ORIGINAL ARTICLE

A COMPARATIVE STUDY OF ANTICONVULSANT EFFECT OF FLUNARIZINE AND DILTIAZEM WITH SODIUM VALPROATE ON EXPERIMENTAL MODELS OF EPILEPSY IN ALBINO RATS

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ABSTRACT: BACKGROUND: It is a known fact that calcium ions are involved in pathogenesis of seizures. Hence current study was undertaken to evaluate the anticonvulsant effect of calcium channel blockers flunarizine, diltiazem and compare their efficacy with that of sodium valproate, the broad spectrum anticonvulsant in MES and PTZ induced seizures in albino rats. **MATERIALS AND METHODS:** Albino rats were treated with diltiazem 7.5mg/kg, 15mg/kg, flunarizine 7.5mg/kg,15mg/kg and sodium valproate 250mg/kg bodyweight intraperitoneally and the effects were observed in MES and PTZ models of epilepsy. The parameters observed in MES model were, duration of HLTE phase. Convulsive phase, and post ictal depressive phase. In PTZ model duration of seizure latency, duration of convulsion, and duration post ictal depression were observed. **RESULTS:** our study demonstrated that flunarizine affords protection against convulsions induced in both models, with its efficacy almost approaching that of sodium valproate whereas protection by diltiazem was not significant in both models. **CONCLUSION:** Flunarizine has significant, while diltiazem has no statistically significant anticonvulsant activity as compared to sodium valproate.

KEYWORDS: Epilepsy, flunarizine, diltiazem, sodium valproate, MES model, PTZ model.

ABBREVIATIONS: MES – maximal electro shock seizures, PTZ - pentylene tetrazole, HLTE – hind limb tonic extension, PID - post ictal depression.

INTRODUCTION: Calcium ions are involved in generation of impulse, its propagation, and contraction of heart. These properties of calcium ions have made the calcium channel blockers the first line of drugs in many of the cardiovascular disorders.[1]

These calcium ions are also involved in many of the central nervous system disorders like stroke and epilepsy. They play a prominent role in occurrence of exicitoxicity, a key event in stroke[2]. In epilepsy ca^{+2} ions are involved in both seizure initiation and propagation. It is the influx of extracellular ca^{+2} ions into the neuron through the calcium channels in the beginning that leads to the opening of the voltage gated sodium channels. Influx of Na^{+} ions then causes generation of repetitive action potential causing seizures.[3] So if we can prevent the influx of ca^{+2} ions into the neuron by blocking calcium channels with the help of calcium channel blockers, the seizure activity can be prevented.

Thus we took up this study wherein we evaluate and compare as well, the anticonvulsant activity of calcium channel blockers diltiazem and flunarizine[4] with sodium valproate, the broad spectrum anticonvulsant having multiple mechanisms of action,[5,6] in MES and PTZ induced seizures in albino rats.

Materials and Methods: diltiazem in the dose of 7.5mg\kg, and 15mg\kg, flunarizine in the dose of 7.5mg\kg, and 15 mg\kg, sodium valproate in the dose of 250mg\kg were used, the solvent used was propylene glycol, for all the three drugs. The dose of diltiazem and sodium valproate was calculated from human doses extrapolating onto albino rats depending on the body surface area. The dose of flunarizine was calculated from the dose used in a previous study,[7] conducted on mice, again the dose was converted accordingly depending on the body surface area.

Albino wistar rats of either sex of average weight 150gm to 250gm which were in bred in central animal house of JJMMC were used for experimentation. The study was done after getting the clearance of institutional animal ethical committee.

All the animals were allowed food and water ad libitum both being withdrawn just prior to experiment. The animals were housed in polypropylene cage under standard conditions in dim light and noise free room.

The above test animals were divided into two major groups, one group was subjected to electroshock of 150 m A, 50 HZ for 0.2 seconds, through ear electrodes for MES model.[8] Only those animals that showed convulsive response were used for the study. They were divided into six groups of six each. Remaining group of test animals were used for chemo shock that is PTZ model[9] and they were also divided into six groups of six rats each.

All the test animals were subjected to further study after a gap of 24 hours to prevent any possible kindling effect. All the drugs were given intraperitoneally, except for PTZ used in a dose of 70 mg\kg bodyweight was administered subcutaneously.

All the drugs were given to the predetermined corresponding groups with propylene glycol acting as control. The drugs were given 30 minutes before being subjected to MES stimulus in MES model. Similarly all the drugs were given 30 minutes prior to the administration of PTZ in chemoshock group.

The parameters observed in MES model were, duration of HLTE phase, duration of convulsion and post ictal depression, the parameters observed in PTZ model were duration of seizure latency, duration of convulsion, and post ictal depression.

STATISTICAL ANALYSIS: All the data obtained were tabled as mean and standard error of mean, the data were analysed using one way ANOVA test followed by dunnetts multiple comparisons test.

RESULTS: In MES model, there was highly significant reduction in the duration of HLTE phase, duration of convulsion and postictal depression, with flunarizine in the dose of 15mg\kg, with p value <0.0001, whereas at the dose of 7.5mg\kg the reduction in all the three parameters were significant with a p value < 0.001.

With diltiazem, there was reduction in the duration of all the three parameters in both doses but that was not significant statistically.

The efficacy of flunarizine in the dose of 15mg\kg almost approached that of sodium valproate in respect to all the three parameters. There was no statistical difference between the two drugs at the above mentioned dose of flunarizine with respect to their efficacy.

PTZ model;, even in PTZ model also flunarizine at the dose of 15mg\kg increased latency period of onset of seizures, reduced the duration of convulsion and reduced the post ictal depression phase duration, with a very high significant p value of <.0001, in the dose of 7.5mg\kg it increased the seizure latency period significantly and there was significant reduction in the duration of the other two parameters with p value of <.001

With diltiazem in this model also protection afforded against all the three parameters were not significant statistically

In this model also the efficacy of flunarizine in the dose of 15mg\kg was comparable to that of standard antiepileptic drug sodium valproate.

DISCUSSION: Epilepsy is a common neurological disorder with the prevalence of about 0.3 % to 0.5% in general population[10]. But the now available drugs for epilepsy are not without side effects and about 20% of these seizures are not amenable to these drugs.[11]

As discussed above the calcium ions are also involved in neuronal excitability hence these calcium channel blockers can play a significant role in reducing the excitability of the neurons,they in a way can help in calming down of repetitively firing neurons.

Calcium channel blockers like flunarizine and diltiazem block L and T type of calcium channels with flunarizine having greater potency[1.] Of late it has been found in recent studies that flunarizine blocks Na^+ channels also,[7]

In our study fluanarizine showed dose dependent increase in its potency, and its efficacy almost approaching that of sodium valproate. These observations can be explained by the fact that flunarizine acts by multiple mechanisms and also has got higher affinity for blocking calcium channels. Whereas diltiazem has got no significant efficacy as an anticonvulsant may be because it is less lipophilic and so its ability to cross blood brain barrier is also low owing to its hydrophilic benzothiazepine structure.[12]

CONCLUSION: Flunarizine has got anticonvulsant activity with its efficacy comparable to that of sodium valproate. Flunarizine can be an addition to the prevalent group of anticonvulsants, if proper studies involving human controls are done over this drug.

REFERENCES:

1. Desai SJ, Kulkarni VN, Shankar PS.Therapeutic applications of calcium channel blockers in cardiovascular disorders.indian J Clin Pharnacol Ther 1999; 19; 34-39.
2. Hazell.AS, Exicitotoxic mechanisms in stroke.An update of concepts and treatment strategies. Neurochemistry International 2007 vol 50 (7); 941-953.
3. Heinemann U Hamon, Calcium and Epileptogenesis. Exp Brain Res 1986; 65: 1 – 10.
4. Todd PA, Benfield P. Flunarizine - A reappraisal of its pharmacological properties and therapeutic uses in neurological disorders. Drugs 1989; 38: 481 – 499.
5. Johanessen Lu. Valproate; past, present and future. CNS Drug Rev 2003 p 200 – 216.
6. Meldrum BS, Simiond J. Mechanism of anticonvulsant action of valproate. Drug Neuro Biol 1982; 19: 315 -359.

7. Fischer W, Kittner H, Regenethal R, De Sarro G. Anticonvulsant profile of flunarizine and relation to Na^+ channel blocking effects. Basic And Clinical Pharmacology And Toxicology 2004; 94: 79 – 88.
8. Castel Branco MM, Alves GL, Figueirido IV,Falcao AC. The maximal electroshockseizure (MES) model in the preclinical assessment of potential new antiepileptic drugs. Methods Find Exp Clin Pharmacol 2009; 31(2): 101 – 106.
9. Peter P, Deyn De, Hooge DR, Marescau B, Quan Pe Y. C hemical models of epilepsy with some reference to their applicability in the development of anticonvulsants. Epilepsy Research 1992; 12: 87 – 110.
10. Daniel H Lowenstein.Seizures and Epilepsy, in Harrisons Principles of Internal Medicine-Braunwald,Wilson, Martin. vol 2, 16th edition, pp 2357-2371.
11. Sharma HL, Sharma KK, Antiepileptic Drugs, In principles Of Pharmacology, 2nd edition, Paras Publications 2011, pp529-543.
12. Tripathi KD, Essentials of medical pharmacology, 7th edition. Jaypee publications, 2013, pp 539-557.

Drug (mg\kg), i.p	Duration Of HLTE	Duration Of convulsion	Duration of PID
Control (propylene glycol)	14.16±0.23	34±0.87	47.16±0.26
Sodium valproate 250mg\kg	3.5±0.24	13.66±0.25	18.66±0.56
Flunarizine 7.5mg\kg	8.66±0.67**	23.66±0.50*	24.83±0.20*
Flunarizine 15mg\kg	5.16±0.78**	15.66±0.85**	21.83±0.34**
Diltiazem 7.5mg\kg	12±0.57	31±0.89	42±2.48
Diltiazem 15mg\kg	11.5±061.	29.16±1.62	39.5±2.88

Table 1: Effects of sodium valproate, flunarizine, diltiazem, on MES model

All the values are in mean±SEM.
All the durations are in seconds.
*indicates p value <0.001 i.e significant.
**indicates p value <0.0001 i.e highly significant.

Drug mg\kg, i.p	Seizure latency	Duration of Convulsion	Duration of PID
Control (propylene glycol)	108.66± 5.5	25.83 ± 0.70	81.5 ± 0.99

Sodium valproate 250mg\kg	162.16 ± 2.46	8.83 ± 0.89	17.83 ± 0.95
Flunarizine 7.5mg\kg	140.5 ± 2.04*	16.5± 0.86*	35.16 ± 0.13*
Flunarizine 15mg\kg	155.66 ± 2.68**	10.16 ± 0.32**	20.83 ± 1.04**
Diltiazem 7.5mg\kg	115.33 ±5.85	24.83±0.90	76.66±2.19
Diltiazem 15mg\kg	120.5±3.61	23.33±0.88	73.16±1.5

Table 2: Effects of sodium valproate, flunarizine and diltiazem on PTZ model

PTZ was used in the dose of 70mg\kg s.c.
All the values are in mean±SEM.
All the durations are in seconds.
*indicates p value <0.001 i.e significant.
**indicates p value <0.0001 i.e highly significant.

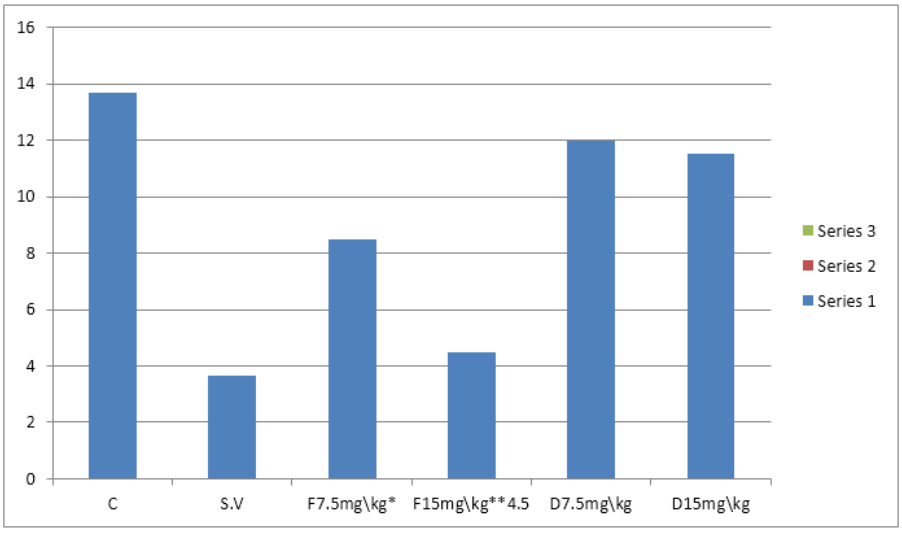

Fig. 1: Showing the effect of sodium valproate, flunarizine, and diltiazem, on duration of HLTE phase in MES model

C –control, s v – sodium valproate, f- flunarizine D- diltiazem.
Y-axis indicates the mean duration in seconds.
*indicates significant p value with p<0.001.
** indicates higly significant p value p<0.0001.

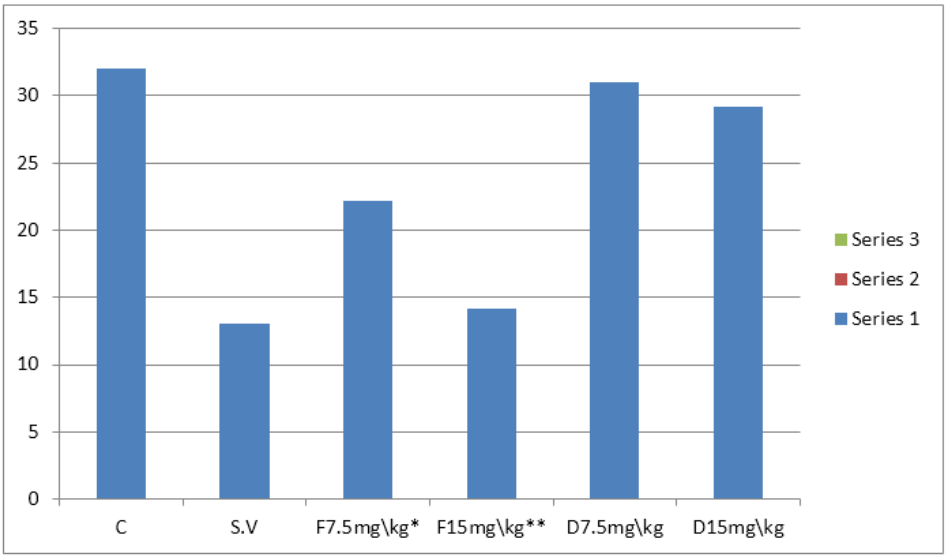

Fig. 2: Showing the effect of sodium valproate, flunarizine, and diltiazem on duration of convulsion in MES model

C-control, sv –sodium valproate, f – flunarizine, D-diltiazem.
Y-axis indicates the mean duration in seconds.

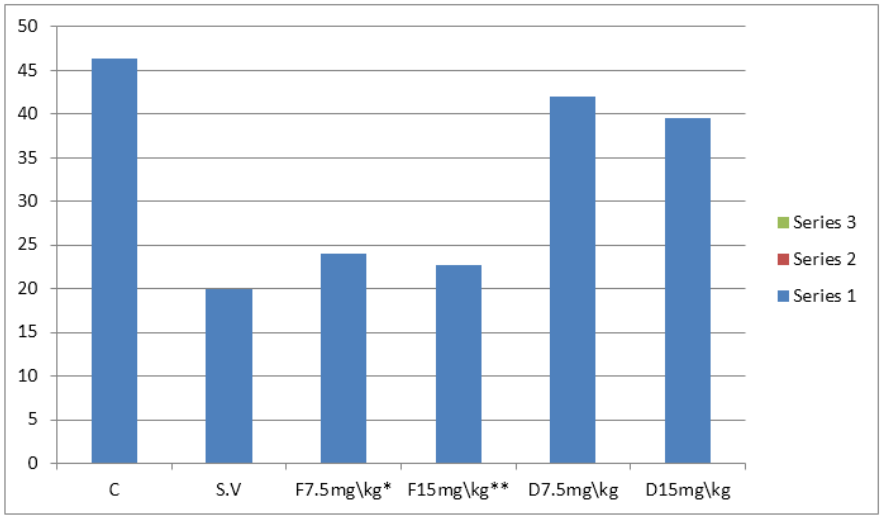

Fig. 3: Showing the effect of sodium valproate, flunarizine, and diltiazem, on duration of PID in MES model

Y-axis indicates the mean duration of the drugs in seconds.

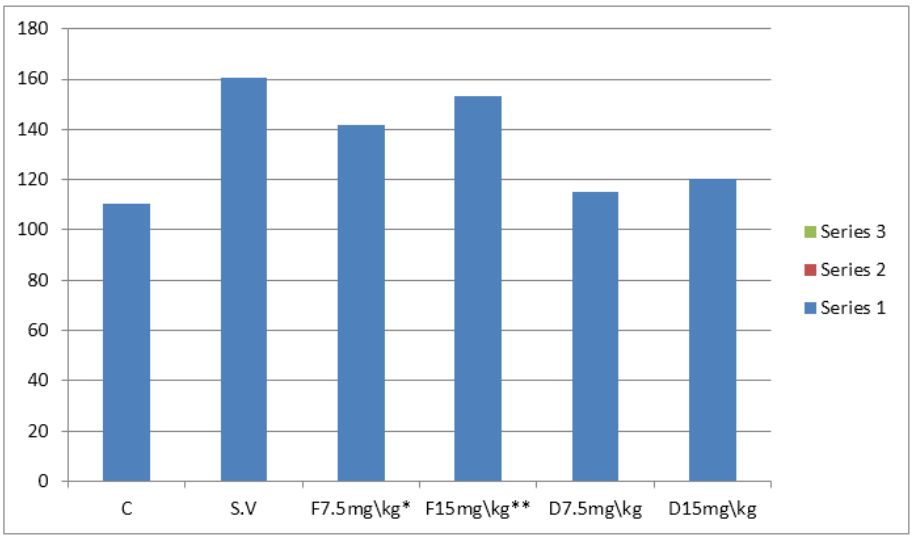

Fig. 4: Effects of sodium valproate, flunarizine, and diltiazem, on duration of seizure latency in PTZ model

Y – axis indicates the mean duration of seizure latency in seconds.

*indicates p value <0.001. i.e significant, ** indicates p value <0.0001 i.e highly significant.

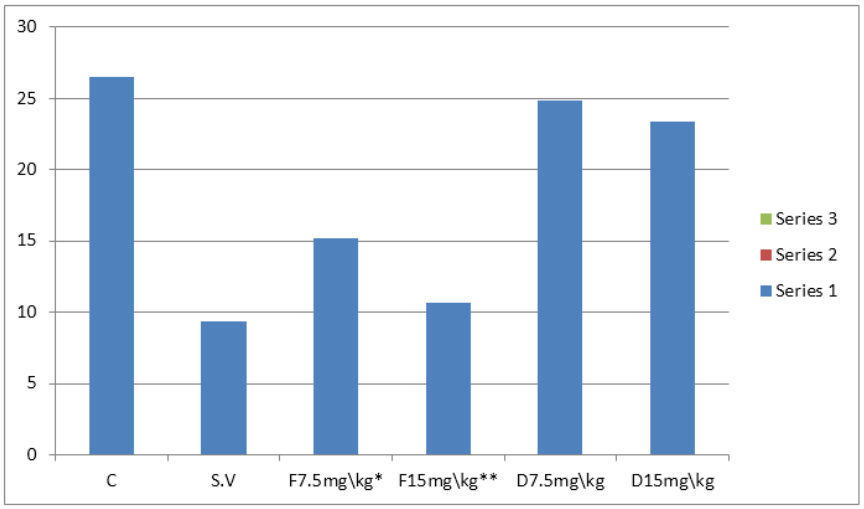

Fig. 5: Showing the effects of sodium valproate, flunarizine, and diltiazem, on duration of convulsion in PTZ model

y- axis indicates the mean duration of convulsion in seconds.

*indicates p value <0.001 i.e significant.

** indicates p value <0.0001 i.e highly significant.

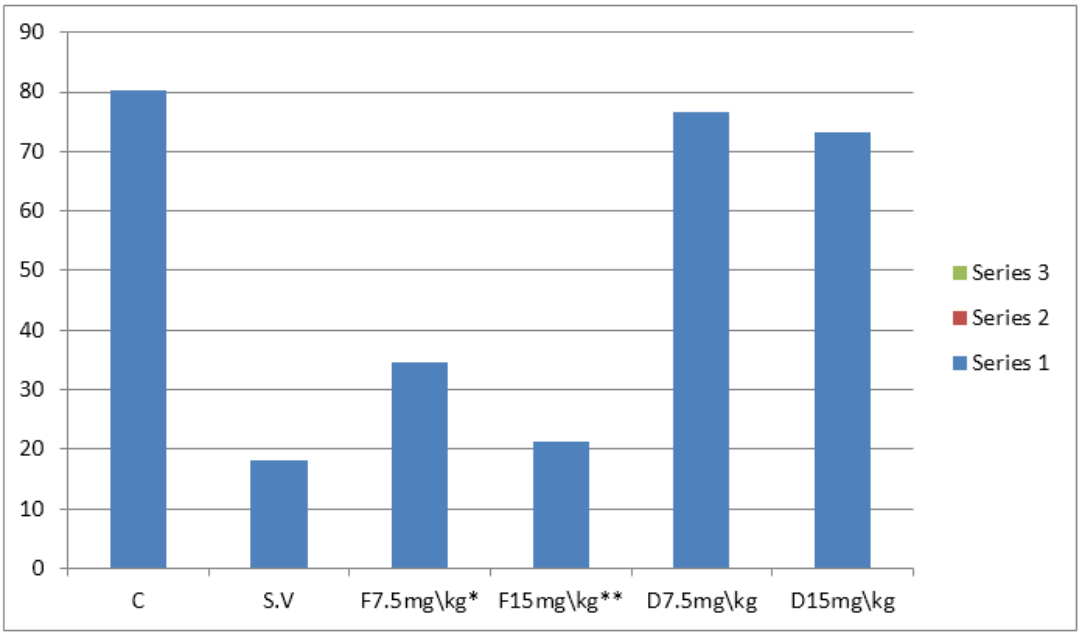

Fig. 6: Showing the effects of sodium valproate, flunarizine, diltiazem, on duration of PID in PTZ model

Y – axis indicates the mean duration of PID in seconds,
*indicates the p value <0.001 i.e significant,
** indicates the p value <0.0001 i.e highly significant.

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