

Synthesis and Anticonvulsant Evaluation of 3-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazol-3-ylamino)-2-(2-methylphenyl)quinazolin-4(3H)-one Derivatives

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ABSTRACT

Epilepsy arise due to discharge of electric current in CNS and it is characterized by repeated seizure because of different factors like social, neurological and environmental or it may be due to genetic or non-genetic. A large number of AED's used to treat epilepsy but all these shows drug resistance and side effects, so research interest continue to find out novel antiepileptic drugs with higher efficiency and less toxicity. A novel series of 3-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazole-3-ylamino)-2-(2-methylphenyl)quinazolin-4(3H)-one was developed, synthesized and evaluated for anticonvulsant activity using two pharmacological models, maximal electroshock seizure (MES) and subcutaneous pentylenetetrazole (scPTZ) models. Spectral data and elemental analysis were used to validate the structure of the synthetic compounds. Synthesized substances have also been tested for their neurotoxicity by rotary apparatus. Both compounds display strong anticonvulsant and neurotoxic activity. 3-(5-(4-fluorophenyl)-4,5-dihydro-1H-pyrazole-3-ylamino)-2-(2-methylphenyl)quinazolin-4(3H)-one, 8(v) was found to be the most successful in maximal electroshock seizure (percentage protection = 73.63 at 150 mg/kg) and subcutaneous pentylenetetrazole induced convulsion model (percentage protection = 75.59 at 150 mg/kg) models and was found to be non-neurotoxic.

Key words: AED's, Quinazolin-4-(3H)-one, Anticonvulsant, MES, scPTZ, Neurotoxicity.

INTRODUCTION

Epilepsy arise due to discharge of electric current in central nervous system and it is characterized by recurrent seizures because of different factors like social, neurological and environmental or it may be due to genetic or non-genetic. 50 million of world population suffering from epilepsy¹ out of which 0.5% population reside in India.² Uncontrolled epilepsy has been linked with excess mortality,³ Cognitive - behavioral dysfunctions⁴ and public and academic drawback.⁵

Anti-epileptic drugs (AEDs) are by far the most popular treatment approved by the World Health Organisation. Immense AEDs, hailing from different groups, are commonly used to control seizures, like phenytoin, carbamazepine, valproic acid, topiramate, gabapentin, felbamate, levetiracetam, etc.⁶ A significant percentage of epileptic patients have acceptable AED enhancements, but they represent drawbacks such as insufficient seizure regulation, unpredictability of affect and failure, low effectiveness, inadequate knowledge on drug

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receptors, etc. and the most important downside is the possibility of drug resistance. Anti-epileptics with better sensitivity and no adverse effects are the most common research area in the field in medicinal chemistry.

Current findings into heterocyclic nitrogen-containing compounds are of considerable importance owing to their utility and a substantial number of therapeutic molecules have been produced with higher efficacy for treatment.⁷ Quinazoline is a combined heterocyclic compound of benzene and pyrimidine which include couple of nitrogen atoms at two consecutive places. This has a broad spectrum of biological activities including anticancer,^{8,9} anti-tumor,¹⁰ antibacterial,¹¹ antifungal,¹² antiviral,¹³ anti-inflammatory,¹⁴ antidepressant,¹⁵ anticonvulsant,¹⁶ etc. Therapeutically known quinazoline-derived compounds include prazosin, gefitinib, erlotinib, tetrodotoxin, alfuzosin, etc.¹⁷ Methaqualone, a quinazoline-containing drug, was first described as sedative hypnotic. Methaqualone encouraged researchers to concentrate on the quinazoline nucleus for the development of novel anti-epileptic drug molecules by various substitutions in specific places. The second and third places of quinazoline are responsible for their anticonvulsant actions,¹⁸ In addition, four pharmacophoric features, i.e. a lipophilic aryl ring, an electron donating molecule, a hydrogen bond group and a hydrophobic-hydrophilic ring, are also crucial for improving their effectiveness.¹⁹⁻²¹

The existing investigation was intended at developing and synthesizing quinazoline derivatives through second and third alteration. Both compounds were tested for anticonvulsant action using the MES²² (Maximal electroshock seizures) and scPTZ²³ (Subcutaneous pentylenetetrazole) models. Derivatives were also tested for their neurotoxicity by the rota rod test.²⁴ The layout and synthetic framework for quinazoline derivatives were shown in Figure 1.

MATERIALS AND METHODS

Here all chemicals and reagents utilized were of a business contingency of laboratory grade obtained from Sigma-Aldrich and Merck and have been used without further detoxification.

Experimental Chemistry

Title compounds 8(i) – 8(xi) have been synthesized in accordance with Scheme 1. 2-Aminobenzoic acid 1 (0.1 mol) was reacted with substituted 2-methyl benzoylchloride 2c (0.2 mol) in the presence of pyridine at a temperature of 0-5°C to produce the first intermediate 2-(2-methyl-4*H*-benzo[d][1,3]oxazin-4-one (3c) which

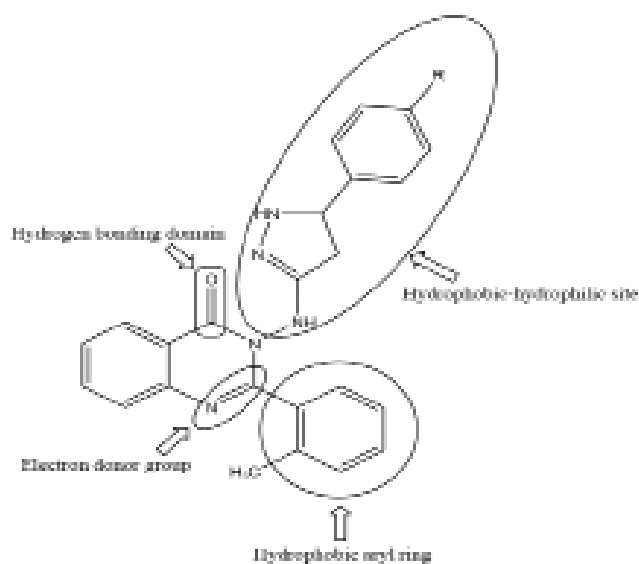
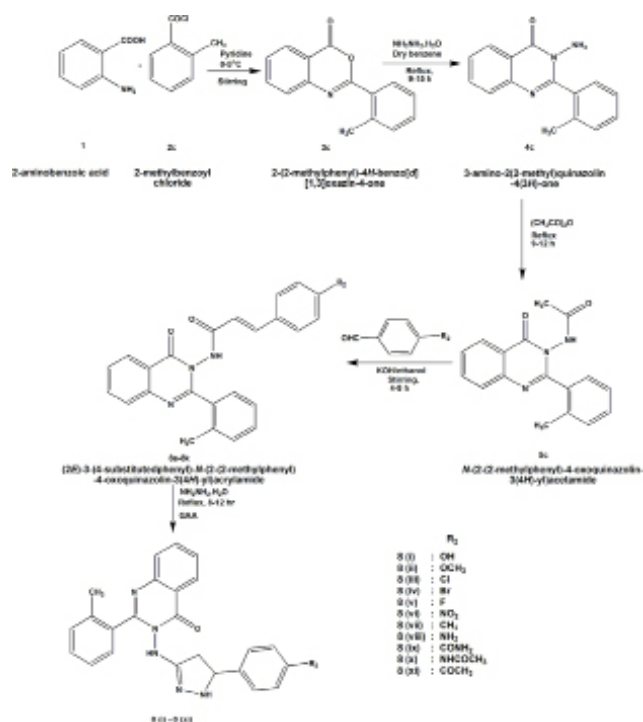


Figure 1: Graphic description of N-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazole-3-ylamino)-2-(2-methylphenyl)quinazolin-4(3H)-one derivatives and their pharmacophoric properties required for anticonvulsant action.



Scheme 1: Synthesis of 3-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazol-3-ylamino)-2-(2-methylphenyl)quinazolin-4(3H)-one.

on further reaction with hydrazine hydrate (0.01 mol) led to the formation of next intermediate 3-amino-2-(2-methylphenyl)quinazolin-4(3*H*)-one (4c). Reaction of compound (4c) (0.01 mol) with acetic anhydride (0.01 mol) in the presence of hydrated sodium acetate and HCl resulted in the formation of the *N*-(2-(2-methylphenyl)-4-oxoquinazolin-3(4*H*)-yl)acetamide (5c). Compound

5c on further reaction with appropriately substituted benzaldehydes led to the synthesis of compounds (8a-8k) which on further cyclization with hydrazine hydrate (0.01 mol) yielded the corresponding final compounds, 3-(5-(4-substituted phenyl)-4,5-dihydro-1H-pyrazol-3-ylamino)-2-(2-methylphenyl)quinazolin-4(3H)-ones [8 (i) – 8 (xi)]. All the synthesized compounds were recrystallized using ethanol.¹

Characterization of both intermediate and final compounds was carried out using various physico-chemical and spectral techniques. The melting point of all the final compounds was reported on the digital melting point apparatus (Kshitij Innovation) using single-end open capillary tubes and is not corrected. The success of the reaction and the purity of the intermediate and final compounds has been confirmed by the thin layer chromatography using silica gel G plates. The FTIR spectra was reported in the SHIMAZDU IR affinity spectrophotometer using the KBr pellet process. The ¹H NMR was reported in the BRUKER FOURIER 300 HD spectrophotometer using CDCl₃ as a solvent. TMS has been used as an internal standard. Mass spectra were recorded on the Varian 1200L mass spectrometer. The data was expressed as values (M+1)⁺. The elementary

analysis (C, H, N) was determined using Carlo-Erba 1160 elemental analyzer. The C, H, N values for the compounds were found to be in the range of ±0.4 percent. The physicochemical and spectral characterization data for the final compounds 8 I – 8 (xi) were provided in Table 1 and Table 2 respectively.

Pharmacological Evaluation

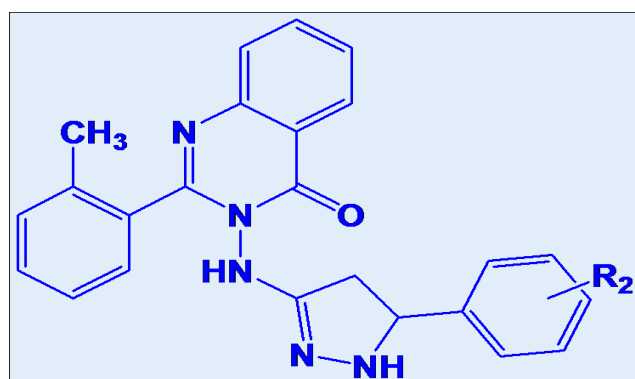
All of the experimental treatments were carried out in compliance with CPCSEA guidelines and licensed by the foundation's IAEC (1205 / c/08 / CPCSEA).

Animals

The entire study used Swiss albino mice weighing between 18-25 g. To order to acclimatize them at laboratory levels, the animals were kept to individual cages for a week. They were given free access to water and food.

Anticonvulsant activity

Final compounds 8 I – 8 (xi) were assessed for anticonvulsant activity that used the maximum electroshock seizure (MES) and subcutaneous pentylenetetrazole (scPTZ) models.^{25,26}



8 (i) – 8 (xi)

MES

The animals were categorized into three groups consisting of six animals each. Testing, control and standard. Both research substances were diluted in 1% CMC solution and delivered intraperitoneally to animals at three doses of 25 mg / kg, 75 mg / kg and 150 mg / kg; 30 min prior to seizures. The reference compound used was phenytoin (25 mg / kg). Health animals got a saline solution of 0.9 per cent. Using an electroconvulsion meter, mice were treated with a 50 mA electroshock for 0.2 s by the couple of ear clip electrodes and the seizure latency was

Table 1: Physico-chemical characterization data for final compounds 8 (i) – 8 (xi).

Comp. Code.	R	Mol. Formula	Mol. Wt.	M. P. (°C)	% Yield
8 (i)	-OH	C ₂₄ H ₂₁ N ₅ O ₂	411.46	200-205	55
8 (ii)	-OCH ₃	C ₂₅ H ₂₃ N ₅ O ₂	425.48	195-199	43
8 (iii)	-Cl	C ₂₄ H ₂₀ ClN ₅ O	429.9	210-214	65
8 (iv)	-Br	C ₂₄ H ₂₀ BrN ₅ O	474.35	187-193	54
8 (v)	-F	C ₂₄ H ₂₀ FN ₅ O	413.45	205-210	45
8 (vi)	-NO ₂	C ₂₄ H ₂₀ N ₆ O ₃	440.45	199-204	61
8 (vii)	-CH ₃	C ₂₅ H ₂₃ N ₅ O	409.48	167-172	59
8 (viii)	-NH ₂	C ₂₄ H ₂₂ N ₆ O	410.47	203-208	50
8 (ix)	-CONH ₂	C ₂₅ H ₂₂ N ₆ O ₂	438.48	211-216	51
8 (x)	-NHCOCH ₃	C ₂₆ H ₂₄ N ₆ O ₂	452.51	206-209	56
8 (xi)	-COCH ₃	C ₂₅ H ₂₂ N ₅ O ₂	424.47	214-218	67

Table 2: Spectral characterization data of the final compounds 8 (i) – 8 (xi).

Comp. Code	IR (KBr, cm ⁻¹)	¹ H NMR (CDCl ₃ , δ in ppm)	Mass (m/e) [M+1] ⁺	Elemental analysis	
				Calculated	Observed
8 (i)	3550 (O-H str), 3452 (N-H str), 3090 (Ar C-H str), 2965 (C-H Aymm. str.), 1690 (C=O str), 1550 (C=C str), 1620 (C=N)	7.4-7.9 (m, 4H, ArH), 7.0 (s, 1H, N-H), 7.13-7.55 (m, 4H, ArH), 2.3 (s, 1H, N-H), 6.5 (s, 1H, N-H), 6.68-6.95 (m, 4H, ArH), 5.0 (s, 1H, -OH), 2.35 (s, 3H, -CH ₃).	412.17	C (70.06%) H (5.14%) N (17.02%) O (7.78%)	C (70.34%) H (5.11%) N (17.45%) O (7.56%)
8 (ii)	3449 (N-H str), 3070 (Ar C-H str), 2950 (C-H Aymm. str.), 1660 (C=O str), 1450 (C=C str), 1612 (C=N), 1060 (C-O-C str), 2835 (O-CH ₃ str).	7.2-7.7 (m, 4H, ArH), 7.6 (s, 1H, N-H), 7.01-7.45 (m, 4H, ArH), 2.8 (s, 1H, N-H), 7.3 (s, 1H, N-H), 6.72-7.01 (m, 4H, ArH), 3.74 (s, 1H, -CH ₃), 2.39 (s, 3H, -CH ₃).	426.19	C (70.57%) H (5.45%) N (16.46%) O (7.52%)	C (70.63%) H (5.23%) N (16.57%) O (7.63%)
8 (iii)	3452 (N-H str), 3100 (Ar C-H str), 2970 (C-H Aymm. str.), 1655 (C=O str), 1590 (C=C str), 1616 (C=N), 750 (-Cl).	7.8-7.13 (m, 4H, ArH), 9.0 (s, 1H, N-H), 7.09-7.50 (m, 4H, ArH), 3.0 (s, 1H, N-H), 7.4 (s, 1H, N-H), 7.08-7.24 (m, 4H, ArH), 2.4 (s, 3H, -CH ₃).	430.14	C (67.05%) H (4.69%) N (16.29%) O (3.72%) Cl (8.25)	C (67.45%) H (4.87%) N (16.45%) O (3.54%) Cl (8.67%)
8 (iv)	3450 (N-H str), 3085 (Ar C-H str), 2960 (C-H Aymm. str.), 1675 (C=O str), 1555 (C=C str), 1611 (C=N), 690 (-Br).	7.7-7.12 (m, 4H, ArH), 8.4 (s, 1H, N-H), 7.14-7.57 (m, 4H, ArH), 2.0 (s, 1H, N-H), 7.8 (s, 1H, N-H), 7.01-7.17 (m, 4H, ArH), 2.30 (s, 3H, -CH ₃).	475.08	C (60.77%) H (4.25%) N (14.76%) O (3.37%) Br (16.84%)	C (60.89%) H (4.33%) N (14.43%) O (3.64%) Br (16.95%)
8 (v)	3348 (N-H str), 3090 (Ar C-H str), 2952 (C-H Aymm. str.), 1680 (C=O str), 1465 (C=C str), 1614 (C=N), 1050 (-F).	7.1-7.6 (m, 4H, ArH), 8.1 (s, 1H, N-H), 7.0-7.41 (m, 4H, ArH), 2.2 (s, 1H, N-H), 8.0 (s, 1H, N-H), 7.11-7.27 (m, 4H, ArH), 2.40 (s, 3H, -CH ₃).	414.17	C (69.72%) H (4.88%) F (4.60%) N (16.94%) O (3.87%)	C (69.89%) H (4.45%) F (4.71%) N (16.34%) O (3.91%)
8 (vi)	3450 (N-H str), 3090 (Ar C-H str), 2965 (C-H Aymm. str.), 1670 (C=O str), 1490 (C=C str), 1625 (C=N), 1500 (Ar-NO ₂ str)	7.3-7.8 (m, 4H, ArH), 7.8 (s, 1H, N-H), 4.0 (s, 1H, N-H), 2.37 (s, 3H, C-H), 2.30 (t, 3H, C-H)	441.16	C (63.15%) H (4.42%) N (18.41%) O (14.02%)	C (63.38%) H (4.58%) N (18.21%) O (14.34%)
8 (vii)	3445 (N-H str), 3110 (Ar C-H str), 2954 (C-H Aymm. str.), 1665 (C=O str), 1550 (C=C str), 1615 (C=N),	7.2-7.7 (m, 4H, ArH), 7.6 (s, 1H, N-H), 7.01-7.45 (m, 4H, ArH), 2.8 (s, 1H, N-H), 7.3 (s, 1H, N-H), 6.72-7.01 (m, 4H, ArH), 3.74 (s, 1H, -CH ₃), 2.39 (s, 3H, -CH ₃)	410.19	C (73.33%) H (5.66%) N (17.10%) O (3.91%)	C (73.03%) H (5.86%) N (17.19%) O (3.80%)
8 (viii)	3455 (N-H str), 3080 (Ar C-H str), 2960 (C-H Aymm. str.), 1660 (C=O str), 1580 (C=C str), 1614 (C=N), 3510 (Ar-NH str).	7.75-7.98 (m, 4H, ArH), 7.3 (s, 1H, N-H), 2.6 (s, 1H, N-H), 2.45 (s, 3H, C-H), 6.0 (d, 2H, N-H).	411.19	C (67.59%) H (5.20%) N (19.71%) O (7.50%)	C (67.46%) H (5.13%) N (19.53%) O (7.62%)
8 (ix)	3460 (N-H str), 3075 (Ar C-H str), 2975 (C-H Aymm. str.), 1690 (C=O str), 1570 (C=C str), 1616 (C=N), 1645 (C=O amide str).	6.70-7.8 (m, 4H, ArH), 6.9 (s, 1H, N-H), 2.0 (s, 1H, N-H), 2.35 (s, 3H, C-H), 4.0 (d, 2H, N-H).	439.18	C (68.48%) H (5.06%) N (19.17%) O (7.30%)	C (68.30%) H (5.32%) N (18.99%) O (7.50%)
8 (x)	3450 (N-H str), 3095 (Ar C-H str), 2960 (C-H Aymm. str.), 1685 (C=O str), 1490 (C=C str), 1618 (C=N), 3510 (Ar-NH str)	6.6-7.9 (m, 4H, ArH), 7.2 (s, 1H, N-H), 2.0 (s, 1H, N-H), 2.35 (s, 3H, C-H), 8.0 (s, 1H, N-H), 2.02 (t, 3H, C-H).	453.20	C (66.65%) H (5.16%) N (17.94%) O (10.24%)	C (66.27%) H (5.25%) N (18.05%) O (10.0%)
8 (xi)	3465 (N-H str), 3120 (Ar C-H str), 2950 (C-H str.), 1677 (C=O str), 1550 (C=C str), 1620 (C=N), 3510(Ar-NH str). 1710 (C=O str).	6.75-7.85 (m, 4H, ArH), 7.3 (s, 1H, N-H), 2.0 (s, 1H, N-H), 2.35 (s, 3H, C-H), 2.55 (s, 3H, C-H).	438.19	C (68.86%) H (5.11%) N (15.44%) O (10.58%)	C (68.92%) H (5.09%) N (15.32%) O (11.06%)

Table 3: Preliminary anticonvulsant screening data of final compounds 8 (i) – 8 (xi) in MES model.

Comp. Code	R ²	Dose (mg/kg)	Onset convulsion threshold (Mean± SEM)	% Protection
8 (i)	-OH	25	3.63±0.48 ^{ns}	38.84
		75	4.36±0.37 ^{ns}	49.08
		150	7.31±0.42 ^{***}	69.63
8 (ii)	-OCH ₃	25	3.03±0.18 ^{ns}	26.73
		75	4.11±0.33 ^{ns}	45.99
		150	6.92±0.57 ^{***}	67.92
8 (iii)	-Cl	25	4.34±0.73 ^{ns}	48.85
		75	5.11±0.93 ^{**}	56.56
		150	8.26±1.04 ^{***}	73.12
8 (iv)	-Br	25	4.13±0.57 ^{ns}	46.25
		75	5.01±0.43 ^{**}	55.69
		150	8.1±1.40 ^{***}	72.59
8 (v)	-F	25	8.42±0.65 ^{***}	50.88
		75	5.31±0.70 ^{**}	58.19
		150	4.52±0.28 [*]	73.63
8 (vi)	-NO ₂	25	3±0.18 ^{ns}	26.00
		75	4.06±0.36 ^{ns}	45.32
		150	6.18±0.55 ^{***}	64.08
8 (vii)	-CH ₃	25	2.99±0.39 ^{ns}	25.75
		75	2.36±0.22 ^{ns}	5.93
		150	3±0.14 ^{ns}	26.00
8 (viii)	-NH ₂	25	3.25±0.33 ^{ns}	31.69
		75	4.15±0.49 ^{ns}	46.51
		150	7.11±0.45 ^{***}	68.78
8 (ix)	-CONH ₂	25	4.04±0.31 ^{ns}	45.05
		75	4.95±0.43 ^{**}	55.15
		150	7.71±1.31 ^{***}	71.21
8 (x)	-NHCOCH ₃	25	2.56±0.24 ^{ns}	13.28
		75	3.11±0.19 ^{ns}	28.62
		150	6.23±0.22 ^{***}	64.37
8 (xi)	-COCH ₃	25	2.41±0.24 ^{ns}	7.88
		75	3.13±0.25 ^{ns}	29.07
		150	6.21±0.26 ^{***}	64.25
Phenytoin (Std.)		25	12.23±1.38 ^{***}	81.85

Level of statistical significance was calculated using Bonferroni's Post test. *P < 0.05, **P < 0.01, ***P < 0.001. All values represented in MEAN ± SEM; n = 6

recorded. The findings of the MES examination for 8 I – 8 (xi) were reported in Table 3.

scPTZ model

The experimental animals were grouped into separate classes, initially handled and controlled in the same manner as seen in the MES process. After 30 min, the animals were treated subcutaneously with a convulsive dosage of PTZ (60 mg / kg). Sodium valproate (200 mg / kg) was used as a reference as p. Upon

administration of PTZ, observations were made 60 min. The findings of the scPTZ model for the synthesized compounds 8 I – 8 (xi) are displayed in Table 4.

Neurotoxicity screening

The substances were tested using rotarod equipment for reduced movement disability. The animals were taught to sit at a pace of 6 rpm to propel rotarod (rod diameter: 3.2 cm). Gave the educated animals an i. Check compound injection p. (Dose: 150 mg / kg). In each

Table 4: Preliminary anticonvulsant screening data of final compounds 8 (i) – 8 (xi) in scPTZ model.

Comp. Code	R ²	Dose (mg/kg)	Clonic convulsion threshold (Mean± SEM)	% Protection
8 (i)	-OH	25	25.37±0.39 ^{ns}	50.34
		75	37.39±2.03 _{ns}	66.30
		150	46.84±1.17 ^{***}	73.10
8 (ii)	-OCH ₃	25	23.84±1.81 ^{ns}	47.15
		75	35.74±0.74 ^{ns}	64.75
		150	44.55±1.62 ^{***}	71.72
8 (iii)	-Cl	25	29.39±2.04 ^{ns}	57.13
		75	40.85±0.44 ^{**}	69.16
		150	50.63±0.97 ^{***}	75.11
8 (iv)	-Br	25	27.52±1.55 ^{ns}	54.22
		75	39.82±1.37 ^{**}	68.36
		150	49.35±1.25 ^{***}	74.47
8 (v)	-F	25	30.73±1.40 ^{***}	59.00
		75	42.71±1.35 ^{**}	70.50
		150	51.62±0.96 [*]	75.59
8 (vi)	-NO ₂	25	22.17±1.29 ^{ns}	43.17
		75	34.84±2.21 ^{ns}	63.83
		150	43.45±1.50 ^{***}	71.00
8 (vii)	-CH ₃	25	18.85±1.48 ^{ns}	33.16
		75	30.73±1.11 ^{ns}	59.00
		150	37.74±1.79 ^{ns}	66.61
8 (viii)	-NH ₂	25	24.17±1.15 ^{ns}	47.87
		75	36.26±1.52 ^{ns}	65.25
		150	45.82±1.54 ^{***}	72.50
8 (ix)	-CONH ₂	25	26.17±1.34 ^{ns}	51.85
		75	38.84±0.40 ^{**}	67.56
		150	47.72±1.28 ^{***}	73.60
8 (x)	-NHCOCH ₃	25	21.98±1.06 ^{ns}	42.68
		75	33.34±0.93 ^{ns}	62.21
		150	42.24±1.73 ^{***}	70.17
8 (xi)	-COCH ₃	25	20.61±1.01 ^{ns}	38.86
		75	32.32±2.45 ^{ns}	61.01
		150	41.12±1.08 ^{***}	69.36
Sodium valproate (Std.)		200	140.3±21.26 ^{***}	91.02

Level of statistical significance was calculated using Bonferroni's Post test. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$. All values represented in MEAN ± SEM; $n = 6$

of the three consecutive experiments, neurotoxicity was demonstrated by the animals' ability to sustain equilibrium on the rod for at least one minute. The animals were checked at intervals of four days, respectively. 0.5 hr, 1 hr, 2 hr and 4 hr. The results were tabulated in Table 5 as tabulated.

The data indicates number of animals toxic/total number of animals used.

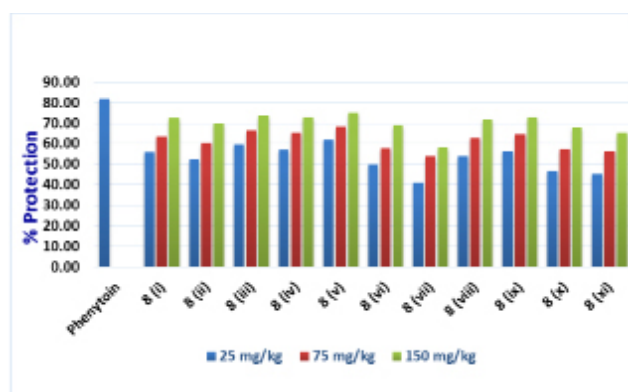
RESULTS AND DISCUSSION

Chemistry

The synthesis of the title compounds 8 I – 8 (xi) was performed according to the reaction sequence described in Scheme 1. FTIR, ¹H NMR, mass spectrometry and basic research examined the intermediates and final compounds. The spectral data obtained were found to be in keeping with the synthesized compound structures.

Table 5: Neurotoxicity screening data for the final compounds 8 (i) – 8 (xi).

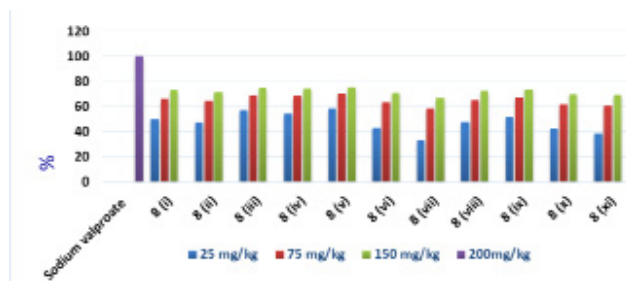
Comp. Code	0.5 h	1 h	2 h	4 h
8 (i)	0/4	0/4	0/4	0/4
8 (ii)	0/4	0/4	1/4	0/4
8 (iii)	0/4	0/4	0/4	0/4
8 (iv)	0/4	0/4	0/4	0/4
8 (v)	0/4	0/4	0/4	0/4
8 (vi)	0/4	0/4	0/4	0/4
8 (vii)	0/4	¼	0/4	2/4
8 (viii)	0/4	¼	2/4	¾
8 (ix)	0/4	0/4	0/4	0/4
8 (x)	0/4	¼	1/4	0/4
8 (xi)	0/4	0/4	1/4	2/4
Phenytoin	0/4	0/4	0/4	0/4

**Figure 2: MES Model-Comparison of percentage protection of all synthesized compounds 8 (i) – 8 (xi) at different test doses.**

Anticonvulsant activity

Maximal electroshock seizure Test

The early findings of the MES screening were expressed in percentage protection format and as Table 3. All compounds at a dosage of 25 mg / kg provided a percentage safety of about 50 percent except for compound 8 (vii). All compounds display percentage security between 60%-75% except compounds 8(vi), 8(x) and 8(xi) showing percentage security < 60%. At 75 mg / kg, compounds 8 (iii), 8 (iv), 8 (v) and 8 (ix) displayed a percentage protection of 70 percent with the exception of compounds 8 (x) and 8 (xi) showing a percentage protection of less than 50 percent. The majority of the compounds displayed safety in the range of 55% -70%. All compounds at a dose of 150 mg / kg showed a percentage protection of about 60% except compounds 8 (xi). The compounds 8 (v) show maximum percentage protection (75.97). The similarity of the upper

**Figure 3: Comparison of percentage protection of final compounds at different doses in scPTZ model.**

limit of onset convulsion and percentage protection of the synthesized compounds was as shown in Figure 2.

Subcutaneous pentylenetetrazole test

Table 4 describes the initial scPTZ screening findings in percentage safety. At a dosage of 25 mg / kg, all compounds displayed a percentage safety of 50 percent with the exception of compounds 8(vii) and 8(xi) and 8(i), 8(iii), 8(iv), 8(v), 8(vi) and 8(ix). The majority of the compounds had protection percentage varying from 50 to 60 percent. Both compounds at a dosage of 75 mg / kg provided a percentage safety of about 60% except compounds 8(vi), 8(vii) and 8(xi). Compound 8 (ix), 8 (v), 8 (iv), 8 (iii) and 8 (ii) displayed 65 percent safety and 60 – 65 percent protection for the rest of the compounds. All compounds at a dosage of 150 mg / kg provided a percentage safety of about 70 percent excluding compounds 8 (vii) and 8 (xi). Compounds 8 (iii) showed 75.41 per cent maximum protection. Comparison of the cutoff point of clonic convulsion and percentage protection of the final compounds was as shown in Figure 3.

Neurotoxicity screening

The test findings for neurotoxicity are as shown in Table 5. We considered both substances to be non-neurotoxic at 0.5 h. Compounds 8 (vii), 8 (viii) and 8 (x) were neurotoxic at 25 per cent and neurotoxic compounds at 1 h. Compounds 8 (ii), 8 (x) and 8 (xi) were 25% neurotoxic and compounds 8 (viii) were 50% neurotoxic at 2 h. Compounds 8 (vii) and 8 (xi) showed 50 per cent and compound 8 (viii) demonstrated neurotoxicity of 75 per cent at 4 h. Compounds 8 (v), 8 (iii), 8 (iv), 8 (vi) and 8 (ix) is non-neurotoxic at all time intervals.

Fluorine aryl substitution showed the maximum anticonvulsant effect in comparison to other substituents. This may be due to fluorine atom is highly electro negative than all other substituent. Compounds which are very less active, has electropositive substituent.

CONCLUSION

In the present study synthesis of sequence or series of 3-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazole-3-ylamino)-2-(2-methylphenyl)quinazole-4(3H)-one tested for anticonvulsant behavior using MES and scPTZ models has been recorded. The Neurotoxicity assessment was tested by the rotary unit. The compounds displayed strong anticonvulsant action. Synthesized compounds 3-(5-(4-fluorophenyl)-4,5-dihydro-1H-pyrazol-3-ylamino)-2-(2-methylphenyl)quinazolinone-4(3H)-one, 8 (v) were found to be the most active in both MES (percentage protection = 73.63 at 150 mg / kg) and scPTZ (percentage protection = 75.59 at 150 mg / kg) models and were considered as non-neurotoxic.

All the compounds displayed moderate to good anticonvulsant activity. Compounds with noteworthy activity can be evaluated for their therapeutic effectiveness using different dose concentrations and can be studied for possible mechanism of action which in turn will be helpful to estimate possible toxicological and safety profile for future investigations.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

ABBREVIATIONS

CNS: Central Nervous System; **AED's:** Anti-epileptic Drugs; **MES:** Maximal Electroshock Seizure; **scPTZ:** Subcutaneous Pentylene Tetrazole; **FTIR:** Fourier Transform Infra-Red spectroscopy; **NMR:** Nuclear Magnetic Resonance; **CPCSEA:** Committee for the Purpose of Control and Supervision of Experiments on Animals; **CMC:** Carboxy Methyl Cellulose; **SEM:** Standard Error Mean; **IAEC:** Institutional animal ethical committee; **%:** Percentage; **Mol. Wt.:** Molecular weight; **Mol. Formula:** Molecular Formula; **IR:** InfraRed spectroscopy; **M. P.:** Melting Point; **TLC:** Thin Layer Chromatography.

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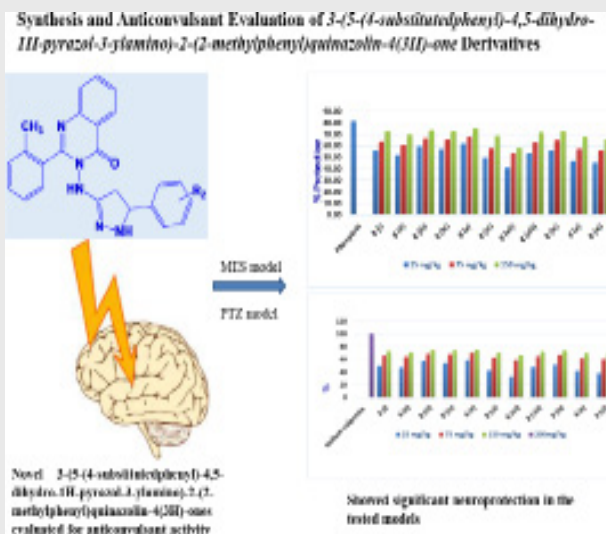
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SUMMARY

- Novel 3-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazol-3-ylamino)-2-(2-methylphenyl) quinazolin-4(3H)-one derivatives of quinazolines were synthesised and evaluated for their physico-chemical characteristics including structural elucidation through various analytical studies i.e. FTIR spectrophotometry, ¹HNMR spectroscopy, elemental analysis, m.p. determination etc.
- The syntheses were carryout through the reaction of Anthranilic acid with benzoyl chloride leading to the formation of 2-phenyl-4H-benzo[d][1,3]oxazin-4-one (compound-1) which was fused with guanidine to produce 4-oxo-2-phenylquinazoline-3-(4H)-carboxamide (compound-2). Further compound-2 was reacted with suitably substituted aromatic or aliphatic aldehydes leading to the formation of different N-(substituted)-4-oxo-2-phenylquinazoline-3-(4H)-carboxamides (SKA-1 to 9). In the final step, the N-(substituted)-4-oxo-2-phenylquinazoline-3-(4H)-carboxamides were cyclised with thioglycolic acid in the presence of catalytic amount of ZnCl₂ leading to the formation of 3-(imino(4-oxo-2-substituted thiazolidin-3-yl)methyl)-2-phenyl quinazoline-4(3H)-ones (SKB -1 to 9).
- The structural elucidation of new compounds was established by the FTIR spectrophotometry and ¹HNMR spectroscopy. IR spectra were recorded on the SHIMAZDU Affinity-I by using potassium bromide (KBr) pellet method while ¹HNMR spectra were recorded on Bruker Advance 400 MHz in DMSO against tetramethylsilane (TMS) as internal standard.
- All synthesized compounds were screened for their Anti convulsant activities by MES (using diazepam as a standard drug) and PTZ induced (using diazepam as a standard drug) convulsive rat models.
- 3-(5-(4-substitutedphenyl)-4,5-dihydro-1H-pyrazole-3-ylamino)-2-(2-methylphenyl)quinazole-4(3H)-one tested for anticonvulsant behavior using MES and scPTZ models has been recorded. The Neurotoxicity assessment was tested by the rotary unit. The compounds displayed strong anticonvulsant action. Synthesized compounds 3-(5-(4-fluorophenyl)-4,5-dihydro-1H-pyrazol-3-ylamino)-2-(2-methylphenyl) quinazoline 4(3H)-one, 8 (v) were found to be the most active in both MES (percentage protection = 73.63 at 150 mg / kg) and scPTZ (percentage protection = 75.59 at 150 mg / kg) models and were considered as non-neurotoxic.

PICTORIAL ABSTRACT



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