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Research Article

SYNTHESIS, CHARECTERIZATION AND ANTI-CONVULSANT ACTIVITY OF SUBSTITUTED 5-(5-SULFANYL-1,3,4- OXADIAZOL-2-YL)BENZENE- 1,2,3-TRIOL DERIVATIVES

Vinod Nakra * [1], Manoj Sharma [2]

[1] Research Scholar, Bhagwant University, Ajmer, Rajasthan, India

[2] Research Supervisor, School of Studies in Pharmaceutical Sciences, Jiwaji University, Gwalior, M.P., India

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ABSTRACT

All solvents were redistilled before use. Reactions were routinely monitored by thin layer chromatography and spots were visualized by exposure to iodine vapour or UV light. A solution of propyl gallate(0.01 mol) in ethanol and hydrazine hydrate (0.01 mol) was refluxed for 4 hours. The excess solvent was distilled off under reduced pressure. The cooled residual mass was washed with distilled water. It was filtered and dried. The crude product was recrystallised from methanol to yield galloylhydrazide, Carbon disulfide (2 ml) was added drop wise to an ice cooled solution of KOH (2g) in ethanol (20 ml) containing the acid hydrazide 4 (0.02 mole), then the reaction mixture was stirred at room temperature 2h . After dilution with ethanol the solid precipitated was washed twice with ether. To the solid obtained (1 g), 10% KOH (20 ml) was added then the reaction mixture was refluxed for 4 hr, cooled, acidified with conc. HCl. The resulting solid was filtered washed with water, dried and crystallized. A mixture of (0.97g, 0.005mol) of 5-(5-sulfanyl- 1,3,4-oxadiazol-2-yl)benzene-1,2,3-triol and (0.005mol) of different aryl or alkyl halides were refluxed in 25ml of pyridine solution for 3.5 hours. The resultant mixture was cooled and poured into crushed ice. The solid mass is thus separated out was dried and recrystallized from ethanol. Synthesized derivatives purity were checked by TLC, Melting point & characterized by FT-IR, Mass, NMR spectroscopic techniques.

Key words: Oxadiazole, Oxadiazole derivatives, convulsion, Anti-convulsant activity.

1. INTRODUCTION:

Oxadiazoles are a class of heterocyclic aromatic chemical compound of the azole family; with the molecular formula $C_2H_2N_2O$. There are four isomers of oxadiazole: Five member heterocyclic compounds show various type of biological activities among than 2,5-disubstituted 1,3,4-oxadiazole are associated with diverse biological activities.[1]Various biological activities like antimicrobial, anti-tubercular, anti-inflammatory, Anticonvulsant[2], Hypnotic , Anesthetic activity[3].1,3,4-oxadiazoles showed antibacterial properties similar to those of well-known sulfonamide drugs. The oxadiazole nucleus with N=C-S linkage exhibits a large number of pharmacological activities.[4]Sulfone derivatives containing heterocyclic moiety are known for their interesting antifungal bioactivities and have attracted considerable attention in pesticide and medicinal formulation. A large number of report on their synthesis and biological activities have appeared during the last three years[5]

1.1 ANTI-CONVULSANT ACTIVITY

SEIZURE [15]: -

Seizures are sudden, transitory, and uncontrolled episodes of brain dysfunction resulting from abnormal discharge of neuronal cells with associated motor, sensory or behavioral changes.

Epilepsy A group of chronic CNS disorders Characterized by recurrent seizures.

Types of Epileptic Seizures

I. Partial (focal) Seizures

A. Simple Partial Seizures

B. Complex Partial Seizures

C. Partial with secondary generalized tonic

II. Generalized Seizures

A. Generalized Tonic-Clonic Seizures

B. Absence Seizures

C. Myoclonic Seizures

Corresponding author: Vinod Nakra

Pathological Basis

- Abnormal electrical discharge in the brain.
- Coordinated activity among neurons depends on a controlled balance between excitation and inhibition.
- Any local imbalance will lead to a seizure
- Imbalances occur between glutamate-mediated excitatory neurotransmission and gamma amino butyric acid (GABA) mediated inhibitory neurotransmission.
- Generalized epilepsy is characterized by disruption of large scale neuro-networks in the higher centers.

1.2 Anticonvulsants (also commonly known as **antiepileptic drugs** or as **antiseizure drugs**) are a diverse group of pharmaceuticals used in the treatment of epileptic seizures. Anticonvulsants are also increasingly being used in the treatment of bipolar disorder, since many seem to act as mood stabilizers, and for the treatment of neuropathic pain. Anticonvulsants suppress the rapid and excessive firing of neurons during seizures. Anticonvulsants also prevent the spread of the seizure within the brain. Some investigators have observed that anticonvulsants themselves may cause reduced IQ in children.[2] However these adverse effects must be balanced against the significant risk epileptic seizures pose to children and the distinct possibility of death and devastating neurological sequel a secondary to seizures. Anticonvulsants are more accurately called **antiepileptic drugs** (abbreviated "AEDs"), and are often referred to as **antiseizure drugs** because they provide symptomatic treatment only and have not been demonstrated to alter the course of epilepsy[16]

Conventional antiepileptic drugs may block sodium channels or enhance γ-aminobutyric acid (GABA) function. Several antiepileptic drugs have multiple or uncertain mechanisms of action. Next to the voltage-gated sodium channels and components of the GABA system, their targets include $GABA_A$ receptors, the GAT-1 GABA transporter, and GABA transaminase. Additional targets include voltage-gated calcium channels, SV_2A, and $\alpha_2\delta$. By blocking sodium or calcium channels, antiepileptic drugs reduce the release of excitatory glutamate, whose release is considered to be elevated in epilepsy, but also that of GABA. This is probably a side effect or even the actual mechanism of action for some antiepileptic drugs, since GABA can itself, directly or indirectly, act proconvulsively. The drug class was the US's 5th-best-selling in 2007.

Some anticonvulsants have shown antiepileptogenic effects in animal models of epilepsy. That is, they either prevent the development of epilepsy or can halt or reverse the progression of epilepsy. However, no drug has been shown in human trials to preventepil epitogenesis (the development of epilepsy in an individual at risk, such as after a head injury).

2. MATERIAL & METHODS:

Chemical and reagents

All solvents were redistilled before use. Reactions were routinely monitored by thin layer chromatographyand spots were visualized by exposure to iodine vapour or UV light. All the synthesized compounds were purified by recrystallization.Melting points were determined by using open capillary method.

Instrumentation

Spectroscopic data were recorded with following instrument;

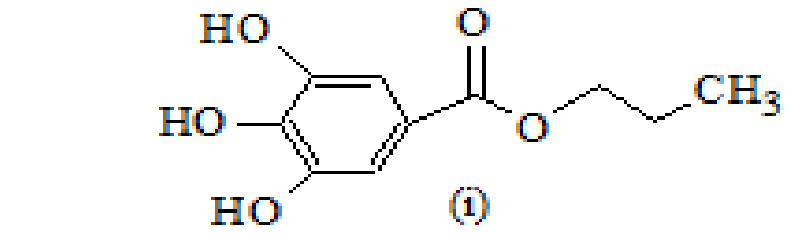
1. Fourier Transform Infra-Red spectra (FTIR) were recorded on Shimadzu FTIR-8400S spectrophotometer using potassium bromide pellets and sodium chloride cell.

2. Nuclear Magnetic Resonance spectra (1H-NMR) were recorded on JEOL-300 MHz spectrophotometer in CDCl3 using TMS as an internal standard. Chemical shifts (δ) are expressed in parts per million (ppm).

3. Mass spectra were recorded on HEWLETT 180017, PACKARD GCD System mass spectrophotometer using electron ionization detector.

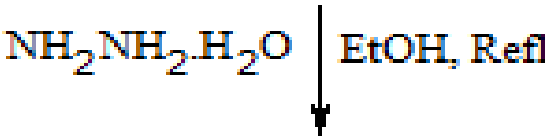
4. Anticonvulsant activity checked by Electro-convulsometer (LABTECH)

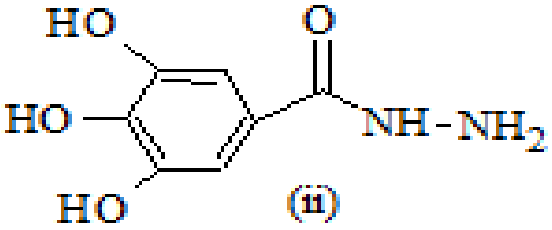
2.1 SYNTHETIC SCHEME:



(i)

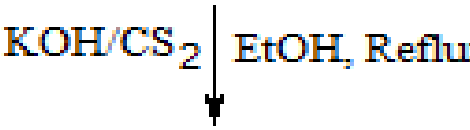
propyl 3,4,5-trihydroxybenzoate /(propyl gallate)

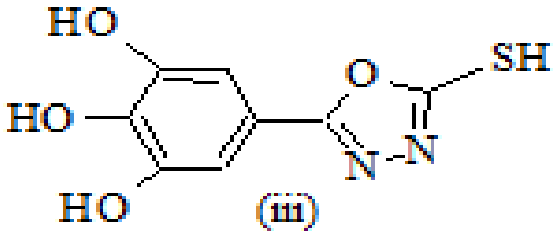




(ii)

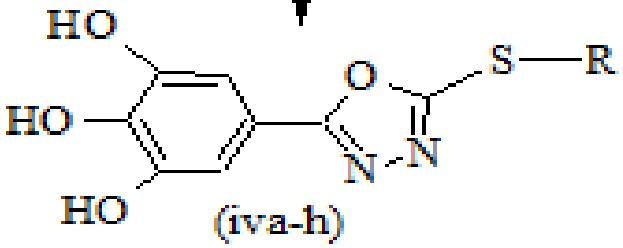
3,4,5-trihydroxybenzohydrazide/(galloyl hydrazide)





(iii)

5-(5-sulfanyl-1,3,4-oxadiazol-2-yl)benzene-1,2,3-triol



(iva-h)

Where **R**=

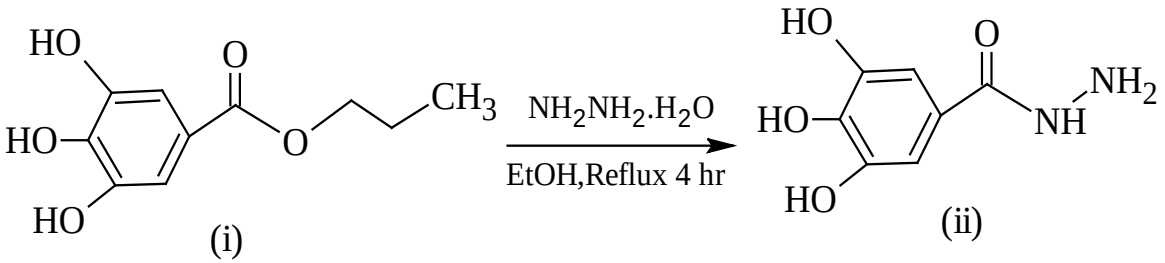
iva=Ar-CH_3**ivb**=Ar-OCH_3**ivc**=Ar-OH **ivd**=Ar-NO_2**ive**=Ar-C_2H_5 **ivf**=CH_3**ivg**= -C_2H_5

ivh= C_3H_7 **ivi**=Ar-NH_2**ivj**=Phenyl

2.3 SYNTHETIC PROCEDURE:

2.3.1:- Step1. Synthesis of3,4,5-trihydroxybenzohydrazide/ galloylhydrazide:-

A solution of propyl gallate(0.01 mol) in ethanol and hydrazine hydrate (0.01 mol) was refluxed for 4 hours. The excess solvent was distilled off under reduced pressure. The cooled residual mass was washed with distilled water. It was filtered and dried. The crude product was recrystallised from methanol to yield galloylhydrazide.



(i) propyl 3,4,5-trihydroxybenzoate or propyl gallate

(ii) 3,4,5-trihydroxybenzohydrazide or galloyl hydrazide

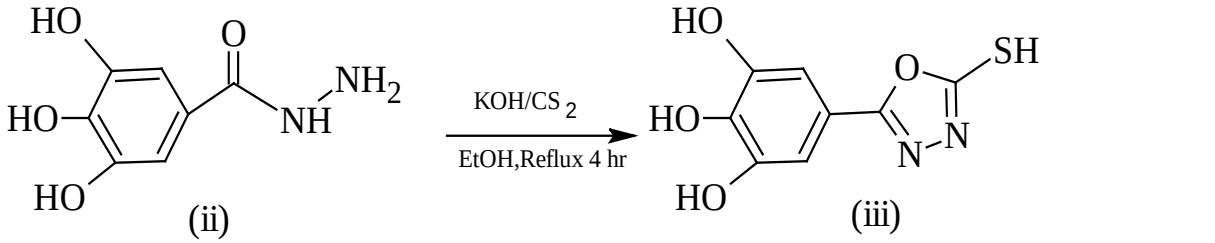
Table 1 physiochemical data of synthesized compound (ii)

COMPOUND	% YIELD	RF VALUE	MOL. FORMULA	**Mol. mass**
II	91.64	0.65	$C_7H_8N_2O_4$	184.14

SOLVENT SYSTEM FOR TLC- ETHYL ACETOACETATE: N-HEXANE (70:30)

2.3.2 Step-2:-**General procedure for preparation of 5-(5-sulfanyl-1,3,4-oxadiazol-2 yl) benzene-1,2,3-triol (iii)**

Carbon disulfide (2 ml) was added drop wise to an ice cooled solution of KOH (2g) in ethanol (20 ml) containing the acid hydrazide 4 (0.02 mole), then the reaction mixture was stirred at room temperature 2h . After dilution with ethanol the solid precipitated was washed twice with ether. To the solid obtained (1 g), 10% KOH (20 ml) was added then the reaction mixture was refluxed for 4 hr, cooled, acidified with conc. HCl. The resulting solid was iltered washed with water, dried and crystallized.



(ii) 3,4,5-trihydroxybenzohydrazide or galloyl hydrazide

(iii) 5-(5-sulfanyl-1,3,4-oxadiazol-2-yl)benzene-1,2,3-triol

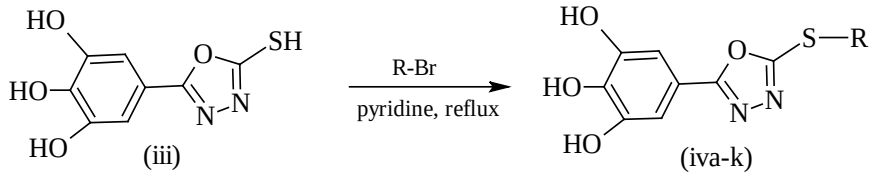
TABLE 2 PHYSIOCHEMICAL DATA OF SYNTHESIZED COMPOUND (III)

COMPOUND	% YIELD	RF VALUE	MOL. FORMULA	**Mol. Mass**
III	72.64	0.71	$C_8H_6N_2O_4S$	226.20

SOLVENT SYSTEM FOR TLC- ETHANOL: N-HEXANE (65:35)

2.3.3:- STEP3: GENERAL PROCEDURE FOR PREPARATION OF 5-[5-(SUBSTITUTEDSUFANYL)-1,3,4-OXADIAZOLE-2-YL]BENZENE-1,2,3-TRIOL (IVA-J)

A mixture of (0.97g, 0.005mol) of 5-(5-sulfanyl- 1,3,4-oxadiazol-2-yl)benzene-1,2,3-triol and (0.005mol) of different aryl or alkyl halides were refluxed in 25ml of pyridine solution for 3.5 hours. The resultant mixture was cooled and poured into crushed ice. The solid mass is thus separated out was dried and recrystallized from ethanol.



5-(5-sulfanyl-1,3,4-oxadiazol-2-yl)benzene-1,2,3-triol

5-[5-(substitutedsulfanyl)-1,3,4-oxadiazol-2-yl]benzene-1,2,3-triol

3. ANTI-CONVULSANT ACTIVITY:

All antiepileptic drugs (AEDs) are rigorously study in animals, particularly rodents, before they are given to patients. Understanding how drugs are screened in animals is useful to the clinician, since the screening process is valuable in predicting the type of seizure in which the drug would be efficacious, as well as determining the mechanism of the drug's anti-seizure action. Indeed, the dramatic discovery of the anti-seizure properties of phenytoin was identified by Merritt and Putnam in 1938 using the electroshock-induced seizure model.

Animals

Animals Albino mice, weighing 18-30 g, were used for experiments. The animals were kept in colony cages (6 mice each), maintained on a standard pellet diet with water, and left for 2 days for acclimatization before the experimental session. The food was withdrawn on the day before the experiment, but free access to water was allowed. All experiments were carried out according to the suggested ethical guidelines for the care of laboratory animals.

Selection of experimental animals:

Healthy Albino wistar male rats weighing between 18-30 g were used for the evaluation of anti-convulsant activity.

Laboratory conditions: The rats were housed comfortably in a group of six in a single clean plastic cage with a metal frame lid on its top. Environmental room should be 22°C (± 3°C) relative humidity was kept at least 30 % and preferably not exceed 70 % other than during room cleaning the aim was maintained between 50-60%. Lighting was artificial, the sequence being 12 hours light and 12 hours.

Food and water: All animals were allowed free access to water and standard palletized laboratory animal diet.

Bedding: In the present study animals were provided with clean paddy husk bedding. Bedding was changed every alternate day to maintain proper hygienic conditions.

The anticonvulsant activity was carried out by maximal electrical shock induced convulsion method in albino mice.

Animals to be use: - Albino mice

No. of animals used per group:-6 mice

Dose of test compound:-0.5ml/100g

Dose of standard drug:-0.5ml/100 (Phenytoin)

Route of administration:-Intra peritoneal (suspended in 1% tween-80 solution)

Requirements:-

Instruments: - Electroconvulsiometer

Chemicals:-Tween-80

Standard drug: - Phenytoin (25 mg/kg) aqueous suspension was prepared using solution of tween-80 as a suspending agent.

Test compounds:-suspension of compounds were prepared and administered intra peritoneal similar to that of standard drug.

Apparatus: -Syringes (1 ml, 2 ml), sample tubes.

Experimental design and procedure:-

Animals were weighed and numbered. Mice were divided into 7 groups of six animals each. Group 1 served as control which was treated with vehicle (2% v/v Tween 80), group 2 was treated with standard drug phenytoin (25 mg/kg, i.p.) and groups 3– 7 were treated with newly synthesized oxadiazole derivatives (25 mg/kg, i. p.). One hour after injection, the animals were subjected to electro shock through ear electrodes of 80 mA for 0.2 sec by electroconvulsiometer and the duration of time for extensor response was noted and the activity was expressed in terms of % protection.

All the results are expressed as mean ± SEM. The % inhibition of epileptic seizures was calculated by using the formula,

Percent (%) protection = VC – VT/VC X 100,

Where, VT- Mean time in test group, VC-Mean time in control group.

3.1 Screening of anti-convulsant activity

Table 3:-Screening of Anti-convulsant activity in Albino Mice (By Maximal electro shock method)

Compound Code	Duration (Mean±SEM,Sec, sec)					% Protection
	Flexion	Extension	Clonus	Stupor	Recovery	
Control	12.75±0.3	15.85±0.23	27.50±0.19	96.0±0.09	Recovered	
Standard (Phenytoin)	9.80±0.08	11.83±0.19	3.07±0.05	1.91±0.03	Recovered	83.95 %
Comp-iv a	10.5±0.12	12.5±0.13	6.5±0.07	22.5±0.02	Recovered	65.8%
Comp-iv b	10.0±0.09	12.0±0.15	5.0±0.14	33.7±0.01	Recovered	60.0%
Comp-ivc	11.2±0.11	15.5±0.25	15.0±0.15	20.5±0.11	Recovered	59.23%
Comp-ivd	10.1±0.09	13.2±0.15	6.1±0.14	32.7±0.01	Recovered	59.90%
Comp-ive	11.1±0.09	14.2±0.15	6.5±0.14	33.9±0.01	Recovered	58.90%
Comp-ivf	9.80±0.08	11.83±0.19	5.0±0.14	13.7±0.01	Recovered	70.52%
Comp-ivg	10.0±0.09	12.0±0.15	5.0±0.14	20.5±0.11	Recovered	60.23%
Comp-ivh	11.2±0.11	15.5±0.25	15.0±0.15	31.9±0.01	Recovered	57.90%
Comp-ivi	10.5±0.12	12.5±0.13	6.5±0.07	32.7±0.01	Recovered	55.25%
Comp-ivj	9.70±0.08	12.80±0.19	3.07±0.05	31.9±0.01	Recovered	76.87%

4. RESULT& DISUSSION:

The pharmacological screening of the synthesized compounds showed anti convulsant activity ranging from 55.25 % to 76.87 % inhibition of epileptic seizures in mice, where as the standard drug Phenytoin showed 83.95 % inhibition of epileptic seizures in mice.

The compound ivf, ivj was found to be nearly potent to Phenytoin which is used as standard drug. Compounds

iva, ivb, ivc, ivd, ive, ivg, ivh and ivi shown less % of inhibition of epileptic seizures in mice than Phenytoin (standard drug).

5. CONCLUSION:

The present work, which has undertaken is bonafied, and novel for the synthesis of oxadiazole derivatives. In this view we have made an attempt in reviewing the literature on substituted oxadiazolefor their medicinal significance with help of chemical abstract, journals and internet sites.

All the synthesized compounds were light creamish to brown coloured crystalline solids. Most of the compounds are freely soluble in chloroform and other solvents like methanol, ethanol. The melting point of the compounds was in the range of 152ºC to 251ºC.

All synthesize compounds were tested for the preliminary tests, physical constants and TLC. All structures of final compound were confirmed by IR and ^{1}HNMR spectra as well as Mass spectra.

The pharmacological screening of the synthesized compounds showed anti-convulsant activity ranging from 55.25 % to 76.87 % inhibition of epileptic seizures in mice, where as the standard drug Phenytoin showed 83.95 % inhibition of epileptic seizures in mice.

The compound ivf, ivj was found to be nearly potent to Phenytoin which is used as standard drug. Compounds iva, ivb, ivc, ivd, ive, ivg, ivh and ivi shown less % of inhibition of epileptic seizures in mice than Phenytoin (standard drug).

REFERENCES:

1. http://en.wikipedia.org/wiki/seizures
2. Loring, David W (1 September 2005). "Cognitive Side Effects of Antiepileptic Drugs in Children". *Psychiatric Times* **XXII** (10).
3. https://www.neurology.wisc.edu/publications/2010%20pubs/stafstrom-4.pdf
4. Rogawski MA, Löscher W (July 2004). "The neurobiology of antiepileptic drugs".*Nature Reviews Neuroscience* **5** (7): 553–64.
5. Rogawski MA, Bazil CW (July 2008). "New molecular targets for antiepileptic drugs: alpha(2)delta, SV2A, and K(v)7/KCNQ/M potassium channels". *CurrNeurol Neurosci Rep* **8** (4): 345–52.
6. Meldrum BS, Rogawski MA (January 2007). "Molecular targets for antiepileptic drug development". *Neurotherapeutics* **4** (1): 18–61.
7. Kammerer, M.; Rassner, M. P.; Freiman, T. M.; Feuerstein, T. J. (2 May 2011). "Effects of antiepileptic drugs on GABA release from rat and human neocortical synaptosomes". *Naunyn-Schmiedeberg's Archives of Pharmacology* **384** (1): 47–57.
8. http://www.joacp.org/index.php?option=com_content&view=article&id=188&catid=1"According to the Washington Post who quoted research from IMS Health, AEDs were the fifth best selling class of drugs in the US in 2007, with sales topping 10 billion dollars. "
9. Kaminski, R. M.; Rogawski, M. A.; Klitgaard, H (2014). "The potential of antiseizure drugs and agents that act on novel molecular targets as antiepileptogenic treatments". *Neurotherapeutics* **11** (2): 385–400.
10. Abou-Khalil BW (2007). "Comparative Monotherapy Trials and the Clinical Treatment of Epilepsy". *Epilepsy currents / American Epilepsy Society* **7** (5): 1279.
11. Plosker, GL (November 2012). "Stiripentol : in severe myoclonic epilepsy of infancy (dravet syndrome)". *CNS Drugs* **26** (11): 993–1001.
12. "Public summary of positive opinion for orpphan opinion for orphan designation of stiripentol for the treatment of severe myoclonic epilepsy in infancy". European Medicines Agency. 30 July 2007. Retrieved 19 May 2013. "Doc.Ref: EMEA/COMP/269/04"
13. Browne TR (May 1976). "Clonazepam .A review of a new anticonvulsant drug". *Arch Neurol* **33** (5): 326–32.
14. Isojärvi, JI; Tokola RA (December 1998). "Benzodiazepines in the treatment of epilepsy in people with intellectual disability". *J Intellect Disabil Res* **42** (1): 80–92.
15. Tomson T; Svanborg E, Wedlund JE (May–Jun 1986). "Nonconvulsive status epilepticus". *Epilepsia* **27** (3): 276–85.
16. ZarghiAfshin, HamediSamaneh, Tootoonifatemeh, "Synthesis and Pharmacological Evaluation of New 2-Substituted-5-{2-[(2-halobenzyl)thio)phenyl}- 1,3,4-oxadiazoles as Anticonvulsant Agents.", 185-201.
17. .Zarghi A, Tabatabai SA, faizi, ahadian A. "synthesis anticonvulsant activity of new 2-substituted benzyloxyphenyl -1,3,4-oxadiazoles,*bioorg lett*. 15: 1863-1865, (2005).
18. Almasirad A, tabatabai SA, faizi M, " synthesis& anticonvulsant activity of new 2-substituted-5-[2-(2-fluorophenoxy)phenyl]-1,3,4-oxadiazole & 1,2,4-triazoles, *Bioorg med. Chem. Lett.* 14: 6057-6059.
19. Tripathi K.D., "essential of medical pharmacology, 5th edition, 2003, page no.167, 627, 759, 698.