Synthesis and Structure–Active Relationship of 1-Aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline Anticonvulsants

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We have previously disclosed that some 6,7-dimethoxyisoquinoline derivatives are able to produce anticonvulsant effects in different animal models of epilepsy. Following these studies this paper describes the synthesis of a small series of new 1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines strictly related to previously reported analogues. This novel series of isoquinolines was designed on the basis of well defined structure–active relationship (SAR) information already acquired for this class of anticonvulsant agents. The pharmacological effects of the new synthesized compounds were evaluated against audiogenic seizures in Dilute Brown non-Agouti (DBA/2) mice. The preliminary pharmacological screening led to the identification of a new active molecule the 2-acetyl-1-(4′-methylphenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (6d) that displayed significant anticonvulsant activity. Computational studies helped to rationalize these obtained pharmacological results.

Key words isoquinoline; anticonvulsant; Dilute Brown non-Agouti mouse; molecular modeling

Epilepsy is commonly considered the result of an imbalance between excitatory and inhibitory "tone" leading to periodic and unpredictable seizures related to an abnormal discharge of cerebral neurons. Epilepsy is one of the most common neurological disorders, affecting *ca.* 1% of the population worldwide, and the incidence increases to 3% by the age of 75 years. Although most people become seizure-free with drug therapy, there is still a significant number of patients (30%) who are resistant to the currently available antiepileptic drugs whether used alone or in combination. The pathophysiology of epilepsy is complex and several mechanisms play a role in epileptogenesis and epileptogenicity.[1)] The efficacy of anticonvulsant drugs may be connected with different molecular targets to reduce the excitability of neurons involved in seizure onset.[2)] The mechanism of action of currently available effective antiepileptics are: a) the induction of a prolonged inactivation of the Na^+ channel; b) the blockade of Ca^{2+} channel currents; c) the enhancement of the inhibitory γ-aminobutyrate (GABA)ergic 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.[3—10)]

In our previous works, some isoquinoline derivatives were found to have anticonvulsant activities in various seizure models interacting with glutamate ionotropic α-amino-3-hydroxyl-5-methyl-4-isoxazole propionate (AMPA) receptor (AMPAR) subtype in a selective and non-competitive fashion.[11—20)] The most active compound of the series was the 2-acetyl-1-(4′-chlorophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**1**, Fig. 1) which showed both in *in vivo* and *in vitro* tests activity stronger than other known AMPAR antagonists such as GYKI 52466, talampanel, and CFM-2 (Fig. 1).[17,21,22)]

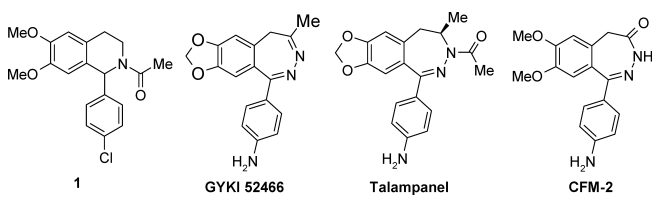

Fig. 1. AMPA Receptor Antagonists as Anticonvulsant Agents

Starting from the "lead compound" **1**, in several previous studies, we examined a large series of its analogues and reported a comprehensive structure–active relationship (SAR) studies concerning the influence of specific substituents on tetrahydroisoquinoline skeleton; we found that the most active derivatives are generally characterized by the presence of 2-acetyl substituent; moreover two methoxy substituents on the benzene fused ring are crucial to produce pharmacological effects.[19)] Furthermore our investigations suggested that the introduction of hydrophobic substituents at 4′-position of C-1 phenyl moiety (*e.g.* halogen atoms) could improve the anticonvulsant efficacy against sound-induced seizures in Dilute Brown non-Agouti (DBA/2) mice. These experimental evidences were confirmed by our computational studies in which we developed a 3-D pharmacophore model for non-competitive AMPAR antagonists.[15)] Herein we report the synthetic approach and the preliminary anticonvulsant screening in DBA/2 mice with the aim to explore other substitutions at the C-1 phenyl group and so to find new anticonvulsant isoquinolines deepening SAR studies. Finally, we tried to rationalize our findings by use a computational approach.

Results and Discussion

As depicted in Chart 1 a small series of 2-acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline derivatives (**6a**—**f**) was prepared following previously reported procedures[17,23)] with slight modifications. In particular, we employed an efficient high-speed microwave-assisted synthetic approach (Chart 1) optimizing the chemical yield and purity as well as greatly reducing the reaction times and solvent

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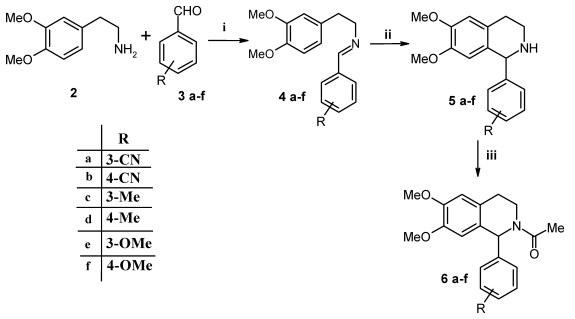
amount. In particular, in solvent-free conditions and starting from amine **2** and suitable aldehyde **3a**—**f** we prepared the imine intermediates **4a**—**f** which, in the same reactor, were transformed into 1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines (**5a**—**f**). Then compounds **5a**—**f** were readily acetylated to give desired final compounds **6a**—**f**. The structures of the obtained compounds were supported by elemental analyses and spectroscopic measurements.

The anticonvulsant effects of the new 2-acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline derivatives (**6a**—**f**) and the corresponding parent compounds **5a**—**f** were evaluated against audiogenic seizures in DBA/2 mice, considered an excellent animal model for generalized epilepsy and for screening new anticonvulsant drugs.[24] Table 1 reports the pharmacological results for compounds **5a**—**f** and **6a**—**f** compared with those of known non-competitive AMPA receptor antagonists such as isoquinoline **1** and 2,3-benzodiazepines (GYKI 52466, CFM-2 and talampanel) as well as of other antiepileptic drugs such as gabapentin and valproate.[25]

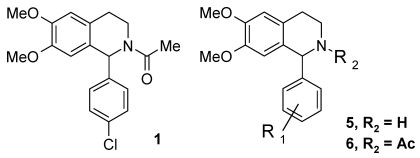


Reagents and conditions: (i) MW, 5 min, 90 °C, 100 W; (ii) TFA, MW, 5 min, 90 °C, 280 W; (iii) Ac_2O, MW, 5 min, 70 °C, 150 W.

Chart 1

The biological results reported in Table 1 suggest that these new synthesized compounds generally possess potency lower than "lead compound" **1**.[12,16] The analysis of ED_{50} values on clonic and tonic phases of the test (see Table 1) highlighted that the most active molecules of this new series of compounds were derivatives **5d** and **6d** bearing 4′-methyl substituent on C-1 phenyl ring. In particular, compound **6d** exhibited an anticonvulsant potency better than that of GYKI 52466, the prototype of non-competitive AMPAR antagonists showing anticonvulsant efficacy. Moreover, the anticonvulsant effects of compound **6d** were comparable to those of active 2,3-benzodiazepine derivatives CFM-2 and talampanel, which has already undergone phase 1 and 2 trials for refractory epilepsy, as well as amyotrophic lateral sclerosis and Parkinson disease.[21,22] It is interesting to note that in this experimental model of epilepsy the observed activity of compound **6d** exceeded that of valproate and gabapentin, two drugs largely used in antiepileptic therapy. On the basis of calculation of $Log D_{7.4}$,[26] predicted from a commercially available program (advanced chemistry development/Lab), we hypothesized that the hydrophobic character of methyl substituent could positively contribute to the blood–brain barrier crossing thus improving the anticonvulsant efficacy. But we also observed a discrepancy of the potency between 3′-methyl-substituted derivative **6c** and 4′-methyl-substituted derivative **6d**, so we decided to superimpose these two compounds in our 3-D pharmacophore model developed for non-competitive AMPAR antagonists.[15] According to the previous reported pharmacophoric hypothesis five structural motifs control AMPAR recognition process; they are two hydro-

Table 1. Anticonvulsant Activity against Audiogenic Seizures in DBA/2 Mice and Estimated Log $D_{7.4}$ Values of Tested Isoquinolines (**1**, **5a**—**f**, and **6a**—**f**)



Compd.	R_1	R_2	ED_{50} mg/kg[a]		Log $D_{7.4}$[b]
			Clonus	Tonus	
1	4-Cl	Ac	1.44 (0.77—1.02)	0.42 (0.31—0.54)	3.40
5a	3-CN	H	>35	>35	1.31
5b	4-CN	H	>35	14.3 (9.67—21.1)	1.25
5c	3-Me	H	15.5 (11.2—21.5)	8.38 (5.20—13.4)	1.76
5d	4-Me	H	13.7 (9.28—20.3)	10.3 (7.98—13.3)	1.88
5e	3-OMe	H	>35	>35	1.38
5f	4-OMe	H	>35	24.0 (14.3—40.1)	1.39
6a	3-CN	Ac	32.3 (28.7—35.3)	19.4 (14.1—26.7)	2.24
6b	4-CN	Ac	31.7 (21.9—45.7)	11.8 (6.78—20.5)	2.24
6c	3-Me	Ac	>35	>35	3.27
6d	4-Me	Ac	6.63 (4.09—10.7)	5.98 (3.93—9.07)	3.27
6e	3-OMe	Ac	30.9 (24.6—38.9)	16.5 (12.1—22.6)	2.72
6f	4-OMe	Ac	>35	25.1 (21.4—29.6)	2.72
CFM-2	—	—	4.66 (2.80—7.46)	3.92 (2.49—5.91)	—
GYKI 52466	—	—	10.5 (7.15—15.3)	7.41 (4.69—11.7)	—
Talampanel	—	—	4.51 (3.40—5.99)	3.27 (2.36—4.52)	
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} 95% confidence limits are given in parentheses. *b*) Log D data at pH=7.4 are predicted from a commercially available program (ACD/Lab).[26]

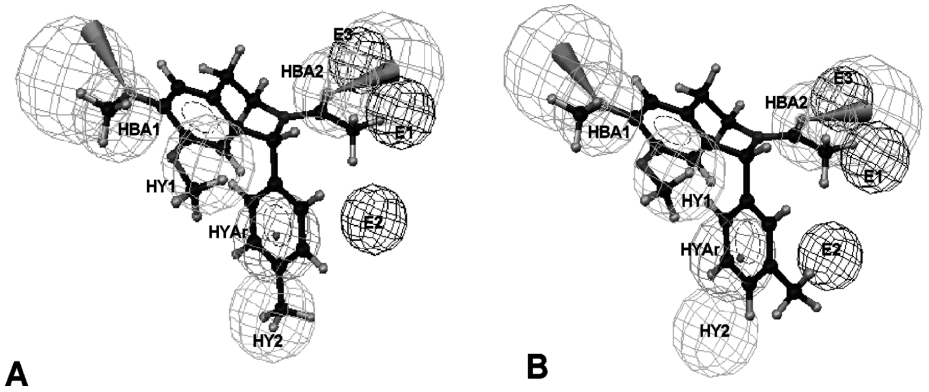


Fig. 2. Alignments of Compounds **6d** (A) and **6c** (B) into the Pharmacophoric Hypothesis (Hydrogen Bond Acceptor, HBA1 and HBA2; Hydrophobic Aromatic Region, HYAr; Hydrophobic Group, HY1 and HY2; Excluded Volumes, E1, E2, E3)

gen-bond acceptors (HBA), one hydrophobic aromatic region (HYAr), and two hydrophobic region (HY); moreover, in this model we described three excluded volumes (E1—E3) as forbidden areas. Figure 2A displays the alignment of the most active derivative compound **6d** revealing that all favorable chemical features were well matched by this molecule and the 4′-methyl substituent fits the HY2 chemical feature; on the contrary in the case of the less active compound **6c** we found that in its best alignment into pharmacophoric hypothesis the HY2 feature was lacking (see Fig. 2B). Considering that the low active pairs of C-3′ and C-4′ substituted 2-acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline derivatives (**6a** and **6b**, **6e** and **6f**) were unable to fit all requested chemical features, their pharmacological efficacy was unaffected by the substituent shift from C-4′ to C-3′ position.

In conclusion, we have identified a new active compound (**6d**) which shows significant anticonvulsant activity. On the basis of our pharmacological data as well as our molecular modeling studies, we also suggest that a methyl group at the 4-position of the C-1 phenyl ring is the best substituent for anticonvulsant efficacy of this new small series of 2-acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines.

Experimental

Chemistry All starting materials and reagents commercially available (Sigma-Aldrich Milan, Italy) were used without further purification. Microwave-assisted reactions were carried out in a CEM focused Microwave Synthesis System. 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. *Rf*=values were determined employing TLC plates and using a $CHCl_3$/MeOH mixture as eluent. ^{1}H- and ^{13}C-NMR spectra were measured in $CDCl_3$ with a Varian Gemini 300 spectrometer; chemical shifts are expressed in δ (ppm) relative to tetramethyl silane (TMS) as internal standard and coupling constants (*J*) in Hz. All exchangeable protons were confirmed by addition of deuterium oxide (D_2O).

General Procedure for the Synthesis of 1-Aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines (5a—f) A mixture of 2-(3′,4′-dimethoxyphenyl)ethylamine (**2**) (1.0 mmol) and suitable benzaldehyde derivative **3a**—**f** (1.2 mmol) was placed in a cylindrical quartz tube (ϕ2 cm), then stirred and irradiated in a microwave oven at 100 W for 5 min at 90 °C; after cooling to room temperature trifluoroacetic acid (2 ml) was added and the mixture was irradiated at 280 W for 5 min at 90 °C. The reaction was quenched by adding water, and the mixture was basified (pH≈8—9) with sodium hydroxide (2 M aqueous) to give a solid. The crude product was powdered by treatment with diethyl ether, filtered and purified by crystallization with EtOH to afford 1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines (**5a**—**f**).

1-(3′-Cyanophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**5a**): Yield: 45%; mp 135—137 °C. *Rf*=0.61 ($CHCl_3$/MeOH; 90 : 10). ^{1}H-NMR ($CDCl_3$) δ: 2.78—3.15 (4H, m, CH_2CH_2), 3.66 (3H, s, OCH_3), 3.89 (3H, s, OCH_3), 5.08 (1H, s, CH), 6.16 (1H, s, ArH), 6.66 (1H, s, ArH), 7.43—7.59 (4H, m, ArH). *Anal.* Calcd for $C_{18}H_{18}N_2O_2$: C, 75.45; H, 6.16; N, 9.52. Found: C, 75.41; H, 6.26; N, 9.33.

1-(4′-Cyanophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**5b**): Yield: 46%; mp 217—219 °C. *Rf*=0.52 ($CHCl_3$/MeOH; 90 : 10). ^{1}H-NMR ($CDCl_3$) δ: 2.85—3.18 (4H, m, CH_2CH_2), 3.63 (3H, s, OCH_3), 3.90 (3H, s, OCH_3), 5.42 (1H, s, CH), 6.07 (1H, s, ArH), 6.67 (1H, s, ArH), 7.44 (2H, d, *J*=8.24, ArH), 7.68 (2H, d, *J*=8.24, ArH). *Anal.* Calcd for $C_{18}H_{18}N_2O_2$: C, 75.45; H, 6.16; N, 9.52. Found: C, 75.55; H, 6.31; N, 9.43.

6,7-Dimethoxy-1-(3′-methylphenyl)-1,2,3,4-tetrahydroisoquinoline (**5c**): Yield: 52%; mp 109—111 C. *Rf*=0.39 ($CHCl_3$/MeOH; 90 : 10). ^{1}H-NMR ($CDCl_3$) δ: 2.33 (3H, s, CH_3), 2.77—3.24 (4H, m, CH_2CH_2), 3.65 (3H, s, OCH_3), 3.88 (3H, s, OCH_3), 5.01 (1H, s, CH), 6.26 (1H, s, ArH), 6.63 (1H, s, ArH), 7.01—7.23 (4H, m, ArH). *Anal.* Calcd for $C_{18}H_{21}NO_2$: C, 76.30; H, 7.47; N, 4.94. Found: C, 76.50; H, 7.61; N, 4.63.

6,7-Dimethoxy-1-(4′-methylphenyl)-1,2,3,4-tetrahydroisoquinoline (**5d**): Yield: 72%; mp 140—142 °C. *Rf*=0.20 ($CHCl_3$/MeOH; 90 : 10). ^{1}H-NMR ($CDCl_3$) δ: 2.34 (3H, s, CH_3), 2.73—3.26 (4H, m, CH_2CH_2), 3.64 (3H, s, OCH_3), 3.87 (3H, s, OCH_3), 5.03 (1H, s, CH), 6.26 (1H, s, ArH), 6.62 (1H, s, ArH) 7.13—7.24 (4H, m, ArH). *Anal.* Calcd for $C_{18}H_{21}NO_2$: C, 76.30; H, 7.47; N, 4.94. Found: C, 76.22; H, 7.34; N, 4.90.

6,7-Dimethoxy-1-(3′-methoxyphenyl)-1,2,3,4-tetrahydroisoquinoline (**5e**): Yield: 62%; mp 97—99 °C. *Rf*=0.43 ($CHCl_3$/MeOH; 90 : 10). ^{1}H-NMR ($CDCl_3$) δ: 2.74—3.26 (4H, m, CH_2CH_2), 3.65 (3H, s, OCH_3), 3.78 (3H, s, OCH_3) 3.88 (3H, s, OCH_3), 5.02 (1H, s, CH), 6.28 (1H, s, ArH), 6.63 (1H, s, ArH) 6.80—7.27 (4H, m, ArH). *Anal.* Calcd for $C_{18}H_{21}NO_3$: C, 72.22; H, 7.07; N, 4.68. Found: C, 72.42; H, 7.11; N, 4.70.

6,7-Dimethoxy-1-(4′-methoxyphenyl)-1,2,3,4-tetrahydroisoquinoline (**5f**): Yield: 40%; mp 94—96 °C. *Rf*=0.25 ($CHCl_3$/MeOH; 90 : 10). ^{1}H-NMR ($CDCl_3$) δ: 2.71—3.24 (4H, m, CH_2CH_2), 3.65 (3H, s, OCH_3), 3.81 (3H, s, OCH_3) 3.87 (3H, s, OCH_3), 5.01 (1H, s, CH), 6.25 (1H, s, ArH), 6.62 (1H, s, ArH), 6.85 (2H, d, *J*=8.51, ArH) 7.17 (2H, d, *J*=8.51, ArH). ^{13}C-NMR ($CDCl_3$) δ: 29.4 and 41.0 (C-3 and C-4), 54.8 (OCH_3), 55.9 (OCH_3), 56.8 (OCH_3), 60.3 (C-1) 111.0 and 111.9 (C-5 and C-8), 114.8 (C-3′,5′), 125.7 and 126.0 (C-4a and C-8a), 129.3 (C-2′,6′), 135.8 (C-1′), 147.5 and 148.8 (C-6, C-7), 158.1 (C-4′). *Anal.* Calcd for $C_{18}H_{21}NO_3$: C, 72.22; H, 7.07; N, 4.68. Found: C, 72.34; H, 7.00; N, 4.61.

General Procedure for the Synthesis of 2-Acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinolines (6a—f) A solution of isoquinoline derivative (**5a**—**f**) (1 mmol) in Ac_2O (1 ml) was irradiated in a microwave oven at 150 W for 5 min at 70 °C; after cooling the reaction was quenched by adding water and extracted with ethyl acetate (3×5 ml). The organic layer was dried over Na_2SO_4, and the solvent was removed until dryness under reduced pressure. The oil residue was powdered with diethylether and crystallized with EtOH to give 2-acetyl-1-aryl-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline derivatives **6a**—**f**.

2-Acetyl-1-(3′-cyanophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**6a**): Yield 88%; mp 205—207 °C. *Rf*=0.74 ($CHCl_3$/MeOH; 98 : 2). ^{1}H-

NMR ($CDCl_3$) δ: 2.19 (3H, s, CH_3), 2.79—3.84 (4H, m, CH_2CH_2), 3.77 (3H, s, OCH_3), 3.91 (3H, s, OCH_3), 6.46 (1H, s, CH), 6.69 (1H, s, ArH), 6.87 (1H, s, ArH), 7.40—7.51 (4H, m, ArH). *Anal.* Calcd for $C_{20}H_{20}N_2O_3$: C, 71.41; H, 5.99; N, 8.33. Found: C, 71.33; H, 5.80; N, 8.62.

2-Acetyl-1-(4′-cyanophenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**6b**): Yield: 69%; mp 96—98 °C. *Rf*=0.66 ($CHCl_3$/MeOH; 98 : 2). ^{1}H-NMR ($CDCl_3$) δ: 2.18 (3H, s, CH_3), 2.72—3.72 (4H, m, CH_2CH_2), 3.76 (3H, s, OCH_3), 3.89 (3H, s, OCH_3), 6.46 (1H, s, CH), 6.68 (1H, s, ArH), 6.87 (1H, s, ArH), 7.36 (2H, d, *J*=8.34, ArH), 7.57 (2H, d, *J*=8.34 Hz, ArH). ^{13}C-NMR ($CDCl_3$) δ: 21.4 (CH_3), 31.4 and 42.0 (C-3 and C-4), 53.3 (C-1), 55.8 (OCH_3), 56.4 (OCH_3), 110.9 and 111.9 (C-5 and C-8), 112.3 (CN), 118.9 (C-4′), 125.7 and 126.0 (C-4a and C-8a), 129.9 (C-2′,6′), 132.7 (C-3′,5′), 146.9 (C-1′), 148.3 and 149.2 (C-6 and C-7), 168.2 (CO). *Anal.* Calcd for $C_{20}H_{20}N_2O_3$: C, 71.41; H, 5.99; N, 8.33. Found: C, 71.66; H, 5.90; N, 8.30.

2-Acetyl-1-(3′-methylphenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**6c**): Yield: 60%; mp 167—169 °C. *Rf*=0.63 ($CHCl_3$/MeOH; 98 : 2). ^{1}H-NMR ($CDCl_3$) δ: 2.17 (3H, s, CH_3), 2.30 (3H, s, CH_3), 2.77—3.71 (4H, m, CH_2CH_2), 3.76 (3H, s, OCH_3), 3.90 (3H, s, OCH_3), 6.52 (1H, s, CH), 6.66 (1H, s, ArH), 6.86 (1H, s, ArH), 6.97—7.16 (4H, m, ArH). *Anal.* Calcd for $C_{20}H_{23}NO_3$: C, 73.82; H, 7.12; N, 4.30. Found: C, 73.60; H, 7.22; N, 4.26.

2-Acetyl-1-(4′-methylphenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**6d**): Yield: 58%; mp 184—186 °C. *Rf*=0.50 ($CHCl_3$/MeOH; 98 : 2). ^{1}H-NMR ($CDCl_3$) δ: 2.16 (3H, s, CH_3), 2.32 (3H, s, CH_3), 2.72—3.68 (4H, m, CH_2CH_2), 3.75 (3H, s, OCH_3), 3.89 (3H, s, OCH_3), 6.51 (1H, s, CH), 6.65 (1H, s, ArH), 6.86 (1H, s, ArH), 7.07—7.15 (4H, m, ArH). ^{13}C-NMR ($CDCl_3$) δ: 21.3 ($CO\underline{CH_3}$), 24.3 (CH_3) 31.5 and 42.0 (C-3 and C-4), 55.3 (C-1), 55.8 (OCH_3), 56.4 (OCH_3), 110.9 and 111.7 (C-5 and C-8), 125.8 and 126.5 (C-4a and C-8a), 127.9 (C-2′,6′), 129.7 (C-3′,5′), 138.2 (C-1′), 139.0 (C-4′), 148.3 and 149.2 (C-6 and C-7), 168.2 (CO). *Anal.* Calcd for $C_{20}H_{23}NO_3$: C, 73.82; H, 7.12; N, 4.30. Found: C, 74.00; H, 7.15; N, 4.31.

2-Acetyl-1-(3′-methoxyphenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**6e**): Yield: 61%; mp 187—189 °C. *Rf*=0.31 ($CHCl_3$/MeOH; 98 : 2). ^{1}H-NMR ($CDCl_3$) δ: 2.17 (3H, s, CH_3), 2.71—3.69 (4H, m, CH_2CH_2), 3.76 (3H, s, OCH_3), 3.76 (3H, s, OCH_3), 3.89 (3H, s, OCH_3), 6.53 (1H, s, CH), 6.65 (1H, s, ArH), 6.61—7.22 (5H, m, ArH). ^{13}C-NMR ($CDCl_3$) δ: 21.4 ($CO\underline{CH_3}$), 31.4 and 42.3 (C-3 and C-4), 54.8 (OCH_3), 55.6 (OCH_3), 56.7 (OCH_3), 57.1 (C-1), 110.2 and 111.8 (C-5 and C-8), 112.0 (C-2′), 112.8 (C-4′), 120.9 and 132.7 (C-5′,6′), 125.7 and 126.0 (C-4a and C-8a), 146.7 (C-1′), 148.2 and 149.4 (C-6 and C-7), 162.0 (C-3′) 168.5 (CO). *Anal.* Calcd for $C_{20}H_{23}NO_4$: C, 70.63; H, 6.79; N, 4.10. Found: C, 70.68; H, 6.66; N, 4.45.

2-Acetyl-1-(4′-methoxyphenyl)-6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (**6f**): Yield: 66%; mp 107—109 °C. *Rf*=0.33 ($CHCl_3$/MeOH; 98 : 2). ^{1}H-NMR ($CDCl_3$) δ: 2.16 (3H, s, CH_3), 2.69—3.65 (4H, m, CH_2CH_2), 3.75 (3H, s, OCH_3), 3.78 (3H, s, OCH_3), 3.89 (3H, s, OCH_3), 6.51 (1H, s, CH), 6.65 (1H, s, ArH), 6.79 (1H, s, ArH), 6.82 (2H, d, *J*=8.44 Hz, ArH), 7.17 (2H, d, *J*=8.44 Hz, ArH). *Anal.* Calcd for $C_{20}H_{23}NO_4$: C, 70.63; H, 6.79; N, 4.10. Found: C, 70.70; H, 6.60; N, 4.31.

Pharmacology. Testing of Anticonvulsant Activity All experiments were performed with DBA/2 mice which are genetically susceptible to sound-induced seizures. DBA/2 mice (8—12 g; 21—25-d-old) were purchased from Harlan Italy (Corezzana, 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). Individual mice were 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 and drug-treated mice 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.[20]

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 was determined for each dose of compound administered, and dose–response curves were fitted using a computer program by Litchfield and Wilcoxon's method.[27]

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References

1) Bialer M., White H. S., *Nat. Rev. Drug Discov.*, **9**, 68—82 (2010).
2) Meldrum B. S., Rogawski M. A., *Neurotherapeutics*, **4**, 18—61 (2007).
3) Mayer M. L., *Curr. Opin. Neurobiol.*, **15**, 282—288 (2005).
4) Rogawski M. A., *Epilepsy Res.*, **68**, 22—28 (2006).
5) Catarzi D., Colotta V., Varano F., *Med. Res. Rev.*, **27**, 239—278 (2007).
6) Rogawski M. A., *Epilepsy Res.*, **69**, 273—294 (2006).
7) Palmer C. L., Cotton L., Henley J. M., *Pharmacol. Rev.*, **57**, 253—277 (2005).
8) De Sarro G., Gitto R., Russo E., Ibbadu G. F., Barreca M. L., De Luca L., Chimirri A., *Curr. Top. Med. Chem.*, **5**, 31—42 (2005).
9) Chimirri A., De Sarro G., Quartarone S., Barreca M. L., Caruso R., De Luca L., Gitto R., *Pure Appl. Chem.*, **76**, 931—939 (2004).
10) Stefan H., Feuerstein T. J., *Pharmacol. Ther.*, **113**, 165—183 (2007).
11) Barreca M. L., Gitto R., Quartarone S., De Luca L., De Sarro G., Chimirri A., *J. Chem. Inf. Comput. Sci.*, **43**, 651—655 (2003).
12) Gitto R., Barreca M. L., De Luca L., De Sarro G., Ferreri G., Quartarone S., Russo E., Constanti A., Chimirri A., *J. Med. Chem.*, **46**, 197—200 (2003).
13) Ferreri G., Chimirri A., Russo E., Gitto R., Gareri P., De Sarro A., De Sarro G., *Pharmacol. Biochem. Behav.*, **77**, 85—94 (2004).
14) Gitto R., Caruso R., Orlando V., Quartarone S., Barreca M. L., Ferreri G., Russo E., De Sarro G., Chimirri, A., *Farmaco*, **59**, 7—12 (2004).
15) De Luca L., Gitto R., Barreca M. L., Caruso R., Quartarone S., Citraro R., De Sarro G., Chimirri A., *Arch. Pharm.* (Weinheim), **339**, 388—400 (2006).
16) Gitto R., Caruso R., Pagano B., De Luca L., Citraro R., Russo E., De Sarro G., Chimirri A., *J. Med. Chem.*, **49**, 5618—5622 (2006).
17) Gitto R., Ficarra R., Stancanelli R.,. Guardo M, De Luca L., Barreca M. L., Pagano B., Rotondo A., Bruno G., Russo E., De Sarro G., Chimirri A., *Bioorg. Med. Chem.*, **15**, 5417—5423 (2007).
18) Gitto R., De Luca L., Pagano B., Citraro R., De Sarro G., Costa L., Ciranna L., Chimirri A., *Bioorg. Med. Chem.*, **16**, 2379—2384 (2008).
19) Chimirri A., De Luca L., Ferro S., De Sarro G., Ciranna L., Gitto R., *ChemMedChem*, **4**, 917—922 (2009).
20) Gitto R., Pagano B., Citraro R., Scicchitano F., De Sarro G., Chimirri A., *Eur. J. Med. Chem.*, **44**, 1349—1354 (2009).
21) Howes J. F., Bell C., *Neurotherapeutics*, **4**, 126—129 (2007).
22) Traynor B. J., Bruijn L., Conwit R., Beal F., O'Neill G., Fagan S. C., Cudkowicz M. E., *Neurology*, **67**, 20—27 (2006).
23) Gao M., Kong D., Clearfield A., Zheng Q. H., *Bioorg. Med. Chem. Lett.*, **16**, 2229—2233 (2006).
24) Chapman A. G., Croucher M. J., Meldrum B. S., *Arzneim.-Forsch.*, **34**, 1261—1264 (1984).
25) Donato Di Paola E., Gareri P., Davoli A., Gratteri S., Scicchitano F., Naccari C., De Sarro G., *Epilepsy Res.*, **75**, 112—121 (2007).
26) Advanced Chemistry Development, ACD/Lab 2004, in www.acdlabs.com/.
27) Litchfield J. T. Jr., *J. Pharmacol. Exp. Ther.*, **97**, 399—408 (1949).