Available online at www.sciencedirect.com
ScienceDirect
European Journal of Medicinal Chemistry 43 (2008) 135–141
EUROPEAN JOURNAL OF MEDICINAL CHEMISTRY
http://www.elsevier.com/locate/ejmech

Original article

Synthesis and CNS depressant activity of some novel 3-[5-substituted 1,3,4-thiadiazole-2-yl]-2-styryl quinazoline-4(3*H*)-ones

Varsha Jatav [a], Pradeep Mishra [a], Sushil Kashaw [a,*], J.P. Stables [b]

[a] *Pharmaceutical Chemistry Division, Department of Pharmaceutical Sciences, Dr. H.S.G. University, Sagar, M.P. 470003, India*
[b] *Preclinical Pharmacology Section, Epilepsy Branch, National Institutes of Health, Bethesda 20892-9020, USA*

Received 14 November 2006; received in revised form 11 February 2007; accepted 12 February 2007
Available online 25 February 2007

Abstract

A series of novel 3-[5-substituted phenyl-1,3,4-thiadiazole-2-yl]-2-styryl quinazoline-4(3*H*)-ones were synthesized and evaluated for anticonvulsant, sedative-hypnotic and CNS depressant activities. After i.p. injection to mice at doses of 30, 100, and 300 mg/kg body weight 2-styrylquinazolin-4(3*H*)-one derivatives were examined in the maximal electroshock induced seizures (MES) and subcutaneous pentylenetetrazole (scPTZ) induced seizure models in mice. The neurotoxicity was assessed using the rotorod method. Out of eighteen compounds only **4a**, **4d**, **4e**, **4j** and **4k** showed anticonvulsant activity in one or more test models. All except **4e** and **4f** exhibited significant sedative-hypnotic activity via actophotometer screen. CNS depressant activity screened with the help of the forced swim pool method resulted into some potent compounds. From the experimental observation it can be concluded that synthesized compounds exhibited relatively better sedative-hypnotic and CNS depressant activities.

Keywords: 4(3*H*)-Quinazolinones; MES; Subcutaneous pentylenetetrazole induced seizure

1. Introduction

One of the most frequently encountered heterocycles in medicinal chemistry is 4(3*H*)-quinazolinone with wide applications including anticonvulsant, sedative, tranquilizer, analgesic, antimicrobial, anesthetic, anticancer, antihypertensive, anti-inflammatory, diuretic and muscle relaxant properties [1–4]. Literature survey revealed that the presence of substituted aromatic ring at position 3 and methyl group at position 2 are necessary requirement for the central nervous system (CNS) depression and anticonvulsant activities. In spite of the fact that literally hundreds of quinazolinones related to 2-methyl-3-*o*-tolyl-4(3*H*)-quinazolinone (methaqualone) have been synthesized and tested for central nervous system (CNS) depression and anticonvulsant activities, none of the drugs currently in use contain the 4(3*H*)-quinazolinone ring system. Among the few reports in the literature our attention was drawn to the earlier discovery by Boltze et al. [5] and Wolfe et al. [6] that modification of methyl group by some other chemical moiety yielded structural analogues with anticonvulsant activity. Medicinal chemists over the years have substituted different heterocyclic rings at position 3 of the 4(3*H*)-quinazolinone to get potent CNS acting drugs. 1,3,4-Thiadiazoles nucleus itself exhibits anticonvulsant, sedative-hypnotic and CNS neurotoxicity activities [7]. In hope of getting synergistic response of 4(3*H*)-quinazolinone nucleus itself, substitution of 1,3,4-thiadiazoles nucleus at third position and chemically modifying second position of 4(3*H*)-quinazolinone, the present paper reports on the synthesis, anticonvulsant, neurotoxicity, CNS depressant activity and behavioral study of 18 new 3-(1′3′4′thiadiazol-2-yl)-2-styrylquinazolin-4(3*H*)-ones.

2. Chemistry

The synthesis of 3-(1′3′4′-Thiadiazolyl)-2-methylquinazolin-4(3*H*)-one is accomplished as shown in Figs. 1 and 2.

* Corresponding author. Tel.: +91 7582240757; fax: +91 7582244094.
E-mail address: sushilkashaw@gmail.com (S. Kashaw).

0223-5234/$ - see front matter © 2007 Elsevier Masson SAS. All rights reserved.
doi:10.1016/j.ejmech.2007.02.004

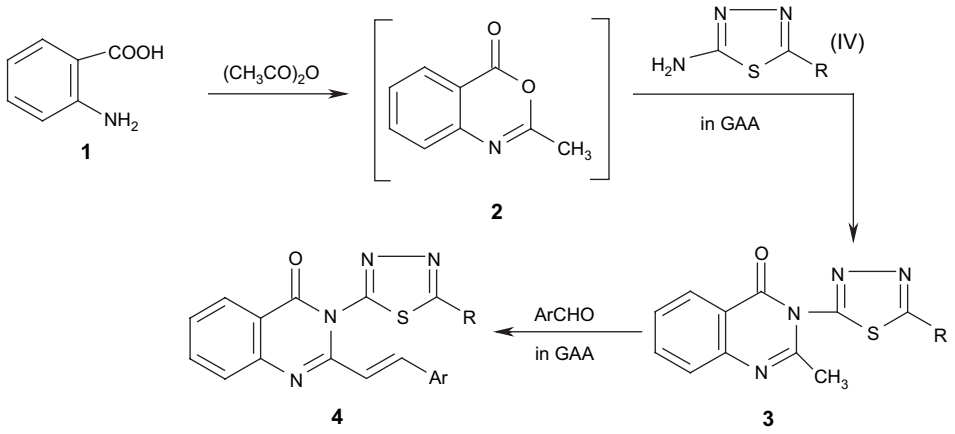


Fig. 1. Scheme for the synthesis of title compounds.

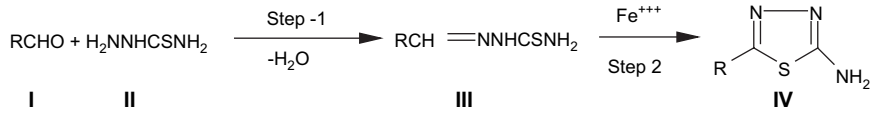
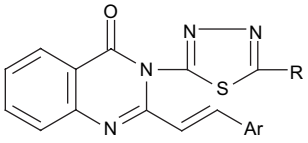


Fig. 2. Scheme for the synthesis of 2-amino-5-aryl-1,3,4-thiadiazoles.

3-(1′3′4′-Thiadiazolyl)-2-styrylquinazolin was synthesized by 3-step procedure, whereas 3-(1′3′4′-thiadiazolyl)-2-styryl-quinazolin **4** obtained by refluxing equimolar amount of 3-(1′3′4′-thiadiazolyl)-2-methyl quinazolin and aromatic aldehyde in glacial acetic acid. The structures of the new compounds were elucidated by analytical and spectroscopic measurements.

The IR spectra showed the C=O peak at 1701 cm^{-1}, C=O stretching at 1580 and C=C stretching (Alkene) vibration at 1630 cm^{-1}. The ^{13}C NMR depicted spectrum at (C-2) 162.4,

Table 1
Physical data of 3-[5-substituted 1,3,4-thiadiazole-2-yl]-2-styryl quinazoline-4(3*H*)-ones



Code No.	Ar	R	Yield (%)	M.p. (°C)	Molecular formula[a]	R_f	Log p[b]
4a	$-C_6H_5$	$-C_6H_5$	50	198	$C_{24}H_{18}N_4OS$	0.61	3.68
4b	$-C_6H_5$	p-$OCH_3C_6H_4$	48	200	$C_{25}H_{20}N_4O_2S$	0.66	3.73
4c	$-C_6H_5$	p-$CH_3C_6H_4$	47	208	$C_{25}H_{20}N_4OS$	0.71	4.17
4d	$-C_6H_5$	p-ClC_6H_4	42	220	$C_{24}H_{17}ClN_4OS$	0.84	4.39
4e	$-C_6H_5$	m-ClC_6H_4	43	220	$C_{24}H_{17}ClN_4OS$	0.81	4.39
4f	$-C_6H_5$	$-CH{=}CHC_6H_4$	25	206	$C_{26}H_{20}N_4OS$	0.58	3.87
4g	p-$OCH_3C_6H_4$	$-C_6H_5$	30	232	$C_{25}H_{20}N_4O_2S$	0.63	3.59
4h	p-$OCH_3C_6H_4$	p-$OCH_3C_6H_4$	35	230	$C_{26}H_{22}N_4O_3S$	0.59	3.65
4i	p-$OCH_3C_6H_4$	p-$CH_3C_6H_4$	30	226	$C_{26}H_{22}N_4O_2S$	0.58	4.09
4j	p-$OCH_3C_6H_4$	p-ClC_6H_4	32	>250[c]	$C_{25}H_{17}ClN_4O_2S$	0.85	4.31
4k	p-$OCH_3C_6H_4$	m-ClC_6H_4	38	>250[c]	$C_{25}H_{17}ClN_4O_2S$	0.82	4.31
4l	p-$OCH_3C_6H_4$	$-CH{=}CHC_6H_4$	27	225	$C_{27}H_{20}N_4O_2S$	0.79	3.87
4m	p-$CH_3C_6H_4$	$-C_6H_5$	30	206	$C_{25}H_{18}N_4OS$	0.75	4.17
4n	p-$CH_3C_6H_4$	p-$OCH_3C_6H_4$	33	226	$C_{26}H_{22}N_4O_2S$	0.76	4.23
4o	p-$CH_3C_6H_4$	p-$CH_3C_6H_4$	35	224	$C_{26}H_{20}N_4OS$	0.63	4.67
4p	p-$CH_3C_6H_4$	p-ClC_6H_4	38	212	$C_{25}H_{17}ClN_4OS$	0.81	4.89
4q	p-$CH_3C_6H_4$	m-ClC_6H_4	38	210	$C_{25}H_{17}ClN_4OS$	0.83	4.89
4r	p-$CH_3C_6H_4$	$-CH{=}CHC_6H_4$	28	242	$C_{27}H_{20}N_4OS$	0.78	4.37

[a] Elemental analyses for C, H, N were within ±0.4% of the theoretical value.
[b] Log p was generated using hyperchem.
[c] Melting point of the compound at their decomposition.

(C-4) 168.3, (C-11) 112 and (C-12) 136. Thin layer chromatography (TLC) was run throughout the reaction to optimize the reaction for purity and completion. The physicochemical data for the newly synthesized compounds are presented in Table 1.

3-(1′3′4′-Thiadiazolyl)-2-methyl quinazolinones [8–10] were obtained by refluxing 2-methylbenzoxazin-4(3*H*)-one **2** [11] with the amine derivative **IV** according to Fig. 1. The amino derivative **IV** [12–15] was obtained by the oxidative cyclization of thiosemicarbazone **III** (through condensation of aromatic aldehyde **I** and thiosemicarbazide **II**) in the presence of ferric chloride according to the scheme given in Fig. 2.

3. Pharmacology

The new derivatives obtained from the reaction sequence were injected intraperitoneally into mice and evaluated in the maximal electroshock (MES), subcutaneous pentylenetetrazole (scPTZ) and neurotoxicity screens, using doses of 30, 100 and 300 mg/kg at two different time intervals. These data are presented in Table 2. These compounds were also screened for their CNS behavioral activity in mice using actophotometer and Porsolt's swim pool test in rats and results are presented in Tables 3 and 4.

Table 2
Anticonvulsant activity and minimal motor impairment of 3-[5-substituted 1,3,4-thiadiazole-2-yl]-2-styryl quinazoline-4(3*H*)-ones

Code no.	Intraperitoneal injection in mice[a]					
	MES screen		scPTZ screen		Neurotoxicity screen	
	0.5 h	4 h	0.5 h	4 h	0.5 h	4 h
4a	30	–	–	–[b]	–	–
4b	–	–	–	–	–	–
4c	–	–	–	–	–	–
4d	100	100	300	300	–	–
4e	–	100	–	–	300[c]	–
4f	–	–	–	–	300	–
4g	–	–	–	–	100[d]	–
4h	–	–	–	–	300	–
4i	–	–	300	–	–	–
4j	–	300	300[e]	300	–	–
4k	–	300	–	–	–	–
4l	–	–	–	–	–	–
4m	–	–	–	–	–	–
4n	–	–	–	–	–	–
4o	–	–	–	–	–	––
4p	–	300	–	–	–	–
4q	–	–	300	300	–	–
4r	–	–	–	–	–	–
Phenytoin[f]	30	30	–	–	100	100
Carbamazepine[f]	30	100	100	300	100	300
Sodium vaporate[f]	–	–	300	–	–	–
Phenobarbitals[f]	100	30	30	300	100	300
Ethosuximide[f]	–	–	300	–	–	–

[a] Doses of 30, 100 and 300 mg/kg were administered. The figure in the table indicates the minimum dose whereby bioactivity was demonstrated in half or more of the mice. The animal was examined 0.5 and 4 h after injections were made the dash(–) indicates an absence of activity at maximum dose administered (300 mg/kg).
[b] Died during test at 300 mg/kg without seizure.
[c] Neurotoxicity at 100 mg/kg (0.25 h, 1 h).
[d] Loss of righting reflux.
[e] At 100 mg/kg after 0.25 h, 3/5 and after 1 h 4/5 mice were protected.
[f] Data from Refs. [9–11].

4. Results and discussions

Initial anticonvulsant activity and neurotoxicity data for the quinazolinone analogs are reported in Table 2, along with the literature data on phenytoin, carbamazepine, sodium valproate, phenobarbital and ethosuximide [16–18]. In this series all the quinazolinone analogs showed more potent sedative-hypnotic and CNS depressant activities than anticonvulsant activity. In the earlier reports it was highlighted that the presence of electron rich atom/group attached at the *para* position of the aryl ring showed increased potency in the MES screen [19,20]. Compounds **4a**, **4d**, **4e**, **4j**, **4k** and **4p** were found to exhibit anticonvulsant activity in MES screen, however, compound **4a** showed potency similar to standard drug (phenytoin, carbamazepine) without any neurotoxicity. All the synthesized compounds were active in MES screen for a long duration of time (after 4 h). Compound **4d** displayed activity in the MES screen after 0.5 h (100 mg/kg) and 4 h (100 mg/kg) while it was active at both 0.5 h (300 mg/kg) and 4 h (300 mg/kg) in the scPTZ test. This compound exhibited rapid onset of action and long duration of activity. Compounds **4b**, **4c**, **4f**, **4g**, **4h**, **4l**, **4m**, **4n**, **4o** and **4r** did not show any activity in MES as well as in scPTZ after 0.5 h. Compounds **4e**, **4f**, **4g** and **4h** showed neurotoxicity after 0.5 h at 300 mg/kg body weight.

Table 3
Behavioral study of 3-[5-substituted 1,3,4-thiadiazole-2-yl]-2-styryl quinazoline-4(3*H*)-ones

Compound[a]	Activity score[b]			% Inhibition
	Control (24 h prior)	Post treatment		
		0.5 h After	1 h After	
4a	559.32 ± 20.78	472.31 ± 26.78	249.45 ± 11.69	55
4b	416.04 ± 11.26	381.82 ± 9.69	266.36 ± 11.89	36
4c	442.24 ± 10.42	531.00 ± 17.22NS	467.00 ± 21.11NS	–
4d	418.10 ± 9.97	250.62 ± 10.30	132.04 ± 12.16	68
4e	454.16 ± 10.62	319.57 ± 9.66	165.52 ± 12.70	63
4f	454.72 ± 20.52	372.87 ± 2.21	489.92 ± 11.63NS	–
4g	467.63 ± 8.89	419.24 ± 6.12	210.24 ± 12.62	55
4h	442.82 ± 21.24	308.13 ± 7.25	180.30 ± 12.05	59
4i	398.46 ± 26.18	315.46 ± 4.56	142.19 ± 13.28	64
4j	410.52 ± 10.62	306.98 ± 21.13	155.31 ± 17.30	62
4k	324.76 ± 27.73	292.39 ± 29.87	110.45 ± 11.25	66
4l	468.62 ± 13.38	362.37 ± 11.02	228.32 ± 26.72	51
4m	420.86 ± 30.22	353.52 ± 6.77	185.15 ± 31.79	55
4n	397.92 ± 10.80	272.19 ± 2.11	137.19 ± 32.84	65
4o	328.16 ± 32.96	291.39 ± 1.02	92.20 ± 34.91	71
4p	435.72 ± 22.66	373.56 ± 2.35	153.08 ± 10.90	64
4q	376.34 ± 32.75	298.67 ± 7.35	128.94 ± 26.37	65
4r	384.52 ± 15.66	310.32 ± 2.44	123.32 ± 38.13	67
Phenytoin[c]	546.40 ± 31.12	251.02 ± 12.32	164.10 ± 30.11	70

[a] The compound were tested at a dose of 100 mg/kg (i.p.).
[b] Each score represents the mean ± SEM of six mice, significantly different from the control score at <0.02 and NS indicate not significant at $p > 0.02$ (student's t test).
[c] Tested at 25 mg/kg p.o.

Table 4
CNS study on 3-[5-substituted 1,3,4-thiadiazole-2-yl]-2-styryl quinazoline-4(3*H*)-ones by forced swim pool test

Compound[a]	Immobility time[b] (s)	
	Control (24 h prior)	Post treatment (60 min after)
PEG	174.67 ± 14.72	178 .53 ± 12.32 NS
4a	135.42 ± 9.87	200.30 ± 11.69
4b	97.12 ± 6.29	130.37 ± 10.32
4c	154.62 ± 11.23	172.00 ± 12.49NS
4d	88.65 ± 10.92	163.30 ± 12.06
4e	108.19 ± 7.31	197.60 ± 12.94
4f	125.63 ± 8.81	172.72 ± 11.62
4g	114.33 ± 7.24	120.83 ± 10.91NS
4h	57.66 ± 11.19	110.60 ± 12.72
4i	187.19 ± 10.68	208.72 ± 10.12
4j	101.04 ± 12.05	182.24 ± 11.24
4k	134.38 ± 13.29	179.07 ± 12.86
4l	107.97 ± 12.62	118.43 ± 11.10NS
4m	63.42 ± 12.16	120.56 ± 10.98
4n	119.37 ± 11.89	187.16 ± 12.36
4o	136.18 ± 11.69	210.70 ± 13.42
4p	49.10 ± 13.72	74.63 ± 13.63
4q	134.93 ± 17.30	198.05 ± 14.08
4r	172.17 ± 13.89	218.29 ± 10.93
Carbamazepine[c]	138.82 ± 15.09	240.30 ± 14.10

[a] The compounds were tested at a dose of 100 mg/kg (i.p.).
[b] Each value represent the mean ± SEM of six rats significantly different from the control at $p < 0.05$ and NS denotes not significant at $p < 0.05$ (student's t test).
[c] Tested at 30 mg/kg (i.p.).

The most active compound in the scPTZ test, a test used to identify compound that elevates seizure threshold, were **4j** and **4q**.

Experimental results indicated that our compound exhibited better sedative-hypnotic and CNS depressant activities as compared to anticonvulsant activity. All the compounds were screened for behavior study and CNS depressant activity. In the behavioral study using actophotometer scoring technique, compounds showed decrease in locomotor activity between 36% and 71% where 36% was the lowest and 71% was the maximal decrease in locomotor activity when compared to phenytoin as reported in Table 3. Compound **4o** was equipotent to phenytoin in decreasing % locomotor activity. All the compounds except **4b** exhibited more than 50% decrease in locomotor activity ($p < 0.02$) after 1 h. Generally compounds possessing higher log *p* value showed higher decrease in locomotor activity. Bulkier compounds are more lipophilic and can cross blood brain barrier to exert their effect at CNS. In a similar study using swim pool test, the immobility time after administration of the test compounds were compared with carbamazepine (Table 4). Except for **4c**, **4g** and **4l** other tested compounds were found to exhibit potent CNS depressant activity ($p < 0.05$) as indicated by increased immobility time.

Present study explored that substitution of 1,3,4-thiadiazoles at third position and styryl moiety at second position of 4(3*H*)-quinazolinone leads to the development of new chemical entities with potent sedative-hypnotic and CNS depressant activities as compared to anticonvulsant activity.

5. Experimental protocol

5.1. Chemistry

Melting points were determined in one end open capillary tubes on a Buchi 530 melting point apparatus and are uncorrected. Infrared (IR) spectra were recorded for the compounds on Perkin Elmer Spectrum RXI Spectrophotometer in KBr. ^{13}C nuclear magnetic resonance (^{13}C NMR) and ^{1}H nuclear magnetic resonance (^{1}H NMR) spectra were recorded for the compounds on Advance bruker (300 MHz) instrument. Chemical shifts are reported in parts per million (ppm) using tetramethylsilane (TMS) as an internal standard. Elemental analysis (C, N and S) was undertaken with Elemental vario EL III Carlo Erba 1108 analyzer. The purity of the compounds was confirmed by thin layer chromatography using silica gel glass plates and a solvent system of benzene:ethanol (8:2). The spots were developed in iodine chamber and visualized under ultra violet lamp. The log *p* values were determined using Hyperchem and Chemdraw software.

5.1.1. Synthesis of 2-amino 5-aryl 1′3′4′-thiadiazole [5–8]

2-Amino-5-aryl 1′3′4′-thiadiazole was synthesized following two steps.

Step 1: synthesis of thiosemicarbazones

Aromatic aldehyde **I** (0.2 M) in warm alcohol (300 mL) and thiosemicarbazide **II** (0.2 M) in warm water (300 mL) were mixed slowly with continuous stirring. The product separated immediately on cooling which was filtered with suction, dried and recrystallised in 75% ethanol to yield **III**. Physicochemical properties are presented in Table 5.

Table 5
Physico-chemical data of thiosemicarbazone R–CH=NNHCSNH$_2$

S. No.	R	Yield	Solvent recrystallize	M.P. (°C)	Molecular formula	Molecular weight	IR (cm^{-1})	^{1}H NMR (ppm)
1	C_6H_5	92%	Aq. ethanol (50%)	159	$C_8H_9N_3S$	179.25	3350 (NH_2), 3320 (NH)	2.0 (NH_2), 2.0 (NH)
2	*p*-$OCH_3C_6H_4$	85%	Ethanol	170	$C_9H_4N_3OS$	209.27	3362 (NH_2), 3315 (NH)	2.0 (NH_2), 2.0 (NH), 3.73 (CH_3)
3	*p*-$CH_3C_6H_4$	95%	Aq. ethanol (50%)	160	$C_9H_4N_3S$	193.27	3294 (NH_2), 3140 (NH)	2.0 (NH_2), 2.0 (NH), 2.35 (CH_3)
4	*p*-ClC_6H_4	90%	Ethanol	207	$C_8H_8ClN_3S$	213.69	3315 (NH_2), 3326 (NH)	2.0 (NH_2), 2.0 (NH)
5	*m*-ClC_6H_4	90%	Aq. ethanol (50%)	206	$C_8H_8ClN_3S$	213.69	3340 (NH_2), 3326 (NH)	2.0 (NH_2), 2.0 (NH)
6	-CH=CHC_6H_4 (1 2)	80%	Ethanol	185	$C_{10}H_{11}N_3S$	205.28	3350 (NH_2), 3321 (NH)	2.0 (NH_2), 5.6 (CH^1), 6.6 (CH^2)

Step-2: synthesis of 2-amino-5-aryl 1′3′4′-thiadiazoles

Thiosemicarbazone **III** (0.05 M) was suspended in 300 ml warm water, $FeCl_3$ (0.15 M) in 300 ml water was added quantitatively, slowly with constant stirring. The contents were heated at 80–90 °C for 45 min. Solution was filtered hot and then citric acid (0.11 M) and sodium citrate (0.05 M) were added. The resulting mixture was divided into 4 parts and each part was neutralized separately with ammonia (10%). The required amine separated out, filtered with suction, dried and recrystallised with appropriate solvent. Physicochemical properties are reported in Table 6.

5.1.2. Synthesis of 2-methyl-3-(1′3′4′-thiadiazole-2′-yl)-4(3H)-quinazolinone[21]

Anthranilic acid **1** (0.01 M) and acetic anhydride were refluxed under anhydrous condition for 4 h. Excess of acetic anhydride was distilled off under reduced pressure. To the mixture obtained, amines **IV** (0.01 M) in glacial acetic acid was added and refluxed for 4 h. Obtained reaction mixture was poured into crushed ice and left overnight. The solid which separated out was filtered, washed thoroughly with cold distilled water, dried and recrystallised from hot ethanol. The yield, melting point and other physical properties of synthesized compound are recorded in Table 7.

5.1.3. Synthesis of title compound

The title compounds were synthesized by following the procedure reported earlier [11,22–26]. A solution of **3** (0.01 M) and opportune benzaldehyde (0.01 M) were reacted with glacial acetic acid (10 ml) and refluxed for 12 h. The solid **4** which separated out was filtered with suction and recrystallised from dimethylformamide to give pure compound. The physical data of the styryl quinazolinone are given in Table 1. The IR spectra, ^{13}C NMR spectra and ^{1}H NMR spectra of the title compounds are as follows:

4a IR (cm^{-1}) 1693 (C=O), 1650 (C=C) Alkene, 1530 (C=N), 1317 (CN), 688 (CS); ^{13}C NMR (300 MHz, δ) 168 (C-4), 162.4 (C-2), 112 (C-11), 136 (C-12), 136.5 (C-1″), 135 (C-18); ^{1}H NMR (300 MHz, δ) 5.16 (d, 1H, olefinic CH, $J = 15.2$ Hz), 6.6–7.92 (a set of signals, 14H, aromatic protons and olefinic CH).

4b IR (cm^{-1}) 1700 (C=O), 1611 (C=C) Alkene, 1520 (C=N), 1313 (CN), 675 (CS); ^{13}C NMR (300 MHz, δ) 161 (C-2), 161.5 (C-4), 114.7 (C-11), 136 (C-12), 136.5 (C-1″), 134 (C-13), 55.4 (C_A); ^{1}H NMR (300 MHz, δ) 3.73 (s, 3H, CH_3), 5.74 (d, 1H, olefinic CH, $J = 15.5$ Hz), 6.83–8.00 (a set of signals, 13H, aromatic protons and olefinic CH).

4c IR (cm^{-1}) 1691 (C=O), 1640 (C=C) Alkene, 1530 (C=N), 1275 (CN), 773 (CS); ^{13}C NMR (δ) 165 (C-2), 168 (C-4), 112 (C-11), 136 (C-12), 133.5 (C-1″), 134.9 (C-13), 21.4 (C_A); ^{1}H NMR (300 MHz, δ) 2.35 (s, 3H, CH_3), 5.84 (d, 1H, olefinic CH, $J = 15.2$ Hz), 6.12–7.85 (a set of signals, 13H, aromatic protons and olefinic CH).

4d IR (cm^{-1}) 1700 (C=O), 1668 (C=C) Alkene, 1591 (C=N), 1326 (CN), 752 (CS); ^{13}C NMR (δ) 165 (C-2), 168 (C-4), 112 (C-11), 136.4 (C-12), 134.6 (C-1″), 135 (C-13); ^{1}H NMR (300 MHz, δ) 5.52 (d, 1H, olefinic CH, $J = 15.2$ Hz), 6.80–7.48 (a set of signals, 13H, aromatic protons and olefinic CH).

4e IR (cm^{-1}) 1700 (C=O), 1614 (C=C) Alkene, 1555 (C=N), 1326 (CN), 760 (CS); ^{13}C NMR (δ) 160.2 (C-2), 168.8 (C-4), 112 (C-11), 136 (C-12), 137.9 (C-1″), 134 (C-13); ^{1}H NMR (300 MHz, δ) 5.12 (d, 1H, olefinic CH, $J = 15.4$ Hz), 6.41–7.49 (a set of signals, 13H, aromatic protons and olefinic CH).

4f IR (cm^{-1}) 1737 (C=O), 1610 (C=C) Alkene, 1532 (C=N), 1269 (CN), 733 (CS); ^{13}C NMR (δ) 167 (C-2), 168.9 (C-4), 112 (C-4), 136 (C-12), 134 (C-1″), 134 (C-13), 125 (C_A), 130 (C_B); ^{1}H NMR (300 MHz, δ) 5.60 (d, 1H, olefinic CH, $J = 15.6$ Hz), 6.99 (d, 2H, olefinic CH), 6.78–8.01 (a set of signals, 14H, aromatic protons and olefinic CH).

4g IR (cm^{-1}) 1700 (C=O), 1620 (C=C) Alkene, 1542 (C=N), 1274 (CN), 740 (CS); ^{13}C NMR (δ) 158.8 (C-2), 168.6 (C-4), 112 (C-11), 136 (C-12), 136.5 (C-1″), 134.5 (C-13), 56 (C_A); ^{1}H NMR (300 MHz, δ) 3.84 (s, 3H, CH_3), 5.82 (d, 1H, olefinic CH, $J = 14.4$ Hz), 6.72–7.94 (a set of signals, 13H, aromatic protons and olefinic CH).

4h IR (cm^{-1}) 1739 (C=O), 1642 (C=C) Alkene, 1542 (C=N), 1334 (CN), 759 (CS); ^{13}C NMR (δ) 167 (C-2), 168.6 (C-4), 112 (C-11), 136 (C-12), 128.8 (C-1″), 131.4 (C-13), 55.5 (C_A), 56 (C_B); ^{1}H NMR (300 MHz, δ) 3.63 (s, 3H, CH_3), 3.72 (s, 3H, CH_3), 5.76 (d, 1H, olefinic CH, $J = 15.0$ Hz), 6.60–7.86 (a set of signals, 12H, aromatic protons and olefinic CH).

4i IR (cm^{-1}) 1701 (C=O), 1637 (C=C) Alkene, 1530 (C=N), 1267 (CN), 774 (CS); ^{13}C NMR (δ) 165 (C-2), 168 (C-4), 112 (C-11), 136 (C-12), 133.5 (C-1″), 131.4 (C-13), 21.4 (C_A), 56 (C_B); ^{1}H NMR (300 MHz, δ) 2.32 (s, 3H, CH_3), 3.74 (s, 3H, CH_3), 5.85 (d, 1H, olefinic CH, $J = 15.2$ Hz), 6.74–7.10 (a set of signals, 12H, aromatic protons and olefinic CH).

4j IR (cm^{-1}) 1734 (C=O), 1634 (C=C) Alkene, 1542 (C=N), 1293 (CN), 658 (CS); ^{13}C NMR (δ) 167 (C-2), 168.7 (C-4), 112 (C-11), 135.8 (C-12), 134.6 (C-1″), 131.4 (C-13), 56 (C_A); ^{1}H NMR (300 MHz, δ) 3.80 (s, 3H, CH_3), 5.62 (d, 1H, olefinic CH, J = 15.6 Hz), 6.51–7.89 (a set of signals, 12H, aromatic protons and olefinic CH).

4k IR (cm^{-1}) 1700 (C=O), 1630 (C=C) Alkene, 1560 (C=N), 1348 (CN), 591 (CS); ^{13}C NMR (δ) 165 (C-2), 168.6 (C-4), 112 (C-11), 136 (C-12), 137.9 (C-1″), 131.6 (C-13), 56 (C_A); ^{1}H NMR (300 MHz, δ) 3.46 (s, 3H, CH_3), 5.43 (d, 1H, olefinic CH, $J = 15.4$ Hz), 6.65–7.96 (a set of signals, 12H, aromatic protons and olefinic CH).

4l IR (cm^{-1}) 1708 (C=O), 1608 (C=C) Alkene, 1505 (C=N), 1278 (CN), 620 (CS); ^{13}C NMR (δ) 167 (C-2), 168 (C-4), 112 (C-11), 136.5 (C-12), 134.9 (C-1″), 131.6 (C-13), 124.2 (C_A), 129.2 (C_B), 56 (C_C); ^{1}H NMR (300 MHz, δ) 3.57 (s, 3H, CH_3), 5.66 (d, 1H, olefinic CH, $J = 15.2$ Hz), 7.01 (d, 2H, olefinic CH), 6.72–8.12 (a set of signals, 13H, aromatic protons and olefinic CH).

4m IR (cm^{-1}) 1701 (C=O), 1693 (C=C) Alkene, 1562 (C=N), 1274 (CN), 761 (CS); ^{13}C NMR (δ) 164 (C-2), 165 (C-4), 112 (C-11), 136 (C-12), 136.5 (C-1″), 131.9 (C-13), 20.9 (C_A); ^{1}H NMR (300 MH_z, δ) 2.36 (s, 3H, CH_3), 5.60 (d, 1H, olefinic CH, $J = 15.2$ Hz), 6.66–7.90 (a set of signals, 13H, aromatic protons and olefinic CH).

4n IR (cm^{-1}) 1703 (C=O), 1609 (C=C) Alkene, 1559 (C=N), 1251 (CN), 615 (CS); ^{13}C NMR (δ) 161.4 (C-2), 168 (C-4), 112 (C-11), 136 (C-12), 128.5 (C-1″), 131.9 (C-13), 55.4 (C_A), 22 (C_B); ^{1}H NMR (300 MHz, δ) 2.30 (s, 3H, CH_3), 3.74 (s, 3H, CH_3), 5.64 (d, 1H, olefinic CH, J = 15.2 Hz), 6.68–7.89 (a set of signals, 12H, aromatic protons and olefinic CH).

4o IR (cm^{-1}) 1612 (C=O), 1650 (C=C) Alkene, 1520 (C=N), 1313 (CN), 615 (CS); ^{13}C NMR (δ) 165 (C-2), 168 (C-4), 112 (C-11), 136 (C-12), 134.6 (C-1″), 131.4 (C-13), 50.5 (C_A), 20.9 (C_B); ^{1}H NMR (300 MHz, δ) 2.30 (s, 3H, CH_3), 2.34 (s, 3H, CH_3), 5.58 (d, 1H, olefinic CH, $J = 15.4$ Hz), 6.87–8.10 (a set of signals, 12H, aromatic protons and olefinic CH).

4p IR (cm^{-1}) 1693 (C=O), 1610 (C=C) Alkene, 1590 (C=N), 1316 (CN), 667 (CS); ^{13}C NMR (δ) 158.6 (C-2), 168.7 (C-4), 112 (C-11), 136 (C-12), 135.1 (C-1″), 131.6 (C-13), 22.4 (C_A), 131.9 (C_B); ^{1}H NMR (300 MHz, δ) 2.40 (s, 3H, CH_3), 5.76 (d, 1H, olefinic CH, $J = 15.2$ Hz), 6.54–7.94 (a set of signals, 12H, aromatic protons and olefinic CH).

4q IR (cm^{-1}) 1695 (C=O), 1610 (C=C) Alkene, 1530 (C=N), 1322 (CN), 618 (CS); ^{13}C NMR (δ) 160.1 (C-2), 168.7 (C-4), 112 (C-11), 136 (C-12), 137.9 (C-1″), 131.1 (C-13), 12.3 (C_A); ^{1}H NMR (300 MHz, δ) 2.32 (s, 3H, CH_3), 5.67 (d, 1H, olefinic CH, $J = 15.6$ Hz), 6.76–7.49 (a set of signals, 12H, aromatic protons and olefinic CH).

4r IR (cm^{-1}) 1708 (C=O), 1600 (C=C) Alkene, 1520 (C=N), 1311 (CN), 633 (CS); ^{13}C NMR (δ) 161.6 (C-2), 168.6 (C-4), 112 (C-11), 136.4 (C-12), 135.4 (C-1″), 131.9 (C-13), 124.8 (C_A), 130.8 (C_B), 22.4 (C_C); ^{1}H NMR (300 MHz, δ) 2.35 (s, 3H, CH_3), 5.63 (d, 1H, olefinic CH, $J = 15.4$ Hz), 6.93 (d, 2H, olefinic CH), 6.70–7.99 (a set of signals, 13H, aromatic protons and olefinic CH).

5.2. Pharmacology

The anticonvulsant evaluation was undertaken using reported procedure [27–30]. Male albino mice (CF-1 strain or swiss, 18–25 g) and rats (Sprague–Dawley or Wistar, 100–150 g) were used as experimental animals. The tested compounds were suspended in polyethylene glycol 400.

5.2.1. Anticonvulsant screening

Initially all the compounds were administered i.p. in a volume of 0.01 ml/g body weight for mice and 0.004 ml/g body weight for rats at doses of 30, 100, 300 mg/kg to one to four animals. Activity was established using the MES and scPTZ test and these data are presented in Table 2.

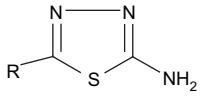
5.2.2. Neurotoxicity screening

Minimal motor impairment [31] was measured in mice by the rotorod test. The mice were trained to stay on an accelerating rotorod that rotated at six revolutions per minute. The rod diameter was 3.2 cm. Neurotoxicity was indicated by the inability of the animal to maintain equilibrium on the rod for at least 1 min in each of the three trials.

5.2.3. Behavioral testing

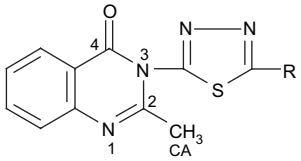
The titled compounds (100 mg/kg) were screened for their behavioral effect using actophotometer [32] at 30 min and 1 h after drug administration. The behavior of animals inside the photocell was recorded as a digital score. Increased scores suggest good behavioral activity. The activity of the compounds were at maximum at 1 h, therefore, the activity values at 1 h were used to calculate % decrease in locomotor activity. The control group animals were administered PEG 400. The observations are tabulated as Table 3.

Table 6
Physico-chemical data of 2-amino-5-aryl 1,3,4-thiadiazole



S. No.	R	Yield	Solvent recrystallize	M.P. (°C)	Molecular formula	Molecular weight	**IR** (cm^{-1})	^{1}H NMR (ppm)
1	C_6H_5	65%	Aq. ethanol (25%)	224	$C_8H_7N_3S$	177.23	3496 (NH_2)	4.0 (NH_2)
2	*p*-$OCH_3C_6H_5$	60%	Aq. ethanol (50%)	210	$C_9H_9N_3OS$	207.26	3500 (NH_2)	4.0 (NH_2), 3.73 (OCH_3)
3	*p*-$CH_3C_6H_4$	62%	Aq. ethanol (25%)	215	$C_9H_9N_3S$	191.26	3450 (NH_2)	4.0 (NH_2), 2.35 (CH_3)
4	*p*-ClC_6H_4	70%	Aq. ethanol (50%)	227	$C_8H_6ClN_3S$	211.67	3452 (NH_2)	4.0 (NH_2)
5	*m*-ClC_6H_4	70%	Aq. ethanol (50%)	226	$C_8H_6ClN_3S$	211.67	3466 (NH_2)	4.0 (NH_2)
6	-CH=CHC_6H_4 (1 2)	60%	Aq. ethanol	220	$C_8H_8N_3S$	202.27	3490 (NH_2)	4.0 (NH_2), 6.9 (CH^1), 6.9 (CH^2)

Table 7
Physicochemical properties of 3-[5-substituted 1,3,4-thiadiazole-2-yl] quinazoline-4(3*H*)-ones



S. No.	R	Yield	M.P. (°C)	Molecular formula	Molecular weight	IR (cm^{-1})	^{13}C NMR (ppm)
1	C_6H_5	60%	172–174	$C_{17}H_{12}N_4SO$	320.38	1693 (C=O)	164 (C-2), 173 (C-4), 17.4 (C_A)
2	p-$OCH_3C_6H_4$ (C_B)	60%	185–187	$C_{18}H_{14}N_4SO_2$	350.40	1701 (C=O)	164 (C-2), 173 (C-4), 17.4 (C_A), 56 (C_B)
3	p-$CH_3C_6H_4$ (C_B)	58%	180–182	$C_{18}H_{14}N_4SO$	334.40	1700 (C=O)	164 (C-2), 173 (C-4), 17.4 (C_A), 20.9 (C_B)
4	*p*-ClC_6H_4	55%	189–190	$C_{17}H_{11}N_4SOCl$	354.82	1695 (C=O)	164 (C-2), 173 (C-4), 17.4 (C_A)
5	*m*-ClC_6H_4	55%	188–190	$C_{17}H_{11}N_4SOCl$	354.5	1705 (C=O)	164 (C-2), 173 (C-4), 17.4 (C_A)
6	CH=CHC_6H_4 (C_B C_C)	50%	176–177	$C_{19}H_{13}N_4SO$	346.41	1692 (C=O)	164 (C-2), 173 (C-4), 17.4 (C_A), 124.8 (C_B), 130.8 (C_C)

5.2.4. CNS depressant activity

The forced swim pool method described earlier [33] was followed. Wistar rats were placed in chamber (diameter 45 cm, height 20 cm) containing water up to a height of 15 cm at 25 ± 2 °C. Two swim sessions were conducted an initial 15 min pretest, followed by a 5 min test session 24 h later. The animals were drug administrated (100 mg/kg) the test compound i.p. 30 min before the test session. The period of immobility (passive floating without struggling, making only those movements which are necessary to keep its head above the surface of water) during the 5 min test period was measured. The results are presented in Table 4.

Acknowledgement

The authors acknowledge the assistance of the Antiepileptic Drug Development Program, Epilepsy Branch, Preclinical Pharmacology Section, NIH, USA.

References

[1] W.L.F. Armarego, Adv. Heterocycl. Chem. 24 (1979) 1–62.
[2] L. Fisnerova, B. Brunova, Z. Kocfeldova, J. Tikalova, E. Maturova, J. Grimova, Collect. Czech. Chem. Commun. 56 (1991) 2373–2381.
[3] S. Saxena, M. Verma, A.K. Saxena, K. Shanker, Indian J. Pharm. Sci. 53 (1991) 48–52.
[4] D. Gravier, J.P. Dupin, F. Casadebaig, G. Hou, M. Boisseau, H. Bernard, Pharmazie 47 (1992) 91–94.
[5] K.H. Boltze, H.D. Dell, H. Lehwald, D. Loranz, M. Ruberg-schweer, Arzneim.-Forsch./Drug Res. 13 (1963) 688–692.
[6] J.F. Wolfe, T.L. Rathman, M.C. Sleevi, J.A. Campbell, T.D. Greenwood, J. Med. Chem. 33 (1990) 161–166.
[7] S.K. Jain, P. Mishra, Asian J. Chem. 12 (2000) 1341–1343.
[8] V. Jatav, S.K. Jain, S.K. Kashaw, P. Mishra, Indian J. Pharm. Sci. 07 (2006) 360–362.
[9] P. Mishra, V. Jatav, S.K. Kashaw, J. Indian Chem. Soc. 83 (2006) 1165–1170.
[10] C. Parkanyi, H.L. Yuan, B.H.E. Stromberg, A. Evenzahav, J. Heterocycl. Chem. 29 (1992) 749–753.
[11] D. Raffa, G. Daidone, B. Maggio, S. Cascioferro, F. Plescia, D. Schillaci, IL Farmaco 59 (2004) 215–221.
[12] G. Young, W. Eyre, J. Chem. Soc. 54 (1901) 162.
[13] J. Bernstein, H.L. Yale, K. Losee, M. Holsing, J. Martins, W.A. Lott, J. Am. Chem. Soc. 73 (1957) 906.
[14] G. Maffii, E. Testa, R. Ettorre, IL Farmaco 13 (1958) 187.
[15] V.R. Range, V.R. Srinivasan, Indian J. Chem. 8 (1970) 509.
[16] R.J. Porter, J.J. Cereghino, G.D. Gladding, B.J. Hessie, H.J. Kupferberg, B. Seoville, B.G. White, Cleve. Clin. Q. 51 (1984) 293–305.
[17] J.R. Dimmock, R.N. Puthucode, J.M. Smith, M. Hetherington, J.W. Quail, U. Pugazhenthi, T. Lechler, J.P. Stables, J. Med. Chem. 39 (1996) 3984–3997.
[18] P.T. Flaherty, T.D. Greenwood, A.L. Manhein, J.F. Wolfe, J. Med. Chem. 39 (1996) 1509–1513.
[19] J.R. Dimmock, S.N. Pandeya, J.W. Quail, U. Pugazhenthi, T.M. Allen, G.Y. Kao, J. Balzarini, Clercq E. De, Eur. J. Med. Chem. 30 (1995) 303–314.
[20] J.R. Dimmock, K.K. Sidhu, S.D. Tumber, S.K. Basran, M. Chen, J.W. Quail, J. Yang, I. Rozas, D.F. Weaver, Eur. J. Med. Chem. 30 (1995) 287–301.
[21] M.T. Bogert, H.A. Seil, J. Am. Chem. Soc. 10 (1905) 1305.
[22] D. Raffa, G. Daidone, B. Maggio, S. Cascioferro, F. Plescia, D. Schillaci, IL Farmaco 59 (2004) 451–455.
[23] M. Shrimali, R.D. Kalsi, R.S. Dixit, J.P. Barhwal, Arzneim.-Forsch./Drug Res. 41 (1991) 514–519.
[24] D. Raffa, M.C. Edler, G. Daidone, B. Maggio, M. Merickech, S. Plescia, D. Schillaci, R. Bai, E. Hamel, Eur. J. Med. Chem. 39 (2004) 299–304.
[25] V. Murugan, C.C. Thomas, G.V.S. Ramasarma, E.P. Kumar, B. Suresh, Indian J. Pharm. Sci. 65 (2003) 386–389.
[26] J.B. Jiang, D.P. Hesson, B.A. Dusak, D.L. Dexter, G.J. Kang, E. Hamel, J. Med. Chem. 33 (1990) 1721–1728.
[27] E.A. Swinyard, J.H. Woodhead, H.S. White, M.R. Franklin, General Principles: Experimental, Selection, Quantification and Evaluation of Anticonvulsants in Antiepileptic Drugs, Raven press, New york, 1989, 85–102.
[28] H.S. White, M. Johnson, H.H. Wolf, H.J. Kupferberg, Ital J. Neurol. Sci. 16 (1995a) 73–77.
[29] H.S. White, J.H. Woodhead, M.R. Franklin, General Principals: Experimental Selection, Quantification and Evaluation of Antiepileptic Drugs, Antiepileptic Drug, Raven press, New York, 1995, 99–110.
[30] E.A. Swinyard, L.D. Clark, J.T. Miyahara, H.H. Wolf, J. Physiol 132 (1961) 97–102.
[31] M.S. Dunham, T.A. Miya, J. Am. Pharm. Ass. Sci. 46 (1957) 208–209.
[32] J.R. Boissoer, P. Simon, Arch. Int. Pharmacodyn. Ther. 158 (1965) 212–214.
[33] R.D. Porsolt, G. Anton, N. Blanet, M. Jalbre, Eur. J. Pharm. 47 (1978) 379–386.