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Short communication

Solution-phase parallel synthesis and evaluation of anticonvulsant activity of *N*-substituted-3,4-dihydroisoquinoline-2(1*H*)-carboxamides

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Abstract

In our previous studies we identified several isoquinoline derivatives displaying potent anticonvulsant effects in different animal models of epilepsy. With the aim to exploit the main structure–activity relationships (SAR) for this class of compounds we planned a solution-phase parallel synthesis (SPPS) of new *N*-substituted-3,4-dihydroisoquinoline-2(1*H*)-carboxamides exploring the effect of introduction of different (cyclo)alkyl groups at carboxamide moiety linked to *N*-2 atom of isoquinoline scaffold. The pharmacological effects were evaluated against audiogenic seizures in DBA/2 mice and, even if some new derivatives were more active than valproate, the designed modifications did not improve the anticonvulsant efficacy with respect to their precursors.

Keywords: Anticonvulsant; Isoquinoline; DBA/2 mice; Solution-phase parallel synthesis (SPPS)

1. Introduction

Epilepsy affects approximatily 1% of the world's population. Despite the introduction of many new antiepileptic drugs (AEDs) and remarkable strides in this research field, more than a third of patients with epilepsy remain refractory to available treatments and more effective therapies are needed. Continued efforts are being made in the development of antiepileptic drugs employing a range of strategies, including modification of the structure of existing drugs, targeting novel molecular substrates and non-mechanism-based drug screening of compounds in traditional and newer animal models [1,2].

The mechanism of action of currently available effective antiepileptics are: the induction of a prolonged inactivation of the Na^+ channel; the blockade of Ca^{2+} channel currents; the enhancement of the inhibitory GABAergic neurotransmission or the modulation of excitatory glutamatergic neurotransmission [3]. With respect to this last pharmacological target, extensive studies have demonstrated that competitive and non-competitive antagonists of the ionotropic glutamate receptors (iGluRs) show promise in terms of their therapeutic potential for the prevention and treatment of the epilepsy [4].

In our previous works, a large series of isoquinolines were found to have anticonvulsant activities in various seizure models interacting with glutamate ionotropic AMPA receptor (AMPAR) subtype in a selective and non-competitive fashion [5–12]. The most active compound of the series was 2-acetyl-1-(4′-chlorophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**1**, Fig. 1) which showed the strongest activity with respect to other known AMPAR antagonists (e.g. **GYKI52466**, Fig. 1) when compared to both in vivo and in vitro tests [4,13–15].

Starting from this ''lead compound'' we explored the effect of the modification of both acetyl moiety at *N*-2 position, and the substituent on the C-1 phenyl group, as well as methoxy groups on the benzene fused ring. Our computational studies [5–7] suggested that the carbonyl moiety linked to the *N*-2

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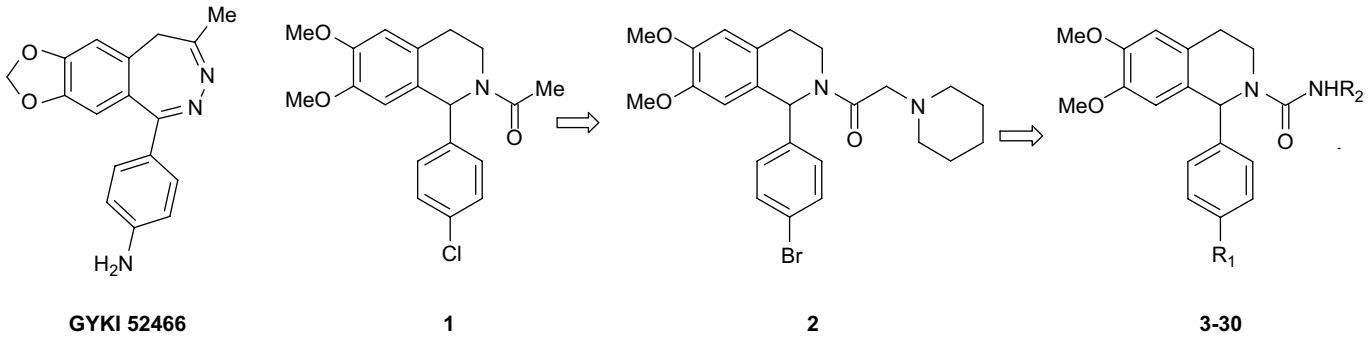


Fig. 1.

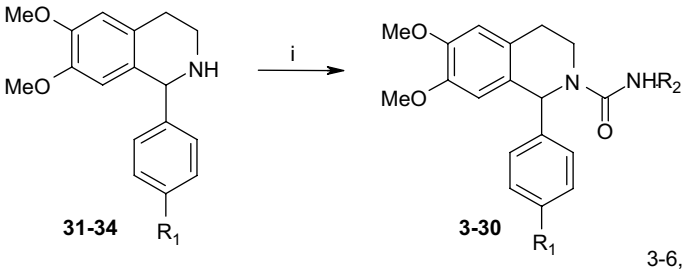
position of tetrahydroisoquinoline system plays an important role in the process of AMPAR recognition and we found that the nature of fragment linked to carbonyl function significantly influenced the pharmacological profile in terms of both pharmacokinetic and pharmacodynamic effects. Furthermore, the results of the pharmacological evaluation of some *N*-acetamide derivatives [e.g. **2**, 1-(4′-bromophenyl)-6,7-dimethoxy-2-(piperidin-1-yl-acetyl)-3,4-dihydroisoquinoline] demonstrated that the substituent at 4′-position of phenyl ring also modulated the anticonvulsant potency and the halogens appeared to have a better impact on anticonvulsant activity probably because of their lipophilic effects [10].

On the basis of these findings and with the aim to gain more insights into the structure–activity relationships (SAR) we herein describe a simple solution-phase parallel synthesis of new *N*-(cyclo)alkyl-3,4-tetrahydroisoquinoline-2(1*H*)-carboxamide derivatives (**3–30**) in which we explored if the presence of carboxamide frame could be a feature able to improve the anticonvulsant efficacy against sound-induced seizures in DBA/2 mice. Moreover we inserted on nitrogen atom of carboxamide moiety different (cyclo)alkyl fragments in an attempt to decipher the impact of the bulkiness as well as the lipophilicity on pharmacological properties. Finally, we investigated the influence of the nature of halogen substituent at 4′-position of the C-1 aryl group and compared the activity with that of the corresponding unsubstituted derivatives.

2. Results and discussion

As depicted in Scheme 1, a small library of *N*-substituted-1-aryl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1*H*)-caboxamide derivatives (**3–30**) was prepared starting from different 1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines (**31–34**) [7] and various alkylisocyanates. An efficient solution-phase parallel synthesis was set up at room temperature employing a Buchi Syncore reactor and the target compounds **3–30** were obtained in good yields. The structures of the compounds obtained were supported by elemental analyses and spectroscopic measurements (^{1}H NMR).

The anticonvulsant effects of the synthesized *N*-substituted-1-aryl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1*H*)-caboxamide derivatives (**3–30**) were evaluated against audiogenic seizures in DBA/2 mice, which is considered an excellent animal model for generalized epilepsy and for screening new anticonvulsant drugs [16]. Table 1 reports the pharmacological results compared with those of antiepileptic drugs such as gabapentin and valproate as well as isoquinolines **1–2** and **GYKI 52466** known as non-competitive AMPA receptor antagonists.



	R_1
3, 7, 11, 15, 19, 23, 27, 31	H
4, 8, 12, 16, 20, 24, 28, 32	Br
5, 9, 13, 17, 21, 25, 29, 33	Cl
6, 10, 14, 18, 22, 26, 30, 34	F

3-6, R_2 = ethyl
7-10, R_2 = n-propyl
11-14, R_2 = i-propyl
15-18, R_2 = n-butyl
19-22, R_2 = t-butyl
23-26, R_2 = cyclopentyl
27-30, R_2 = cyclohexyl

Scheme 1. Reagents and conditions: (i) R_2NCO, CH_2Cl_2, TEA, r.t., 90 min.

Table 1
Anticonvulsant activity against audiogenic seizures in DBA/2 mice

Compound	ED_{50} (mg/kg)[a]		log P[b]
	Clonus	Tonus	
1	2.25 (1.70–2.97)	1.22 (0.90–1.56)	3.40
2	5.99 (2.87–12.4)	3.86 (1.90–7.86)	4.93
3	32.5 (24.7–42.8)	11.9 (8.77–16.1)	3.41
4	>35	>35	4.18
5	30.4 (22.3–41.5)	14.2 (7.78–25.8)	4.00
6	>35	>35	3.46
7	>35	18.1 (12.6–26.1)	3.94
8	>35	31.2 (19.8–54.0)	4.71
9	>35	23.4 (16.9–32.3)	4.53
10	>35	26.9 (15.4–47.2)	3.99
11	>35	>35	3.75
12	>35	23.6 (17.9–31.1)	4.53
13	>35	26.1 (21.3–31.9)	4.35
14	>35	17.3 (11.8–25.5)	3.81
15	31.7 (23.0–43.8)	11.0 (7.84–15.5)	4.47
16	>35	15.5 (9.12–25.2)	5.24
17	>35	16.5 (16.4–26.0)	5.06
18	>35	13.7 (7.76–24.1)	4.52
19	27.5 (22.2–34.0)	13.9 (7.65–25.4	4.10
20	>35	16.9 (9.30–30.8)	4.88
21	30.6 (23.7–39.4)	13.8 (8.60–23.6)	4.70
22	>35	18.7 (14.1–24.7)	4.16
23	>35	15.1 (9.84–23.2)	4.37
24	>35	14.7 (11.0–19.6)	5.14
25	>35	12.1 (7.91–18.7)	4.97
26	27.7 (19.2–40.2)	7.24 (4.42–11.8)	4.42
27	26.9 (21.5–33.7)	9.38 (6.46–13.6)	4.94
28	>35	19.3 (10.2–36.4)	5.71
29	>35	14.2 (9.67–20.8)	5.53
30	>35	>35	4.99
GYKI 52466	10.5 (7.15–15.3)	7.41 (4.69–11.7)	–
Gabapentin	20.3 (13.7–30.2)	9.90 (7.81–12.6)	–
Valproate	>35	21.5 (16.1–28.7)	–

[a] All data were calculated according to the method of Litchfield and Wilcoxon. At least 32 animals were used to calculate each ED_{50}. Confidence limits (±95%) are given in parentheses.
[b] log P data are predicted from a commercially available program (ACD/Lab).

The biological results presented in Table 1 show that these compounds generally possess potency lower than other isoquinoline analogs **1**–**2** previously synthesized by us [7,10].

The analysis of reported ED_{50} values on clonic phase of the test (see Table 1) shows that the most active derivatives of this new series of compounds were **19**, **26** and **27** in which the anticonvulsant potency in the clonic phase of the test was approximately 2.5 fold lower than **GYKI 52466**, the prototype of non-competitive AMPAR antagonists showing anticonvulsant efficacy [13,14,17,18]. Nevertheless, their activity was higher than that of valproate and comparable to that of gabapentin two drugs largely used in antiepileptic therapy [19].

Moderate activity against sound-induced seizures characterized compounds **3**, **5**, **15**, **21** whereas the other studied compounds were inactive at highest tested doses (i.e. clonic phase $ED_{50} > 35$ mg/kg).

The main structure–activity relationship (SAR) considerations suggest that the different degree of anticonvulsant efficacies do not seem directly connected neither with the lengthening nor the bulkiness of the (cyclo)alkyl chain linked to the carboxamide group. Considering the effect of the 4′-substituent on the C-1 aryl group, we observed that the unsubstituted derivatives **3**, **15**, **19** and **27** are generally more active than the halogen-substituted analogs. In an attempt to correlate the rank order of potency with their lipophilic properties, we also estimated [20] the log P values (Table 1) but no significant correlation was observed.

In conclusion an efficient solution-phase parallel synthesis for the preparation of new *N*-substituted-1-aryl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1*H*)-caboxamide derivatives (**3**–**30**) as potential anticonvulsant agents was set up. Unfortunately, even if some synthesized compounds showed anticonvulsant activity comparable to other well-known anticonvulsants, the planned modifications generally afforded molecules (**3**–**30**) with lower activity than the previously reported isoquinoline derivatives (e.g. **1** and **2**). Considering the structure–activity relationships so far obtained for this class of compounds and analyzing the contribution of different groups at *N*-2 position to the anticonvulsant efficacy we could speculate that both the size and the nature of the substituents modulate the activity.

3. Experimental section

3.1. Chemistry

Melting points were determined on a Stuart SMP10 apparatus and are uncorrected. Elemental analyses (C, H, N) were carried out on a Carlo Erba Model 1106 Elemental Analyzer and the results are within ±0.4% of the theoretical values. Merck silica gel 60 F_{254} plates were used for analytical TLC. ^{1}H NMR spectra were measured in $CDCl_3$ with a Varian Gemini 300 spectrometer; chemical shifts are expressed in δ (ppm) relative to TMS as internal standard and coupling constants (J) in Hz.

3.1.1. General procedure for the synthesis of N-(cyclo)alkyl-1-aryl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamides (3–30)

To a solution of the 1-aryl-6,7-dimethoxy-3,4-dihydroisoquinoline derivative (**31**–**34**) (0.001 mol) prepared as previously reported [7] in CH_2Cl_2 (5 ml) was added suitable alkylisocyanate (0.0015 mol) and triethylamine (TEA) (0.001 mol) in the Buchi Syncore reactor. The mixture was stirred for 90 min at room temperature. After removal of the solvent under reduced pressure, the residue was powdered and then crystallized with diethyl ether to give target products as white solid (**3**–**30**).

3.1.1.1. N-Ethyl-1-phenyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (3). Yield 90%. Mp. 158–160 °C. ^{1}H NMR ($CDCl_3$): 1.13 (3H, t, $J = 7.0$, CH_3), 2.63–3.57 (4H, m, CH_2CH_2), 3.30 (2H, q, $J = 7.0$, CH_2), 3.78 (3H, s, CH_3O-6), 3.87 (3H, s, CH_3O-7), 4.47 (1H, bs, NH), 6.34 (1H, s, H-1), 6.63 (1H, s, H-5), 6.66 (1H, s, H-8), 7.22–7.28 (5H, m, ArH). Anal. $C_{20}H_{24}N_2O_3$ (C, H, N).

*3.1.1.2. 1-(4′-Bromophenyl)-N-ethyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (**4**).* Yield 95%. Mp. 129–131 °C. ^{1}H NMR ($CDCl_3$): 1.15 (3H, t, $J = 7.0$, CH_3), 2.63–3.52 (4H, m, CH_2CH_2), 3.30 (2H, q, $J = 7.0$, CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.46 (1H, bs, NH), 6.38 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 7.11 (2H, d, $J = 8.5$), 7.38 (2H, d, $J = 8.5$). Anal. $C_{20}H_{23}BrN_2O_3$ (C, H, N).

*3.1.1.3. 1-(4′-Chlorophenyl)-N-ethyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (**5**).* Yield 96%. Mp. 144–146 °C. ^{1}H NMR ($CDCl_3$): 1.14 (3H, t, $J = 7.0$, CH_3), 2.64–3.54 (4H, m, CH_2CH_2), 3.35 (2H, q, $J = 7.0$, CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.45 (1H, bs, NH), 6.39 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 7.17 (2H, d, $J = 8.5$), 7.24 (2H, d, $J = 8.5$). Anal. $C_{20}H_{23}ClN_2O_3$ (C, H, N).

*3.1.1.4. 1-(4′-Fluorophenyl)-N-ethyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (**6**).* Yield 92%. Mp. 139–141 °C. ^{1}H NMR ($CDCl_3$): 1.14 (3H, t, $J = 7.0$, CH_3), 2.65–3.54 (4H, m, CH_2CH_2), 3.33 (2H, q, $J = 7.0$, CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.45 (1H, bs, NH), 6.39 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 6.92–7.22 (4H, m, ArH). Anal. $C_{20}H_{23}FN_2O_3$ (C, H, N).

*3.1.1.5. 6,7-Dimethoxy-1-phenyl-N-propyl-3,4-dihydroisoquinolin-2(1H)-carboxamide (**7**).* Yield 90%. Mp. 141–143 °C. ^{1}H NMR ($CDCl_3$): 0.88 (3H, t, $J = 7.4$, CH_3), 1.47–1.54 (2H, m, CH_2), 2.68–3.63 (4H, m, CH_2CH_2), 3.19–3.27 (2H, m, CH_2N), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.49 (1H, bs, NH), 6.31 (1H, s, H-1), 6.63 (1H, s, H-5), 6.66 (1H, s, H-8), 7.23–7.28 (5H, m, ArH). Anal. $C_{21}H_{26}N_2O_3$ (C, H, N).

*3.1.1.6. 1-(4′-Bromophenyl)-6,7-dimethoxy–N-propyl-3,4-dihydroisoquinolin-2(1H)-carboxamide (**8**).* Yield 91%. Mp. 135–137 °C. ^{1}H NMR ($CDCl_3$): 0.91 (3H, t, $J = 7.4$, CH_3), 1.49–1.56 (2H, m, CH_2), 2.65–3.54 (4H, m, CH_2CH_2), 3.18–3.26 (2H, m, CH_2N), 3.80 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.47–4.49 (1H, m, NH), 6.37 (1H, bs, H-1), 6.60 (1H, s, H-5), 6.66 (1H, s, H-8), 7.12 (2H, d, $J = 8.2$), 7.39 (2H, d, $J = 8.2$). Anal. $C_{21}H_{25}BrN_2O_3$ (C, H, N).

*3.1.1.7. 1-(4′-Chlorophenyl)-6,7-dimethoxy-N-propyl-3,4-dihydroisoquinolin-2(1H)-carboxamide (**9**).* Yield 88%. Mp. 127–129 °C. ^{1}H NMR ($CDCl_3$): 0.91 (3H, t, $J = 7.1$, CH_3), 1.49–1.56 (2H, m, CH_2), 2.65–3.55 (4H, m, CH_2CH_2), 3.23–3.26 (2H, m, CH_2N), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.49 (1H, bs, NH), 6.39 (1H, s, H-1), 6.60 (1H, s, H-5), 6.67 (1H, s, H-8), 7.17 (2H, d, $J = 8.2$), 7.24 (2H, d, $J = 8.2$). Anal. $C_{21}H_{25}ClN_2O_3$ (C, H, N).

*3.1.1.8. 1-(4′-Fluorophenyl)-6,7-dimethoxy-N-propyl-3,4-dihydroisoquinolin-2(1H)-carboxamide (**10**).* Yield 80%. Mp. 121–123 °C. ^{1}H NMR ($CDCl_3$): 0.91 (3H, t, $J = 7.4$, CH_3), 1.49–1.56 (2H, m, CH_2), 2.66–3.55 (4H, m, CH_2CH_2), 3.20–3.28 (2H, m, CH_2N), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.48 (1H, bs, NH), 6.38 (1H, s, H-1), 6.60 (1H, s, H-5), 6.67 (1H, s, H-8), 6.93–7.23 (4H, m, ArH). Anal. $C_{21}H_{25}FN_2O_3$ (C, H, N).

*3.1.1.9. 6,7-Dimethoxy-N-isopropyl-1-phenyl-3,4-dihydroisoquinolin-2(1H)-carboxamide (**11**).* Yield 79%. Mp. 141–143 °C. ^{1}H NMR ($CDCl_3$): 1.12 and 1.16 (6H, 2d, $J = 6.3$, CH_3), 2.67–3.62 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.87 (3H, s, CH_3O-7), 3.95–4.07 (1H, m, CH), 4.28 (1H, bs, NH), 6.30 (1H, s, H-1), 6.62 (1H, s, H-5), 6.66 (1H, s, H-8), 7.22–7.29 (5H, m, ArH). Anal. $C_{21}H_{26}N_2O_3$ (C, H, N).

*3.1.1.10. 1-(4′-Bromophenyl)-N-isopropyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (**12**).* Yield 84%. Mp. 128–130 °C. ^{1}H NMR ($CDCl_3$): 1.14 and 1.18 (6H, d, $J = 6.6$, CH_3), 2.64–3.51 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 3.99–4.05 (1H, m, CH), 4.27 (1H, bs, NH), 6.37 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 7.11 (2H, d, $J = 8.5$), 7.39 (2H, d, $J = 8.5$). Anal. $C_{21}H_{25}BrN_2O_3$ (C, H, N).

*3.1.1.11. 1-(4′-Chlorophenyl)-N-isopropyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (**13**).* Yield 92%. Mp. 133–135 °C. ^{1}H NMR ($CDCl_3$): 1.14 and 1.18 (6H, d, $J = 6.3$, CH_3), 2.63–3.51 (4H, m, CH_2CH_2), 3.80 (3H, s, CH_3O-6), 3.83 (3H, s, CH_3O-7), 3.97–4.06 (1H, m, CH), 4.26 (1H, bs, NH), 6.38 (1H, s, H-1), 6.60 (1H, s, H-5), 6.67 (1H, s, H-8), 7.17 (2H, d, $J = 8.5$), 7.24 (2H, d, $J = 8.5$). Anal. $C_{21}H_{25}ClN_2O_3$ (C, H, N).

*3.1.1.12. 1-(4′-Fluorophenyl)-N-isopropyl-6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-carboxamide (**14**).* Yield 93%. Mp. 135–137 °C. ^{1}H NMR ($CDCl_3$): 1.14 and 1.18 (6H, d, $J = 6.3$, CH_3), 2.65–3.54 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 3.99–4.06 (1H, m, CH), 4.27 (1H, bs, NH), 6.37 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 6.93–7.23 (4H, m, ArH). Anal. $C_{21}H_{25}FN_2O_3$ (C, H, N).

*3.1.1.13. N-Butyl-6,7-dimethoxy-1-phenyl-3,4-dihydroisoquinoline-2(1H)-carboxamide (**15**).* Yield 78%. Mp. 114–116 °C. ^{1}H NMR ($CDCl_3$): 0.90 (3H, t, $J = 7.1$, CH_3), 1.24–3.31 (6H, m, $CH_2CH_2CH_2$), 2.68–3.64 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.48 (1H, bs, NH), 6.31 (1H, s, H-1), 6.62 (1H, s, H-5), 6.66 (1H, s, H-8), 7.22–7.28 (5H, m, ArH). Anal. $C_{22}H_{28}N_2O_3$ (C, H, N).

*3.1.1.14. 1-(4′-Bromophenyl)-N-butyl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**16**).* Yield 77%. Mp. 132–134 °C. ^{1}H NMR ($CDCl_3$): 0.92 (3H, t, $J = 7.1$, CH_3), 1.23–3.31 (6H, m, $CH_2CH_2CH_2$), 2.63–3.52 (4H, m, CH_2CH_2), 3.80 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.48 (1H, bs, NH), 6.37 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 7.11 (2H, d, $J = 8.2$), 7.39 (2H, d, $J = 8.2$). Anal. $C_{22}H_{27}BrN_2O_3$ (C, H, N).

*3.1.1.15. 1-N-Butyl-(4′-chlorophenyl)-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**17**).* Yield 88%. Mp. 130–132 °C. ^{1}H NMR ($CDCl_3$): 0.92 (3H, t, $J = 7.1$, CH_3), 1.27–3.31 (6H, m, $CH_2CH_2CH_2$), 2.65–3.53 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.47 (1H, bs, NH), 6.38 (1H, s, H-1), 6.60 (1H, s, H-5), 6.66 (1H, s, H-8), 7.17 (2H, d, $J = 8.5$), 7.24 (2H, d, $J = 8.5$). Anal. $C_{22}H_{27}ClN_2O_3$ (C, H, N).

*3.1.1.16. 1-N-Butyl-(4′-fluorophenyl)-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**18**).* Yield 93%. Mp. 129–131 °C. ^{1}H NMR ($CDCl_3$): 0.92 (3H, t, $J = 7.1$, CH_3), 1.29–3.29 (6H, m, $CH_2CH_2CH_2$), 2.66–3.55 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.47 (1H, bs, NH), 6.38 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 6.93–7.23 (4H, m, ArH). Anal. $C_{22}H_{27}FN_2O_3$ (C, H, N).

*3.1.1.17. N-tert-butyl-6,7-dimethoxy-1-phenyl-3,4-dihydroisoquinoline-2(1H)-carboxamide (**19**).* Yield 90%. Mp. 166–168 °C. ^{1}H NMR ($CDCl_3$): 1.33 (9H, s, CH_3), 2.66–3.63 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.87 (3H, s, CH_3O-7), 4.38 (1H, bs, NH), 6.23 (1H, s, H-1), 6.62 (1H, s, H-5), 6.66 (1H, s, H-8), 7.21–7.31 (5H, m, ArH). Anal. $C_{22}H_{28}N_2O_3$ (C, H, N).

*3.1.1.18. 1-(4′-Bromophenyl)-N-tert-butyl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**20**).* Yield 85%. Mp. 188–190 °C. ^{1}H NMR ($CDCl_3$): 1.36 (9H, s, CH_3), 2.62–3.52 (4H, m, CH_2CH_2), 3.80 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.36 (1H, bs, NH), 6.32 (1H, s, H-1), 6.60 (1H, s, H-5), 6.66 (1H, s, H-8), 7.10 (2H, d, $J = 8.2$), 7.39 (2H, d, $J = 8.5$). Anal. $C_{22}H_{27}BrN_2O_3$ (C, H, N).

*3.1.1.19. N-tert-Butyl-1-(4′-chlorophenyl)-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**21**).* Yield 73%. Mp. 178–180 °C. ^{1}H NMR ($CDCl_3$): 1.36 (9H, s, CH_3), 2.62–3.52 (4H, m, CH_2CH_2), 3.80 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.36 (1H, bs, NH), 6.33 (1H, s, H-1), 6.60 (1H, s, H-5), 6.66 (1H, s, H-8), 7.16 (2H, d, $J = 8.5$), 7.24 (2H, d, $J = 8.5$). Anal. $C_{22}H_{27}ClN_2O_3$ (C, H, N).

*3.1.1.20. N-tert-Butyl-1-(4′-fluorophenyl)-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**22**).* Yield 78%. Mp. 150–152 °C. ^{1}H NMR ($CDCl_3$): 1.35 (9H, s, CH_3), 2.63–3.55 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.37 (1H, bs, NH), 6.32 (1H, s, H-1), 6.60 (1H, s, H-5), 6.67 (1H, s, H-8), 6.93–7.22 (4H, m, ArH). Anal. $C_{22}H_{27}FN_2O_3$ (C, H, N).

*3.1.1.21. N-Cyclopentyl-6,7-dimethoxy-1-phenyl-3,4-dihydroisoquinoline-2(1H)-carboxamide (**23**).* Yield 93%. Mp. 157–158 °C. ^{1}H NMR ($CDCl_3$): 1.25–1.98 (8H, m, cycloalkyl), 2.69–3.64 (4H, m, CH_2CH_2), 3.78 (3H, s, CH_3O-6), 3.87 (3H, s, CH_3O-7), 4.11–4.18 (1H, m, CH), 4.41 (1H, bs, NH), 6.27 (1H, s, H-1), 6.61 (1H, s, H-5), 6.66 (1H, s, H-8), 7.23–7.31 (5H, m, ArH). Anal. $C_{23}H_{28}N_2O_3$ (C, H, N).

*3.1.1.22. 1-(4′-Bromophenyl)-N-cyclopentyl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**24**).* Yield 79%. Mp. 169–171 °C. ^{1}H NMR ($CDCl_3$): 1.25–2.02 (8H, m, cycloalkyl), 2.65–3.52 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 3.95–4.16 (1H, m, CH), 4.41 (1H, bs, NH), 6.36 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 7.11 (2H, d, $J = 8.2$), 7.39 (2H, d, $J = 8.5$). Anal. $C_{23}H_{27}BrN_2O_3$ (C, H, N).

*3.1.1.23. 1-(4′-Chlorophenyl)-N-cyclopentyl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**25**).* Yield 94%. Mp. 156–158 °C. ^{1}H NMR ($CDCl_3$): 1.25–2.02 (8H, m, cycloalkyl), 2.63–3.52 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 3.93–4.18 (1H, m, CH), 4.40 (1H, bs, NH), 6.36 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 7.17 (2H, d, $J = 8.5$), 7.24 (2H, d, $J = 8.5$). Anal. $C_{23}H_{27}ClN_2O_3$ (C, H, N).

*3.1.1.24. N-Cyclopentyl-6,7-dimethoxy-1-(4′-fluorophenyl)-3,4-dihydroisoquinoline-2(1H)-carboxamide (**26**).* Yield 85%. Mp. 141–143 °C. ^{1}H NMR ($CDCl_3$): 1.25–2.03 (8H, m, cycloalkyl), 2.66–3.55 (4H, m, CH_2CH_2), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 3.93–4.18 (1H, m, CH), 4.42 (1H, bs, NH), 6.35 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 6.93–7.23 (4H, m, ArH). Anal. $C_{23}H_{27}FN_2O_3$ (C, H, N).

*3.1.1.25. N-Cyclohexyl-6,7-dimethoxy-1-phenyl-3,4-dihydroisoquinoline-2(1H)-carboxamide (**27**).* Yield 80%. Mp. 182–184 °C. ^{1}H NMR ($CDCl_3$): 1.01–1.95 (10H, m, cycloalkyl), 2.67–3.74 (5H, m, CH_2CH_2 and CH), 3.78 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.35 (1H, bs, NH), 6.29 (1H, s, H-1), 6.62 (1H, s, H-5), 6.66 (1H, s, H-8), 7.23–7.31 (5H, m, ArH). Anal. $C_{24}H_{30}N_2O_3$ (C, H, N).

*3.1.1.26. 1-(4′-Bromophenyl)-N-cyclohexyl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**28**).* Yield 75%. Mp. 181–183 °C. ^{1}H NMR ($CDCl_3$): 1.04–1.95 (10H, m, cycloalkyl), 2.63–3.74 (4H, m, CH_2CH_2), 3.06–3.14 (1H, m, CH), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.34 (1H, bs, NH), 6.36 (1H, s, H-1), 6.59 (1H, s, H-5), 6.67 (1H, s, H-8), 7.12 (2H, d, $J = 8.2$), 7.39 (2H, d, $J = 8.2$). Anal. $C_{24}H_{29}BrN_2O_3$ (C, H, N).

*3.1.1.27. 1-(4′-Chlorophenyl)-N-cyclohexyl-6,7-dimethoxy-3,4-dihydroisoquinoline-2(1H)-carboxamide (**29**).* Yield 74%. Mp. 176–178 °C. ^{1}H NMR ($CDCl_3$): 1.04–1.95 (10H, m, cycloalkyl), 2.65–3.74 (4H, m, CH_2CH_2), 3.05–3.13 (1H, m, CH), 3.79 (3H, s, CH_3O-6), 3.88 (3H, s, CH_3O-7), 4.34 (1H, bs, NH), 6.37 (1H, s, H-1), 6.60 (1H, s, H-5), 6.67 (1H, s, H-8), 7.17 (2H, d, $J = 8.2$), 7.24 (2H, d, $J = 8.2$). Anal. $C_{24}H_{29}ClN_2O_3$ (C, H, N).

*3.1.1.28. N-Cyclohexyl-6,7-dimethoxy-1-(4′-fluorophenyl)-3,4-dihydroisoquinoline-2(1H)-carboxamide (**30**).* Yield 94%. Mp. 175–177 °C. ^{1}H NMR ($CDCl_3$): 1.03–1.94 (10H, m, cycloalkyl), 2.66–3.68 (5H, m, CH_2CH_2 and CH), 3.78 (3H, s,

CH_3O-6), 3.87 (3H, s, CH_3O-7), 4.32 (1H, bs, NH), 6.36 (1H, s, H-1), 6.59 (1H, s, H-5), 6.66 (1H, s, H-8), 6.92–7.25 (4H, m, ArH). Anal. $C_{24}H_{29}FN_2O_3$ (C, H, N).

3.2. Pharmacology

3.2.1. Testing of anticonvulsant activity against audiogenic seizures in DBA/2 mice

All experiments were performed with DBA/2 mice which are genetically susceptible to sound-induced seizures. DBA/2 mice (8–12 g; 22–25-days-old) were purchased from Harlan Italy (Corezzano, Italy). Groups of 10 mice of either sex were exposed to auditory stimulation 30 min following administration of vehicle or each dose of drugs studied. The compounds were given intraperitoneally (i.p.) (0.1 mL/10 g of body weight of the mouse) as a freshly-prepared solution in 50% dimethylsulfoxide (DMSO) and 50% sterile saline (0.9% NaCl). The mice were individually placed under a hemispheric perspex dome (diameter 58 cm), and 60 s were allowed for habituation and assessment of locomotor activity. Auditory stimulation (12–16 kHz, 109 dB) was applied for 60 s or until tonic extension occurred, and induced a sequential seizure response in control DBA/2 mice, consisting of an early wild running phase, followed by generalized myoclonus and tonic flexion and extension sometimes followed by respiratory arrest. The control mice and drug-treated ones were scored for latency to and incidence of the different phases of the seizures. The experimental protocol and all the procedures involving animals and their care were conducted in conformity with the institutional guidelines and the European Council Directive of laws and policies.

3.2.2. Statistical analysis

Statistical comparisons between groups of control and drug-treated animals were made using Fisher's exact probability test (incidence of the seizure phases). The ED_{50} values of each phase of audiogenic seizures were determined for each dose of compound administered, and dose-response curves were fitted using a computer program by Litchfield and Wilcoxon's method [21].

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References

[1] E. Perucca, C. French, M. Bialer, Development of new antiepileptic drugs: challenges, incentives, and recent advances, Lancet Neurol. 6 (2007) 793–804.
[2] M.A. Rogawski, Molecular targets versus models for new antiepileptic drug discovery, Epilepsy Res. 68 (2006) 22–28.
[3] M.A. Rogawski, Diverse mechanisms of antiepileptic drugs in the development pipeline, Epilepsy Res. 69 (2006) 273–294.
[4] G. De Sarro, R. Gitto, E. Russo, G.F. Ibbadu, M.L. Barreca, L. De Luca, A. Chimirri, AMPA receptor antagonists as potential anticonvulsant drugs, Curr. Top. Med. Chem. 5 (2005) 31–42.
[5] M.L. Barreca, R. Gitto, S. Quartarone, L. De Luca, G. De Sarro, A. Chimirri, Pharmacophore modeling as an efficient tool in the discovery of novel noncompetitive AMPA receptor antagonists, J. Chem. Inf. Comput. Sci. 43 (2003) 651–655.
[6] L. De Luca, R. Gitto, M.L. Barreca, R. Caruso, S. Quartarone, R. Citraro, G. De Sarro, A. Chimirri, 3D pharmacophore models for 1,2,3,4-tetrahydroisoquinoline derivatives acting as anticonvulsant agents, Arch. Pharm. (Weinheim) 339 (2006) 388–400.
[7] R. Gitto, M.L. Barreca, L. De Luca, G. De Sarro, G. Ferreri, S. Quartarone, E. Russo, A. Constanti, A. Chimirri, Discovery of a novel and highly potent noncompetitive AMPA receptor antagonist, J. Med. Chem. 46 (2003) 197–200.
[8] R. Gitto, M.L. Barreca, E. Francica, R. Caruso, L. De Luca, E. Russo, G. De Sarro, A. Chimirri, Synthesis and anticonvulsant properties of 1,2,3,4-tetrahydroisoquinolin-1-ones, Arkivoc (2004) 170–180.
[9] R. Gitto, R. Caruso, V. Orlando, S. Quartarone, M.L. Barreca, G. Ferreri, E. Russo, G. De Sarro, A. Chimirri, Synthesis and anticonvulsant properties of tetrahydroisoquinoline derivatives, Farmaco 59 (2004) 7–12.
[10] R. Gitto, R. Caruso, B. Pagano, L. De Luca, R. Citraro, E. Russo, G. De Sarro, A. Chimirri, Novel potent anticonvulsant agent containing a tetrahydroisoquinoline skeleton, J. Med. Chem. 49 (2006) 5618–5622.
[11] E. Arstad, R. Gitto, A. Chimirri, R. Caruso, A. Constanti, D. Turton, S.P. Hume, R. Ahmad, L.S. Pilowsky, S.K. Luthra, Closing in on the AMPA receptor: Synthesis and evaluation of 2-acetyl-1-(4′-chlorophenyl)-6-methoxy-7-[(11)*C*]methoxy-1,2,3,4-tetrahydro isoquinoline as a potential PET tracer, Bioorg. Med. Chem. 14 (2006) 4712–4717.
[12] G. Ferreri, A. Chimirri, E. Russo, R. Gitto, P. Gareri, A. De Sarro, G. De Sarro, Comparative anticonvulsant activity of *N*-acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline derivatives in rodents, Pharmacol. Biochem. Behav. 77 (2004) 85–94.
[13] G. DeSarro, A. Chimirri, A. DeSarro, R. Gitto, S. Grasso, P. Giusti, A.G. Chapman, GYKI 52466 and related 2,3-benzodiazepines as anticonvulsant agents in DBA/2 mice, Eur. J. Pharmacol. 294 (1995) 411–422.
[14] S.D. Donevan, M.A. Rogawski, GYKI 52466, a 2,3-benzodiazepine, is a highly selective, noncompetitive antagonist of AMPA/kainate receptor responses, Neuron 10 (1993) 51–59.
[15] R. Gitto, M.L. Barreca, L. De Luca, A. Chimirri, New trends in the development of AMPA receptor antagonists, Expert Opin. Ther. Pat. 14 (2004) 1199–1213.
[16] A.G. Chapman, M.J. Croucher, B.S. Meldrum, Evaluation of anticonvulsant drugs in DBA/2 mice with sound-induced seizures, Arzneimittelforschung 34 (1984) 1261–1264.
[17] K.K. Borowicz, A.M. Duda, Z. Kleinrok, S.J. Czuczwar, Interaction of GYKI 52466, a selective non-competitive antagonist of AMPA/kainate receptors, with conventional antiepileptic drugs in amygdala-kindled seizures in rats, Pol. J. Pharmacol. 53 (2001) 101–108.
[18] R. Jakus, M. Graf, R.D. Ando, B. Balogh, I. Gacsalyi, G. Levay, S. Kantor, G. Bagdy, Effect of two noncompetitive AMPA receptor antagonists GYKI 52466 and GYKI 53405 on vigilance, behavior and spike-wave discharges in a genetic rat model of absence epilepsy, Brain Res. 1008 (2004) 236–244.
[19] E. Donato Di Paola, P. Gareri, A. Davoli, S. Gratteri, F. Scicchitano, C. Naccari, G. De Sarro, Influence of levetiracetam on the anticonvulsant efficacy of conventional antiepileptic drugs against audiogenic seizures in DBA/2 mice, Epilepsy Res. 75 (2007) 112–121.
[20] Advanced Chemistry and Development, ACD/Lab, 2004, <www.acdlabs.com>.
[21] J.T. Litchfield Jr., A method for rapid graphic solution of time-percent effect curves, J. Pharmacol. Exp. Ther. 97 (1949) 399–408.