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SYNTHESIS AND *IN VIVO* ANTICONVULSANT EVALUATION OF 2-CHLOROQUINOLINYL HYDRAZONE DERIVATIVES

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Epilepsy is a neurological condition which is characterized by unprovoked seizures resulting from abnormal discharge of cerebral neurons and affecting at least 50 million people worldwide. There is continuing demand for new anticonvulsant agents as it has not been possible to control every kind of seizure with existing antiepileptic drugs. Moreover, conventional antiepileptic drugs exhibited unfavorable side effect profile and failure to adequately control seizures (1–3).

Searching for compounds with potential anticonvulsant activity we focused our attention on various hydrazones and semicarbazones which have been found to display significant anticonvulsant activity (4–6). These compounds were believed to interact at two locations on a putative binding site designated as hydrogen binding domain and hydrophobic binding area as proposed by Dimmock et al. (Figure 1). In their study it is also reported that replacement of aryl group with other hydrophobic

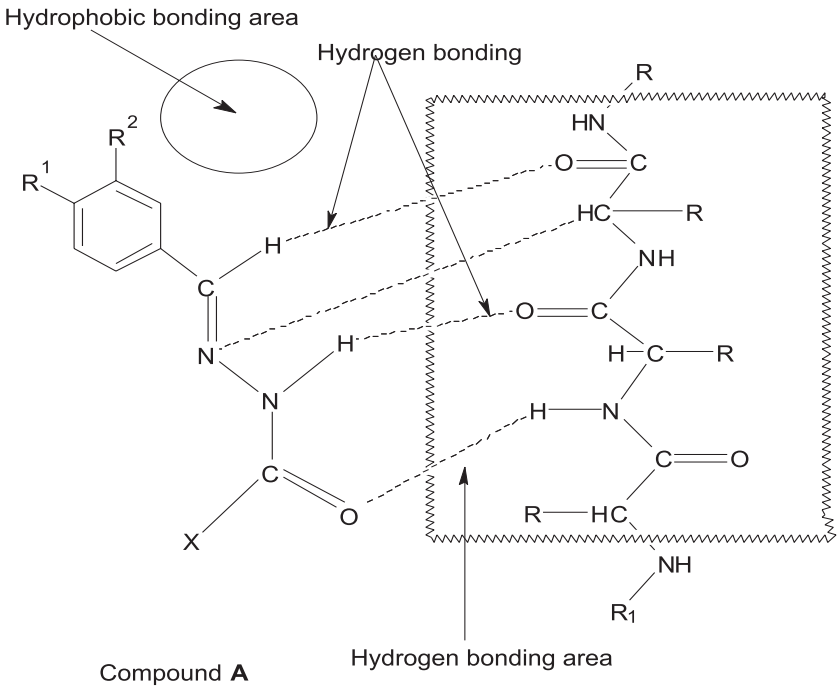

Figure 1. Proposed interaction of compound A (aryl hydrazones, semicarbazones and thiosemicarbazones) at a binding site (7)

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moieties result in a derivatives with retention of anticonvulsant activity (7–11). Moreover, the quinoline derivatives previously have been proven to exhibit potent anticonvulsant activity (12, 13). The main objective of the study was to prepare a number of analogues of compound **A** by employing 2-chloroquinoline as a hydrophobic moiety, in continuation of our research for bioactive molecules based on quinoline derivatives (14). We report herein synthesis and *in vivo* anticonvulsant activity of some new 2-chloroquinolinyl-hydrazones (**3a-n**). The evaluation of anticonvulsant activity was carried out in mice by two models of seizures, *viz*. maximal electroshock (MES) and subcutaneous pentylenetetrazole (scPTZ).

EXPERIMENTAL

Melting points were determined by the open capillary method using electrical melting point apparatus and are uncorrected. The IR spectra were recorded as KBr (discs) on Bio Rad FT-IR spectrophotometer. ^{1}H- and ^{13}C-NMR spectra were recorded on Bruker DPX 300 MHz spectrophotometer using TMS as an internal standard. The mass spectra were recorded on JEOL SX102/DA-6000 mass spectrometer and elemental analysis was carried out on Vario-EL III CHNOS elemental analyzer and were within the range of ± 0.4%. Thin layer chromatography (TLC) was performed to check the purity of the compounds, spots being visualized under iodine vapors.

Synthesis of 2-chloro-3-formylquinoline 2 (15).

N,N-Dimethylformamide (0.125 mol) was cooled to 0^{o}C in a flask equipped with a drying tube and phosphoryl chloride (0.35 mol) was added dropwise with stirring. To this solution was added appropriately substituted N-arylacetamide **1** (0.05 mol) and after 5 min the solution was heated under reflux for 16.5 h at 75^{o}C. The reaction mixture was cooled and poured into ice-water (300 mL) and stirred for 30 min at 0–10^{o}C. The precipitate formed was filtered and washed with cold water, dried, and recrystallized from ethyl acetate yielding light yellow shiny needle shaped crystals of 2-chloroquinoline-3-carboxaldehyde.

IR (KBr, cm^{-1}): 1690 (C=O), 1619 (C=C), 1591 (C=N), 753 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d$_6$, δ, ppm): 7.65–7.70 (t, 1H, *J* = 7.49 Hz, H-6), 7.88–7.93 (t, 1H, *J* = 7.18, Hz H-7), 8.00–8.02 (d, 1H, *J* = 8.09 Hz, H-5), 8.08–8.11 (d, 1H, J = 8.44 Hz, H-8), 8.78 (s, 1H, H-4), 10.52 (s, 1H, CHO). ^{13}C-NMR (75 MHz, DMSO-d$_6$, δ, ppm): 125.93, 126.12, 127.39, 127.75, 129.65, 133.31, 140.99, 148.28, 188.71 (CHO). FAB-MS: m/z 192 (M)$^{+}$, 194 (M+2).

The required various substituted acyl hydrazines were synthesized according to the method reported in the literature (16).

Synthesis of compounds (**3a-n**)

To a solution of compound **2** (0.005 mol) in 20 mL of absolute ethanol, equimolar amount of substituted acyl hydrazines / semicarbazide / thiosemicarbazide (0.005 mol) was added and the mixture was refluxed for 4–6 h. On cooling, solid was obtained which was filtered, washed with hot methanol, dried and recrystallized from ethanol and DMF mixture to give final compounds (**3a-n**). The spectral data of compounds **(3a-n)** are given below.

***N'*-[(2-Chloroquinolin-3-yl)methylidene]benzohydrazide** (**3a**)

IR (KBr, cm^{-1}): 3260 (N-H), 1659 (C=O), 1625 (C=N), 1579 (C=C), 755 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d$_6$, δ, ppm): 7.54–7.63 (m, 3H, H-2', 3', and 4'), 7.68–7.73 (t, 1H, *J* = 7.39 Hz, H-6), 7.85-7.90 (t, 1H, *J* = 7.44 Hz, H-7), 7.95–7.99 (m, 3H, H-5, 2' and 6'), 8.22–8.25 (d, 1H, *J* = 7.08 Hz, H-8), 8.82 (s, 1H, H-4), 8.94 (s, 1H, CH=N), 12.22 (s, 1H, CONH). ^{13}C-NMR (75 MHz, DMSO-d$_6$, δ, ppm): 126.13, 126.81, 127.60, 127.76, 128.50, 128.93, 131.99, 133.01, 135.61, 142.79, 147.10, 148.46, 157.17, 169.24 (C=O). FAB-MS: m/z 310 (M)$^{+}$, 312 (M+2).

***N'*-[(2-Chloroquinolin-3-yl)methylidene]-4-methylbenzohydrazide** (**3b**)

IR (KBr, cm^{-1}): 3284 (N-H), 1660 (C=O), 1637 (C=N), 1585 (C=C), 749 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d$_6$, δ, ppm): 2.40 (s, 3H, CH_3), 7.35–7.38 (d, 2H, *J* = 7.98 Hz, H-3' and 5'), 7.68–7.73 (t, 1H, *J* = 7.39 Hz, H-6), 7.84–7.89 (m, 3H, H-2', 6' and 7), 7.97–8.00 (d, 1H, *J* = 8.37 Hz, H-5), 8.22–8.25 (d, 1H, *J* = 8.0 Hz, H-8), 8.79 (s, 1H, H-4), 8.97 (s, 1H, CH=N), 12.18 (s, 1H, CONH). ^{13}C-NMR (75 MHz, DMSO-d$_6$, δ, ppm): 21.43 (CH_3), 125.88, 126.36, 127.93, 128.08, 128.74, 129.13, 131.53, 133.26, 134.10, 135.93, 143.41, 146.84, 148.15, 159.09, 168.34 (C=O). FAB-MS: m/z 324 (M)$^{+}$, 326 (M+2).

***N'*-[(2-Chloroquinolin-3-yl)methylidene]-4-methoxybenzohydrazide** (**3c**)

IR (KBr, cm^{-1}): 3203 (N-H), 1665 (C=O), 1620 (C=N), 1580 (C=C), 756 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d$_6$, δ, ppm): 3.85 (s, 3H, OCH_3)

7.08–7.11 (d, 2H, *J* = 8.76 Hz, H-3' and 5'), 7.68–7.73 (t, 1H, *J* = 7.45 Hz, H-6), 7.84–7.89 (t, 1H, *J* = 7.63 Hz, H-7), 7.95–8.00 (m, 3H, H-2', 5 and 6') 8.22–8.25 (d, 1H, *J* = 8.01 Hz, H-8), 8.91 (s, 1H, H-4), 8.96 (s, 1H, CH=N), 12.14 (s, 1H, CONH). ^{13}C-NMR (75 MHz, DMSO-d_6, δ, ppm): 55.73 (OCH_3), 125.98, 126.46, 127.35, 127.91, 128.56, 129.07, 130.69, 133.77, 136.24, 142.36, 147.12 149.21, 159.98, 170.28 (C=O). FAB-MS: m/z 340 $(M)^+$, 342 (M+2).

4-Chloro-*N'*-[(2-chloroquinolin-3-yl)methylidene]benzohydrazide (3d)

IR (KBr, cm^{-1}): 3233 (N-H), 1659 (C=O), 1628 (C=N), 1582 (C=C), 763 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 7.56–7.59 (d, 2H, *J* = 7.78 Hz H-3' and 5'), 7.70–7.75 (t, 1H, *J* = 7.32 Hz, H-6,), 7.85–7.91 (m, 3H, H-2', 6' and 7), 7.98–8.01 (d, 1H, *J* = 8.17 Hz, H-5), 8.23–8.26 (d, 1H, *J* = 7.93 Hz, H-8), 8.89 (s, 1H, H-4), 8.97 (s, 1H, CH=N), 12.15 (s, 1H, CONH).

2,4-Dichloro-*N'*-[(2-chloroquinolin-3-yl)methylidene]benzohydrazide (3e).

IR (KBr, cm^{-1}): 3209 (N-H), 1669 (C=O), 1633 (C=N), 1589 (C=C), 751 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 7.57–7.72 (m, 3H, H-3', 5' and 6), 7.85–7.90 (t, 1H, *J* = 7.71 Hz, H-7), 7.97–8.03 (m, 2H, H-6' and 5), 8.25–8.28 (d, 1H, *J* = 8.12 Hz, H-8), 8.87 (s, 1H, H-4), 8.94 (s, 1H, CH=N), 12.14 (s, 1H, CONH).

4-Bromo-N'-[(2-chloroquinolin-3-yl)methylidene]benzohydrazide (3f)

IR (KBr, cm^{-1}): 3274 (N-H), 1670 (C=O), 1630 (C=N), 1591 (C=C), 759 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 7.53–7.56 (d, 2H, *J* = 7.82 Hz, H-3' and 5'), 7.69–7.74 (t, 1H, *J* = 7.51 Hz, H-6), 7.84–7.91 (m, 3H, H-2', 6' and 7), 7.97–8.00 (d, 1H, *J* = 8.05 Hz, H-5), 8.24–8.27 (d, 1H, *J* = 8.01 Hz, H-8), 8.91 (s, 1H, H-4), 8.98 (s, 1H, CH=N), 12.18 (s, 1H, CONH).

***N'*-[(2-Chloroquinolin-3-yl)methylidene]-4-fluorobenzohydrazide (3g)**

IR (KBr, cm^{-1}): 3210 (N-H), 1662 (C=O), 1635 (C=N), 1582 (C=C), 759 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 7.59–7.72 (m, 3H, H-3', 5' and 6), 7.81–7.88 (m, 3H, H-2', 6' and 7), 7.97–8.00 (d, 1H, *J* = 8.23 Hz, H-5), 8.23–8.26 (d, 1H, *J* = 8.10 Hz, H-8), 8.90 (s, 1H, H-4), 8.97 (s, 1H, CH=N), 12.18 (s, 1H, CONH).

***N'*-[(2-Chloroquinolin-3-yl)methylidene]-4-nitrobenzohydrazide (3h)**

IR (KBr, cm^{-1}): 3260 (N-H), 1673 (C=O), 1625 (C=N), 1579 (C=C), 766 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 7.55–7.58 (d, 2H, *J* = 7.83 Hz, H-3' and 5'), 7.67–7.72 (t, 1H, *J* = 7.49 Hz, H-6), 7.83–7.88 (m, 3H, H-2', 6' and 7), 7.99–8.02 (d, 1H, *J* = 7.93 Hz H-5), 8.25–8.28 (d, 1H, *J* = 8.17 Hz, H-8), 8.91 (s, 1H, H-4), 8.97 (s, 1H, CH=N), 12.19 (s, 1H, CONH).

***N'*-[(2-Chloroquinolin-3-yl)methylidene]-3-methylbenzohydrazide (3i)**

IR (KBr, cm^{-1}): 3265 (N-H), 1669 (C=O), 1631 (C=N), 1578 (C=C), 757 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 2.39 (s, 3H, CH_3), 7.37–7.41 (m, 2H, H-4' and 5'), 7.66–7.71 (t, 1H, *J* = 7.25 Hz, H-6), 7.84–7.88 (m, 3H, H-2', 6' and 7), 7.98–8.01 (d, 1H, *J* = 8.42 Hz, H-5), 8.22–8.25 (d, 1H, *J* = 8.08 Hz, H-8), 8.79 (s, 1H, H-4), 8.96 (s, 1H, CH=N), 12.15 (s, 1H, CONH).

***N'*-[(2-Chloroquinolin-3-yl)methylidene]-2-phenylacetohydrazide (3j)**

IR (KBr, cm^{-1}): 3260 (N-H), 1671 (C=O), 1629 (C=N), 1593 (C=C), 759 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 3.67 (s, 2H, CH_2), 7.43–7.57 (m, 5H, H-2', 3', 4', 5', and 6'), 7.68–7.73 (t, 1H, *J* = 7.39 Hz, H-6), 7.86–7.91 (t, 1H, *J* = 7.41 Hz, H-7), 7.98–8.01 (d, 1H, *J* = 8.27 Hz, H-5), 8.23–8.26 (d, 1H, *J* = 7.93 Hz, H-8), 8.78 (s, 1H, H-4), 8.97 (s, 1H, CH=N), 12.11 (s, 1H, CONH).

2-(4-Chlorophenyl)-N'-[(2-chloroquinolin-3-yl)-methylidene]acetohydrazide (3k)

IR (KBr, cm^{-1}): 3252 (N-H), 1668 (C=O), 1638 (C=N), 1589 (C=C), 762 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 3.69 (s, 2H, CH_2), 7.54–7.73 (m, 5H, H-2', 3', 5', 6' and 6), 7.87–7.92 (t, 1H, *J* = 7.36 Hz, H-7), 7.98–8.01 (d, 1H, *J* = 8.31 Hz, H-5), 8.22–8.25 (d, 1H, *J* = 7.89 Hz, H-8), 8.80 (s, 1H, H-4), 8.97 (s, 1H, CH=N), 12.15 (s, 1H, CONH).

N'-[(2-Chloroquinolin-3-yl)methylidene]-2-phenoxyacetohydrazide (3l)

IR (KBr, cm^{-1}): 3210 (N-H), 1665 (C=O), 1640 (C=N), 1591 (C=C), 758 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 4.73 (s, 2H, OCH_2), 7.11–7.20 (m, 3H, H-3', 4' and 5'), 7.39–7.42 (d, 2H, *J* = 7.28 Hz, H-2' and 6'), 7.67–7.72 (t, 1H, *J* =

7.57 Hz, H-6), 7.83–7.88 (t, 1H, *J* = 7.29 Hz, H-7), 7.94–7.97 (d, 1H, *J* = 8.09 Hz, H-5), 8.20–8.23 (d, 1H, *J* = 7.93 Hz, H-8), 8.89 (s, 1H, H-4), 8.94 (s, 1H, CH=N), 12.20 (s, 1H, CONH).

2-[(2-Chloroquinolin-3-yl)methylidene]hydrazinecarboxamide (3m)

IR (KBr, cm^{-1}): 3243 (N-H), 1685 (C=O), 1632 (C=N), 1591 (C=C), 751 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 5.03 (s, 2H, NH_2), 7.64–7.69 (t, 1H, *J* = 7.53 Hz, H-6), 7.89–7.94 (t, 1H, *J* = 7.26 Hz, H-7), 8.00–8.02 (d, 1H, *J* = 8.07 Hz, H-5), 8.19–8.22 (d, 1H, *J* = 8.04 Hz, H-8), 8.77 (s, 1H, H-4), 9.06 (s, 1H, CH=N), 11.39 (s, 1H, CONH).

2-[(2-Chloroquinolin-3-yl)methylidene]hydrazinecarbothioamide (3n)

IR (KBr, cm^{-1}): 3260 (N-H), 1347 (C=S), 1625 (C=N), 1579 (C=C), 748 (C-Cl). ^{1}H-NMR (300 MHz, DMSO-d_6, δ, ppm): 5.27 (bs, 2H, NH_2), 7.63–7.68 (t, 1H, *J* = 7.48 Hz, H-6), 7.89–7.94 (t, 1H, *J* = 7.47 Hz, H-7), 8.01–8.03 (d, 1H, *J* = 8.21 Hz, H-5), 8.21–8.24 (d, 1H, *J* = 8.09 Hz, H-8), 8.78 (s, 1H, H-4), 9.01 (s, 1H, CH=N), 11.43 (s, 1H, CONH).

Pharmacology

Anticonvulsant screening (17, 18).

Male albino mice weighing 25–30 g were used as experimental animals. Test compounds were suspended in PEG 200 for MES and scPTZ screening. The animals were maintained at an ambient temperature 22 ± 1°C, in groups of six per cage under standard laboratory conditions and allowed to free access to food and water.

Maximal Electroshock Test (MES test)

Test compounds were administered as an *i.p.* injection at dose level of 100 and 300 mg/kg and the anticonvulsant activity was assessed after 0.5 h and 4 h intervals of administration. Maximal electroshock seizures were elicited in mice by delivering 60 Hz, 50 mA electrical stimuli for 0.2 s *via* ear clip electrodes. Abolition of hind limb tonic extensor component of the seizure in half or more of the animals is defined as protection.

Subcutaneous pentylenetetrazole (scPTZ) seizure test

The scPTZ test primarily identifies compounds that raise seizure threshold. The scPTZ test utilizes a dose of pentylenetetrazole (85 mg/kg). This produces clonic seizures lasting for a period of at least five seconds in 97 percent (CD_{97}) of animals tested. Solutions of test compounds were administered as an *i.p.* injection at dose levels of 100 and 300 mg/kg. At the anticipated time of testing (0.5 h and 4.0 h) the convulsant (PTZ) was administered subcutaneously and animals were observed over a 30 min period. Absence of clonic spasm in half or more of the animals in the observed time period indicated a compound's ability to abolish the effect of pentylenetetrazole on seizure threshold.

Neurotoxicity screening (19)

The minimal motor impairment was measured in mice by the rotarod test. The mice were trained to stay on an accelerating rotarod of diameter 3.2 cm that rotates at 10 rpm. Neurotoxicity was indicated by the inability of the animal to maintain equilibration on the rod for at least one minute in each of the three trials. The dose at which 50% of the animals enabled to balance themselves and fell off the rotating rod was determined.

RESULTS AND DISCUSSION

Chemistry

The synthesis of compounds **(3a-n)** was undertaken as shown in Scheme 1. The required 2-chloroquinoline-3-carboxaldehyde **2** was prepared by the action of DMF/$POCl_3$ (Vilsmeier-Haack reaction) on N-aryl acetamide **1**. The various hydrazones, semicarbazones and thiosemicarbazones (**3a-n)** were synthesized by condensation of 2-chloroquinoline-3-aldehyde with substituted acyl hydrazines, semicarbazide and thiosemicarbazide in absolute ethanol, in a yield ranging between 63 to 86%. All the compounds were characterized by IR and ^{1}H-NMR along with ^{13}C-NMR and FAB-MS spectra of some selected compounds. The IR spectra of final compounds (**3a-n**) showed absorption bands at 1659 to 1685 cm^{-1} appearing due to presence of C=O functional group, while band observed at 1620 to 1640 cm^{-1} corresponds to C=N linkage. The NH band was observed in the range of 3203 to 3284 cm^{-1}. In ^{1}H-NMR spectra the synthesis of the compounds **(3a-n)** was confirmed on the basis that aldehydic proton, which was observed at δ 10.52 ppm in compound **2,** disappeared and a new signal was found arising due to the azomethine group (CH=N) present in final compounds. The signal due to CH=N (azomethine) which was present in all the compounds was observed as a singlet varying from δ values 8.94–9.06 ppm integrating for one proton. The CONH proton resonated at δ values between 11.39 to 12.22 ppm as singlet to broad singlet and this signal was absent when ^{1}H-NMR was recorded in the

Table 1. Physicochemical data of compounds **(3a-n)**.

Comp. no.	X/Y	R	Yield (%)[a]	M. p. (°C)	Molecular formula	Mol. wt.	C log P [b]	Rf [c]
3a	–	H	71	220–223	$C_{17}H_{12}ClN_3O$	309.74	3.57 ± 0.36	0.62
3b	–	4-CH_3	68	193–195	$C_{18}H_{14}ClN_3O$	323.77	4.03 ± 0.36	0.59
3c	–	4-OCH_3	70	207	$C_{18}H_{14}ClN_3O_2$	339.77	3.44 ± 0.48	0.62
3d	–	4-Cl	65	236–237	$C_{17}H_{11}Cl_2N_3O$	344.19	4.33 ± 0.38	0.67
3e	–	2,4-$Cl)_2$	72	272	$C_{17}H_{10}Cl_3N_3O$	379.63	4.52 ± 0.40	0.58
3f	–	4-Br	68	258–260	$C_{17}H_{11}BrClN_3O$	388.64	4.51 ± 0.45	0.65
3g	–	4-F	73	245–246	$C_{17}H_{11}ClFN_3O$	327.74	3.79 ± 0.45	0.59
3h	–	4-NO_2	63	215	$C_{17}H_{11}ClN_4O_3$	354.74	3.19 ± 0.50	0.64
3i	–	3-CH_3	73	189	$C_{18}H_{14}ClN_3O$	323.77	4.03 ± 0.36	0.66
3j	CH_2	H	78	190–191	$C_{18}H_{14}ClN_3O$	323.77	4.76 ± 0.58	0.64
3k	CH_2	4-Cl	76	217	$C_{18}H_{13}Cl_2N_3O$	358.22	5.36 ± 0.58	0.62
3l	CH_2O	H	70	198	$C_{18}H_{14}ClN_3O_2$	339.77	4.60 ± 0.59	0.67
3m	O	H	83	248–250	$C_{11}H_9ClN_4O$	248.66	2.28 ± 0.33	0.58
3n	S	H	86	266–267	$C_{11}H_9ClN_4S$	264.73	2.87 ± 0.33	0.61

[a] After recrystallization from ethanol : DMF (8:2, v/v); [b] calculated from ACD lab software 12.0 version; [c] solvent for TLC: toluene : ethyl acetate : formic acid (5 : 4 : 1, v/v/v).

Table 2. Anticonvulsant and neurotoxicity screening data of compounds **(3a-n).**

Compd. no.	Intraperitoneal injection in mice[a]					
	MES screening		scPTZ screening		Neurotoxicity screen	
	0.5 h	4.0 h	0.5 h	4.0 h	0.5 h	4.0 h
3a	300	—	300	—	—	—
3b	300	—	—	—	—	—
3c	300	—	—	—	—	—
3d	100	300	300	300	300	—
3e	100	100	100	300	300	300
3f	Nt	Nt	Nt	Nt	Nt	Nt
3g	100	300	100	300	300	—
3h	300	—	300	—	—	—
3i	Nt	Nt	Nt	Nt	Nt	Nt
3j	—	—	—	—	Nt	Nt
3k	300	—	—	—	300	—
3l	—	—	—	—	Nt	Nt
3m	100	300	300	—	300	—
3n	100	300	300	300	300	—
Phenytoin	30	30	—	—	100	100
Carbamazepine	30	100	100	300	100	300

[a]Doses of 100 and 300 mg/kg of the compound were administered and the protection and neurotoxicity were measured after 0.5 and 4 h. The figures indicate the minimal dose required to cause protection or neurotoxicity in 50% or more of the animals. The dash (—) indicates the absence of anticonvulsant activity or neurotoxicity. Nt denotes not tested.

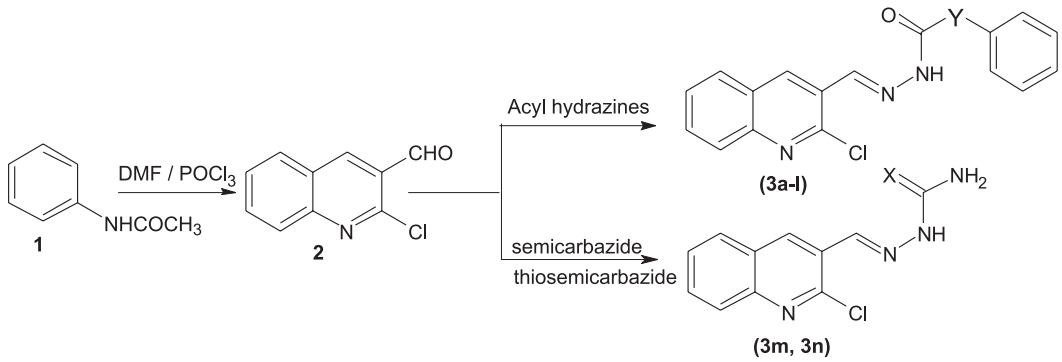

Figure 2. Scheme for synthesis of 2-chloroquinolinyl-hydrazone derivatives **(3a-n)**.

presence of D_2O. The aromatic protons of the benzoyl ring (3'/5') appeared as a doublet in compounds **3b**, **3c, 3d, 3f, 3h**, i.e., (*para*-disubstituted product) with a coupling constant of $J = 7.78$ to 8.76 Hz, while the 2'/6' protons of the benzoyl ring were observed as multiplets merged with the quinolinyl protons and they resonated at slightly more down field value which may be due to strong electron withdrawing effect of carbonyl group. In compounds **3a-n**, protons of the quinoline ring were observed as follows. The H-4 proton of the quinoline ring was observed as singlet at down field δ values 8.78–8.91 ppm and H-6 and H-7 appeared as triplets at δ values from 7.68–7.75 ppm (J = 7.32–7.51 Hz) and 7.84–7.92 ppm (J = 7.36-7.68 Hz), respectively. Sometimes the triplet arising due to presence of H-7 proton was found merged with aromatic protons of the benzoyl ring, e.g., in compounds **3b**, **3f, 3g, 3h and 3i**. The H-5 and H-8 protons of the quinoline appeared as doublet at δ values 7.97 to 8.02 ppm (J = 7.93–8.42 Hz) and 8.22 to 8.26 ppm (J = 8.01–8.26 Hz), respectively. The ^{13}C-NMR spectra of some of the selected compounds (**3a, 3b, 3c)** were also recorded. The carbon signal due to (-CHO) of intermediate **2** was observed at δ values 188.71 ppm, however, this signal was found to be absent in compounds **3a, 3b, 3c** but a new signals at δ values 157.17, 159.09 and 159.98 ppm arised due to the presence of CH=N, respectively, which further supported, in addition to ^{1}H-NMR spectra, the synthesis of compounds **3a-n**. The C=O carbon of CONH appeared at δ 168.34–170.28 ppm. The mass spectra of some selected compounds (**3a, 3b and 3c)** were recorded using FAB-MS technique. The molecular ion peaks of these selected compounds were observed as M^+ and M+2 peaks at *m/z* 310/312, *m/z* 324/326 and *m/z* 340/342, respectively. These data were found in agreement with the structures assigned to these compounds.

Anticonvulsant activity

All the newly syntheszied compounds (**3a**-**3n**) were tested *in-vivo* in order to evaluate their anticonvulsant activity. Both MES and scPTZ model of anticonvulsant activity were employed and data have been summarized in Table 2. The 2-chloroquinolinyl hydrazones derived from substituted benzoic acid hydrazide (**3a-3i**), which showed anti-MES activity at dose levels of 100 to 300 mg/kg, include **3a, 3b, 3c, 3d, 3e, 3g,** and **3h.** This is indicative of their ability to prevent seizure spread. Among them, compounds **3d, 3e, 3g** (0.5 and 4.0 h) exhibited protection against MES model at dose level of 100 mg/kg and only compound **3e** demonstrated anticonvulsant activity after 4.0 h of administration. A majority of these compounds (**3a, 3d, 3e, 3g, 3h**) also exhibited anticonvulsant activity in scPTZ screening at a dose of 100 to 300 mg/kg. This model identifies compounds that elevate seizure threshold. Only compound **3e** and **3g** showed anti-scPTZ activity at a dose of 100 mg/kg after 0.5 h and compounds **3d, 3e, 3g** showed anti-scPTZ activity at dose level of 300 mg/kg after 4.0 h. Anticonvulsant activity was found absent in 2-chloroquinolinyl hydrazones of substituted phenyl/phenoxy acetic acid hydrazides (**3j, 3l**), whereas compound **3k** showed anti-MES activity at a dose of 300 mg/kg (0.5 h). The semicarbazone and thiosemicarbazone (**3m, 3n**) exhibited good anti-MES activity at a dose of 100 mg/kg (0.5 h) and 300 mg/kg (4.0 h), whereas in scPTZ model they showed anticonvulsant activity at a dose of 300 mg/kg (0.5 h). Neurotoxicity screening of compounds revealed that compounds **3d, 3e, 3g, 3k, 3m and 3n** were neuro-

toxic at anticonvulsant dose of 300 mg/kg and compounds **3a, 3b, 3c, 3h** did not show neurotoxicity at a maximal administered dose of 300 mg/kg.

CONCLUSION

Examination of the *in-vivo* anticonvulsant activity profile for differently substituted 2-chloroquinolinyl hydrazones (**3a**-**3n**) against electrical and chemical model of convulsions, provide a preliminary SAR, which may be summarized as follows. Among the compounds tested, those with electron withdrawing group (Cl, F) in the benzoyl ring (**3d**, **3e**) showed good anticonvulsant activity and were long acting. Anticonvulsant activity decreases or abolishes when a spacer like CH_2 or CH_2O was introduced between the carbonyl group and the phenyl ring in compounds **3j**, **3k**, **3l**. Replecement of phenyl ring in componds **3m**, **3n** with amino group results in retention of the activity. Hence, it appears that 2-chloroquinoline could be employed as a hydrophobic domain in hydrazone motif (compound A in Fig. 1) for the development of new class of anticonvulsant agents. However, further investigation and optimization of molecules are needed before their possible use as anticonvulsant agents.

REFERENCES

1. Malawska B.: Curr. Topics Med. Chem. 5, 69 (2005).
2. Shindikar A.V., Khan F., Viswanathan C.L.: Eur. J. Med. Chem. 41, 786 (2006).
3. Bell G.S., Sander J.W.: Seizure 11 (Suppl. A.) 306 (2002).
4. Agarwal N.: J. Pharm. Pharmaceut. Sci. 7, 260 (2004).
5. Popp F.D.: Eur. J. Med. Chem. 24, 313 (1989).
6. Ragavendran J.V., Sriram D., Patel S.K., Reddy I.V., Bharathwajan N., Stables J., Yogeeswari P.: Eur. J. Med. Chem. 42, 146 (2007).
7. Dimmock J.R., Vashishthaa S.C., Stables J.P.: Eur. J. Med. Chem. 35, 241 (2000).
8. Sridhar S.K., Pandeya S.N., Stables J.P., Ramesh A.: Eur. J. Pharm. Sci. 16, 129 (2002).
9. Dimmock J.R., Pandeya S.N., Stables J.P., Quail J.W., Pugazhenthi U. et al.: Eur. J. Med. Chem. 30, 303 (1995).
10. Dimmock J.R., Vashishthaa S.C., Stables J.P.: Pharmazie 50, 823 (1995).
11. Pandeya S.N., Agarwal A.K., Singh A., Stables J.P.: Acta Pharm. 53, 15 (2003).
12. Guan L-P., Jin Q-H., Tian G-R. Chai K-Y., Quan Z-S.: J. Pharm. Pharmaceut. Sci. 10, 254 (2007).
13. Muruganantham N., Sivakumar R., Anbalagan N., Gunasekaran V., Leonard J.T.: Biol. Pharm. Bull. 27, 1683 (2004).
14. Bawa S., Kumar S.: Indian J. Chem. 48B, 142 (2009).
15. Meth-Cohn O., Narine B., Tarnowski B.: J. Chem. Soc. Perkin Trans. I 1520 (1981).
16. Vogel's Textbook of practical organic chemistry, 5th edn., pp. 1007 and 1269, Addison-Wesley Longman Inc., UK 2006.
17. Krall R.I., Penry J.K., White B.G., Kupferberg H.J., Swinyard E.A.: Epilepsia 19, 409 (1978).
18. Loscher W., Schmidt D.: Epilepsy Res. 17, 95 (1994).
19. Dunham N.W., Miya T.A.: J. Am. Pharm. Assoc. Sci. Ed. 46, 208 (1957).

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