Full Paper

Synthesis and Anticonvulsant Activity of 5-Phenyl-[1,2,4]-triazolo[4,3-*a*]quinolines

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A series of novel 5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline derivatives was synthesized by the cyclization of 2-chloro-4-phenyl-1,2-dihydronaphthalene with formohydrazide. The starting material 2-chloro-4-phenyl-1,2-dihydronaphthalene was synthesized from ethyl-3-oxo-3-phenylpropanoate and substituted aniline. Their anticonvulsant activities were evaluated by the maximal electroshock (MES) test and their neurotoxicity was evaluated by the rotarod neurotoxicity test (Tox). The maximal electroshock test showed that 7-hexyloxy-5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline **4f** was found to be the most potent compound with an ED_{50} value of 6.5 mg/kg and a protective index (PI = ED_{50}/TD_{50}) value of 35.1, which was much higher than the PI of the reference drug phenytoin.

Keywords: Anticonvulsant activity / Maximal electroshock / Synthesis / [1,2,4]Triazolo[4,3-*a*]quinoline

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Introduction

Epilepsy, a ubiquitous disease characterized by recurrent seizures, inflicts more than 60 million people worldwide according to epidemiological studies [1]. For epilepsy treatment, nearly 95% of clinically available drugs were approved before 1985 and they could provide satisfactory seizure control for 60–70% of the patients. These drugs, however, also cause notable adverse side effects such as drowsiness, ataxia, gastrointestinal disturbance, hepatotoxicity, megaloblastic anaemia [2–4], and even life-threatening conditions [5]. Therefore, the continued search for safer and more effective new antiepileptic drugs (AEDs) is imperative and challenging in medicinal chemistry.

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Abbreviations: maximal electroshock (MES); structure-activity relationships (SAR)

In our previous study [6], a series of derivatives of 5-substituted-phenyl-4,5-dihydro-1,2,4-triazolo[4,3-*a*]quinolines was first found to have anticonvulsant activities, but they showed weaker anticonvulsant effects than the reference drug phenytoin in the maximal electroshock (MES) test. It is possible that the phenyl ring in the 5-position of the compound **I** can not conjugate with the triazole nucleus. In order to obtain compounds with better anticonvulsant activity, according to structure-activity relationships (SAR), we noted that two structural modifications of compound **I**, *i. e.* the introduction of a double bond into the 4- and 5-position, and the additional introduction of a methoxy group in the 7-position on the phenyl ring, which in turn gave compound **II** and **4a**, respectively, resulted in markedly enhance anticonvulsant activity. On the basis of our recent investigation [7, 8] on the relationship of the anticonvulsant activity and the structure of 5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline **II**, we thought that the triazole ring C might be the main receptor-binding structure. So, increasing the electron-cloud density at the triazole nucleus might enhance compound binding with the receptor. The introduction of a double bond into the 4- and 5-position could possibly pro-

mote the conjugation of the phenyl ring in the 5-position of ring B with the triazole nucleus, which enhances electron-cloud density at the triazole nucleus. The other aromatic ring A enhances the hydrophobicity of the target compounds, thus make them more permeable to the blood-brain barrier and enhance their anticonvulsant activity. Therefore, we thought that the presence of A- and C-rings was essential for the anticonvulsant activity.

Furthermore, to understand the structure-activity relationship of ring A, we designed and synthesized a novel series of substituted ring-A derivatives of 5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline. Their anticonvulsant activities were evaluated by the maximal electroshock (MES) test and their neurotoxicity was evaluated by the rotarod neurotoxicity test (Tox).

Results and discussion

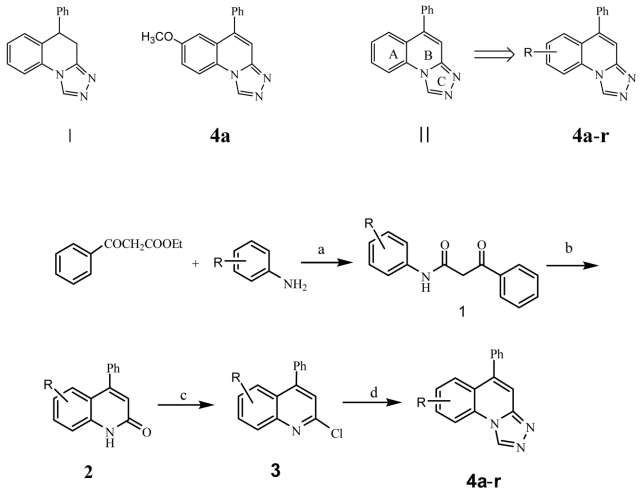
Synthesis

The synthesis of the target compounds **4a – r** were accomplished according to the reaction sequence illustrated in Scheme 1. Compounds **1** were prepared according to a reported procedure [9]. Briefly, they were obtained by acylation of ethyl-3-oxo-3-phenylpropanoate and substituted aniline with good yield. Compounds **2** were prepared with compounds **1** in the presence of polyphosphoric acid as the catalyst with high yield [9, 10]. Compounds **3** were obtained [11, 12] by chlorization of compounds **2** with phosphorus oxychloride ($POCl_3$). Then, 5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline **4a – r** were prepared by cyclizing compounds **3** with formic hydrazide [11]. In the first step reaction, the reaction time of compounds **1** with groups like -OCH_3, -OC_2H_5, -CH_3 etc. was 1 h, and with groups like -F, -Cl, and –Br, it was 7 h. In the last step reaction, the reaction time of compounds **4a – r** with groups like -OCH_3, -OC_2H_5, -CH_3 etc. was 7 h, and with groups like -F, -Cl, and –Br, it was 40 h. These differences in reaction time might be due to the electron-donating (-OCH_3, -CH_3) and electron-absorbing (-F, -Cl) property of the R groups which increases or decreases, respectively, the electron-cloud density of the N atom near the phenyl ring or quinoline ring.

Pharmacological evaluations

Pharmacological tests of the 5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline **4a – r** were conducted at the Epilepsy Branch of the National institute of Neurological Disorders and Stroke (NINDS) following the protocol adopted by the Antiepileptic Drug Development (ADD) program [13, 14].

The results of preliminary (phase I) screening of **4a – r** are summarized in Table 1. All synthesized compounds



R:

4a = 7-OCH_3	**4e** = 7-OC_5H_{11}	**4i** = 7-CH_3	**4m** = 7-$OCH_2C_6H_5$	**4q** = 7-Br
4b = 7-OC_2H_5	**4f** = 7-OC_6H_{13}	**4j** = 9-CH_3	**4n** = 7,8-$(OCH_3)_2$	**4r** = 6,9-Cl_2
4c = 7-OC_3H_7	**4g** = 7-OC_7H_{15}	**4k** = 7,8-OCH_2O-	**4o** = 7-F	
4d = 7-OC_4H_9	**4h** = 7-OC_8H_{17}	**4l** = 6,9-$(OCH_3)_2$	**4p** = 7-Cl	

Reagents and conditions: (a) 1 h or 7 h, 140 – 150°C; (b) PPA, 140°C, 30 min; (c) $POCl_3$, 7 h, 120°C; (d) $HCONHNH_2$, *n*-butyl alcohol, 10 h or 40 h, 140 – 150°C.

Scheme 1. The synthesis route of compounds **4a – r**.

exhibited strong anticonvulsant activity at a dose of 30 mg/kg against MES-induced seizures. In contrast, in the former study [6], only two target compounds showed anticonvulsant activity at a dose of 30 mg/kg.

In the phase-II pharmacology test (Table 2), in the former study [6, 8], the compound **II** (ED_{50} = 28.4 mg/kg) exhibited better activity than compound **I** (ED_{50} = 54.8 mg/kg). This increase in anticonvulsant activity, as we have mentioned earlier, might be due to the conjugation of the phenyl ring in the 5-position and the triazole nucleus through the introduction of a double bond into the 4- and 5-position of compound **I**, which increased electron-cloud density at the triazole nucleus and then enhanced the compound binding with its receptor. Similarly, the fact that the anticonvulsant activity of compound **4a** (ED_{50} = 9.2 mg/kg) was stronger than that of compound **II** (ED_{50} = 28.4 mg/kg) could be explained by the introduction of a methoxyl group in the 7-position on the phenyl ring of compound **II** which, through conjugation of the lone-pair electron at the methoxyl group, increased electron-cloud density at triazole nucleus and enhanced compound binding with its receptor.

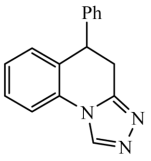
All the compounds in **4a – r** had anti-MES activities better than the compound **I** and all except one, **4n**, of these compounds had anti-MES activities better than the compound **II**. The following SARs were gained through ana-

Table 1. Phase-I mouse anticonvulsant activity data in mice (i. p.)[a].

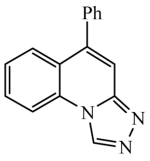
Compound	R	MES[b]	
		0.5 h	4 h
4a	7-OCH_3	30[c]	30
4b	7-OCH_2CH_3	30	–[d]
4c	7-$O(CH_2)_2CH_3$	30	–
4d	7-$O(CH_2)_3CH_3$	30	–
4e	7-$O(CH_2)_4CH_3$	30	–
4f	7-$O(CH_2)_5CH_3$	30	30
4g	7-$O(CH_2)_6CH_3$	30	–
4h	7-$O(CH_2)_7CH_3$	30	–
4i	7-CH_3	30	–
4j	9-CH_3	30	30
4k	7,8-OCH_2O-	30	–
4l	6,9-$(OCH_3)_2$	30	–
4m	7-$OCH_2C_6H_5$	30	–
4n	7,8-$(OCH_3)_2$	30	–
4o	7-F	30	–
4p	7-Cl	30	–
4q	7-Br	30	–
4r	6,9-Cl_2	30	–

[a] All of tested compounds were dissolved in Polyethylene glycol-400.
[b] The maximal electroshock test was carried out 30 min after administration of the test compounds.
[c] Doses are denoted in milligrams per kilogram.
[e] = no activity at 300 mg/kg.

Table 2. Phase-II quantitative anticonvulsant data in mice (test drug administered i. p.).



I



II

Compound	MES, ED_{50}[a]	TD_{50}[b]	PI[c] (TD_{50}/ED_{50})
I	54.8(46.3–64.8)[d]	141.6 (123.7–161.6)	2.6
II	28.4 (21.1–38.5)	126.8 (105.5–152.4)	4.5
4a	9.2 (7.0–12.1)	152.1(127.6–181.2)	16.6
4b	23.8 (20.0–28.8)	91.3 (75.9–109.8)	3.8
4c	24.3 (22.2–26.7)	105.7 (87.9–127.1)	4.3
4d	25.5 (21.3–26.7)	126.8 (108.2–148.6)	5.0
4e	25.8 (17.9–30.8)	169.9 (141.6–203.8)	6.6
4f	6.5 (4.6–9.2)	228.2 (189.7–274.2)	35.1
4g	19.8 (16.4–23.9)	152.2 (126.7–183.0)	7.7
4h	21.2 (17.8–25.7)	118.0 (98.4–141.5)	5.6
4i	25.5 (21.4–30.3)	101.9 (84.9–122.2)	4.0
4j	11.0 (8.4–14.5)	122.3 (102.0–146.7)	11.1
4k	14.5 (10.0–20.9)	77.2 (70.0–86.1)	5.3
4l	18.3 (15.0–22.4)	101.4 (89.7–116.6)	5.5
4m	23.7 (19.5–28.4)	131.4 (109.3–158.0)	5.5
4n	35.3 (29.4–42.4)	63.4 (52.7–76.2)	1.8
4o	15.4 (12.9–18.3)	126.7 (107.6–149.2)	8.2
4p	19.0 (15.8–22.9)	163.8 (136.6–196.5)	8.6
4q	24.5 (21.0–28.6)	109.5 (91.1–131.6)	4.5
4r	23.8 (20.0–28.8)	76.1 (63.3–91.5)	3.2
Phenytoin	9.5 (8.1–10.4)	65.5 (52.5–72.9)	6.9

[a] ED_{50}-median effective dose required to assure anticonvulsant protection in 50% animals.
[b] TD_{50}-median toxic dose eliciting minimal neurological toxicity in 50% animals.
[c] PI protective index (TD_{50} / ED_{50}).
[d] 95% confidence limits given in parentheses.

lyzing the activities of synthesized compounds **4a**–**r**. First, the length of the alkyl chain appeared to have a direct impact on the anticonvulsant activity of the 7-alkyloxyl derivatives. As shown in Table 2, the compound **4a**, with a methoxyl group at the 7-position, showed stronger activity (ED_{50} = 9.2 mg/kg). Compounds **4b**–**4e**, which contain an alkyloxyl group bearing two to five carbon atoms at the 7-position, exhibited weaker activity (ranging from 23.8 to 25.8 mg/kg) than compound **4a**. Compound **4f**, with a hexyloxy-group substitution at the 7-position, exhibited the strongest anticonvulsant activity among **4a**–**r** (ED_{50} = 6.5 mg/kg) which was better than the reference drug phenytoin in the MES test. It also had a very low neurotoxicity (TD_{50} = 228.2 mg/kg) and thus a high PI value of 35.1 was gained. However, compounds with a 7-alkyloxyl chain having more than six carbon atoms, **4g** and **4h**, had lower activity than **4f** (ED_{50} = 19.8 and 21.2 mg/kg, respectively).

When the substitution was a member of the halogen family, the activity order was 7-F > 7-Cl > 7-Br, and the position of atom Cl on the phenyl ring influenced the anticonvulsant activity, with the potency order of the two Cl-substituted derivatives being 7-Cl > 6,9-Cl_2.

Comparing a 7-substituted compound containing a phenyl ring, **4m**, two methyl-substituted compounds, **4i** and **4j**, and three compounds substituted at two positions, **4k**, **4l**, **4n**, indicated that these different substitutions contributed to the anticonvulsant activity in the order of 9-CH_3 (ED_{50} = 11.0 mg/kg) > 7,8-OCH_2O– > 6,9-$(OCH_3)_2$ > 7-$OCH_2C_6H_5$ > 7-CH_3 > 7,8-$(OCH_3)_2$ (ED_{50} = 35.3 mg/kg), indicating that the position of substitution influences the activity. The 9-methyl-substituted derivative **4j** (ED_{50} = 11.0 mg/kg) exhibited stronger anti-MES activity than the 7-methyl-substituted derivative **4i** (ED_{50} = 25.5 mg/kg). Similarly, substitution with two methoxyl groups at positions 6 and 9 (compound **4l**, ED_{50} = 18.3 mg/kg) rather than positions 7 and 8 (**4n**, ED_{50} = 35.3 mg/kg) resulted in higher anti-MES activity. The SAR in the above two pairs of compounds might be explained by the fact that the former compounds increased electron-cloud density at the triazole nucleus.

Conclusions

In conclusion, a new series of 7-substituted-5-phenyl-[1,2,4]-triazolo[4,3-*a*]quinoline derivatives was synthesized and the pharmacological properties were evaluated. It was found that these compounds exhibited remarkable anticonvulsant activity. Especially, compound **4f** not only has stronger anticonvulsant activity, but also has markedly lower neurotoxicity than the reference drug phenytion, and, thus, a larger protective index was observed for this compound in the MES test.

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The authors have declared no conflict of interest.

Experimental

Chemistry

Melting points were determined in open capillary tubes and were uncorrected. IR spectra were recorded (in KBr) on a FT-IR1730 (Bruker, Switzerland), ^{1}H-NMR spectra were measured on a AV-300 (Bruker), ^{13}C-NMR spectra were measured on a AV-300 (Bruker), and all chemical shifts were given in ppm relative to tetramethysilane. Microanalyses of C, N, and H were performed using a Heraeus CHN Rapid Analyzer (Heraeus, Hanau, Germany). Mass spectra were measured on an HP1100LC (Agilent Technologies, USA). The major chemicals were purchased from Alderich Chemical Corporation. All other chemicals were the analytical grade.

*2-Chloro-4-phenylquinoline **3***

4-Phenylquinolin-2(1*H*)-one (1.00 g, 0.04 mol) was placed in a round-bottomed flask, to which 10 mL $POCl_3$ was added. The mixture was refluxed for 7 h in a nitrogen atmosphere. After removing the solvent under reduced pressure, the residue was dissolved in 30 mL of water, and then extracted with CH_2Cl_2. The CH_2Cl_2 layer was dried over anhydrous $MgSO_4$. Evaporation of the solvents gave a crude product that was purified by recrystallization in hexane, and a yellow solid was obtained in a yield of 81%. ^{1}H-NMR (CDCl3, ppm): 4.01 (s, 3H, -OCH_3), 7.95 (s, 1H, =CH), 8.08 – 8.13 (m, 3H, -C_6H_3), 7.41 – 7.56 (m, 5H, -C_6H_5); MS m/z: 269 [M + 1]. Anal. Calcd. for $C_{16}H_{12}ClNO$: C, 71.25; H, 4.48; N, 5.19. Found: C, 71.14; H, 4.36; N, 4.95.

*General procedure for the synthesis of compounds II and **4a–r***

2-Chloro-4-phenylquinoline (0.04 mol) and formohydrazide (0.04 mol) were dissolved in *n*-butyl alcohol in a round-bottomed flask, and the mixture was refluxed for 20 – 40 h in a nitrogen atmosphere. Solvents were removed under reduced pressure, and the residue was extracted twice with 30 mL dichloromethane. The dichloromethane layer was washed three times with water (3 × 30 mL) and dried over anhydrous MgSO4. After removing the solvents, the products was purified by silica gel column chromatography (dichloromethane / methanol = 20 : 1).

5-Phenyl-[1,2,4]-triazolo[4,3-a]quinoline II

Yield: 78%; m.p.: 164 – 166°C IR (KBr) cm^{-1}: 1613 (C=N), 1297 (C-N), 1243, 1024 (C-O-C), 1151 (N-N); ^{1}H-NMR ($CDCl_3$ ppm) : 7.51 (s, 1H, =CH), 7.81 – 8.09 (m, 4H, -C_6H_4), 7.54 – 7.77 (m, 5H, -C_6H_5), 9.31 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 114.04, 115.85, 123.40, 126.29, 128.39, 128.71, 128.75, 129.36, 129.94, 130.48, 134.56, 137.40, 141.91, 147.94; MS m/z: 246 [M + 1]. Anal..Calcd. for $C_{16}H_{11}N_3$: C, 78.35; H, 4.52; N, 17.13. Found: C, 78.21; H, 4.39; N, 17.05.

*7-Methoxy-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline **4a***

Yield: 64%; m.p.: 120 – 122°C; IR (KBr) cm^{-1}: 1610 (C=N), 1300 (C-N), 1250, 1025 (C-O-C), 1146 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 3.79 (s, 3H, -OCH_3), 7.46 (s, 1H, =CH), 6.99 – 7.99 (m, 3H, -C_6H_3), 7.22 – 7.59 (m, 5H, -C_6H_5), 9.22 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 55.67, 110.84, 114.56, 115.02, 117.04, 117.85, 124.80, 127.06, 128.52, 129.81, 134.21, 137.52, 141.47, 147.76, 157.58; MS m/z: 276[M + 1]. Anal. Calcd. for $C_{17}H_{13}N_3O$: C, 74.17; H, 4.76; N, 15.26. Found: C, 74.12; H, 4.64; N, 15.12.

*7-Ethoxy-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline **4b***

Yield: 58%; m.p.: 125 – 127°C; IR (KBr) cm^{-1}: 1612 (C=N), 1301 (C-N), 1249, 1023 (C-O-C), 1148 (N-N). ^{1}H-NMR ($CDCl_3$, ppm): 1.34 (t, *J* = 7.2 Hz, 3H, -CH_3), 3.93 (t, *J* = 6.9 Hz, 2H, -CH_2-O-), 7.47 (s, 1H, =CH), 7.10 – 7.97 (m, 3H, -C_6H_3), 7.27 – 7.55 (m, 5H, -C_6H_5), 9.22 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 14.36, 55.67, 110.84, 114.56, 115.02, 117.04, 117.85, 124.80, 127.06, 128.52, 129.81, 134.21, 137.52, 141.47, 147.76, 157.58; MS m/z: 290 [M + 1]. Anal. Calcd. for $C_{18}H_{15}N_3O$: C, 74.72; H, 5.23; N, 14.52. Found: C, 74.69; H, 5.14; N, 14.39.

*5-Phenyl-7-propoxy-[1,2,4]-triazolo[4,3-a]quinoline **4c***

Yield: 56%; m.p.: 129 – 132°C; IR (KBr) cm^{-1}: 1612 (C=N), 1301 (C-N), 1251, 1024 (C-O-C), 1147 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 0.99 (t, *J* = 6.9 Hz, 3H, -CH_3), 1.76 (m, 2H, -CH_2), 3.94 (m, *J* = 6.6 Hz, 2H, -CH_2-O-), 7.48 (s, 1H, =CH), 7.03 – 8.01 (m, 3H, -C_6H_3), 7.21 – 7.49 (m, 5H, -C_6H_5), 9.29 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 10.47, 22.40, 69.97, 111.49, 114.29, 115.76, 117.02, 118.27, 124.58, 127.46, 128.75, 129.24, 129.91, 137.51, 141.73, 147.57, 157.21; MS m/z: 304 [M + 1]. Anal. Calcd. for $C_{19}H_{17}N_3O$: C, 75.23; H, 5.65; N, 13.85. Found: C, 75.12; H, 5.54; N, 13.72.

*7-Butoxy-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline **4d***

Yield: 57%; m.p.: 172-174°C; IR (KBr) cm^{-1}: 1611 (C=N), 1302 (C-N), 1251, 1024 (C-O-C), 1148 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 0.97 (t, *J* = 7.5 Hz, 3H, -CH_3), 1.32 – 1.77 (m, 4H, -$(CH_2)_2$), 3.98 (t, *J* = 6.6 Hz, 2H, -CH_2-O-), 7.48 (s, 1H, =CH), 6.99 – 7.98 (m, 3H, -C_6H_3), 7.21 – 7.50 (m, 5H, -C_6H_5), 9.23 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 13.80, 20.28, 31.12, 68.24, 108.38, 111.55, 115.77, 116.96, 121.49, 123.98, 125.58, 128.82, 129.25, 137.62, 140.60, 141.44, 157.19; MS m/z: 318 [M + 1]. Anal. Calcd. for $C_{20}H_{19}N_3O$: C, 75.69; H, 6.03; N, 13.24. Found: C, 75.57; H, 5.90; N, 13.09.

*7-(Pentyloxy)-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline **4e***

Yield: 55%; m.p.: 185 – 187°C; IR (KBr) cm^{-1}: 1608 (C=N), 1296 (C-N), 1249, 1021 (C-O-C), 1143 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 0.91 (t, *J* = 6.9 Hz, 3H, -CH_3), 1.34-1.78(m, 6H, -$(CH_2)_3$), 3.92(t, *J* = 6.3 Hz, 2H, -CH_2-O-), 7.47 (s, 1H, =CH), 7.32 – 7.96 (m, 3H, -C_6H_3), 7.23-7.51 (m,

5H, -C_6H_5), 9.23 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 13.96, 22.40, 28.08, 28.70, 68.66, 111.52, 112.21, 117.92, 119.58, 124.15, 124.95, 128.51, 129.26, 131.09, 133.12, 136.81, 138.19, 144.91, 148.57; MS m/z: 332 [M + 1]. Anal. Calcd. for $C_{21}H_{21}N_3O$: C, 76.11; H, 6.39; N, 12.68. Found: C, 75.89; H, 6.14; N, 12.52.

7-(Hexyloxy)-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4f***

Yield: 56%; m.p.: 147 – 148°C; IR (KBr) cm^{-1}: 1609 (C=N), 1298 (C-N), 1251, 1022 (C-O-C), 1148 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 0.92 (t, *J* = 7.2 Hz, 3H, -CH_3), 1.34 – 1.72 (m, 8H, -$(CH_2)_4$), 4.03 (t, *J* = 6.6 Hz, 2H, -CH_2-O-), 7.48 (s, 1H, =CH), 7.21 – 7.98 (m, 3H, -C_6H_3), 7.28 – 7.55 (m, 5H, -C_6H_5), 9.21 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 14.04, 22.58, 25.67, 29.06, 31.57, 68.58, 111.55, 114.52, 116.95, 118.29, 123.39, 124.81, 128.75, 128.86, 129.07, 129.29, 135.54, 137.62, 141.57, 157.22; MS m/z: 346 [M + 1]. Anal. Calcd. for $C_{22}H_{23}N_3O$: C, 76.49; H, 6.71; N, 12.16. Found: C, 76.36; H, 6.58; N, 12.02.

7-(Heptyloxy)-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4g***

Yield: 51%; m.p.: 156 – 158°C; IR (KBr) cm^{-1}: 1659 (C=N), 1301 (C-N), 1253, 1027 (C-O-C), 1149 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 0.92 (t, *J* = 7.5 Hz, 3H, -CH_3), 1.28 – 1.78 (m, 10H, -$(CH_2)_5$), 3.96 (t, *J* = 6.9 Hz, 2H, -CH_2-O-), 7.47 (s, 1H, =CH), 6.99 – 7.98 (m, 3H, -C_6H_3), 7.24 – 7.51 (m, 5H, -C_6H_5), 9.22 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$; ppm): 14.05, 22.57, 28.68, 28.97, 31.73, 32.24, 68.83, 109.15, 111.43, 115.40, 115.96, 119.34, 121.21, 123.39, 125.08, 129.09, 129.88, 135.37, 135.75, 142.97, 149.20. MS m/z: 360 [M + 1]. Anal. Calcd. for $C_{23}H_{25}N_3O$: C, 76.85; H, 7.01; N, 11.69. Found: C, 76.62; H, 6.87; N, 11.85.

7-(Octyloxy)-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4h***

Yield: 53%; m.p.: 150 – 152°C; IR (KBr) cm^{-1}: 1612 (C=N), 1301 (C-N), 1251, 1027 (C-O-C), 1149 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 0.96 (t, *J* = 7.4 Hz, 3H, -CH_3), 1.25 – 1.71 (m, 12H, -$(CH_2)_6$), 3.92 (t, *J* = 6.6 Hz, 2H, -CH_2-O-), 7.50 (s, 1H, =CH), 6.94 – 7.88 (m, 3H, -C_6H_3), 7.20 – 7.39 (m, 5H, -C_6H_5), 9.37 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 14.11, 22.66, 25.99, 29.21, 29.32, 29.71, 31.80, 68.56, 108.25, 111.46, 115.76, 116.45, 119.81, 121.51, 123.98, 125.28, 128.83, 129.26, 134.47, 136.35, 142.57, 149.53; MS m/z: 374 [M + 1]. Anal. Calcd. for $C_{24}H_{27}N_3O$: C, 77.18; H, 7.29; N, 11.25. Found: C, 77.02; H, 7.14; N, 11.07.

7-Methyl-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4i***

Yield: 62%; m.p.: 102 – 104°C; IR (KBr) cm^{-1}: 1611 (C=N), 1297 (C-N), 1251, 1023 (C-O-C), 1148 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 2.44(s, 3H, -CH_3), 7.49 (s, 1H, =CH), 7.51 – 7.96 (m, 3H, -C_6H_3), 7.27 – 7.49 (m, 5H, -C_6H_5), 9.25 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 21.39, 114.0, 115.65, 123.41, 127.94, 128.11, 128.66, 129.35, 131.12, 134.54, 136.38, 137.58, 138.07, 141.89, 148.47; MS m/z: 260 [M + 1]. Anal. Calcd. for $C_{17}H_{13}N_3$: C, 78.74; H, 5.05; N, 16.20. Found: C, 78.67; H, 4.94; N, 16.11.

9-Methyl-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4j***

Yield: 63%; m.p.: 148 – 150°C; IR (KBr) cm^{-1}: 1611 (C=N), 1328(C-N), 1251, 1024 (C-O-C), 1147 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 3.02 (s, 3H, -OCH_3), 7.48 (s, 1H, =CH), 7.35 – 7.67 (m, 3H, -C_6H_3), 7.38 – 7.52 (m, 5H, -C_6H_5), 9.59 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 27.04, 114.45, 125.18, 125.74, 126.62, 126.82, 128.57, 128.71, 129.37, 131.07, 133.16, 137.48, 138.10, 142.45, 148.87. MS m/z: 260 [M + 1]. Anal. Calcd. for $C_{17}H_{13}N_3$: C, 78.74; H, 5.05; N, 16.20. Found: C, 78.67; H, 4.94; N, 16.11.

7,8-Dimethylenedioxy-5-phenyl-[1,2,4]-triazolo[4,3-a]-quinoline ***4k***

Yield: 75%; m.p.: 184 – 186°C; IR (KBr) cm^{-1}: 1615 (C=N), 1302 (C-N), 1249, 1022 (C-O-C), 1149 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 6.14(s, 2H, -OCH_2O), 7.46 (s, 1H, =CH), 7.14 – 7.34 (m, 2H, -C_6H_2), 7.36 – 7.54 (m, 5H, -C_6H_5), 9.14 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 96.15, 102.54, 106.15, 112.07, 118.61, 126.44, 128.80, 129.24, 133.79, 137.84, 141.69, 146.83, 148.07, 149.81; MS (m/z): 290 [M + 1]. Anal. Calcd. for $C_{16}H_9Cl_2N_3$: C, 70.58; H, 3.83; N, 14.53. Found: C, 70.43; H, 3.70; N, 14.42.

6,9-Dimethoxy-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4l***

Yield: 71%; m.p.: 138 – 140°C; IR (KBr) cm^{-1}: 1614 (C=N), 1302 (C-N), 1247, 1023 (C-O-C), 1144 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 3.90 (s, 3H, -OCH_3), 4.06 (s, 3H, -OCH_3), 7.48 (s, 1H, =CH), 6.91 – 7.21 (m, 2H, -C_6H_2), 7.51 – 8.15 (m, 5H, -C_6H_5), 9.80 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 56.23, 56.58, 108.87, 111.75, 116.37, 122.75, 125.44, 126.86, 128.62, 129.37, 131.0, 138.98, 140.59, 142.44, 144.06, 151.25; MS m/z: 306 [M + 1]. Anal. Calcd. for $C_{18}H_{15}N_3O_2$: C, 70.81; H, 4.95; N, 13.76. Found: C, 70.76; H, 4.80; N, 13.65.

7-Benzyl-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4m***

Yield: 49%; m.p.: 119 – 121°C; IR (KBr) cm^{-1}: 1658 (C=N), 1297 (C-N), 1248, 1021 (C-O-C), 1150 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 5.30 (s, 2H, -$OCH_2C_6H_5$), 7.50 (s, 1H, =CH), 6.99 – 7.98 (m, 3H, -C_6H_3), 7.21 – 7.53 (m, 10H, -$2C_6H_5$), 9.22 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 70.67, 112.47, 112.74, 117.15, 123.06, 124.05, 125.91, 126.11, 126.84, 127.48, 128.75, 128.86, 129.22, 130.39, 133.21, 137.64, 139.61, 142.08, 154.34; MS m/z: 336 [M + 1]. Anal. Calcd. for $C_{23}H_{17}N_3$: C, 82.36; H, 5.11; N, 12.53. Found: C, 82.22; H, 5.04; N, 12.41.

7,8-Dimethoxy-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4n***

Yield: 70%; m.p.: 94 – 96°C; IR (KBr) cm^{-1}: 1611 (C=N), 1298 (C-N), 1248, 1022 (C-O-C), 1144 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 3.80 (s, 3H, -OCH_3), 4.13 (s, 3H, -OCH_3), 7.48 (s, 1H, =CH), 6.93 – 7.50 (m, 2H, -C_6H_2), 7.18 – 7.70 (m, 5H, -C_6H_5), 9.42 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 56.37, 56.66, 108.62, 111.85, 116.89, 117.90, 125.39, 128.21, 129.90, 133.84, 137.85, 141.38, 145.70, 147.97, 151.39, 152.89; MS m/z: 306 [M + 1]. Anal. Calcd. for $C_{18}H_{15}N_3O_2$: C, 70.81; H, 4.95; N, 13.76. Found: C, 70.72; H, 4.75; N, 13.62.

7-Fluoro-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4o***

Yield: 38%; m.p.: 197 – 199°C; IR (KBr) cm^{-1}: 1613 (C=N), 1301 (C-N), 1253, 1027 (C-O-C), 1149 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 7.47 (s, 1H, =CH), 7.37 – 8.17 (m, 3H, -C_6H_3), 7.30 – 7.55 (m, 5H, -C_6H_5), 9.26 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 116.83, 115.18, 117.51, 124.43(d, $^2J_{C\text{-}F}$ = 23.3 Hz), 125.71, 126.72, 127.31, 128.86, 129.73, 131.19, 136.82, 141.28(d, $^3J_{C\text{-}F}$ = 8.3 Hz), 141.66, 147.23, 161.53(d, $^1J_{C\text{-}F}$ = 241.5 Hz); MS m/z: 264 [M + 1]. Anal. Calcd. for $C_{16}H_{10}FN_3$: C, 72.99; H, 3.83; N, 15.96. Found: C, 72.86; H, 3.68; N, 15.82.

7-Chloro-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4p***

Yield: 36%; m.p.: 204 – 206°C; IR (KBr) cm^{-1}: 1614 (C=N), 1302 (C-N), 1252, 1026 (C-O-C), 1148 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 7.49 (s, 1H, =CH), 7.62 – 8.01 (m, 3H, -C_6H_3), 7.22 – 7.50 (m, 5H, -C_6H_5), 9.30 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 108.07, 110.82, 121.18, 124.29, 126.31, 127.67, 128.68, 129.16, 129.38, 130.71, 131.99, 134.53, 136.56, 139.44, 148.17; MS m/z: 280 [M + 1]. Anal. Calcd.

for $C_{16}H_{10}ClN_3$: C, 68.70; H, 3.60; N, 15.02. Found: C, 68.65; H, 3.51; N, 14.96.

7-Bromo-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4q***

Yield: 41%; m.p.: 198 – 201°C; IR (KBr) cm^{-1}: 1613 (C=N), 1305 (C-N), 1253, 1027 (C-O-C), 1149 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 7.48 (s, 1H, =CH), 7.76 – 8.19 (m, 3H, -C_6H_3), 7.23 – 7.54 (m, 5H, -C_6H_5), 9.59 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 114.97, 118.46, 124.98, 127.47, 128.14, 128.77, 129.32, 129.60, 130.32, 132.86, 135.66, 136.76, 140.73, 147.48; MS m/z: 324 [M + 1]. Anal. Calcd. for $C_{16}H_{10}BrN_3$: C, 59.28; H, 3.11; N, 12.96. Found: C, 58.93; H, 3.04; N, 12.87.

6,9-Dichloro-5-phenyl-[1,2,4]-triazolo[4,3-a]quinoline ***4r***

Yield: 32%; m.p.: 192 – 194°C; IR (KBr) cm^{-1}: 1611 (C=N), 1301 (C-N), 1252, 1026 (C-O-C), 1149 (N-N); ^{1}H-NMR ($CDCl_3$, ppm): 7.47 (s, 1H, =CH), 7.54 – 7.97 (m, 3H, -C_6H_2), 7.29 – 7.51 (m, 5H, -C_6H_5), 9.28 (s, 1H, H-1); ^{13}C-NMR ($CDCl_3$, ppm): 117.47, 122.24, 124.65, 126.23, 127.94, 129.63, 130.65, 132.47, 134.86, 138.01, 139.32, 145.67, 152.12; MS m/z: 314 [M + 1]. Anal. Calcd. for $C_{16}H_9Cl_2N_3$: C, 61.17; H, 2.89; N, 13.38. Found: C, 60.12; H, 2.69; N, 13.22.

Pharmacology

The MES test and the rotarod test were carried out by the standard described in the Antiepileptic Drug Development Program (ADD) of the National Institutes of Health (USA) [13, 14]. All compounds were tested for anticonvulsant activities with C57B/6 mice in the 18 – 25 g weight range purchased from the Laboratory of Animal Research, College of Pharmacy, Yanbian University. The tested compounds were dissolved in polyethylene glycol-400.

In phase-I screening (Table 1), each compound was administered at the dose levels of 30, 100, and 300 mg/kg for evaluating the anticonvulsant activity, and its neurotoxicity was measured at 30-min and 4-h intervals after administration. Anticonvulsant efficacy was measured in the MES test. In the MES test, seizures were elicited with a 60 Hz alternating current of 50 mA intensity in mice. The current was applied via corneal electrodes for 0.2 s. The protection against the spread of MES-induced seizures was defined as the abolition of the hind leg and tonic maximal extension component of the seizure. Anticonvulsant drug-induced neurologic deficit was detected in mice by using the rotorod ataxia test.

The pharmacologic parameters estimated in phase-I screening were quantified for compounds **4a – r** in phase-II screening (Table 2). Anticonvulsant activity was expressed in terms of the median effective dose (ED_{50}), and neurotoxicity was expressed as the median toxic dose (TD_{50}). For the determination of the ED_{50} and TD_{50} values, groups of ten mice were given a range of intraperitoneal doses of the tested compound until at least three points were established in the range of 10 – 90% seizure protection or minimal observed neurotoxicity. From the plot of this data, the respective ED_{50} and TD_{50} values, 95% confidence intervals, slope of the regression line, and the standard error of the slope were calculated by means of a computer program written at National Institute of Neurological Disorders and Stroke.

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