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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis and biological evaluation of some new quinazolin-4(3*H*)-ones derivatives as anticonvulsants

Deepak Gupta · Rajiv Kumar · Ram Kumar Roy · Adish Sharma · Israr Ali · Md. Shamsuzzaman

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Abstract Epilepsy is the most common neurological disorder known, affecting around 1 % of the world's population, characterized by recurrent seizure attack. A new series of 2-phenyl-3-(3-(substituted-benzylideneamino)-quinazolin-4(3*H*)-one derivatives (**6a**–**h**) was synthesized through condensation of anthranilic acid (**1**) and benzoyl chloride to give 2-phenyl-benzo[d][1,3]oxazin-4-one (**2**). Compound **2** was refluxed with hydrazine hydrate and yielded intermediate **3.** Further, the intermediate **3** was dehydrated with catalytic amount of GAA and yielded 3-amino-2-phenyl-1*H*-quinazolin-4-one (**4**). Compound **4** was further treated with 3-hydroxy benzaldehyde and small amount of GAA to afford schiff base derivative **5.** Finally, the Schiff base was treated with various alkyl halide to provide desired compounds **6a**–**h** and structures of the final compounds were confirmed on the basis of their FTIR, NMR, and Mass spectral data. Anticonvulsant activity was evaluated by the maximal electroshock test, further their minimum motor impairment and CNS depressant effect were evaluated by the rotorod motor impairment and Porsolt's force swim tests, respectively. The results showed that 2-phenyl-3-(3-(propoxybenzylideneamino)-3*H*-quinazolin-4-one (**6c**) is the most promising compound with the lowest side effects.

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Introduction

Epilepsy is a chronic and often progressive disorder characterized by the periodic and unpredictable occurrence of epileptic seizures that are caused by abnormal discharge of cerebral neurons. Epilepsy is not a disease, but a syndrome of different cerebral disorders of the CNS. This syndrome is characterized by paroxysmal, excessive, and hyper synchronous discharges of large numbers of neurons (Loscher, 1998) and epilepsy treatment, nearly 95 % of clinically available drugs were approved before 1985 and they could provide satisfactory seizure control for 60–70 % of patients. These drugs, however, also cause notable adverse side effects such as drowsiness, ataxia, gastrointestinal disturbance, hepatotoxicity, and megaloblastic anemia (Leppik, 1994; Perucca, 1996; Lin and Kadaba, 1997) and even life threatening conditions (Al-Soud *et al*., 2003). Research to find more effective and safer antiepileptic drugs are, therefore, imperative and challenging in medicinal chemistry.

In medicinal chemistry, quinazolin-4(3*H*)-one is one of the most versatile nucleus with a wide range of biological properties including antihypertensive (Alagarsamy and Pathak, 2007), anti-inflammatory (Kumar *et al*., 2007; Alagarsamy *et al*., 2008), anticonvulsant (Archana and Kumar, 2002; Zappalà *et al*., 2003; Jatav *et al*., 2008; Kashaw *et al*., 2009), antimicrobial (Grover and Kini, 2006; Pandey *et al*., 2009), antitumor (Bavetsias *et al*., 2002; Al-Rashood *et al*., 2006), and anti-diabetic (Malamas and Millen, 1991) activities. Quinazolin-4(3*H*)-one and their derivatives are also building block for approximately 150 naturally occurring alkaloids isolated from a number of

families of the plant kingdom, from microorganisms and animals.

Schiff base derivatives are an emerging new class of potent anticonvulsants (Sridhar *et al*., 2002; Verma *et al*., 2004), antibacterial, antifungal, and anti-HIV (Karthikeyan *et al*., 2006; Pandeya *et al*., 1999a, b, c) agents.

Materials and methods

Chemistry

Raw materials for the synthesis were supplied by Spectrochem chemicals (India) and S.D. Fine Chemicals (India). Melting points of newly synthesized compounds were determined in open glass capillary and are uncorrected. IR (KBr) spectra were recorded on a Nicolet 5PC FTIR spectrophotometer (λ-max in cm^{-1}) and ^{1}H NMR and ^{13}C NMR spectra were recorded on a Brucker Model-300 NMR Spectrometer in DMSO-d_6 using tetramethylsilane (TMS) as the internal reference (chemical shifts in δ ppm). Chemical shifts are reported in ppm downfield from TMS. Mass spectra were recorded on Jeol SX- 102/DA-6000 spectrometer. The reaction monitoring was performed on Silica gel-GF coated aluminum plate using iodine vapors and UV light as visualizing agents. Evaporation of solvents was performed at reduced pressure, using a Buchi rotary evaporator. Solid products were purified by recrystallization and column chromatography.

Synthesis of 2-phenyl-benzo[d][1,3]oxazin-4-one ***(2)***

Anthranilic acid (**1**) (0.0365 mol) was dissolved in pyridine (15 mL) in dry and clean rbf with guard tube. Benzoyl chloride (0.547 mol) was added slowly while maintaining the temperature at 0–5 °C with proper stirring. The reaction was further stirred at room temperature for 15 min and it was slowly converted into semi solid paste; further it was diluted with saturated $NaHCO_3$ solution. The white solid thus obtained was filtered and dried. The compound was used in the next step without further purification, Yield: 75 %; m.p. 143–148 °C.

Synthesis of 3-amino-2-hydroxy-2-phenyl-2,3-dihydroquinazolin-4(1H)-one ***(3)***

Compound **2** (0.01255 mol) was dissolved in 20 mL ethanol in dry and clean rbf with condenser and guard tube. Hydrazine hydrate (0.99 %) (0.015 mol) was added to it at room temperature and the reaction mixture was refluxed for 2 h. The progress of reaction was monitored by aluminum coated TLC plate using 100 % ethyl acetate solvent system. After completion of reaction it was poured into ice-cold water to yield white solid which was filtered and dried. The dried compound was purified by crystallization with ethyl acetate. It gives yellow color with ninhydrin reagent. Yield 70 %; R_f value: 0.3; m.p. 144–146 °C; IR (KBr, cm^{-1}): 3365 (NH), 3400 (br, OH), 1690 (C=O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): 4.63 (br s, 2H, NH_2, D_2O exchangeable), 7.09–8.44 (m, 9H, ArH), 10.27 (br s, 1H, NH, D_2O exchangeable), 12.43 (s, 1H, OH, D_2O exchangeable); MS: *m/z* 255.9 $(M+1)^+$.

Synthesis of 3-amino-2-phenyl-1H-quinazolin-4-one ***(4)***

Compound **4** (0.010 mol) was dissolved in 20 mL ethanol in dry and clean rbf with condenser and guard tube. GAA (2 mL) was added to it at room temperature and the reaction mixture was refluxed for 4 h. The progress of reaction was monitored by aluminum coated Merck TLC plate using 50 % ethyl-hexane solvent system. After completion of reaction it was poured into ice-cold water to yield white solid which was filtered and dried, it was purified by crystallization with ethyl acetate. It gives orange color with ninhydrine reagent. Yield: 65 %; R_f value: 0.6; m.p. 140–145 °C; IR (KBr, cm^{-1}): 3343 (NH), 1699 (C=O); ^{1}HNMR (300 MHz, $CDCl_3$) δ (ppm): 5.00 (br s, 2H, NH_2, D_2O exchangeable), 7.49–8.32 (m, 9H, ArH); MS: *m/z* 238.2 $(M+1)^+$.

Synthesis of 3[(3-hydroxy-benzylidene)-amino]-2-phenyl-3H-quinazolin-4-one ***(5)***

In a clean rbf, compound (**4**) (0.0021 mol) and conc. HCl (1 mL) was taken. It was stirred at room temperature for 5 min. Water (15 mL), sodium acetate (0.0021 mol), and ethanol (15 mL) were added to the reaction mixture and it was further stirred 10 min. 3-Hydroxy-benzaldehyde (0.0021) was dissolved in ethanol (10 mL) and added into the reaction mixture. The clear solution changed into turbid suspension which was further stirred for 1 h. The progress of reaction was monitored by aluminum coated Merck TLC plate using 50 % ethyl-hexane solvent system. The reaction mixture was poured into ice-cold water to yield white solid which was filtered and dried. The compound was recrystallized with ethanol. Yield: 70 %; R_f value: 0.3; m.p. 160–165 °C; IR (KBr, cm^{-1}): 3423 (OH), 1680 (C=O), 1630 (C=N); ^{1}H NMR (300 MHz, DMSO-d_6): δ 10.26 (s, 1H, OH, D_2O exchangeable), 9.20 (s, 1H, CH=N), 8.26–6.85 (m, 13H, ArH); ^{13}C NMR (75 MHz, DMSO-d_6): δ 168.2 (C=O), 161.2 (C–O), 158.1, 158.0 (N=C), 154.2, 146.2, 133.2, 130.9, 130.2, 128.5, 128.0, 126.9, 126.7, 126.1, 124.0, 123.0, 119.0, 120.2 115.8; MS: *m/z* 342.2 $(M+1)^+$.

*Synthesis of 3-[(3-substituted-benylidene)-amino]-2-phenyl-quinazolin-4(3H)-one **(6a–h)***

3-(3-Hydroxybenzylideneamino)-2-phenyl-quinazolin-4(3*H*)-one (**5**) (0.00145 mol) was dissolved in 20 mL DMF in dry and clean rbf with condenser and guard tube. Anhydrous K_2CO_3 (0.00217 mol) was added and the reaction mixture was stirred at 0–5 °C for 15 min. Alkyl halide (0.00217 mol) was added at same temperature and stirred at room temperature for 1–3 h. The progress of reaction was monitored by aluminum coated Merck TLC plate using 60 % ethyl-hexane solvent system. After completion of reaction, it was poured into ice-cold water to yield solid, which was purified and dried. The compound was purified by recrystallization in ethyl acetate and hexane.

*3-[(3-Methoxy-benzylidene)-amino]-2-phenyl-3H-quinazolin-4-one **(6a)*** Yield: 70 %; R_f value: 0.4; m.p. 145–147 °C; IR (KBr, cm^{-1}): 1682 (C=O), 1624 (C=N), 1235 (C–O); ^{1}H NMR (300 MHz, DMSO-d_6): δ 8.99 (s, 1H, CH=N), 8.30–6.85 (m, 13H, ArH), 3.91 (m, 3H, OCH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 169.7 (C=O), 164.7 (C–O), 163.9, 158.6 (N=C), 154.1, 146.4, 134.1, 130.1, 130.0, 128.9, 128.6, 127.1, 126.7, 126.2, 124.7, 121.4, 121.0, 117.2, 115.8, 68.1 (OCH); MS: *m/z* 356 $(M+1)^+$; Anal. Calcd for $C_{22}H_{17}N_3O_2$: C, 74.35, H, 4.82, N, 11.82; Found: C, 74.34; H, 4.80; N, 11.80 %.

*3-[(3-Ethoxy-benzylidene)-amino]-2-phenyl-3H-quinazolin-4-one **(6b)*** Yield: 75 %; m.p.: 150–152 °C; IR (KBr, cm^{-1}): 1682 (C=O), 1624 (C=N), 1230 (C–O); ^{1}H NMR (300 MHz, DMSO-d_6): δ 9.24 (s, 1H, CH=N), 8.25–6.94 (m, 13H, ArH), 4.15 (q, 2H, OCH_2), 1.35 (t, 3H, $J = 6.6$ Hz, CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 169.5 (C=O), 163.9 (C–O), 162.9, 158.2 (N=C), 153.8, 146.0, 139.8, 130.2, 130.0, 128.8, 128.6, 127.4, 126.5, 126.2, 124.0, 121.4, 121.2, 117.5, 115.3, 70.1 (OCH), 16.1(CH_3); MS: *m/z* 370 $(M+1)^+$; Anal. Calcd for $C_{23}H_{19}N_3O_2$: C, 74.78, H, 5.18, N, 11.37; Found: C, 74.70; H, 5.23; N, 11.42 %.

*2-Phenyl-3-[(3-propoxy-benzylidene)-amino]-3H-quinazolin-4-one **(6c)*** Yield: 70 %; R_f value: 0.4; m.p.: 160–162 °C; IR (KBr, cm^{-1}): 1699 (C=O), 1620 (N=C), 1230 (C–O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): δ 8.98 (s, 1H, CH=N), 8.36–6.87 (m, 13H, ArH), 3.98 (t, 2H, $J = 7.0$ Hz OCH_2), 1.78–1.88 (m, 2H, CH_2), 1.04 (t, 3H, $J = 7.2$ Hz, CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 169.4 (C=O), 163.5 (C–O), 162.0, 158.0 (N=C), 153.4, 146.2, 139.3, 130.1, 130.0, 128.9, 128.6, 127.2, 126.5, 126.3, 124.0, 121.9, 121.3, 116.9, 114.8, 70.3 (OCH), 22.0 (CH_2), 11.1(CH_3); MS: *m/z* 384 $(M + 1)^+$; Anal. Calcd for $C_{24}H_{21}N_3O_2$: C, 75.18, H, 5.52, N, 10.96; Found: C, 75.02; H, 5.53; N, 10.96 %.

*3-[(3-Iso-propoxy-benzylidene)-amino]-2-phenyl-3H-quinazolin-4-one **(6d)*** Yield: 69 %; R_f value: 0.4; m.p.: 155–157 °C. IR (KBr, cm^{-1}): 1625 (N=C), 1680 (C=O), 1230 (C–O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): δ 9.23 (s, 1H, CH=N), 8.27–6.94 (m, 13H, ArH), 4.70 (m, 1H, OCH), 1.30 (d, 6H, $J = 6.0$ Hz, $2CH_3$); ^{13}C NMR (75 MHz, DMSO-d_6): δ 169.5 (C=O), 163.0, 162.5 (C–O), 158.1 (N=C), 153.6, 146.2, 139.2 130.0, 129.9, 128.9, 128.6, 127.1, 126.6, 126.3, 124.1, 122.0, 121.8, 116.8, 114.0, 71.0 (OCH), 22.8 $(CH_3)_2$; MS: *m/z* 384 $(M+1)^+$; Anal. Calcd for $C_{24}H_{21}N_3O_2$: C, 75.18, H, 5.52, N, 10.96; Found: C, 75.00; H, 5.54; N, 10.99 %.

*Synthesis of 3-[3-(allyloxybenzylidene)-amino]-2-phenyl-3H-quinazolin-4-one **(6e)*** Yield: 60 %; R_f value: 0.4; m.p.: 160–162 °C; IR (KBr, cm^{-1}): 1685 (C=O), 1623 (C=N), 1245 (C–O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): δ 9.35 (s, 1H, CH=N), 8.41–7.01 (m, 13H, ArH), 6.04–6.13 (m, 1H, CH=), 5.32–5.48 (m, 2H, $= CH_2$), 4.8 (q, 2H, $J = 5.2$ Hz, OCH_2); ^{13}C NMR (75 MHz, DMSO-d_6): δ 170.0 (C=O), 162.7 (C–O), 162.8, 159.9 (N=C), 153.0, 146.6, 139.0, 135.0 (CH=), 130.3, 130.1, 128.2, 128.1, 127.3, 126.6, 126.0, 124.1, 121.8, 121.0, 119.3 (=CH_2), 117.9, 114.9, 70.6 (OCH_2); MS: *m/z* 382 $(M+1)^+$; Anal. Calcd for $C_{24}H_{19}N_3O_2$: C, 75.57, H, 5.02, N, 11.02; Found: C, 75.53; H, 5.03; N, 11.07 %.

*3-(3-(But-3-enyloxy)benzylidene-amino)-2-phenyl-3H-quinazolin-4-one **(6f)*** Yield: 65 %; R_f value: 0.6; m.p.: 140–145 °C; IR (KBr, cm^{-1}): 1688 (C=O), 1626 (C=N), 1245 (C–O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): δ 9.26 (s, 1H, CH=N), 8.26–6.85 (m, 13H, ArH), 5.85 (m, 1H, CH=C), 5.01 (m, 2H, =CH_2), 3.99 (t, 2H, OCH_2), 2.54 (m, 2H, CH_2); ^{13}C NMR (75 MHz, DMSO-d_6): δ 167.4 (C=O), 162.8, 160.9 (C–O), 157.6 (N=C), 153.4, 146.9, 136.0 136.3 (CH=), 130.0, 129.9, 128.3, 128.1, 127.4, 126.3, 126.0, 124.2, 121.3, 121.0, 119.2, 118.0 (CH_2), 115.0, 68.9 (OCH_2), 36.3 (CH_2); *m/z* 395 $(M+1)^+$; Anal. Calcd for $C_{25}H_{21}N_3O_2$: C, 75.93, H, 5.35, N, 10.63; Found: C, 75.99; H, 5.37; N, 10.66 %.

*3-[(3-Butoxy-benzylidene)-amino]-2-phenyl-3H-quinazolin-4-one **(6g)*** Yield: 68 %; R_f value: 0.3; m.p.: 160–165 °C; IR (KBr, cm^{-1}): 1643 (N=C), 1679 (C=O), 1245 (C–O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): 9.26 (s, 1H, N=CH); 8.26–6.97 (m, 13H, ArH), 4.09 (t, 2H, $J = 6.4$ Hz, OCH_2), 1.75(m, 2H, $OCH_2\underline{CH_2}$), 1.47(m, 2H, $\underline{CH_2}CH_3$), 0.93 (t, 3H, $J = 7.2$ Hz, CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 169.2 (C=O), 163.8 (C–O), 162.1,

158.0 (N=C), 153.5, 146.0, 139.2, 130.2, 130.0, 128.8, 128.7, 127.1, 126.4, 126.8, 124.2, 121.2, 121.0, 117.0, 114.5, 69.3 (OCH), 32.0 (CH_2), 18.4 (CH_2) 12.1(CH_3); MS: *m/z* 398 $(M+1)^+$; Anal. Calcd for $C_{25}H_{23}N_3O_2$: C, 75.54, H, 5.83, N, 10.57; Found: C, 75.50; H, 5.87; N, 10.53 %.

3-[(3-Benzyloxy-benzylidene)-amino]-2-phenyl-3H-quinazolin-4-one ***(6h)*** Yield: 65 %; m.p.: 95–97 °C. IR (KBr, cm^{-1}): 1643 (N=C), 1685 (C=O), 1246 (C–O); ^{1}HNMR (300 MHz, DMSO-d_6) δ (ppm): 9.30 (s, 1H, N=CH), 8.25-6.99 (m, 18H, ArH), 5.25 (s, 2H, OCH_2); ^{13}C NMR (75 MHz DMSO-d_6): δ 168.7 (C=O), 166.7 (C–O), 163.0, 155.6 (N=C), 154.1, 146.3, 136.9, 134.1, 130.1, 130.0, 128.9, 128.6, 128.2, 127.1, 127.0, 126.9, 126.7, 126.2, 124.7, 121.4, 121.2, 116.0, 115.8, 68.9 (OCH_2); MS: *m/z* 384 $(M+1)^+$; Anal. Calcd for $C_{28}H_{21}N_3O_2$: C, 77.94, H, 4.91, N, 9.74; Found: C, 77.99; H, 4.96; N, 9.72 %.

Pharmacology

Anticonvulsant screening

The anticonvulsant evaluations were performed using reported procedures (Krall *et al*., 1978; Porter *et al*., 1984). Male albino mice (CF-1 strain, 18-25 g) were used as experimental animal. The compounds were suspended in polyethylene glycol (PEG-400). All experimental protocols were carried out with permission from the Institutional Animal Ethics Committee (IAEC). Animals were obtained from the Central Animal House Facility, DR. K. N. Modi Institute of Pharmaceutical Education and Research, Modinagar, Uttar Pradesh, India.

Electroshock seizure method (MES)

Maximal seizures were induced by the application of electrical current to the brain via corneal electrodes. For all tests based on MES convulsions, 60 Hz of alternating current (50 mA) was delivered for 0.2 s by corneal electrodes which had been primed with an electrolyte solution containing an anesthetic agent (0.5 % tetracaine HCl). Abolition of the hind limb tonic extensor spasm was recorded as a measurement of anticonvulsant activity.

Rotorod motor impairment screening

Motor impairment is measured in mice by the rotorod test. The mice are trained to stay on an accelerating rotorod that rotates at six revolutions per minute. The rod diameter is 3.2 cm. Motor impairment was indicated by the inability of the animal to maintain equilibrium on the rod for at least 1 min in each of the trials.

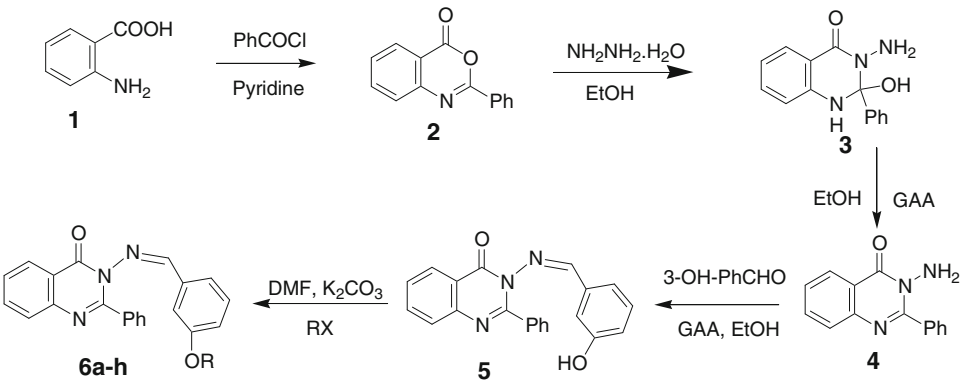
CNS depression study

The forced swim method (Porsolt's swim pool test) is followed to study CNS depression (Porsolt *et al*., 1978). Mice are placed in a chamber (diameter 45 cm, height: 20 cm) containing water up to a height of 15 cm at 25 ± 2 °C. Two swim sessions are conducted, an initial 15 min pre-test, followed by a 5 min test session 24 h later. The animals are administered an i.p. injection (30 mg/kg) of the test compounds 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 are measured.

Results and discussion

Chemistry

Compounds (**5** and **6a–h**) were prepared according to Scheme 1. Synthesis of 2-phenyl-benzo[d][1,3]oxazin-4-one (**2**)



Scheme 1 Synthesis of quinazolinone derivatives **5** and **6a–h**

a; R = CH_3, **b;** R = C_2H_5, **c;** R = C_3H_7, **d;** R = iso-propyl, **e;** R = allyl, **f;** R = but-3-enyl, **g;** R = n-butyl **h;** R = benzyl.

involves condensation of anthranilic acid (**1**) and benzoyl chloride in the presence of pyridine. Compound **2** was refluxed with hydrazine hydrate in ethanol and yielded intermediate **3.** Further, the intermediate **3** was dehydrated with catalytic amount of GAA in ethanol and yielded 3-amino-2-phenyl-1*H*-quinazolin-4-one (**4**) with a good yield. Compound **4** was treated with 3-hydroxy benzaldehyde and small amount of GAA in ethanol to afford schiff base derivative **5.** Finally, synthesis of 3-[(3-substituted-benylidene)-amino]-2-phenyl-3*H*-quinazolin-4-one (**6a-h**) were carried out by reacting 3-[(3-Hydroxy-benzylidene)-amino]-2-phenyl-3*H*-quinazolin-4-one (**5**) with various alkyl halide in the presence of anhydrous K_2CO_3 in DMF. The intermediates (**3**, **4**, and **5**) and final compounds **6a–h** were confirmed by FT-IR, ^{1}HNMR, and mass spectroscopy.

Yogeeswari *et al*. (2005) have proposed the four crucial structural components for anticonvulsant activity like (i) aryl ring center or the lipophilic group (A), (ii) an electron donor atom (D), (iii) a hydrogen bond acceptor (HA), and (iv) a hydrogen bond donor (HD). The proposed four-point pharmacophore model resembles with synthesized quinazolinone-schiff base derivatives and that of standard anticonvulsants as depicted in Fig. 1.

The above-proposed model assists us to perform an effective and simple synthesis of 2-phenyl-3-(3-(substituted-benzylideneamino)-quinazolin-4(3*H*)-one, where quinazolin-one would be an aryl ring center and to explore if the inclusion of schiff base (might be biosteric replacement with aryl semicarbazone) at the 3rd position could be an additional feature as pharmacophoric hybrid that is able to improve the pharmacological profile of quinazolinone compounds. Thus, the substitution by schiff base moiety would be synergistic. The alkyloxy and phenyl moieties (in 2nd position) were also introduced in the structure to increase the lipophilicity of the molecule. Therefore, in the present project first we sought to synthesis the designed molecules and then their anticonvulsant evaluation; finally establishing the structure activity relationship.

Pharmacology

Anticonvulsant activity and motor impairment

Synthesized Quinazolinone derivatives were screened for their anticonvulsant activity against standard models MES (maximal electroshock seizure test) for their ability to reduce seizure spread. Motor impairment screening was also carried out by rotorod test method. The anticonvulsant activity was reported after 0.5 and 4.0 h time intervals at dose levels of 30, 100, and 300 mg/kg body weight. The CNS depressant was studied at a dose level of 100 mg/kg body weight. Phenytoin and Carbamazepine were used as standard drugs.

The anticonvulsant data of this series **(5**, **6a–h)** are summarized in Table 1; compound **6h** showed protection at maximum dose (300 mg/kg) after 4.0 h while compounds **6d** and **6e** showed protection at a maximum dose (300 mg/kg) after 0.5 and 4.0 h. Compounds **6f** and **6g** showed protection at a 100 mg/kg at 4.0 h and at a dose of 300 mg/kg at 0.5 h. Compounds **5** and **6a** showed protection at a 100 mg/kg at 4.0 h. Compound **6b** showed the activity at the dose of 100 mg/kg at 0.5 and 4.0 h. Compound **6c** showed the activity at the dose of 100 mg/kg at

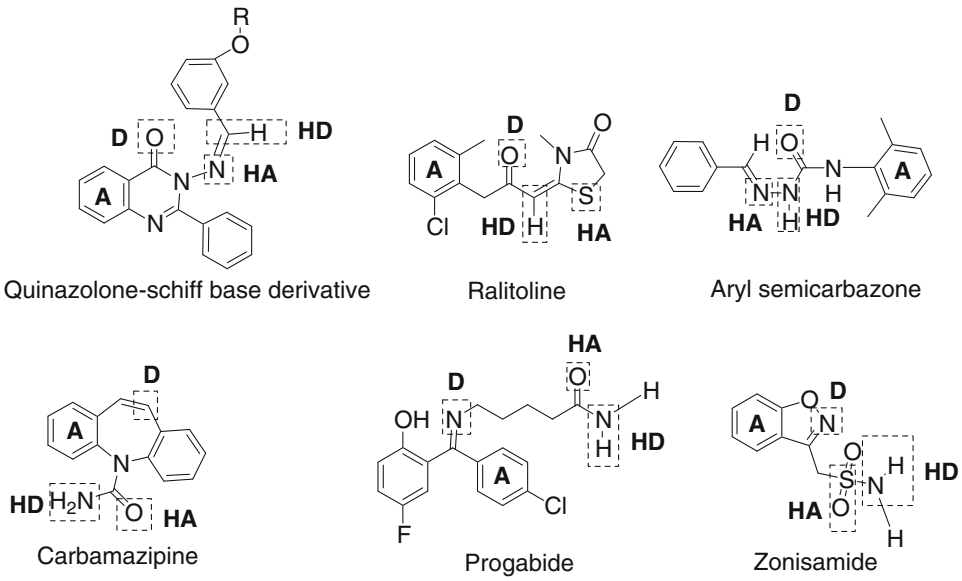


Fig. 1 Development of four-point pharmacophore model, ***R***-aryl ring center or lipophilic group; **D**-An electron donor atom; **HA**-A hydrogen bond acceptor; **HD**-A hydrogen bond donor

Table 1 Anticonvulsant and motor impairment screening of synthesized compounds (**5**, **6a–h**)

Compound	R	MES[a]		Motor impt.[a]	
		0.5 h	4.0 h	0.5 h	4.0 h
5	H	–	100	–	–
6a	CH_3	–	100	–	–
6b	CH_2CH_3	100	100	–	–
6c	*n*-$CH_2CH_2CH_3$	100	30	–	–
6d	$CH(CH_3)_2$	300	300	–	–
6e	CH_2=$CHCH_2$	300	300	–	–
6f	*n*-$CH_2(CH_2)_2CH_3$	300	100	–	–
6g	CH_2=$CHCH_2CH_2$	300	100	–	–
6h	$CH_2C_6H_5$	–	300	–	300
Control		–	–	–	–
Phenytoin[b]		30	30	–	100
Carbamazepin[b]		30	30	300	100

[a] Doses of 30, 100, and 300 mg/kg were administered; the figures in the table indicate the minimum dose whereby bioactivity was demonstrated in half or more of mice. The animals were examined 0.5 and 4.0 h after injection was made. The *dash* indicates an absence of activity at maximum dose administered

[b] Activity reported (show 100 % protection at respective dose)

0.5 h and at dose 30 mg/kg at 4.0 h. The compounds **6b** and **6c** having R = ethyl and *n*-propyl groups, respectively, were found to be the most active of the series showing activity both at 0.5 and 4.0 h at lower doses of 100 mg/kg and 30 mg/kg, respectively. In the rotarod motor impairment screening, compounds did not show any motor impairment even at even maximum dose of 300 mg/kg except compound **6h**, having R = benzyl group, which showed motor impairment at 300 mg/kg dose after 4.0 h. This motor impairment might be due to the presence of bulkier aromatic group.

CNS depressant activity

After anticonvulsant activity some significant compounds were selected and tested for their CNS depressant activity. CNS depressant effect of the compounds was determined by Porsolt's force swim pool method. The CNS depressant activity was done at a dose level of 100 mg/kg body weight, and Carbamazepine was used as the standard drug.

Selected compounds i.e., **6b**, **6c**, and **6f** having significant anticonvulsant activities were tested for their CNS depressant effect. The compounds showed 29.60, 52.18, and 110.81 % increase in immobility time with respect to control whereas the standard drug carbamazepine showed 56.35 % increase in the immobility time (Table 2). Thus it was observed that compound **6b** (29.60 %) and **6c** (52.18) showed less CNS depressant effect in comparison to carbamazepine.

Table 2 CNS depressant study of the selected compounds by Porsolt's forced swim pool test

Compound[a]	R	Duration of immobility(s) (mean ± SEM)[b]	% Increased of immobility
6b	CH_2CH_3	68.3 ± 1.16*	29.60
6c	$CH_2CH_2CH_3$	80.2 ± 1.15*	52.18
6f	$CH_2(CH_2)_2CH_3$	111.1 ± 1.89*	110.81
Carbamazepine		82.4 ± 0.94	56.35
Control		52.7 ± 0.70	–

[a] The compounds were tested at a dose of 100 mg/kg (i.p.). Each value represents the mean ± SEM of six mice. The CNS depressant effect was compared with respect to the standard drug. * $p < 0.0001$

[b] Data was analyzed by unpaired student's *t* test

The most active compounds of this study were found to be **6b** and **6c** as they are effective in both 0.5 and 4.0 h time interval at lower dose 100 and 30 mg/kg, respectively, and show no motor impairment effect. The compounds **6b** and **6c** also showed reduced CNS depressant effect in comparison to the standard drug carbamazepine.

Conclusion

On the basis of the above observation it can be concluded that as the aliphatic straight chain increases from H to *n*-propyl in compound **5**, **6a–c**, and **6f**, lipophilicity of compound also increases that lead to gradually enhancement in the blood brain barrier (BBB) crossing capacity of the compounds and potency increase up to n-propyl gradually than decrease in *n*-butyl, it may be due to excess of lipophilicity of *n*-butyl derivative. However, when *R* was replaced with branched chain, unsaturated chain, or bulkier groups like *iso*-propyl, allyl, benzyl, etc. groups, the potency decreased, which might be due to steric hindrance to binding the additional bulkier groups and lipophilicity of the compounds also decreases on incorporation of saturated chain which leads to reduction in the BBB crossing capacity of the compounds and, therefore, anticonvulsant activity decrease of having branched, unsaturated, and bulkier groups.

On the bases of the above study, the activity may be due to the presence of adequate long and straight aliphatic chain i.e., ethyl and propyl that provide adequate lipophilicity and well fitted to receptor site.

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