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ANTIEPILEPTIC EFFECT OF TELMISARTAN AND VALSARTAN ALONE AND IN COMBINATION WITH ETHOSUXIMIDE ON PENTYLENETETRAZOL INDUCED EXPERIMENTAL MODEL OF EPILEPSY IN ALBINO MICE

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Keywords:

Pentylenetetrazol, Telmisartan, Valsartan, Ethosuximide, Renin angiotensin system

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ABSTRACT: Epilepsy is a common neurological disorder in clinical practice requiring newer targets for its efficient treatment. Recently, brain renin-angiotensin system has gained immense importance because of its involvement in seizure regulation. Hence, the present study was planned to evaluate whether angiotensin II AT1 receptor antagonists possess anticonvulsant activity in experimental animals. Forty- eight Swiss albino mice were divided into 8 groups: Control - distilled water (5ml/kg), Standard – ethosuximide 200mg/kg, T1- telmisartan 5mg/kg, T2- telmisartan 8mg/kg, V1- valsartan 10mg/kg, V2- valsartan 20mg/kg, E+T2- ethosuximide 200mg/kg+ telmisartan 8mg/kg, E+V2- ethosuximide 200mg/kg+ valsartan 20mg/kg. After 1 hour of administration of control, test and standard drugs intraperitonally, convulsions were induced by administering pentylenetetrazol (60mg/kg- s.c). The time of onset of myoclonic spasm and clonic convulsion were the parameters recorded. The results were analysed by one-way- ANOVA followed by Dunnett's t test. Telmisartan 8mg/kg, valsartan 10mg/kg, valsartan 20mg/kg significantly increased the latency of onset of myoclonic spasm and clonic convulsion ($p< 0.05$) as compared to control. The combinations of (ethosuximide + telmisartan 8mg/kg) and (ethosuximide + valsartan 20mg/kg) also significantly increased the latencies of onset of myoclonic spasm and clonic convulsion ($p<0.05$) as compared to control. Conclusively, angiotensin receptor antagonist *viz.* telmisartan and valsartan showed anticonvulsant activity in PTZ seizure models. Both telmisartan and valsartan potentiated the antiepileptic activity of ethosuximide. So, dose of ethosuximide can be reduced in epileptic patients receiving telmisartan or valsartan for other clinical conditions.

INTRODUCTION: Epilepsy is a chronic disorder of brain function characterized by the recurrent and unpredictable occurrence of seizures. Approximately 1% of the world's population has epilepsy. Seizures that occur in people with epilepsy are transitory alterations in behaviour, sensation or consciousness caused by abnormal, synchronized electrical discharge in the brain. Tests in rodent models have identified most antiseizure drugs. The pentylenetetrazol (PTZ) test, in which animals receive a dose of the chemical convulsant PTZ (an antagonist of $GABA_A$ receptors) has been used for screening of drugs effective in petit mal epilepsy or absence seizures [1, 2].

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Angiotensin AT1 receptor antagonists are widely used in clinical practice for treatment of hypertension, congestive heart failure or diabetic nephropathy [3]. It is known that besides the peripheral renin-angiotensin system, all components (precursors and enzymes) of this system are present in the brain [4].

Among them, angiotensin II is the main component of this system. It has been demonstrated that angiotensin II administered intracerebroventricularly increased the threshold of pentylenetetrazol, bicuculline and picrotoxin-induced seizures and attenuated the intensity of clonic convulsions induced by pentylenetetrazol and 3-mercaptopropionic acid in mice [5]. Taking into consideration, the significant role of renin angiotensin aldosterone system in the brain, we sought to evaluate the influence of ARBs *i.e.* telmisartan and valsartan on the seizure protective action of ethosuximide.

METHODS:

Study design: Experimental animal-based study.
Sample size: n = 6 for 8 groups. Total N = 48

Groupings:

C: Control
E: Ethosuximide 200mg/kg
T_1: Telmisartan 5mg/kg
T_2: Telmisartan 8mg/kg
V_1: Valsartan 10mg/kg
V_2: Valsartan 20mg/kg

$E+T_2$: Ethosuximide 200mg/kg + Telmisartan 8mg/kg.
$E+V_2$: Ethosuximide 200mg/kg + Valsartan 20mg/kg.

Experimental Animals: Swiss albino mice of either sex weighing 20-30 g were procured from the animal house of JNIMS, Imphal, for experimental purposes. They were housed in departmental polypropylene cages under a standard laboratory diet and water *ad libitum* and acclimatized for 10 days. 12 hours dark-light cycle was maintained. The animals were fasted for 8 hours before the experiment, and care was taken to avoid coprophagy. Each mouse was used only once. The animals were grouped in 8 groups with 6 animals in each group. The protocol containing the materials and methods for the present study was approved by the Institutional Animal Ethics Committee (IAEC) of the Regional Institute of Medical Sciences (Registration No.: 1596/GO/a/12/CPCSEA).

Drugs: Angiotensin AT1 receptor antagonists, telmisartan (Abbott, India) and valsartan (Novartis, India), and antiepileptic drug ethosuximide (Parke Davies, India) were used. All drugs were suspended in 1% solution of Tween 80 (Sigma, USA) in distilled water and were injected intraperitonally (i.p) in a volume of 5ml/kg body weight.

Experimental Model: Anticonvulsant activity was tested using PTZ model. PTZ dissolved in distilled water at a dose of 60mg/kg was used. 60 min after drug administration, a seizure was induced by subcutaneous injection of PTZ at the nape of the neck at a volume of 5ml/kg. The animals were observed for 30mins after the PTZ challenge for onset of myoclonic spasm and clonic convulsion. The time of onset of myoclonic spasm and clonic convulsion was noted in seconds.

Statistical Analysis: The values were expressed as mean± SEM. The statistical analysis was carried out by SPSS software. One-way analysis of variance (ANOVA) followed by post hoc Dunnett's t-test was applied. P values <0.05 were considered significant.

RESULTS: The results of the test are tabulated below in **Table 1.**

TABLE 1: EFFECT OF TELMISARTAN AND VALSARTAN ON PTZ-INDUCED SEIZURES IN MICE, ALONE AND IN COMBINATION WITH ETHOSUXIMIDE

Groups	Onset of myoclonic spasm in secs (Mean±Sem)	Onset of clonic convulsion in sec. (Mean± Sem)
Control, C	179.50 ± 14.63	295.67 ± 30.29
Ethosuximide, E	300.00 ± 20.45*	585.83 ± 65.27
Telmisartan, 5mg/kg T1	238.83 ± 14.33	359.83 ± 22.78
Telmisartan, 8mg/kg T2	375.33 ± 27.93*#	509.83 ± 20.66*
Valsartan, 10 mg/kg V1	278.17 ± 19.30*	433.67 ± 15.61
Valsartan, 20mg/kg V2	424.17 ± 23.24*#	559.17 ± 23.87*
E+T2	397.50 ± 24.52*#	925.00 ± 126.13*#
E+ V2	452.83 ± 14.11*#	836.67 ± 49.09*#
One Way F Anova, d.f, P	22.34, 7,40 < 0.0001	15.33, 7,40 <0.0001

*p < 0.05 compared to control, #p < 0.05 as compared to ethosuximide.

The latencies of onset of myoclonic spasm and clonic convulsion in the mice treated with the vehicle were 179.50± 14.63 seconds and 295.67± 30.29 seconds, respectively. The latencies of onset of myoclonic spasm in the groups treated with telmisartan 5 mg/kg and 8 mg/kg were 283.83± 14.33 seconds and 375.33± 27.93 seconds, respectively. The prolongation in the latency of myoclonic spasm with telmisartan 8mg/kg was found to be significant ($p< 0.05$) compared to the control. The latencies of onset of myoclonic spasm in the groups treated with valsartan 10mg/kg and 20mg/kg were 278.17±19.30 seconds and 424.17± 23.24 seconds, respectively. This prolongation in the latency of myoclonic spasm was found to be significant ($p< 0.05$) compared to the control. The latencies of onset of clonic convulsion treated with telmisartan 5mg/kg, telmisartan 8mg/kg, valsartan 10mg/kg, and valsartan 20mg/kg were 359.83± 22.78 seconds, 509.83± 20.66 seconds, 433.67± 15.61 seconds and 559.17± 23.87 seconds respectively. The prolongation in the latencies was statistically significant ($p< 0.05$) only in the groups treated with telmisartan 8mg/kg and valsartan 20mg/kg compared to the control.

The latencies of onset of myoclonic spasm and clonic convulsion treated with ethosuximide 200mg/kg were 300.00±20.45 seconds and 585.83±67.27 seconds, respectively, and these were found to be statistically significant ($p< 0.05$) as compared to control. The latencies of onset of myoclonic spasm when ethosuximide is combined with telmisartan 8mg/kg and valsartan 20mg/kg were 397.50± 24.52 seconds and 452.83± 14.11 seconds, respectively. These latencies were found to be significant compared to control and ethosuximide ($p< 0.05$). The onset of clonic convulsion latencies, when ethosuximide is combined with telmisartan 8mg/kg and valsartan 20mg/kg, were found to be 925.00±126.13 seconds and 836.67 ± 49.09 seconds, respectively. These increases in the latencies of onset of clonic convulsion were found to be significant as compared to control and ethosuximide.

DISCUSSION: The result of the present study suggests that angiotensin receptor blockers *viz.* telmisartan and valsartan alone, possess antiepileptic activity against pentylenetetrazol-induced seizure in mice. Preventing seizures induced by pentylenetetrazol in laboratory animals is the most commonly used preliminary screening test for characterizing potential anticonvulsant drugs. It represents a valid model for human generalized petit mal (absence seizure) type seizures. The mechanism of the convulsant action of PTZ seems to be related to the inhibitory function of the neurotransmitter gamma-aminobutyric acid (GABA).

PTZ has been shown to have an affinity for the chloride-ionophore of the postsynaptic GABA receptor ionophore complex and to antagonize GAB Aergic function [6]. Our results agree with previous data illustrating the positive role of renin angiotensin aldosterone system inhibition in different models of seizures. AT1 receptor antagonist, telmisartan, and olmesartan in a dose-dependent manner showed an increase in antiepileptic activity [7].

An early study reported a significant decrease in seizure severities when epileptic animals were treated with clinically used doses of enalapril, an ACE inhibitor and losartan, an AT1 receptor blocker [8]. There is also evidence that the brain RAS mediates seizure susceptibility. Angiotensin II (Ang II) administered intracerebroventricularly increased the threshold of pentylenetetrazol-, bicuculline-, andpicrotoxin-induced seizures and attenuated the intensity of clonic convulsions induced by pentylenetetrazol and 3-mercaptopropionic acid in mice [5].

Knowing that both telmisartan and valsartan are able to cross the blood-brain barrier [9, 10] and the blockade of angiotensin II action may indirectly interfere in the modulation of excitatory and inhibitory components of CNS, this can be proposed as a possible mechanism of action by which the angiotensin receptor blockers impaired the epileptic seizures described in this study. The present study also indicates that the protective action of antiepileptic drug ethosuximide against PTZ-induced seizures in mice is potentiated by co-administration of angiotensin receptor blockers *viz.* telmisartan and valsartan. The concept of rational polytherapy developed within recent years is based on the assumption that combining some antiepileptic drugs may result in supraadditive

(synergistic) efficacy and infra-additive (antagonists) toxicity resulting in an enhanced efficacy/toxicity profile [11]. The mechanism of the anticonvulsive activity of ethosuximide is poorly understood.

In contrast to other antiepileptic drugs, ethosuximide was not found to augment GABA-ergic transmission and even slightly antagonized γ-amino butyric acid (GABA) mediated events recorded from cultured cortical neurons of the rats [12]. Furthermore, this antiepileptic drug did not affect the veratridine-stimulated uptake of calcium into slices of cortex from the rat nor the release of glutamate and Aspartate [13]. However, recent findings showed that ethosuximide reduced a low threshold calcium current in the thalamic neurons at therapeutic concentration [14]. It is of important that this calcium current may be involved in the genesis of abnormal activity, characteristic for petit mal epilepsy [15]. The mechanism behind potentiation of antiepileptic activity of ethosuximide by telmisartan and valsartan might be because of pharmacodynamics or pharmacokinetic interactions which needs further investigations.

CONCLUSION: From the present study, it may be assumed that the dose of ethosuximide may be reduced in an epileptic patient receiving telmisartan or valsartan for some other conditions. But further studies on experimental animals and human beings are necessary to support the present work.

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CONFLICTS OF INTEREST: None declared.

REFERENCES:

1. Porter RJ and Rogawski MA: Antiseizure drugs. In: Bertram G. Katzung Basic and Clinical Pharmacology.14th Ed. University of California, San Francisco: McGraw-Hill; 2018; 409-439.
2. Gaikwad P, Adak V and Shete RV: The Screening models for antiepileptic drugs: A Review. JDDT 2021; 11(2): 175-8.
3. Capric V, PriyanChandrakumar H, Celenza-Salvatore J and N. Makaryus A: The Role of the Renin-Angiotensin-Aldosterone System in Cardiovascular Disease: Pathogenetic Insights and Clinical Implications. Renin-Angiotensin Aldosterone System 2021; 24.
4. Jackson L, Eldahshan W, Fagan SC and Ergul A: Within the Brain: The Renin Angiotensin System. International Journal of Molecular Sciences 2018; 19(3): 876.
5. Krasniqi S and Daci A: Role of the Angiotensin Pathway and its Target Therapy in Epilepsy Management. International J of Molecular Sciences 2019; 20(3): 726.
6. Phalak C and Sadar S: Animal models used in the screening of antiepileptic drugs. Int J Pharm Sci & Res 2022; 13(2): 612-20.
7. Pushpa VH, Padmaja SK, Suresha RN, Jayanthi MK, Ashwini V and Vaibhani PS: Evaluation and comparison of anticonvulsant activity of telmisatanand olmesartan in experimentally induced animal models of epilepsy. J Clin Diagn Res 2014; 8(10): 08-11.
8. Pereira MG, Becari C, Oliveira JA, Salgado MC, Garcia-Cairasco N and Costa-Neto CM: Inhibition of the renin-angiotensin system prevents seizures in a rat model of epilepsy. Clin Sci (Lond) 2010; 119(11): 477-482.
9. Cai L, Li W, Zeng R, Cao Z, Guo Q, Huang Q, Liu K. Valsartan alleviates the blood-brain barrier dysfunction in db/db diabetic mice. Bioengineered 2021; 12(1): 9070-9080.
10. Goldzik L and Santistiban MM: blood brain barrier crossing renin-angiotensin system drugs: considerations for dementia and cognitive decline. Hypertension 2021; 78: 644-646.
11. Park KM, Kim SE and Lee BI: Antiepileptic Drug Therapy in Patients with Drug-Resistant Epilepsy. J Epilepsy Res 2019; 9(1): 14-26.
12. Barnes DM and Dichter MA: Effects of ethosuximide and tetramethylsuccinimide on cultured cortical neurons. Neurology 1984; 34(5): 620-625.
13. Crowder JM and Bradford HF: Common anticonvulsants inhibit Ca2+ uptake and amino acid neurotransmitter release in vitro. Epilepsia 1987; 28(4): 378-382.
14. Shills GJ and Rogawski MA: Mechanisms of action of currently used antiseizure drugs. Neuropharmacology 2020; 168: 107966
15. Weiss N and Zamponi GW: T-type channel druggability at a crossroads. ACS Chem. Neuros 2019; 10(3): 1124-1126.

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