Journal of Pharma Research
Available online through
www.jprinfo.com

Research Article
ISSN: 2319-5622

Evaluation of Antiepileptic and Nootropic Activity of *Momordica Sahyadrica*

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Received on: 31-10-2014; Revised and Accepted on: 17-11-2014

ABSTRACT

Epilepsy is a common and diverse set of chronic neurological disorders characterized by seizures. Epilepsy results from abnormal, excessive (or) hyper synchronous neuronal activity in brain. Epilepsy is usually controlled but cannot be cured with medication. Despite the use of available antiepileptic drugs, many patients with epilepsy fail to experience seizure control. Moreover, many patients suffer with the strong side effects including decreased cognitive abilities and memory loss either temporary or permanent. So there is a strong need for the development of new and improved therapies for Anti Epileptics and cognitive abilities. The present report is an investigation of Anti Epileptic and Nootropic activity of Momordica Sahyadrica.The hydroalchoholic extract of Momordica Sahyadrica was subjected to Phytochemical screening to find out the presence of alkaloids, flavonoids, tannins, proteins, phenols and absence of carbohydrates and glycosides.,acute toxicity study as per OECD guideline No. 423 to find its Lethal dose and then screened for antiepileptic activity on ISONIAZD induced seizures and Pentylenetetrazole induced seizures models and Nootropic activity on Scopolamine induced dementia using Elevated Plus Maze model in wistar rats. Acute toxicity of extract was non toxic up to the recommended dose 2000mg/kg body weight orally as per OECD guidelines No.423. The study reported that MSE has shown significant delay in seizures induced by PTZ and ISONIAZD models and has shown comparative results with standard drugs. Similarly Wistar rats with Scopolamine induced dementia were tested with Elevated Plus Maze model for Nootropic activity.MSE dose dependently has improved memory significantly.

Keywords: *Anti Epileptic activity, Nootropic activity, Momordica Sahyadrica, Elevated Plus Maze, ISONIAZD, Pentylenetetrazole.*

INTRODUCTION

1. Epilepsy:

Epilepsy is a chronic disorder characterized by recurrent seizures, which may vary from a brief lapse of attention or muscle jerks, to severe and prolonged convulsions [1]. The seizures are caused by sudden, usually brief, excessive electrical discharges in a group of brain cells (neurons).

According to latest study by WHO, there are over 50 million sufferers in the world today, of which 85% lie in developing countries. In India, studies have reported the prevalence rate of epilepsy varying from 1710 to 9780 cases per million populations [2].

The cause of epilepsy is unknown in over half of afflicted patients, and the underlying mechanisms of the development of epilepsy,a process referred to as epileptogenesis,have yet to be clearly identified. However, the generation of seizure disorders in humans has been connected to various phenomena.

Table No. 1: General causes of seizures

Neonates	**1. Perinatal Hypoxia, Ischemia 2.Intracranial Hemorrhage 3.Metabolic Disturbances**
Infants & Children (Upto 12 Yrs)	1.Febrile Seizures 2.Genetic Disorders
Adolescents (12-18 Yrs)	1.Trauma 2. Genetic 3. Infection
Young Adults (18-35 Yrs)	1.Trauma 2. Alcohol Withdrawal
Older Adults (>35 yrs)	1. CVS Disease 2. Tumour

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Throughout the years, antiepileptic drug discovery has progressed from centuries of ignorance and ineffective treatments, to a period of discovery by serendipity and random screening, and finally, to the modern era of rational drug design.

2. Cognition:

Cognition in a broad sense means processing of information. It denotes a relatively high level of processing of specific information including thinking, memory, perception, motivation, skilled movements and language. Learning and memory both can be conceived as a psychological process, as well as change in synaptic neural connectivity [3]. Learning is defined as the ability to alter behavior on the basis of experience [4]. Cognitive deficits have long been recognized as severe and consistent neurological disorders associated with numerous psychiatric and neurodegenerative diseases, such as Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's disease (HD), Down's syndrome. Pick's disease, trauma, chronic insomnia and Attention deficit disorders [3].

3. Dementia:

Dementia is a syndrome of failing memory and other intellectual functions with little or no disturbance in consciousness.

Dementia is the most common age related neurological illness. Although it affects only 1% of the geriatrics, the incidence of dementia doubles with every five years of age, so that 30 to 40% of the individuals are affected by it at the age of 85 [5].

Memory disorders are becoming more prevalent due to various factors such as natural (ageing, physical and mental stress), environmental (excess levels of carbon monoxide, carbon dioxide, methyl mercury in atmosphere, aluminum in foods), iatrogenic (electroconvulsive shock therapy and use of certain CNS depressants) [6].

4. Nootropics:

Nootropics are referred to as “smart drugs”, ”memory enhancers”, ”cognitive enhancers”, “smart nutrients”.Nootropics represent a new class of psychotropic agents with selective facilitatory effect on integrative functions of the central nervous system, particularly on intellectual performance, learning capacity

and memory [7]. Nootropic substances include drugs, nutrients and herb with cognitive enhancing effects.

Ayurveda, an ancient Indian system of medicine had reported various plants with the caliber of Neuroprotective activity and the plant *Momrdica sahyadrica* is one as such recommended as a Neuroprotective, anti-convulsant agent and memory enhancing agent for its nootropic activity.

MATERIALS AND METHODS

1. Plant Material:

The plant material consists of dried powdered fruits of *Momordica Sahyadrica*, belongs to the family Cucurbitaceae.

2. Plant Collection And Authentication Certificate:

The fruits of *Momordica Sahyadrica* were collected from Western Ghats, Kerala, India during the month of November 2013. The plant was identified and authenticated by V.Chelladurai, Research Officer-Botony,Central Council for Research in Ayurveda & Siddha, Govt. of India

3. Drugs:

Momordica sahyadrica, PTZ, Isoniazide,Diazepam Phynetoin,Mentat, Scopalamine.

4. Extraction:

The dried fruits of *Momordica Sahyadrica* plant are coarsely powdered and are successively extracted by continuous hot percolation method using Soxhlet apparatus. The solvent used was a mixture of ethanol:water in the ratio of 7:3. About 100gm of powder was extracted with 300ml of solvent.Extraction process was repeated thrice to obtain the required amount of drug. Hydro alchoholic extraction yielded sufficiently good quantity of the product. The extract was concentrated to dryness under controlled temperature between 50-60^{o}C and stored in a dessicator for further use. The extract was tested for Phytochemical and Pharmacological screening.

% yield =(weight of extract / weight of dried fruits powder) * 100.

Table No. 2: % of Yield

Solvent	Weight in gm		% of Yield
	Momordica Sahyadrica Fruit powder	Extract	
Ethanol + Water(7:3)	300	10.08	3.36

Fig. 1: Soxhlet apparatus

5. Phytochemical Screening:

5.1. Alkaloid Test:

Dragendorff's Test: To 1 ml of the extract, add 1 ml Dragendorff's reagent, and orange red precipitate indicates the presence of alkaloids.

Glycoside Test:

Baljet Test: To 1 ml of the test extract add 1 ml sodium picrate solution and the yellow to orange colour reveals the presence of glycosides.

5.2. Flavonoid Test:

To 1 ml of the extract, add 1 ml ferric chloride. The formation of brown colour confirms the presence of flavonoids.

5.3. Tannins & Phenolic Test:

To 1 ml of extract, add few ml of gelatin solution, a white precipitate reveals the presence of tannins and phenolic compounds.

5.4. Proteins &Aminoacids Test:

Ninhydrin Test: Add two drops of freshly prepared 0.2 percent Ninhydrin reagent to the extract solution and heat. Development of a blue colour reveals the presence of proteins and amino acids.

5.5. Carbohydraes:

Benedict's Test: to 5ml of Benedict's reagent, add 1 ml of extract solution and boil for 2 minutes and cool. Formation of a red precipitate shows the presence of carbohydrates.

6 Acute Toxicity Study:

Four groups of male wistar rats were taken, of which one of the groups is considered as control and given normal saline. The remaining three groups were administered with the hydroalcoholic fruit extract of *Momordica Sahyadrica* in graded doses of 50 mg/kg, 300 mg/kg and 2000 mg/kg *p.o.,* respectively as per OECD guidelines 423. The animals were kept under observation for change in behavior or death up to 14 days following the plant extract administration.

7. Pharmacological Screening of Antiepileptic Activity:

7.1. Isoniazid induced convulsions:

Principle: Isoniazid is known to produce convulsions by inhibiting the GABA synthesis.By comparing the results of MSE with standard AED Diazepam we can evaluate its Antiepileptic properties.

Procedure: Adult male Wistar rats (180-250g) will be divide into four groups each containing six animals.

Group I has been administered with vehicle 1% CMC solution and served as control.

Group II has been administered with Diazepam (2mg/kg B.W) alone and served as standard control.

Group III and IV has been administered with MSE at low and high doses resp.

Anticonvulsant activity of MSE was evaluated using Isoniazid induced convulsions model in rats.

MSE was administered in varying doses (200, 400 mg/kg B.W) and diazepam (2mg/kg B.W) was administered IP before 30 min the oral dosing of INH(300 mg/kg BW) to induce clonic seizures and mice were observed for onset of convulsions. Each group undergoes pre-treatment for 7 days. Report will be noted by the observation of behavioral studies. The time of onset of seizures was recorded in both protected and non-protected rats. The group which received only vehicle was considered as control [8].

7.2. PTZ induced Convulsion:

Principle: Pentylenetetrazole is a CNS Stimulant. It Produces jerky type of clonic convulsion in rats and mice similar to petit mal type of

convulsion in man.It causes direct depolarization of central neurons.PTZ also interfere with GABA-ergic inhibition to cause seizures.By giving MSE to one group and standard AED Phynetoin to other group we will compare the results.

Procedure: Adult male Wistar rats (180-250g) will be divide into four groups each containing six animals.

Group I has been administered with vehicle 1% CMC solution and served as control.

Group II has been administered with Phynetoin (80 mg/kg B.W) alone and served as standard control.

Group III and IV has been administered with MSE at low and high doses resp.

Anticonvulsant activity of MSE was evaluated using PTZ induced convulsions model in rats.

MSE was administered IP in varying doses (200, 400mg/kg BW) 30 min before the i.p. injection of PTZ (80 mg/kg) and rats were observed for onset of convulsions[9].One group received vehicle while other group received phenytoin (25 mg/kg, IP) as a reference standard. Each group undergoes pre-treatment for 7 days. Report will be noted by the observation of behavioral studies. The animals were observed for onset of convulsion and HLTE duration after PTZ administration.

8. Pharmacological Screening Nootropic Activity:

Principle: On day 1, Wistar rats were trained to move from open arms to closed arms of the Elevated Plus Maze apparatus and its memory of acquired training was tested and compared with induction of MSE and nootropic Mentat for 7 days.

Procedure: Adult male Wistar rats (180-250g) will be divide into five groups each containing four animals.

Group I has been administered with vehicle 1% CMC solution and served as control.

Group II has been administered with Mentat (3mg/kg B.W) alone and served as standard control.

Group III has been administered with Scopolamine (0.3 mg/kg B.W) alone and served as Negative control without any drug treatment.

Group IV has been administered with MSE (400mg/kg BW) along with scopolamine.

Group V has been administered with Mentat (3mg/kg B.W) along with scopolamine.

The maze was elevated to a height of 25cm, the animals were individually placed at the end of either of the open arms and the time taken for the animal to move from open to closed arm (Transfer latency, TL) was taken as the criterion of task. The animals were allowed to explore the apparatus for 30 seconds.

After 24 hours of first exposure, TL was again noted on day 1 of the study for determining the acquisition. Five minutes later the animals of Group 3, 4 and 5 received Scopolamine (0.3mg/kg body weight) and then were dosed with respective drug and returned to their home cage.

The animals were subjected to induction followed by drug treatment for up to day 7 and on 7th day the animals were subjected to the retention test 25min. after the last dose, for evaluating the transfer latency keeping the time period of 60 seconds as cut off criterion [10,11].

Fig. 2: Elevated Plus Maze Test setup

RESULTS

1. % Yield of MSE:

The dried fruits of *Momodrica sahyadrica* were extracted with ethanol & water (Hydro-alcohol) by hot percolation method. The % yield of *Mamordica Sahyadrica* fruit extract was found to be 3.36%.

2. Preliminary Phytochemical investigations:

The active constituents viz; Alkaloid, Glycoside, Flavonoids, Tannins, Phenolics, Carbohydrates, Proteins and Amino acids were estimated from the hydro alcoholic extract of *Mamordica Sahyadrica*, and the results of the active constituents present are depicted in **Table 3**.

Table No. 3: Qualitative Phytochemical investigation of MSE

Sl. No.	TEST	RESULT
1.	Alkaloid	+
2.	Glycoside	-
3.	Flavonoids	+
4.	Tannins	+
5.	Phenolic compounds	+
6.	Proteins &aminoacids	+
7.	Carbohydrates	-

"+" represents Present; "-" represents Absent

Table No. 4: Acute toxicity studies

Sl. No	Treat-ment	Dose (mg/Kg)	Weight of the animal (Day 1)	Weight of the animal (Day 14)	Signs of toxic-ity	Onset of toxic-ity	Reve-rsible or Irrev-ersible	Dur-ation
1	*MSE*	50	150 gm	158 gm	NIL	NIL	NIL	14 Days
2	*MSE*	50	150 gm	152 gm	NIL	NIL	NIL	14 Days
3	*MSE*	50	150 gm	154 gm	NIL	NIL	NIL	14 Days
4	*MSE*	300	150 gm	153 gm	NIL	NIL	NIL	14 Days
5	*MSE*	300	150 gm	151 gm	NIL	NIL	NIL	14 Days
6	*MSE*	300	150 gm	155 gm	NIL	NIL	NIL	14 Days

7	*MSE*	2000	140 gm	143 gm	NIL	NIL	NIL	14 Days
8	*MSE*	2000	140 gm	146 gm	NIL	NIL	NIL	14 Days
9	*MSE*	2000	140 gm	141 gm	NIL	NIL	NIL	14 Days

3. Acute Toxicity Studies:

In toxicity study three groups of male wistar rats were administered with the hydro-alcoholic extract of *Momodrica sahyadrica* (MSE) fruit in graded doses of 50 mg/kg, 300 mg/kg and 2000 mg/kg *p.o.,* respectively. The animals were kept under observation for change in behavior or death up to 14 days following the plant extract administration. It was observed that the test extract was not lethal to the rats even at the 2000 mg/kg doses. Hence, 1/10th (200mg/kg) and 1/5th (400mg/kg) of this dose were selected for further study.

4. Evaluation Of Antiepileptic Activity:

Antiepileptic activity of MSE was evaluated using Isoniazid induced convulsions and Pentylenetetrazole induced convulsion model in rats.

4.1. Isoniazid induced convulsions:

Isoniazid induced convulsions in animals was significantly minimized by MSE dose dependently as shown in **Table 5**.

Table No. 5: Effect of pre-treated MSE on ISONIAZID induced convulsions in Rats

Group	Treatment	Onset of convulsions (min)	No. of animals showing convulsions
Normal control	CMC suspension	43.17±1.78	06
Standard Control	Diazepam	93.06±2.72***	03
Test group (Dose 1)	MSE (200mg/kg)	64.19±4.87**	06
Test group (Dose 2)	MSE (400mg/kg)	79.25±5.23***	04

Values represented in (Mean ± SD, n=6), ns Non Significant, ** p <0.001, ***p<0.0001

Values are expressed as Mean ± SD of animals. Statistical significance test for comparision were done by ANOVA, followed by Dunnet's multiple comparision test.

Comparisons were done between: Group I vs Group II, III, IV :

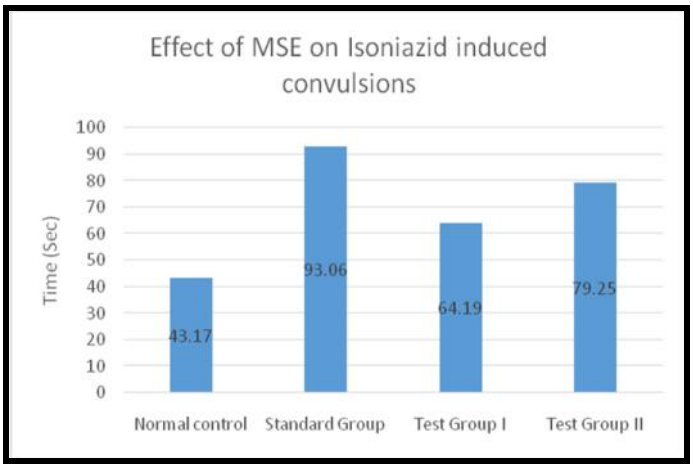

Fig. 3: Effect of MSE on ISONIAZID induced convulsions

Evaluation:

Pre-treatment of *Momordica Sahyadrica* fruit extract on groups III and IV animals has significantly and comparably delayed the ISONIAZD induced convulsions as of group II animals treated with standard anti epileptic drug.

4.2. PTZ induced convulsions:

PTZ induced convulsions in animals was significantly minimized by MSE dose dependently as shown in **Table 6**.

Table No. 6: Effect of pre-treated MSE on Pentylenetetrazole Induced Convulsions in Rats

Sl. No.	Group	Dose (mg/kg)	Onset of time (sec)	Duration of Hind Limb Tonic Extention phase (sec)
1.	Normal control	10	60.19 ± 1.85	38.18 ± 2.43
2.	Standard Control	25	0	0
3.	Test group (Dose 1)	200	64.36 ± 1.5***	32.1 ± 1.36***
4.	Test group (Dose 2)	400	71.14 ± 2.32***	23.18 ± 1.64***

Values represented in (Mean ± SD, n=6), ns Non Significant, ** p <0.001, ***p<0.0001

Values are expressed as Mean ± SD of animals. Statistical significance test for comparision were done by ANOVA, followed by Dunnet's multiple comparision test.

Comparisons were done between: Group I vs Group III, IV:

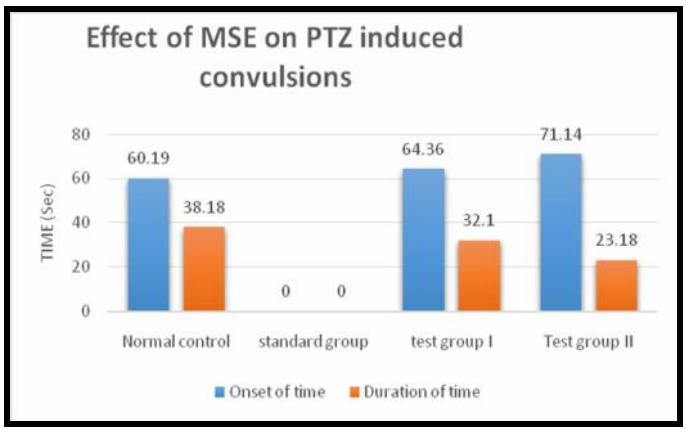

Fig. 4: Effect of MSE on PTZ induced convulsions

Evaluation:

Pre-treatment of *Momordica Sahyadrica* fruit extract on groups III and IV animals has significantly delayed and decreased the PTZ induced convulsions when compared to group I animals.

Phynetoin,an anti epileptic drug given to group II animals has completely controlled the convulsions and so the group II animals has not showed any significant convulsions.

5. Investigation Of Nootropic activity:

Nootropic activity of MSE was evaluated using Transfer latency model in rats.

Table No. 7: Effect of MSE on Scopolamine Induced Dementia in Rats

Sl. No	Group	Sub group	Transfer Latency	
			Day 1	Day 7
1.	Normal Control	CMC Suspension	28.59 ± 3.27	08.89 ± 3.58
2.	Positive Control	Mentat (3mg/kg)	27.85 ± 4.86	19.79 ± 2.14
3.	Negative Control	Scopolamine Induced (0.3mg/kg)	23.62 ± 1.99	52.29 ± 8.6**
4.	Treatment group I	Scopolamine + *Momordica Sahyadrica* (400mg/ kg)	28.97 ± 5.61	17.22 ± 2.26**
5.	Treatment group II	Scopolamine+ Mentat	29.06 ± 2.48	13.43 ± 1.76**

Values represented in (Mean ± SD, n=4), ns Non Significant, *P<0.01, ** p <0.001

Values are expressed as Mean ± SD of animals. Statistical significance test for comparision were done by ANOVA, followed by Dunnet's multiple comparision test.

Comparisons were done between: a) Group I vs Group III; b) Group III vs Group IV, V:

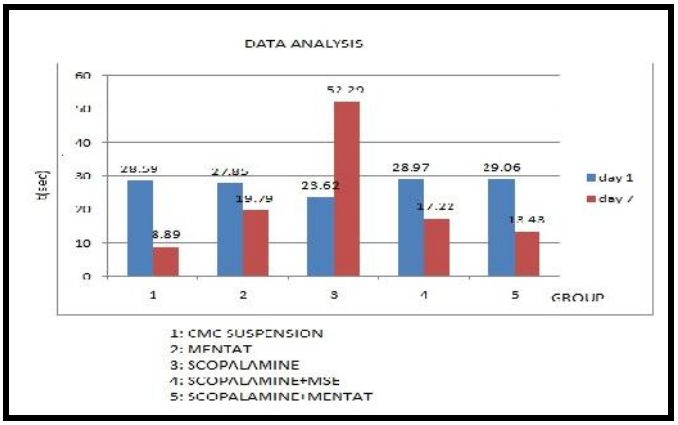

Fig. 5: Effect of MSE on Scopolamine Induced Dementia in Rats

Transfer Latency time in rats with scopolamine induced dementia has significantly decreased with treatment of *Momordica Sahyadrica* fruit extract and showed comparative results with that of standard nootropic, mentat.

DISCUSSION

In this present study, on the basis of results tabulated in **Table 5**, it is found that treatment with MSE on rats significantly delayed the onset of clonic convulsion in ISONIAZID induced epilepsy. Currently used standard anticonvulsant drug diazepam effective in therapy of generalized tonic-clonic seizures, have shown strong anticonvulsant action in ISONIAZID induced convulsions test. Since, MSE significantly delayed generalized tonic-clonic seizures in ISONIAZID induced convulsions in comparative to standard drug Diazepam; it suggests the presence of anticonvulsant compounds.

Similarly, on the basis of results tabulated in **Table 6**, we found that treatment with MSE on PTZ induced rats significantly reduce the duration of convulsion and delayed the onset of clonic convulsion. PTZ may cause seizures by inhibiting chloride ion channel associated with $GABA_A$ receptors. Currently used standard anticonvulsant drug diazepam effective in therapy of generalized tonic-clonic seizures, have shown strong anticonvulsant action in PTZ induced convulsions test. Since, MSE significantly delayed generalized tonic-clonic seizures in PTZ induced convulsions in comparative to standard drug Phynetoin; it suggests the antiepileptic efficacy against the above mentioned seizures type in man.

The test results tabulated in **Table 7**, stated that MSE in rats pretreated with scopolamine,has significantly decreased the transfer latency time.This suggests that like mentat ,MSE also enhance cognition and memory.

CONCLUSION

On the basis of results tabulated in **Tables 5, 6 & 7**, it may be concluded that fruit extract of *Momordica Sahyadrica* shows anticonvulsant potential and also facilitated learning and memory by reversing scopolamine induced dementia.

However, further research is necessary to determine the components involved and their mechanism of action in bringing about the desirable pharmacological effect.

ACKNOWLEDGEMENTS

We wish to thank Dr. Vijaya Kuchana, Principal, TKR College of Pharmacy for her valuable suggestions throughout the course of my research..

REFERENCES:

1. Fisher R.S, Van Emde Boas W, Blume W, Elger C, Genton P, Lee P, *Engel. J. Epilepsia,* **2005**; 46(4): pp 470-472.
2. Gupta YK, Malhotra. J. *Indian J. Physiol. Pharmacol*., **2000**; pp 44-48.
3. Reddy D S. *Indian J. Pharmacol*., **1997**; 29(4): pp 208-221.
4. Jain A K. Textbook of Physiology.2nd ed., Avichal Publishing Company, Sirmour (HP): **2007**; p. 1058-1073.
5. Ellen Y S, Kathryn M U. *Am. J. Health Syst. Pharm.,* **1997**; 54(1): pp 2805-10.
6. Annapurna A, Ramakrishna V, Murali K K, Krishnakumar V, Prasad K B. *Indian J. Pharm. Sci*., **2004**; 66(1): pp 31-35.
7. Rodrigues V, Rao MS, Karnath S, Rao GM. *Ind. J. Pharmacol.*, **1999**: 31(1);pp 69.
8. Bhattacharya S K, Seth S D. Text Book of Pharmacology. 2nd Edition, Elsevier, New Delhi: **1999**; p. 496-499.
9. Trivedi J K. *Indian J. Psychiatry,* **2006**; 48(1): 10-20.
10. Lakshmikantha Y, Padmalatha Rai, Ashajyothi Chavan. *IJPCBS*, **2012**; 2(4): pp 538-544.
11. Kulkarni SK, Verma A. *Indian Drugs*, **1993**; 30(3): pp 97-107.

How to cite this article:

N. Vanaja et al.,: Evaluation of Antiepileptic and Nootropic Activity of *Momordica Sahyadrica*. J. Pharm. Res., 2014; 3(10): 231-236.

Conflict of interest: The authors have declared that no conflict of interest exists.

Source of support: Nil