1 and T2.

PTZ method

Seizure latency

Analysis of results compared with control group suggest that there is an increase in the mean duration of seizure latency in group T3 (100 mg/kg of aqueous extract of Hydrocotyle asiatica) and it is statistically significant ($p < 0.001$). In groups S2 (sodium valproate 300mg/kg) and T4 (300 mg/kg of aqueous extract of Hydrocotyle asiatica), there is no seizure and hence no seizure latency and it is statistically significant ($p < 0.001$). Analysis of results compared with standard group suggest that there is statistically significant difference in group T3 ($p < 0.001$) while no significant difference in group T4.

Tonic hind limb flexion

Comparison of mean duration of tonic hind limb flexion with control group indicates that there is no significant difference between control group and group T3 while in groups S2 and T4, there is abolition of tonic hind limb flexion which is statistically significant ($p < 0.001$).

Comparison of mean duration of tonic hind limb flexion of standard group with test groups T3 and T4 indicates that there is significant difference between group S2 and group T3 ($p < 0.01$), while there is no significant difference between group S2 and group T4.

Tonic hind limb extension

Comparison of mean duration of tonic hind limb extension of control group with test groups and standard indicates that there is no significant difference between group C2 and group T3 while in groups S2 and T4, there is abolition of tonic extensor phase which is statistically significant ($p < 0.001$). Comparison of mean duration of tonic hind limb extension of standard group S2 with test groups T3 and T4 indicates that there is significant difference between group S2 and group T3 ($p < 0.001$), while no significant difference between group S2 and group T4. The abolition of tonic hind limb extension has occurred in all mice in group T4 while in group T3 there is no abolition of tonic hind limb extension. Since abolition of tonic hind limb extension is considered suggestive of protection against MES convulsions, the aqueous extract of Hydrocotyle asiatica has anticonvulsant effect against PTZ convulsions at a dose of 300 mg/kg. This effect is comparable to that of sodium valproate in this study.

Clonus

Comparison of mean duration of clonus of control group with test and standard groups indicates that there is a decrease in the mean duration of clonus in group T3 and this is statistically significant ($p < 0.01$) while there is no clonic phase in groups S1 and T4 which is statistically significant ($p < 0.001$).

Post-ictal depression

Comparison of mean duration of post-ictal depression of control group C2 with test and standard groups indicates that there is decrease in the mean duration of post-ictal depression in group T3 ($p < 0.001$) while there is no post-ictal depression in groups S2 and T4 which is statistically significant ($p < 0.001$). Comparison of mean duration of post-ictal depression of standard group S2 with test groups indicates that there is significant difference between groups S2 and T3 ($p < 0.001$), while no significant difference between S2 and T4. The most significant effect of phenytoin is its ability to modify the pattern of MES. By contrast, phenytoin does not inhibit clonic seizures evoked by pentylenetetrazole.[2] Like phenytoin, valproate inhibits tonic hindlimb extension in MES and kindled seizures at nontoxic doses. Valproic acid at subtoxic doses inhibits clonic motor seizures induced by PTZ. Its efficacy in diverse models parallels its efficacy against absence as well as partial and generalized tonic clonic seizures in humans[2]. It is apparent from the results that the test compound, aqueous extract of Hydrocotyle

asiatica at a dose of 300 mg/kg, has shown similarity to valproic acid in this experimental study. Like valproic acid, it has protected the experimental animals against both MES and PTZ convulsions. It has abolished the tonic hind limb flexion and extension in all but one mouse subjected to MES and in all mice subjected to PTZ convulsions. *In vitro*, valproate can stimulate the activity of the GABA synthetic enzyme, glutamic acid decarboxylase, and inhibit GABA degradative enzymes, GABA transaminase and succinic semialdehyde dehydrogenase.[2] Hence it may be proposed that the mode of action of aqueous extract of Hydrocotyle asiatica may involve blockade of voltage gated sodium channel, calcium channel and GABA metabolism. Hydrocotyle asiatica has recently been shown to have an anti-lipid peroxidative and antiepileptic activity in the lithium pilocarpine model of status epilepticus. In a recent study it has been found that hydrocotyle asiatica causes perceptible changes in the cholinergic system as one of the facets of its anticonvulsant activity.[15] Centella asiatica significantly prevented the cognitive impairment and attenuated the oxidative stress induced by PTZ kindling.[20]

CONCLUSION

The test compound at a dose of 100 mg/kg has abolished tonic hind limb extension in 3 out of 6 animals in MES while no abolition in PTZ convulsions. Hence this dose is not adequate to prevent experimentally induced MES and PTZ convulsions. At a dose of 300 mg/kg body weight, the aqueous extract of Hydrocotyle asiatica has shown statistically significant anticonvulsant effect against both MES and PTZ convulsions. Thus the aqueous extract of Hydrocotyle asiatica has shown efficacy in both MES and PTZ convulsions in mice in the present study. Since the clinical correlates of MES seizures are tonic clonic convulsions and correlates of PTZ seizures are absence seizures, the aqueous extract of Hydrocotyle asiatica is likely to be useful in the treatment of tonic clonic and absence seizures. Further detailed study of the active principle/s of this plant is worth pursuing in this regard.

REFERENCES

1. Trescher WH, Lesser RP: The Epilepsies. In: Bradley WG, Daroff RB, Fenichel GM, Marsden CD (eds.). Neurology in clinical practice; vol 2; 5th edition; Massachusetts, Butterworth-Heinemann; 2008: 1909- 1946.
2. McNamara JO. Pharmacotherapy of the epilepsies. In: Brunton LL, Lazo JS, Parker KL. Goodman & Gilman's The Pharmacological Basis Of Therapeutics; 11th edition; New York, Mc Graw Hill Companies Inc; 2006; 501-525.

3. Lowenstein DH. Seizures and epilepsy. In: Fauci, Braunwald , Kasper, Hauser, Longo, Jameson, Loscalzo; Harrison's Principles Of Internal Medicine; vol 2; 17th edition; New York, Mc Graw Hill companies Inc, 2008: 2498- 2512.
4. Bennett PN, Brown MJ. Epilepsy, parkinsonism and allied conditions. In: Clinical pharmacology; 10th edition; Philadelphia, Churchill Livingstone; 2008: 372-388.
5. Rang HP, Dale MM, Ritter JM, Flower RJ. Antiepileptic drugs. In: Rang and Dale's pharmacology; 6th edition; Philadelphia, Churchill Livingstone; 2007: 575- 587.
6. Ropper AH, Brown RH. Epilepsy and other seizure disorders. In: Adams and Victor's Principles of neurology; 8th edition; New York, McGraw Hill Companies Inc; 2005: 271-301.
7. Nadkarni A K. Hydrocotyla asiatica Linn. In: Dr. K. M. Nadkarni's Indian Materia Medica; vol 1; 3rd edition; Bombay, Popular Book Depot, 1954: 663-664.
8. WHO monographs on selected medicinal plants; vol 1; Geneva,WHO;1999: 77-85.
9. Satoskar RS, Bhandarkar SD, Rege NN. Drugs effective in seizure disorders. In: Pharmacology and pharmacotherapeutics, 20th edition; Mumbai, Popular prakashan, 2007: 122-140.
10. DeLorenzo RJ. The Epilepsies. In: Bradley WG, Daroff RB, Fenichel GM, Marsden CD. Neurology in clinical practice; Vol 2; 1st edition; Massachusetts, Butterworth-Heinemann; 1991: 1443-1477.
11. Foldvary N, Wyllie E. Epilepsy. In: Goetz CG, Pappert EJ. Textbook of clinical neurology; 1st edition; Pennsylvania, W.B.Saunders company; 1999: 1059-1088.
12. Sidiropoulou K, Diamantis A, Magiorkinis E. Hallmarks in the 18th and 19th century epilepsy research." Epilepsy and behavior, 2010; 18: 151-161.
13. Sharpless SK. Hypnotics and sedatives. In: Goodman LS, Gilman A. The pharmacological basis of therapeutics; 4th edition; New York, The Macmillan Company; 1970: 98-120.
14. Toman JEP.Drugs effective in convulsive disorders. In: Goodman LS, Gilman A. The pharmacological basis of therapeutics; 4th edition; New York, The Macmillan Company; 1970: 204-225.
15. Aminoff MJ. Epilepsy. In: McPhee SJ, Papadakis MA. Current medical diagnosis and treatment; 49th edition; New York, McGraw hill companies; 2010: 878-884.
16. Edwards CRW, Bouchier IAD, Haslett C, Chilvers ER. Epilepsy. In: Davidson's Principles and Practice of Medicine; 17th edition; New York, Churchill Livingstone;

1995: 1064-1070.

17. Perkin GD. Epilepsy in later childhood & adults. In: Warrell DA, Cox TM, Firth TD, Benz EJ, Jr. Oxford textbook of medicine; vol 3; 4th edition; New York, Oxford university press; 2003: 1001-1014.
18. Murthy JMK. Seizure disorders. In: Shah SN et al. API textbook of medicine; 8th edition; Mumbai, The Association of Physicians of India; 2008: 1135-1150.
19. Kulkarni C, David J, Joseph T. Involvement of adenosine in postictal events in rats given electroshock. Indian J Physiol Pharmacol; 1993; 38(1): 39-42.
20. During MJ, Spencer DD. Adenosine: A potential mediator of seizure arrest and postictal refractoriness. Ann Neurol; 1992; 32: 618-624