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Synthesis, structure–activity relationships, and anticonvulsant activities of 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives as orally active AMPA receptor antagonists

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ABSTRACT

As part of a program aimed at discovering orally active 2-amino-3-(3-hydroxy-5-methyl-4-isoxazolyl)propionic acid (AMPA) receptor antagonists, we screened our compound library and identified 2-[allyl(4-methylphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (**7**) as a lead compound that inhibited kainate-induced neurotoxicity mediated by AMPA receptors in rat hippocampal cultures. Structure–activity relationship studies of a series of 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives revealed that substituents on the phenyl ring attached to the 2-amino group and the 4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one ring system play an important role in inhibitory activity against kainate-induced neurotoxicity. Several analogs bearing a phenyl group with a 4-substituent or five- or six-membered ring fused at the 3,4-positions exhibited potent inhibitory activity against kainate-induced neurotoxicity. Further, some of these compounds exhibited significant suppression of maximal electroshock seizure in mice following oral administration. Of these compounds, 2-[(4-chlorophenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (**16i**) (YM928) demonstrated the most potent inhibitory effect with an ED_{50} value of 7.4 mg/kg.

1. Introduction

Glutamate is a major excitatory neurotransmitter in the central nervous system (CNS) of vertebrates and plays an essential role in a variety of physiological functions via the activation of postsynaptic ionotropic and metabotropic receptors. Overstimulation of the ionotropic receptors leads to neuronal cell damage and death in epilepsy, cerebral ischemia, and CNS trauma, as well as in various forms of neurodegeneration such as Parkinson's disease and amyotrophic lateral sclerosis.[1] Antagonists of ionotropic glutamate receptors are therefore considered as promising candidates for the development of novel therapeutic agents.[2,3] The ionotropic 2-amino-3-(3-hydroxy-5-methyl-4-isoxazolyl)propionic acid (AMPA) receptor has attracted interest and a number of competitive and noncompetitive antagonists for this receptor have been identified during the past quarter-century (Fig. 1).[4–8]

Quinoxalinediones are the most representative class of competitive antagonist[4] with a number of reported compounds, such as NBQX (**1**),[9] YM90K (**2**),[10] zonampanel (**3**)[11] and others.[12–14] Due to their neuroprotective effects in models of cerebral ischemia, some of these compounds have advanced to clinical trials. Presumably due to their highly polar properties, however, these compounds have rarely exhibited acceptable efficacy following oral administration, which hampers their clinical use for CNS diseases other than stroke. Several different series of compounds have also been described as competitive AMPA receptor antagonists,[15–24] some of which have displayed oral activity in models of seizure or pain.[16,17,21–23] However, none of these have been marketed for use as therapeutic agents.

Approaches to develop novel AMPA receptor antagonists have yielded other classes of compounds that act noncompetitively at the receptor. These compounds are considered to have the advantage of maintaining efficacy even in the presence of the elevated levels of glutamate observed with some neurological disorders.[25,26] Further, these agents are potentially orally active, as the allosteric binding site for these compounds appears less polar than the glutamate binding site.[27,28] The most representative class of noncompetitive antagonists are the 2,3-benzodiazepines,[29,30] such as

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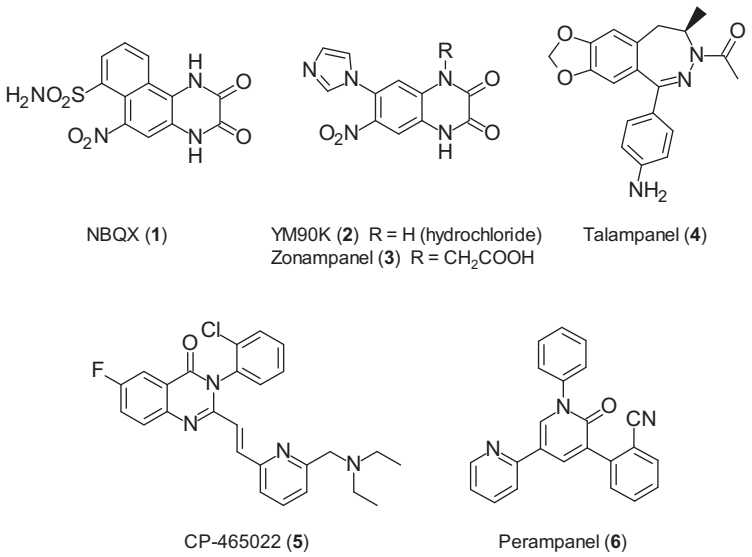
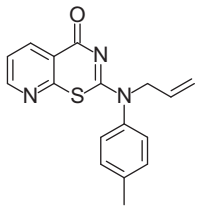


Figure 1. Structures of AMPA receptor antagonists.

talampanel (**4**),[31] which exhibit potent activity in various animal seizure models following oral administration. Talampanel (**4**) has been developed for the treatment of epilepsy, other neurodegenerative disorders, and brain tumors, but it has not yet reached the market. Several recent reports have been published on different series of noncompetitive antagonists, such as CP-465022 (**5**)[32] and perampanel (**6**).[33,34] Perampanel (**6**) was developed by Eisai and launched on the market in 2012 as the first AMPA receptor antagonist for the treatment of epilepsy. However, despite intense research to develop AMPA receptor antagonists, there are only a few drug candidates currently in clinical development.

In an effort to develop a novel class of orally active AMPA receptor antagonists, we screened our compound library to identify inhibitors of kainate-induced neurotoxicity mediated by AMPA receptors in primary rat hippocampal cultures.[35] As a result, we identified 2-[allyl(4-methylphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (**7**) as a lead compound with an IC_{50} value of 9.0 μM (Fig. 2). We then modified the 2-amino group to give the optimal compound 2-[(4-chlorophenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (**16i**) (YM928). As we previously reported, functional characterization of this compound showed that it did not displace [^{3}H]AMPA binding to rat forebrain membrane, and that it blocked AMPA-induced intracellular calcium influx and inward currents in primary rat hippocampal cultures. These results demonstrate that this compound is a noncompetitive AMPA receptor antagonist.[36] We also previously reported the anticonvulsant activity of **16i** in various animal seizure models following oral administration.[37–39] Here, we describe the synthesis, structure–activity relationships (SARs), and anticonvulsant activities in a maximal electroshock seizure (MES) test in mice of a series of 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives, including **16i**.



7

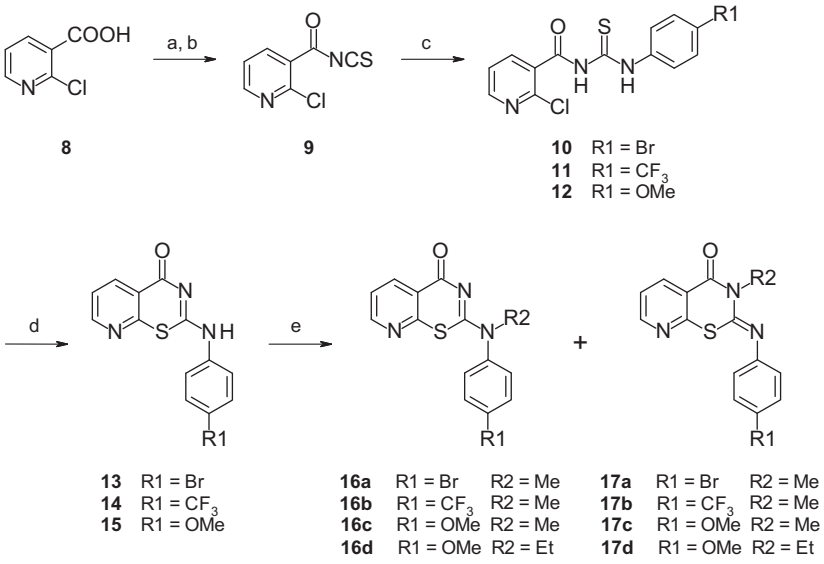
Figure 2. Structure of compound **7**.

2. Chemistry

The synthesis of 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives is outlined in Schemes 1 and 2.[40] The reaction of the acid chloride prepared from 2-chloronicotinic acid (**8**) with ammonium thiocyanate gave 2-chloronicotinoyl isothiocyanate (**9**). Addition of appropriate primary anilines to the isothiocyanate **9** gave *N*-carbamothioyl-2-chloronicotinamides **10**–**12**, which when cyclized in refluxing toluene gave *N*-monosubstituted 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-ones **13**–**15** in 80–92% overall yields. Alkylation of these compounds with appropriate alkyl iodides in the presence of lithium hydride gave the desired *N*,*N*-disubstituted 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-ones **16a**–**d** in 4–11% yields along with corresponding N3-alkylated analogs **17a**–**d** as dominant products in 44–86% yields. Compounds **16a**–**d** and **17a**–**d** were easily separated by column chromatography and structures were confirmed by carbonyl absorption in their infrared (IR) spectra as previously described by Koščik et al.[40] *N*,*N*-Disubstituted 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-ones were amenable to direct and efficient preparation by the addition of appropriate secondary amines to the isothiocyanate **9** at ambient temperature. Compounds **18**–**21** and **16e**–**k**, **m**, **o**–**u** were synthesized by this method in 18–90% yields. The secondary amines used in this step were either commercially available or prepared via known procedures.[41] Aniline derivative **16l** was obtained by hydrogenation of **16k** in 72% yield. Hydrolysis of **16m** gave the benzoic acid derivative **16n** in 91% yield.

As shown in Scheme 3, 4*H*-pyrido[3,4-*e*][1,3]thiazin-4-one derivative **24** was synthesized from 4-chloronicotinic acid (**22**) in 26% yield, and a 7-methyl analog of **16i** (**25**) was synthesized from 2-chloro-6-methylnicotinic acid (**23**) in 39% yield by a similar method to that described in Scheme 2.

For the synthesis of 4*H*-pyrido[4,3-*e*][1,3]thiazin-4-one derivative **30**, *N*-carbamothioyl-3-fluoroisonicotinamide **28** was prepared from 3-fluoroisonicotinic acid (**26**) and cyclized in the presence of sodium hydride at 110 °C in a total yield of 29% as shown in Scheme 4. In a similar manner, benzothiazinone derivative **31** was synthesized starting from 2-fluorobenzoic acid (**27**)



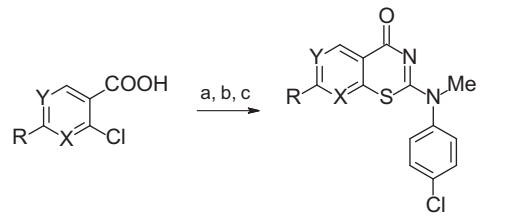
Scheme 1. Reagents and conditions: (a) $SOCl_2$, cat. DMF; (b) NH_4SCN, acetone; (c) primary aniline, acetone; (d) toluene; (e) LiH, DMF, then alkyl iodide.

in a total yield of 64%, and thienothiazinone derivative **34** was synthesized starting from 3-chlorothiophene-2-carboxylic acid (**32**) in a total yield of 42%.

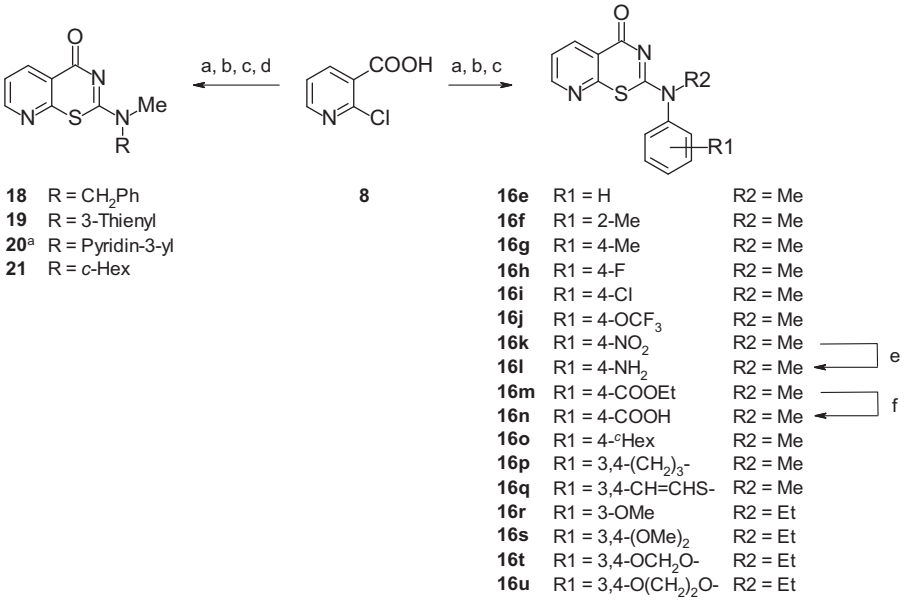
As shown in Scheme 5, benzoxazinone derivative **36** was synthesized via O-cyanation of methyl salicylate (**35**), followed by a treatment with 4-chloro-*N*-methylaniline in the presence of benzoic acid in 71% yield in accordance with a known procedure.[42]

3. Results and discussion

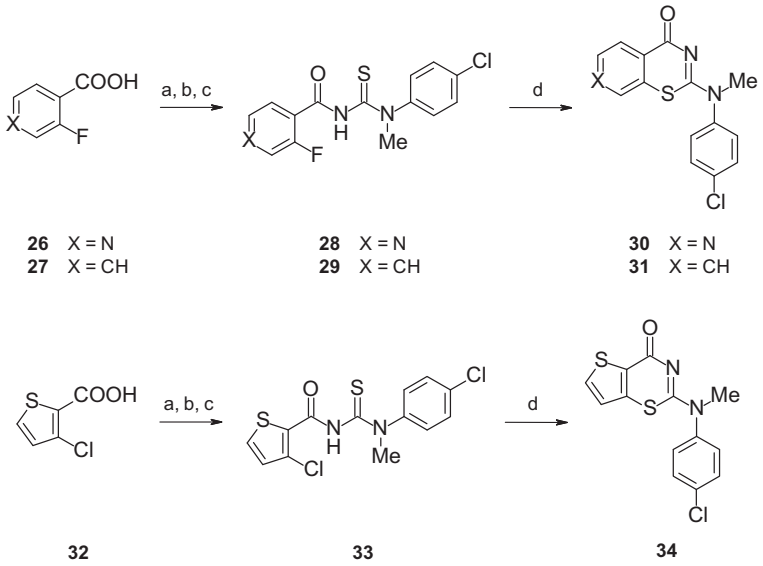
We used kainate-induced neurotoxicity in primary rat hippocampal cultures to assess the inhibitory activities of the synthesized compounds. The structures of the compounds and the results of the assay are summarized in Tables 1–5. Inhibitory activities are presented as the concentration required for 50% inhibition of the neurotoxicity response (IC_{50}; μM) or percent



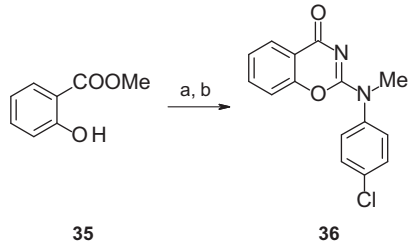
Scheme 3. Reagents and conditions: (a) $SOCl_2$, cat. DMF; (b) NH_4SCN, acetone; (c) 4-chloro-*N*-methylaniline, acetone.



Scheme 2. Reagents and conditions: (a) $SOCl_2$, cat. DMF; (b) NH_4SCN, acetone; (c) secondary amine, acetone; (d) HCl in EtOAc, EtOH for **20**; (e) H_2, Raney-nickel, DMF; (f) aq NaOH, EtOH. [a]HCl salt.

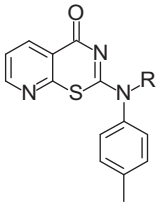


Scheme 4. Reagents and conditions: (a) $SOCl_2$, cat. DMF; (b) NH_4SCN, acetone; (c) 4-chloro-*N*-methylaniline, acetone; (d) NaH, DMF, THF.



Scheme 5. Reagents and conditions: (a) BrCN, Et_3N, acetone; (b) 4-chloro-*N*-methylaniline, PhCOOH, acetone.

Table 1
Inhibitory activities of *N*-substituted 2-[(4-methylphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives against kainate-induced neurotoxicity



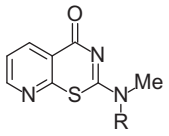
Compound	R	Anti-kainate toxicity IC_{50}[a] (μM)
7	Allyl	9.0
16g	Me	10
Talampanel (**4**)		6.0

[a] Concentration necessary for 50% inhibition.

inhibition of the response at a concentration of 10 μM test compound.

We first examined the effect of an allyl substituent on the 2-amino group of compound **7** (Table 1). Compound **16g** bearing a methyl group retained activity comparable to that of **7**, indicating that the allyl substituent is not critical for inhibitory activity. We therefore conducted further investigations using *N*-methyl analogs, which were readily synthesized from commercially available *N*-methylamines.

Table 2
Inhibitory activities of *N*-substituted 2-(methylamino)-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives against kainate-induced neurotoxicity



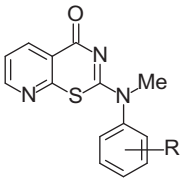
Compound	R	Anti-kainate toxicity % inhibition (at 10 μM)
16e	Ph	43
18	CH_2Ph	0
19	3-Thienyl	28
20[a]	Pyridin-3-yl	18
21	*c*-Hex	16

[a] HCl salt.

We next assessed the effect of a phenyl moiety (Table 2). The phenyl derivative **16e** exhibited moderate activity with 43% inhibition at 10 μM. Insertion of a methylene group between the nitrogen atom and the benzene ring (**18**) resulted in a loss of this activity. The thienyl (**19**), pyridyl (**20**), and cyclohexyl (**21**) analogs each exhibited somewhat weak activity compared to **16e**. In this study, aromatic or aliphatic rings directly attached to the amino group appeared to be tolerated, and the phenyl group was found to be relatively favorable for potent activity.

We then examined the effect of substituents on the phenyl group (Table 3). The 2-methylphenyl analog **16f** had substantially reduced activity, whereas the 4-methylphenyl analog **16g** was nearly as potent as the unsubstituted phenyl analog **16e**. Based on this result, we focused on the effect of 4-substituents. Introduction of halogen atoms resulted in an increase in activity, which was particularly evident in the potent inhibition exerted by the chlorophenyl (**16i**; IC_{50} = 2.0 μM) and bromophenyl (**16a**; IC_{50} = 1.0 μM) derivatives. The (trifluoromethyl)phenyl derivative **16b** also exhibited potent activity, with an IC_{50} value of 2.0 μM, which was 5-fold more potent than the methylphenyl derivative **16g**. Introduction of the methoxy group (**16c**) led to an approximately 3-fold increase in inhibitory activity compared to **16e**, and the trifluoromethoxy group (**16j**) had an effect comparable to that

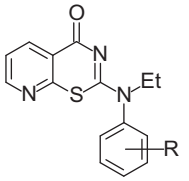
Table 3
Inhibitory activities of 2-[methyl(substituted phenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives against kainate-induced neurotoxicity



Compound	R	Anti-kainate toxicity IC_{50}[a] (μM)
16e	H	(43%)[b]
16f	2-Me	(4%)[b]
16g	4-Me	10
16h	4-F	7.2
16i	4-Cl	2.0
16a	4-Br	1.0
16b	4-CF_3	2.0
16c	4-OMe	3.4
16j	4-OCF_3	2.1
16k	4-NO_2	(38%)[b]
16l	4-NH_2	(19%)[b]
16m	4-COOEt	(29%)[b]
16n	4-COOH	(0%)[b]
16o	4-^{c}Hex	(29%)[b]
16p	3,4-$(CH_2)_3$–	1.3
16q	3,4-CH=CHS–	0.40

[a] Concentration necessary for 50% inhibition.
[b] Percent inhibition at 10 μM concentration.

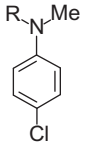
Table 4
Inhibitory activities of 2-[(alkoxyphenyl)(ethyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives against kainate-induced neurotoxicity

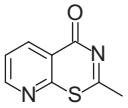
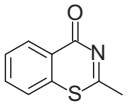
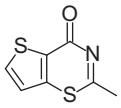
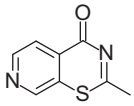
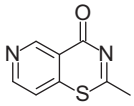
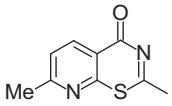
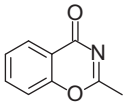


Compound	R	Anti-kainate toxicity IC_{50}[a] (μM)
16r	3-OMe	3.8
16d	4-OMe	2.8
16s	3,4-$(OMe)_2$	(25%)[b]
16t	3,4-OCH_2O–	0.40
16u	3,4-$O(CH_2)_2O$–	0.60

[a] Concentration necessary for 50% inhibition.
[b] Percent inhibition at 10 μM concentration.

Table 5
Inhibitory activities of aryl-fused 2-[(4-chlorophenyl)(methyl)amino]-4*H*-1,3-thiazin-4-one derivatives and benzoxazinone analog **36** against kainate-induced neurotoxicity



Compound	R	Anti-kainate toxicity IC_{50}[a] (μM)
16i		2.0
31		6.5
34		5.4
30		(4%)[b]
24		(2%)[b]
25		(0%)[b]
36		(28%)[b]

[a] Concentration necessary for 50% inhibition.
[b] Percent inhibition at 10 μM concentration.

of the methoxy group (**16c**). Conversely, introduction of the amino group (**16l**) moderately diminished inhibitory activity, and the carboxylphenyl derivative **16n** had markedly decreased potency compared to **16e**, suggesting that hydrophilic substituents are unfavorable in terms of inhibitory activity. A hydrophobic effect rather than an electronic one appears to play a dominant role in inhibitory activity. Interestingly, an 4-amino group on the phenyl ring of 2,3-benzodiazepine derivatives, such as talampanel (**4**), enhanced antagonistic activity,[29] suggesting that the phenyl rings of 4*H*-pyrido[3,2-*e*][1,3]thiazin-4-ones and 2,3-benzodiazepines either occupy the binding pocket in different orientations or bind to distinct sites on the AMPA receptor. In addition, somewhat reduced potency of the ethoxycarbonylphenyl (**16m**) and cyclohexylphenyl (**16o**) derivatives compared to **16e** suggests that the binding space around the 4-position is limited in size. Consistent with the SAR described above, the dihydroindenyl (**16p**) and benzothienyl (**16q**) derivatives bearing hydrophobic and relatively small bicyclic rings exhibited potent inhibition. In particular, **16q** exhibited the most potent inhibitory activity of phenyl and heteroaryl derivatives, with an IC_{50} value of 0.40 μM, which was >25-fold more potent than **16e**.

Given the improved activity of the 4-methoxyphenyl derivative **16c**, we further examined the effect of alkoxy substituents on the phenyl group (Table 4). Reduced hydrophobicity by introduction of lower alkoxy groups might cause a decrease in CNS penetration. The methyl substituent on the 2-amino group was therefore replaced by a more hydrophobic ethyl substituent in this series of compounds. Initially, the ethyl(4-methoxyphenyl)amino derivative **16d** exhibited a closely similar activity relative to corresponding *N*-methyl analog **16c**. Introduction of the 3-methoxy group (**16r**) had an effect comparable to that of the 4-methoxy group (**16d**). As substituents at the 3- and 4-positions each had an independent and favorable effect on potency, the effect of disubstitution was investigated. The activity of the resultant dimethoxyphenyl derivative **16s** unexpectedly decreased. In

Table 6
Anticonvulsant activities of **16h**, **16i**, **16a**, **16j**, and **16t** in MES test in mice

Compound	Anti-kainate toxicity IC_{50}[a] (μM)	ED_{50}[b] (mg/kg, po)
16h	7.2	15.3 (11.7–19.0)
16i	2.0	7.4 (5.9–9.1)
16a	1.0	23.1 (16.2–36.0)
16j	2.1	25.2 (22.1–28.3)
16t	0.40	38.9 (24.4–60.3)
YM90K (**2**)	0.71	>100
Talampanel (**4**)	6.0	4.2 (2.6–7.4)

[a] Concentration necessary for 50% inhibition.
[b] ED_{50} values were calculated by the probit method (95% confidence intervals shown in parentheses).

Table 7
Pharmacokinetic parameters of **16i** in rats

Route	AUC_{inf} (ng·h/mL)	CL_{tot} (mL/h/kg)	Vd_{ss} (mL/kg)	$t_{1/2}$ (h)	C_{max} (ng/mL)	T_{max} (h)	F (%)
iv (1 mg/kg)	542.0	1845.1	1176.0	0.44			
po (1 mg/kg)	472.3			0.58	425.1	0.25	87.1

contrast, the benzodioxolyl derivative **16t** demonstrated strong inhibitory activity, with an IC_{50} value of 0.40 μM, which was equipotent with **16q**. The dihydrobenzodioxinyl derivative **16u** also exhibited relatively potent inhibitory activity nearly equal to that of **16t**. The less sterically-hindered methylenedioxy or ethylenedioxy groups appear favorable for the establishment of potent activity.

We next investigated the effect of the 4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one core (Table 5). Replacement of the pyridine ring of **16i** with a benzene ring (**31**) or thiophene ring (**34**) led to a 3-fold decrease in activity. The 4*H*-pyrido[4,3-*e*][1,3]thiazin-4-one (**30**) and 4*H*-pyrido[3,4-*e*][1,3]thiazin-4-one (**24**) analogs exhibited dramatic reductions in activity relative to **16i**. These results show that the addition of a nitrogen atom at the 6- or 7-position is highly detrimental to inhibitory activity. In addition, the fact that the 7-methyl analog of **16i** (**25**) exhibited no effect at 10 μM suggests that the binding space around this position is extremely limited. Substitution of the sulfur atom of **31** with an oxygen atom (**36**) led to further loss of activity. In this study, the 4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one ring system was the most favorable for the generation of compounds with strong inhibitory activity.

Finally, to investigate the in vivo activity of these 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives, the 4-fluorophenyl (**16h**), 4-chlorophenyl (**16i**), 4-bromophenyl (**16a**), 4-(trifluoromethoxy)phenyl (**16j**), and benzodioxolyl (**16t**) derivatives that exhibited good to excellent potencies regarding kainate-induced neurotoxicity were evaluated for their anticonvulsant activity in MES test in mice (Table 6).[26] YM90K (**2**), one of the representative quinoxalinedione derivatives, exhibited no effect following oral administration. In contrast, all tested 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives exhibited significant anticonvulsant activity following oral administration (ED_{50} = 7.4–38.9 mg/kg). The relatively weak in vivo activity of benzodioxolyl derivative **16t** may be due in part to susceptibility of the methylenedioxy moiety to cytochrome P450 metabolism. Of these compounds, **16i** exhibited the most potent activity with an ED_{50} value of 7.4 mg/kg.[37] As shown in Table 7, **16i** had a favorable pharmacokinetic profile with high oral bioavailability in rats (F = 87%).

4. Conclusions

In our search for novel and orally active AMPA receptor antagonists, we screened our compound library against kainate-induced neurotoxicity and conducted subsequent SAR study for a series of 2-amino-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one derivatives as non-competitive AMPA receptor antagonists. The study revealed that the substitution pattern on the phenyl ring attached to the 2-amino group was crucial for determining the inhibitory activity of the analogs against kainate-induced neurotoxicity and suggested that the hydrophobic interactions and steric hindrances of 4- (and 3-) substituents play dominant roles in inhibitory activity. Potent inhibitory activity was exhibited by compounds bearing chloro (**16i**), bromo (**16a**), trifluoromethyl (**16b**), or trifluoromethoxy (**16j**) substituents on the 4-position of the phenyl group and compounds with a five- or six-membered ring fused at the 3,4-positions of the phenyl group, such as dihydroindenyl (**16p**), benzothienyl (**16q**), benzodioxolyl (**16t**), or dihydrobenzodioxinyl (**16u**) derivatives. In addition, the 4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one ring system plays an important role in potent inhibitory activity. Further, in the MES test in mice, compounds **16h**, **16i**, **16a**, **16j**, and **16t** exhibited significant anticonvulsant activity following oral administration. Of these compounds, **16i** demonstrated the most potent activity with an ED_{50} value of 7.4 mg/kg. Our results indicate that the present compounds, particularly **16i** (YM928), are potential candidates for the treatment of various neurodegenerative diseases based on their AMPA receptor antagonism.

5. Experimental

5.1. Chemistry

Melting points were determined using a Yanaco MP-S3 melting point apparatus and are uncorrected. IR spectra were recorded on a Horiba FT-720 spectrometer as KBr disks. ^{1}H NMR spectra were recorded on a JEOL JNM-EX90, a JEOL JNM-LA300, a JEOL JNM-EX400, a JEOL JNM-A500, or a Bruker Avance III HD 500 spectrometer. ^{13}C NMR spectra were recorded on a Varian VNS-400 or a Bruker Avance III HD 500 spectrometer. Chemical shifts were expressed in δ (ppm) values with tetramethylsilane as an internal standard (NMR descriptions; s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet, and br, broad peak). Mass spectra were recorded on a JEOL JMS-DX300, a JEOL JMS-LX2000, a Hewlett-Packard 5970 MSD, a Fisons TRIO-1000, or a Finnigan MAT TSQ 700 spectrometer. For salts, assignment of ion peaks was based on the basic component. Elemental analyses were conducted using a Yanaco MT-5 microanalyzer (C, H, N) and a Yokogawa IC-7000S ion chromatographic analyzer (halogen and S). Preparative column chromatography was performed using Wakogel C-200 (Wako; 100–200 mesh).

5.1.1. 2-[(4-Bromophenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (13)

A mixture of 2-chloronicotinic acid (**8**, 8.64 g, 54.8 mmol), thionyl chloride (40 ml, 0.55 mol), and 2 drops of *N*,*N*-dimethylformamide (DMF) was heated to reflux for 1.5 h. The reaction mixture was concentrated to dryness, and the residue was dissolved in acetone (30 ml). This solution was added dropwise to a stirred solution of NH_4SCN (4.16 g, 54.7 mmol) in acetone (60 ml). After stirring for 30 min at room temperature, insoluble matter was filtered off, and a solution of 4-bromoaniline (9.41 g, 54.7 mmol) in acetone (35 ml) was added dropwise to the filtrate over a period of 10 min. After stirring for 30 min at room temperature, the reaction mixture was poured into ice water. The resulting precipitate was collected and washed with water to give **10** (19.03 g, 94%). A

mixture of **10** (7.54 g, 20.3 mmol) and toluene (40 ml) was heated to reflux for 4 h. After cooling, the resulting precipitate was collected and washed with toluene to give **13** (6.66 g, 98%) as a beige solid: ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.81–12.42 (m, 1H), 8.72 (br s, 1H), 8.48 (br s, 1H), 7.37–7.96 (m, 4H), 6.93 (br s, 1H); MS (FAB) m/z 334 (M^++1). Anal. Calcd for $C_{13}H_8N_3OSBr$: C, 46.72; H, 2.41; N, 12.57; S, 9.59; Br, 23.91. Found: C, 46.58; H, 2.34; N, 12.58; S, 9.56; Br, 23.60.

Compounds **14** and **15** were prepared in a similar manner.

5.1.2. 2-{[4-(Trifluoromethyl)phenyl]amino}-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (14)

White solid; 80% from **8**; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.61–8.66 (m, 1H), 8.40–8.60 (m, 2H), 7.65 (d, J = 8.5 Hz, 2H), 7.31–7.38 (m, 1H), 7.09 (d, J = 9.0 Hz, 2H); MS (FAB) m/z 324 (M^++1). Anal. Calcd for $C_{14}H_8N_3OSF_3$: C, 52.01; H, 2.49; N, 13.00; S, 9.92; F, 17.63. Found: C, 51.95; H, 2.54; N, 13.02; S, 9.98; F, 18.18.

5.1.3. 2-[(4-Methoxyphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (15)

Pale yellow solid; 81% from **8**; ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.66–11.96 (m, 1H), 8.70 (br s, 1H), 8.47 (d, J = 7.9 Hz, 1H), 7.42–7.80 (m, 2H), 6.90–7.18 (m, 3H), 3.78 (s, 3H); MS (EI) m/z 285 (M^+).

5.1.4. 2-[(4-Bromophenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16a) and 2-[(4-bromophenyl)imino]-3-methyl-2,3-dihydro-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (17a)

To a stirred solution of **13** (900 mg, 2.69 mmol) in DMF (15 ml) was added lithium hydride (24.0 mg, 3.02 mmol), and the mixture was stirred for 30 min at room temperature under an argon atmosphere. To this was added methyl iodide (0.184 ml, 2.96 mmol), and the resulting mixture was stirred for 5 h. Additional lithium hydride (3.0 mg, 0.38 mmol) and methyl iodide (0.022 ml, 0.35 mmol) were added, and the mixture was stirred for 1 h. The mixture was poured into ice water, and the resulting precipitate was collected and washed with water. The crude product thus obtained was chromatographed ($CHCl_3$) to give **16a** (107 mg, 11%) as a white solid and **17a** (803 mg, 86%) as a white solid. Compound **16a**: IR 1641, 1568, 1520, 1483, 1444, 1392, 1308, 1138, 1014, 762 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.56–8.61 (m, 1H), 8.55 (d, J = 7.9 Hz, 1H), 7.50 (d, J = 8.5 Hz, 2H), 7.26–7.33 (m, 1H), 6.79 (d, J = 8.6 Hz, 2H), 3.70 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.78, 164.90, 155.79, 152.79, 140.02, 138.13, 133.82, 129.91, 124.01, 123.42, 119.89, 40.55; MS (FAB) m/z 348 (M^++1). Anal. Calcd for $C_{14}H_{10}N_3OSBr{\cdot}0.25H_2O$: C, 47.67; H, 3.00; N, 11.91; S, 9.09; Br, 22.65. Found: C, 47.82; H, 2.79; N, 11.84; S, 8.93; Br, 22.50. Compound **17a**: IR 1678, 1591, 1574, 1450, 1410, 1346, 1203, 1097, 823, 754 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.59 (dd, J = 4.3, 1.8 Hz, 1H), 8.55 (dd, J = 7.9, 1.8 Hz, 1H), 7.50 (d, J = 8.5 Hz, 2H), 7.23–7.34 (m, 1H), 6.79 (d, J = 8.6 Hz, 2H), 3.70 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 162.22, 154.16, 153.75, 148.34, 146.06, 138.69, 132.67, 122.52, 121.93, 120.43, 117.78, 31.96; MS (EI) m/z 347 (M^+). Anal. Calcd for $C_{14}H_{10}N_3OSBr$: C, 48.29; H, 2.89; N, 12.07; S, 9.21; Br, 22.95. Found: C, 47.95; H, 2.78; N, 12.00; S, 9.31; Br, 23.57.

5.1.5. 2-{Methyl[4-(trifluoromethyl)phenyl]amino}-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16b) and 3-methyl-2-{[4-(trifluoromethyl)phenyl]imino}-2,3-dihydro-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (17b)

The title compounds were prepared from **14** in the same way as **16a** and **17a**. Compound **16b**: White solid; 4%; IR 1647, 1570, 1516, 1394, 1327, 1300, 1180, 1128, 1068, 762 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.68 (dd, J = 7.9, 1.8 Hz, 1H), 8.60 (dd, J = 4.9, 1.8 Hz, 1H), 7.83 (d, J = 8.5 Hz, 2H), 7.52 (d, J = 8.6 Hz, 2H), 7.40 (dd, J = 7.9, 4.9 Hz, 1H), 3.68 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.73, 164.69, 155.57, 152.89, 144.21, 138.20, 132.32, 132.05, 131.79, 131.53, 128.89, 127.75, 127.72, 127.69, 127.66, 126.69, 124.53, 123.56, 122.36, 120.19, 119.92, 40.50; MS (FAB) m/z 338 (M^++1). Anal. Calcd for $C_{15}H_{10}N_3OSF_3$: C, 53.41; H, 2.99; N, 12.46; S, 9.51; F, 16.90. Found: C, 53.60; H, 3.06; N, 12.28; S, 9.29; F, 16.73. Compound **17b**: White solid; 84%; IR 1685, 1595, 1574, 1408, 1350, 1323, 1163, 1124, 1103, 1065, 852, 758 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.59 (dd, J = 4.9, 1.8 Hz, 1H), 8.56 (dd, J = 7.9, 1.8 Hz, 1H), 7.65 (d, J = 8.5 Hz, 2H), 7.31 (dd, J = 7.9, 4.3 Hz, 1H), 7.00 (d, J = 8.6 Hz, 2H), 3.72 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 162.17, 153.93, 153.81, 150.15, 148.49, 138.75, 128.33, 126.93, 126.89, 126.85, 126.81, 126.61, 126.28, 125.63, 122.92, 122.05, 121.02, 120.40, 31.93; MS (EI) m/z 337 (M^+). Anal. Calcd for $C_{15}H_{10}N_3OSF_3{\cdot}0.1H_2O$: C, 53.13; H, 3.03; N, 12.39; S, 9.46; F, 16.81. Found: C, 53.00; H, 2.88; N, 12.39; S, 9.45; F, 17.05.

5.1.6. 2-[(4-Methoxyphenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16c) and 2-[(4-methoxyphenyl)imino]-3-methyl-2,3-dihydro-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (17c)

The title compounds were prepared from **15** in the same way as **16a** and **17a**. Compound **16c**: White solid; 8%; IR 1647, 1572, 1527, 1502, 1446, 1392, 1304, 1250, 1171, 1138, 760 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.67 (dd, J = 7.9, 1.8 Hz, 1H), 8.57 (dd, J = 4.9, 1.8 Hz, 1H), 7.32–7.40 (m, 1H), 7.22–7.30 (m, 2H), 7.03 (d, J = 9.2 Hz, 2H), 3.88 (s, 3H), 3.62 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.95, 165.70, 160.49, 156.26, 152.62, 138.05, 133.55, 129.42, 123.17, 119.90, 115.63, 55.66, 40.75; MS (ESI) m/z 300 (M^++1). Anal. Calcd for $C_{15}H_{13}N_3O_2S{\cdot}0.2H_2O$: C, 59.47; H, 4.46; N, 13.87; S, 10.58. Found: C, 59.67; H, 4.36; N, 13.66; S, 10.51. Compound **17c**: Pale yellow solid; 44%; mp 155–157 °C (from ethyl acetate); IR 1674, 1595, 1574, 1508, 1458, 1408, 1350, 1306, 1246, 1196, 1024, 841, 756 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.50–8.60 (m, 2H), 7.22–7.31 (m, 1H), 6.93 (d, J = 9.2 Hz, 2H), 6.85 (d, J = 7.9 Hz, 2H), 3.83 (s, 3H), 3.71 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 162.34, 156.93, 154.67, 153.60, 148.03, 140.24, 138.57, 121.72, 121.70, 120.52, 114.88, 55.52, 32.05; MS (EI) m/z 299 (M^+). Anal. Calcd for $C_{15}H_{13}N_3O_2S$: C, 60.18; H, 4.38; N, 14.04; S, 10.71. Found: C, 60.29; H, 4.28; N, 14.01; S, 10.70.

5.1.7. 2-[Ethyl(4-methoxyphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16d) and 3-ethyl-2-[(4-methoxyphenyl)imino]-2,3-dihydro-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (17d)

The title compounds were prepared from **15** in the same way as **16a** and **17a** using ethyl iodide instead of methyl iodide. Compound **16d**: White solid; 9%; IR 1647, 1577, 1502, 1439, 1390, 1302, 1248, 1022, 766 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.66 (dd, J = 7.9, 1.8 Hz, 1H), 8.55 (dd, J = 4.9, 1.8 Hz, 1H), 7.35 (dd, J = 7.9, 4.9 Hz, 1H), 7.22 (d, J = 8.5 Hz, 2H), 7.03 (d, J = 9.2 Hz, 2H), 4.12–4.20 (m, 2H), 3.89 (s, 3H), 1.23–1.30 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 170.04, 165.43, 160.59, 156.42, 152.54, 137.99, 131.76, 130.46, 123.10, 120.05, 115.57, 55.66, 47.44, 12.87; MS (EI) m/z 313 (M^+). Anal. Calcd for $C_{16}H_{15}N_3O_2S$: C, 61.32; H, 4.82; N, 13.41; S, 10.23. Found: C, 61.29; H, 4.71; N, 13.40; S, 10.18. Compound **17d**: Slightly yellow solid; 66%; IR 1670, 1599, 1574, 1506, 1448, 1412, 1369, 1323, 1242, 1196, 1099, 756 cm^{-1}; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.56 (dd, J = 4.9, 1.8 Hz, 1H), 8.53 (dd, J = 7.9, 1.8 Hz, 1H), 7.23–7.30 (m, 1H), 6.93 (d, J = 9.2 Hz, 2H), 6.84 (d, J = 9.2 Hz, 2H), 4.40–4.50 (m, 2H), 3.83 (s, 3H), 1.33–1.43 (m, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 161.82, 156.89, 154.71, 153.52, 147.03, 140.34, 138.53, 121.68, 121.66, 120.69, 114.88, 55.53, 40.30, 12.54; MS (EI) m/z 313 (M^+). Anal. Calcd for $C_{16}H_{15}N_3O_2S$: C, 61.32; H, 4.82; N, 13.41; S, 10.23. Found: C, 61.48; H, 4.76; N, 13.47; S, 10.15.

5.1.8. 2-[Benzyl(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (18)

2-Chloronicotinoyl chloride was prepared from 2-chloronicotinic acid (**8**, 1.76 g, 11.2 mmol) and dissolved in acetone (10 ml). This solution was added dropwise to a stirred solution of NH_4SCN (891 mg, 11.7 mmol) in acetone (15 ml) over a period of 5 min. After stirring for 15 min at room temperature, insoluble matter was filtered off, and a solution of *N*-methylbenzylamine (1.42 g, 11.7 mmol) in acetone (15 ml) was added dropwise to the filtrate. After stirring overnight at room temperature, the reaction mixture was poured into ice water. The resulting precipitate was collected, washed with water, and recrystallized from ethyl ether/ethanol to give **18** (1.53 g in two crops, 48%) as a white solid: mp 112–113 °C; ^{1}H NMR (500 MHz, DMSO-d_6, 80 °C) δ 8.71 (dd, J = 4.9, 1.8 Hz, 1H), 8.50 (dd, J = 7.9, 1.8 Hz, 1H), 7.52–7.58 (m, 1H), 7.26–7.41 (m, 5H), 4.98 (s, 2H), 3.24 (s, 3H); ^{13}C NMR (125 MHz, DMSO-d_6, 80 °C) δ 167.49, 163.24, 154.54, 152.84, 137.38, 135.71, 128.68, 127.59, 127.44, 123.90, 120.11, 53.50, 36.57; MS (FAB) m/z 284 (M^++1). Anal. Calcd for $C_{15}H_{13}N_3OS$: C, 63.58; H, 4.62; N, 14.83; S, 11.32. Found: C, 63.62; H, 4.53; N, 14.92; S, 11.48.

Compounds **19**–**21** and **16e**–**k**, **m**, **o**–**u** were prepared in a similar manner.

5.1.9. 2-[Methyl(3-thienyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (19)

White solid; 26% from 2-chloronicotinoyl chloride; mp 231–232 °C (from $CHCl_3$/EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.67 (dd, J = 8.0, 2.0 Hz, 1H), 8.59 (dd, J = 4.8, 2.0 Hz, 1H), 7.48–7.52 (m, 1H), 7.43 (dd, J = 3.2, 1.2 Hz, 1H), 7.38 (dd, J = 8.0, 4.4 Hz, 1H), 7.05 (dd, J = 5.2, 1.2 Hz, 1H), 3.62 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.81, 165.46, 156.11, 152.73, 139.00, 138.12, 127.65, 125.39, 124.02, 123.30, 119.95, 40.45; MS (EI) m/z 275 (M^+). Anal. Calcd for $C_{12}H_9N_3OS_2$: C, 52.34; H, 3.29; N, 15.26; S, 23.29. Found: C, 52.57; H, 3.24; N, 15.53; S, 23.37.

5.1.10. 2-[Methyl(pyridin-3-yl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one hydrochloride (1:1) (20)

The free base of **20** was converted to its hydrochloride salt by crystallization from a mixture of ethanol and 4 M HCl in ethyl acetate: White solid; 59%; ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.99 (s,1H), 8.85 (d, J = 4.8 Hz, 1H), 8.73 (dd, J = 4.0, 1.6 Hz, 1H), 8.53 (dd, J = 8.4, 1.6 Hz, 1H), 8.35 (d, J = 8.4 Hz, 1H), 7.71–8.26 (m, 2H), 7.61 (dd, J = 8.0, 4.8 Hz, 1H), 3.60 (s, 3H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 167.86, 163.95, 154.49, 153.26, 147.10, 146.81, 139.32, 138.92, 137.54, 125.89, 124.20, 119.30; MS (FAB) m/z 271 (M^++1). Anal. Calcd for $C_{13}H_{10}N_4OS{\cdot}HCl{\cdot}0.6H_2O$: C, 49.17; H, 3.87; N, 17.64; S, 10.10; Cl, 11.16. Found: C, 49.11; H, 3.83; N, 17.75; S, 10.06; Cl, 11.01.

5.1.11. 2-[Cyclohexyl(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (21)

Off-white solid; 47%; mp 134–135 °C (from hexane/ethyl ether/ethyl acetate); ^{1}H NMR (400 MHz, DMSO-d_6, 80 °C) δ 8.66–8.72 (m, 1H), 8.47 (dd, J = 7.8, 1.8 Hz, 1H), 7.53 (dd, J = 7.8, 4.6 Hz, 1H), 4.00–5.00 (br, 1H), 3.12 (s, 3H), 1.55–1.89 (m, 7H), 1.33–1.48 (m, 2H), 1.10–1.25 (m, 1H); ^{13}C NMR (125 MHz, DMSO-d_6, 80 °C) δ 167.40, 162.30, 154.69, 152.73, 137.26, 123.78, 120.38, 57.34, 30.96, 29.14, 25.14, 24.71; MS (FAB) m/z 276 (M^++1). Anal. Calcd for $C_{14}H_{17}N_3OS$: C, 61.06; H, 6.22; N, 15.26; S, 11.64. Found: C, 61.02; H, 6.23; N, 15.39; S, 11.79.

5.1.12. 2-[Methyl(phenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16e)

White solid; 79%; mp 174–175 °C (from EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.67 (dd, J = 7.9, 1.8 Hz, 1H), 8.54–8.59 (m, 1H), 7.48–7.59 (m, 3H), 7.31–7.40 (m, 3H), 3.66 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.90, 165.14, 156.08, 152.66, 141.03, 138.08, 130.53, 129.89, 128.17, 123.25, 119.94, 40.65; MS (EI) m/z 269 (M^+). Anal. Calcd for $C_{14}H_{11}N_3OS$: C, 62.43; H, 4.12; N, 15.60; S, 11.91. Found: C, 62.44; H, 4.14; N, 15.61; S, 11.96.

5.1.13. 2-[Methyl(2-methylphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16f)

Slightly yellow solid; 79%; mp 208–209 °C (from EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.68 (dd, J = 7.9, 1.8 Hz, 1H), 8.56 (dd, J = 4.9, 1.8 Hz, 1H), 7.33–7.47 (m, 4H), 7.25 (d, J = 7.9 Hz, 1H), 3.59 (s, 3H), 2.27 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.94, 165.22, 156.31, 152.62, 139.40, 138.16, 136.78, 132.20, 130.38, 128.74, 128.23, 123.23, 120.03, 39.29, 17.24; MS (FAB) m/z 284 (M^++1). Anal. Calcd for $C_{15}H_{13}N_3OS$: C, 63.58; H, 4.62; N, 14.83; S, 11.32. Found: C, 63.62; H, 4.59; N, 14.78; S, 11.34.

5.1.14. 2-[Methyl(4-methylphenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16g)

White solid; 33% from 2-chloronicotinoyl chloride; mp 200–201 °C (from EtOH); ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.67 (dd, J = 7.9, 1.8 Hz, 1H), 8.56 (dd, J = 4.7, 1.8 Hz, 1H), 7.30–7.39 (m, 3H), 7.22 (d, J = 8.3 Hz, 2H), 3.63 (s, 3H), 2.45 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.94, 165.33, 156.21, 152.62, 140.20, 138.41, 138.04, 131.10, 127.85, 123.17, 119.93, 40.66, 21.31; MS (FAB) m/z 284 (M^++1). Anal. Calcd for $C_{15}H_{13}N_3OS$: C, 63.58; H, 4.62; N, 14.83; S, 11.32. Found: C, 63.65; H, 4.76; N, 14.84; S, 11.30.

5.1.15. 2-[(4-Fluorophenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16h)

White solid; 83%; mp 184–185 °C (from EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.64–8.71 (m, 1H), 8.58 (dd, J = 4.8, 2.0 Hz, 1H), 7.32–7.42 (m, 3H), 7.20–7.28 (m, 2H), 3.64 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.82, 165.27, 164.00, 162.00, 155.88, 152.75, 138.14, 136.97, 130.34, 130.27, 123.38, 119.88, 117.72, 117.54, 40.72; MS (FAB) m/z 288 (M^++1). Anal. Calcd for $C_{14}H_{10}N_3OSF$: C, 58.53; H, 3.51; N, 14.63; S, 11.16; F, 6.61. Found: C, 58.42; H, 3.35; N, 14.61; S, 11.20; F, 6.80.

5.1.16. 2-[(4-Chlorophenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16i)

White solid; 87%; mp 189–190 °C (from EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.67 (dd, J = 7.9, 1.2 Hz, 1H), 8.59 (dd, J = 4.9, 1.8 Hz, 1H), 7.52 (d, J = 8.6 Hz, 2H), 7.39 (dd, J = 7.9, 4.9 Hz, 1H), 7.31 (d, J = 8.6 Hz, 2H), 3.63 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.79, 165.00, 155.81, 152.78, 139.49, 138.14, 135.97, 130.81, 129.64, 123.41, 119.89, 40.60; MS (FAB) m/z 304 (M^++1). Anal. Calcd for $C_{14}H_{10}N_3OSCl$: C, 55.36; H, 3.32; N, 13.83; S, 10.56; Cl, 11.67. Found: C, 55.43; H, 3.30; N, 13.84; S, 10.55; Cl, 11.69.

5.1.17. 2-{Methyl[4-(trifluoromethoxy)phenyl]amino}-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16j)

Beige solid; 81%; mp 176–177 °C (from EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.68 (dd, J = 7.9, 1.8 Hz, 1H), 8.59 (dd, J = 4.9, 1.8 Hz, 1H), 7.36–7.44 (m, 5H), 3.65 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.77, 165.01, 155.73, 152.81, 149.84, 139.35, 138.20, 130.07, 123.48, 123.43, 122.82, 121.37, 119.90, 119.31, 117.26, 40.62; MS (FAB) m/z 354 (M^++1). Anal. Calcd for $C_{15}H_{10}N_3O_2SF_3$: C, 50.99; H, 2.85; N, 11.89; S, 9.08; F, 16.13. Found: C, 50.91; H, 2.89; N, 11.98; S, 9.15; F, 16.35.

5.1.18. 2-[Methyl(4-nitrophenyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16k)

Yellow solid; 81%; mp 223–224 °C (from EtOH); ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.69–8.74 (m, 1H), 8.53 (d, J = 7.9 Hz, 1H), 8.43 (d, J = 8.6 Hz, 2H), 7.89 (d, J = 8.5 Hz, 2H), 7.59 (dd, J = 7.9,

4.9 Hz, 1H), 3.58 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 167.98, 163.23, 154.64, 153.28, 147.27, 146.85, 137.55, 129.71, 125.36, 124.19, 119.41; MS (FAB) m/z 315 (M^++1). Anal. Calcd for $C_{14}H_{10}N_4O_3S$: C, 53.50; H, 3.21; N, 17.82; S, 10.20. Found: C, 53.28; H, 3.19; N, 17.84; S, 10.21.

5.1.19. Ethyl 4-[methyl(4-oxo-4*H*-pyrido[3,2-*e*][1,3]thiazin-2-yl) amino]benzoate (16m)

White solid; 88% from 2-chloronicotinoyl chloride; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.67 (dd, J = 7.9, 1.8 Hz, 1H), 8.58 (dd, J = 4.9, 1.8 Hz, 1H), 8.22 (d, J = 8.6 Hz, 2H), 7.45 (d, J = 8.6 Hz, 2H), 7.39 (dd, J = 7.9, 4.9 Hz, 1H), 4.40–4.48 (m, 2H), 3.67 (s, 3H), 1.40–1.47 (m, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.77, 165.31, 164.66, 155.75, 152.81, 144.86, 138.14, 131.78, 131.75, 128.20, 123.44, 119.95, 61.57, 40.55, 14.32; MS (FAB) m/z 342 (M^++1). Anal. Calcd for $C_{17}H_{15}N_3O_3S{\cdot}0.2H_2O$: C, 59.19; H, 4.50; N, 12.18; S, 9.29. Found: C, 59.27; H, 4.33; N, 12.15; S, 9.39.

5.1.20. 2-[(4-Cyclohexylphenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*] [1,3]thiazin-4-one (16o)

White solid; 48% from 2-chloronicotinoyl chloride; mp 192–195 °C (from EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.64–8.70 (m, 1H), 8.56 (dd, J = 4.4, 2.0 Hz, 1H), 7.33–7.39 (m, 3H), 7.24 (d, J = 8.4 Hz, 2H), 3.64 (s, 3H), 2.53–2.65 (m, 1H), 1.74–1.98 (m, 5H), 1.36–1.51 (m, 4H), 1.22–1.35 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.93, 165.34, 156.26, 152.59, 150.21, 138.53, 138.08, 128.86, 127.87, 123.16, 119.96, 44.38, 40.60, 34.37, 26.78, 26.04; MS (FAB) m/z 352 (M^++1). Anal. Calcd for $C_{20}H_{21}N_3OS$: C, 68.35; H, 6.02; N, 11.96; S, 9.12. Found: C, 68.43; H, 6.08; N, 11.89; S, 9.21.

5.1.21. 4-Cyclohexyl-*N*-methylaniline

The title compound was prepared from 4-cyclohexylaniline using the procedure described by Crochet et al.;[41] 64%; ^{1}H NMR (90 MHz, $CDCl_3$) δ 7.05 (d, J = 8.4 Hz, 2H), 6.57 (d, J = 8.6 Hz, 2H), 2.82 (s, 3H), 2.20–2.63 (m, 1H), 0.90–2.10 (m, 10H); MS (EI) m/z 189 (M^+).

5.1.22. 2-[2,3-Dihydro-1*H*-inden-5-yl(methyl)amino]-4*H*-pyrido [3,2-*e*][1,3]thiazin-4-one (16p)

Pale yellow solid; 57% from 2-chloronicotinoyl chloride; mp 157–160 °C (from EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.67 (dd, J = 8.0, 2.0 Hz, 1H), 8.56 (dd, J = 4.8, 2.0 Hz, 1H), 7.33–7.38 (m, 2H), 7.16 (s, 1H), 7.05–7.10 (m, 1H), 3.64 (s, 3H), 2.94–3.02 (m, 4H), 2.17 (quintet, J = 7.6 Hz, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.96, 165.37, 156.34, 152.59, 146.95, 146.43, 139.07, 138.04, 126.06, 125.74, 123.90, 123.12, 119.95, 40.72, 32.90, 32.71, 25.63; MS (FAB) m/z 310 (M^++1). Anal. Calcd for $C_{17}H_{15}N_3OS$: C, 66.00; H, 4.89; N, 13.58; S, 10.36. Found: C, 65.93; H, 4.97; N, 13.50; S, 10.38.

5.1.23. *N*-Methylindan-5-amine

The title compound was prepared from indan-5-amine using the procedure described by Crochet et al.;[41] 56%; ^{1}H NMR (90 MHz, $CDCl_3$) δ 7.04 (d, J = 8.0 Hz, 1H), 6.34–6.59 (m, 2H), 2.63–2.97 (m, 7H), 1.83–2.22 (m, 2H); MS (EI) m/z 147 (M^+).

5.1.24. 2-[1-Benzothiophen-5-yl(methyl)amino]-4*H*-pyrido [3,2-*e*][1,3]thiazin-4-one (16q)

5-Nitro-1-benzothiophene (1.70 g, 9.49 mmol) was hydrogenated in ethanol (34 ml) under 3 atm of H_2 in the presence of 5% Pd/C (0.17 g). After removal of the catalyst by filtration, the filtrate was concentrated in vacuo to give an amino derivative. This compound was methylated using the procedure described by Crochet et al.[41] The crude product thus obtained was treated with 2-chloronicotinoyl isothiocyanate (**9**) prepared from 2-chloronicotinoyl chloride (1.67 g, 9.49 mmol) to give **16q** (546 mg, 18% from 2-chloronicotinoyl chloride) as a white solid by the method described for **18**: mp 214–216 °C (from ethyl acetate/EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.68 (dd, J = 7.9, 1.8 Hz, 1H), 8.53–8.58 (m, 1H), 8.04 (d, J = 8.5 Hz, 1H), 7.82 (d, J = 2.0 Hz, 1H), 7.63 (d, J = 5.4 Hz, 1H), 7.41 (d, J = 5.5 Hz, 1H), 7.34–7.40 (m, 1H), 7.28 (dd, J = 8.5, 2.1 Hz, 1H), 3.72 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.94, 165.54, 156.14, 152.66, 140.93, 140.83, 138.06, 137.53, 129.15, 124.65, 123.88, 123.42, 123.35, 123.22, 119.88, 40.94; MS (FAB) m/z 326 (M^++1). Anal. Calcd for $C_{16}H_{11}N_3OS_2$: C, 59.06; H, 3.41; N, 12.91; S, 19.71. Found: C, 59.03; H, 3.23; N, 12.79; S, 19.48.

5.1.25. 2-[Ethyl(3-methoxyphenyl)amino]-4*H*-pyrido[3,2-*e*] [1,3]thiazin-4-one (16r)

White solid; 83%; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.66 (dd, J = 7.9, 1.8 Hz, 1H), 8.53–8.58 (m, 1H), 7.42–7.48 (m, 1H), 7.32–7.39 (m, 1H), 7.03–7.09 (m, 1H), 6.91 (d, J = 8.5 Hz, 1H), 6.81–6.86 (m, 1H), 4.13–4.25 (m, 2H), 3.86 (s, 3H), 1.29 (t, J = 7.3 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.98, 164.75, 161.10, 156.35, 152.57, 140.33, 138.01, 131.14, 123.14, 121.14, 120.10, 115.59, 114.74, 55.61, 47.43, 12.95; MS (FAB) m/z 314 (M^++1). Anal. Calcd for $C_{16}H_{15}N_3O_2S$: C, 61.32; H, 4.82; N, 13.41; S, 10.23. Found: C, 61.39; H, 4.79; N, 13.42; S, 10.40.

5.1.26. 2-[(3,4-Dimethoxyphenyl)(ethyl)amino]-4*H*-pyrido[3,2-*e*] [1,3]thiazin-4-one (16s)

White solid; 90%; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.66 (dd, J = 7.9, 1.8 Hz, 1H), 8.53–8.59 (m, 1H), 7.32–7.39 (m, 1H), 6.98 (d, J = 8.5 Hz, 1H), 6.90 (dd, J = 8.6, 1.8 Hz, 1H), 6.78 (d, J = 1.8 Hz, 1H), 4.02–4.33 (br, 2H), 3.96 (s, 3H), 3.91 (s, 3H), 1.23–1.34 (m, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.03, 165.38, 156.46, 152.58, 150.33, 138.00, 131.83, 123.12, 121.81, 120.05, 111.77, 56.24, 56.19, 47.47, 12.97; MS (FAB) m/z 344 (M^++1). Anal. Calcd for $C_{17}H_{17}N_3O_3S$: C, 59.46; H, 4.99; N, 12.24; S, 9.34. Found: C, 59.27; H, 4.93; N, 12.15; S, 9.35.

5.1.27. 2-[1,3-Benzodioxol-5-yl(ethyl)amino]-4*H*-pyrido[3,2-*e*] [1,3]thiazin-4-one (16t)

White solid; 65%; mp 184–185 °C (from ethyl ether/EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.66 (dd, J = 7.9, 1.8 Hz, 1H), 8.52–8.59 (m, 1H), 7.36 (dd, J = 7.9, 4.3 Hz, 1H), 6.92 (d, J = 7.9 Hz, 1H), 6.80 (dd, J = 7.9, 1.8 Hz, 1H), 6.75 (d, J = 2.4 Hz, 1H), 6.10 (s, 2H), 4.03–4.26 (br, 2H), 1.27 (t, J = 7.3 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 169.99, 165.34, 156.37, 152.59, 149.05, 148.92, 138.02, 132.75, 123.23, 123.16, 120.07, 109.61, 109.18, 102.25, 47.45, 12.86; MS (EI) m/z 327 (M^+). Anal. Calcd for $C_{16}H_{13}N_3O_3S$: C, 58.70; H, 4.00; N, 12.84; S, 9.80. Found: C, 58.62; H, 4.02; N, 12.80; S, 9.85.

5.1.28. 2-[2,3-Dihydro-1,4-benzodioxin-6-yl(ethyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16u)

Pale yellow solid; 73%; mp 192–193 °C (from EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.65 (dd, J = 7.9, 1.8 Hz, 1H), 8.56 (dd, J = 4.9, 1.8 Hz, 1H), 7.35 (dd, J = 7.9, 4.9 Hz, 1H), 7.00 (d, J = 8.5 Hz, 1H), 6.82 (d, J = 2.4 Hz, 1H), 6.78 (dd, J = 8.5, 2.4 Hz, 1H), 4.21–4.40 (m, 4H), 4.14 (q, J = 7.0 Hz, 2H), 1.27 (t, J = 7.0 Hz, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.03, 165.30, 156.47, 152.54, 144.99, 144.54, 138.00, 132.22, 123.09, 122.15, 120.07, 118.69, 118.11, 64.42, 64.31, 47.41, 12.89; MS (FAB) m/z 342 (M^++1). Anal. Calcd for $C_{17}H_{15}N_3O_3S{\cdot}0.25H_2O$: C, 59.03; H, 4.52; N, 12.15; S, 9.27. Found: C, 58.95; H, 4.34; N, 12.08; S, 9.48.

5.1.29. *N*-Ethyl-2,3-dihydro-1,4-benzodioxin-6-amine

A stirred solution of 2,3-dihydro-1,4-benzodioxin-6-amine (3.01 g, 19.9 mmol) in CH_2Cl_2 (20 ml) was cooled in an ice-water bath and treated dropwise with trifluoroacetic anhydride (4.10 ml, 29.4 mmol) over a period of 5 min. After stirring for

30 min at room temperature, the reaction mixture was concentrated to dryness, and ethyl acetate was added to the residue. The organic phase was washed with water and brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The crude product obtained was dissolved in DMF (35 ml) and cooled in an ice-water bath. To the solution was added lithium hydride (190 mg, 23.9 mmol), and the mixture was stirred for 15 min under an argon atmosphere. To this was added ethyl iodide (1.93 ml, 23.9 mmol), and the resulting mixture was stirred overnight at room temperature. After concentration, the residue was dissolved in ethyl acetate, washed with water and brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The crude product obtained was dissolved in MeOH (50 ml) and treated with 28% NH_3 (30 ml), and the mixture was heated to reflux for 4 h. After concentration, the residue was dissolved in ethyl acetate, washed with water and brine, dried over anhydrous Na_2SO_4, and concentrated in vacuo. The residue was chromatographed (hexane/ethyl acetate 6:1–5:1) to give the title compound (3.33 g, 93%): ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.68 (d, J = 8.6 Hz, 1H), 6.07–6.18 (m, 2H), 4.08–4.25 (m, 4H), 3.10–3.33 (br, 1H), 2.98–3.10 (m, 2H), 1.13–1.24 (m, 3H); MS (EI) m/z 179 (M^+).

5.1.30. 2-[(4-Aminophenyl)(methyl)amino]-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (16l)

The nitro compound **16k** (1.51 g, 4.80 mmol) was hydrogenated in DMF (50 ml) under atmospheric pressure in the presence of Raney-nickel (1.5 g). The reaction mixture was filtered and concentrated in vacuo, and the residue was recrystallized from ethanol to give **16l** (984 mg in two crops, 72%) as a slightly yellow solid: mp 249–251 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.62 (dd, J = 8.0, 2.0 Hz, 1H), 8.54–8.59 (m, 1H), 7.37 (dd, J = 8.0, 4.4 Hz, 1H), 7.06 (d, J = 8.4 Hz, 2H), 6.77 (d, J = 8.8 Hz, 2H), 4.44 (s, 2H), 3.58 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 170.04, 165.97, 156.51, 152.56, 147.86, 138.01, 131.20, 129.15, 123.06, 119.92, 115.94, 40.78; MS (FAB) m/z 285 (M^++1). Anal. Calcd for $C_{14}H_{12}N_4OS$: C, 59.14; H, 4.25; N, 19.70; S, 11.28. Found: C, 58.98, H, 4.32; N, 19.42; S, 11.51.

5.1.31. 4-[Methyl(4-oxo-4*H*-pyrido[3,2-*e*][1,3]thiazin-2-yl)amino]benzoic acid (16n)

To a stirred solution of the ester **16m** (70 mg, 0.20 mmol) in ethanol (0.90 ml) was added 0.25 M NaOH (0.90 ml, 0.23 mmol). After stirring for 5 h at room temperature, the reaction mixture was diluted with water and acidified with 0.5 M HCl. The resulting precipitate was collected and washed successively with water and ethyl ether to give **16n** (61 mg, 91%) as a pale yellow solid: ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.65 (dd, J = 7.6, 2.0 Hz, 1H), 8.60 (dd, J = 5.2, 2.0 Hz, 1H), 8.23 (d, J = 8.4 Hz, 2H), 7.45 (d, J = 8.4 Hz, 2H), 7.38–7.44 (m, 1H), 3.67 (s, 3H); ^{13}C NMR (125 MHz, DMSO-d_6) δ 167.99, 166.35, 163.15, 154.76, 153.11, 144.66, 137.43, 131.57, 131.10, 128.47, 123.97, 119.30; MS (APCI) m/z 314 (M^++1). Anal. Calcd for $C_{15}H_{11}N_3O_3S{\cdot}0.9H_2O$: C, 54.67; H, 3.91; N, 12.75; S, 9.73. Found: C, 54.77; H, 3.75; N, 12.67; S, 9.65.

5.1.32. 2-[(4-Chlorophenyl)(methyl)amino]-4*H*-pyrido[3,4-*e*][1,3]thiazin-4-one (24)

The title compound was prepared in the same manner as described for **18** using 4-chloronicotinic acid (**22**) and 4-chloro-*N*-methylaniline instead of 2-chloronicotinic acid (**8**) and *N*-methylbenzylamine, respectively: Off-white solid; 26%; mp 238–239 °C (from EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.51 (s, 1H), 8.56 (d, J = 5.6 Hz, 1H), 7.55 (d, J = 8.8 Hz, 2H), 7.31 (d, J = 8.4 Hz, 2H), 7.07 (d, J = 5.6 Hz, 1H), 3.61 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 167.93, 162.33, 152.13, 150.92, 143.40, 139.30, 136.20, 130.92, 129.70, 119.58, 116.99, 40.59; MS (FAB) m/z 304 (M^++1). Anal. Calcd for $C_{14}H_{10}N_3OSCl$: C, 55.36; H, 3.32; N, 13.83; S, 10.56; Cl, 11.67. Found: C, 55.15; H, 3.29; N, 13.79; S, 10.70; Cl, 11.58.

5.1.33. 2-[(4-Chlorophenyl)(methyl)amino]-7-methyl-4*H*-pyrido[3,2-*e*][1,3]thiazin-4-one (25)

The title compound was prepared in the same manner as described for **18** using 2-chloro-6-methylnicotinic acid (**23**) and 4-chloro-*N*-methylaniline instead of 2-chloronicotinic acid (**8**) and *N*-methylbenzylamine, respectively: Pale brown solid; 39%; mp 194–196 °C (from EtOH); ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.53 (d, J = 7.9 Hz, 1H), 7.51 (d, J = 8.5 Hz, 2H), 7.30 (d, J = 8.5 Hz, 2H), 7.22 (d, J = 7.9 Hz, 1H), 3.62 (s, 3H), 2.54 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.99, 164.93, 163.01, 155.24, 139.58, 138.22, 135.88, 130.73, 129.73, 123.41, 117.36, 40.43, 24.48; MS (FAB) m/z 318 (M^++1). Anal. Calcd for $C_{15}H_{12}N_3OSCl$: C, 56.69; H, 3.81; N, 13.22; S, 10.09; Cl, 11.16. Found: C, 56.57; H, 3.78; N, 13.08; S, 10.17; Cl, 11.03.

5.1.34. *N*-[(4-Chlorophenyl)(methyl)carbamothioyl]-3-fluoroisonicotinamide (28)

A mixture of 3-fluoroisonicotinic acid (**26**, 1.66 g, 11.8 mmol), thionyl chloride (15 ml, 0.21 mol), and 2 drops of DMF was heated to reflux for 2 h. The reaction mixture was concentrated to dryness, and the residue was dissolved in acetone (10 ml). This solution was added dropwise to a stirred solution of NH_4SCN (984 mg, 12.9 mmol) in acetone (15 ml). After stirring for 5 min at 40 °C, insoluble matter was filtered off, and a solution of 4-chloro-*N*-methylaniline (1.55 g, 10.9 mmol) in acetone (10 ml) was added dropwise to the filtrate over a period of 5 min. After stirring overnight at room temperature, the reaction mixture was poured into ice water and the resulting precipitate was collected. The crude solid was chromatographed ($CHCl_3$/MeOH 99:1–98:2) to give **28** (1.45 g, 41%): ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.63–8.85 (br, 1H), 8.43–8.61 (m, 2H), 7.52–7.75 (br, 1H), 7.39 (d, J = 8.8 Hz, 2H), 7.26 (d, J = 8.4 Hz, 2H), 3.72 (s, 3H); MS (FAB) m/z 324 (M^++1).

5.1.35. *N*-[(4-Chlorophenyl)(methyl)carbamothioyl]-2-fluorobenzamide (29)

The title compound was prepared in the same manner as described for **28** using 2-fluorobenzoic acid (**27**) instead of 3-fluoroisonicotinic acid (**26**): 92%; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.63–8.87 (m, 1H), 7.72–7.93 (br, 1H), 7.40–7.57 (m, 1H), 7.36 (d, J = 8.5 Hz, 2H), 7.15–7.34 (m, 3H), 6.99–7.15 (m, 1H), 3.73 (s, 3H); MS (FAB) m/z 323 (M^++1).

5.1.36. 3-Chloro-*N*-[(4-chlorophenyl)(methyl)carbamothioyl]thiophene-2-carboxamide (33)

The title compound was prepared in the same manner as described for **28** using 3-chlorothiophene-2-carboxylic acid (**32**) instead of 3-fluoroisonicotinic acid (**26**): 73%; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.99 (br s, 1H), 7.51 (d, J = 5.2 Hz, 1H), 7.38 (d, J = 8.8 Hz, 2H), 7.27 (d, J = 8.8 Hz, 2H), 6.93 (d, J = 5.2 Hz, 1H), 3.71 (s, 3H); MS (FAB) m/z 345 (M^++1).

5.1.37. 2-[(4-Chlorophenyl)(methyl)amino]-4*H*-pyrido[4,3-*e*][1,3]thiazin-4-one (30)

To a stirred solution of **28** (1.23 g, 3.80 mmol) in DMF (10 ml) was added a mixture of 60% sodium hydride in mineral oil (167 mg, 4.18 mmol) and tetrahydrofuran (THF) (1 ml), and the mixture was stirred for 10 min at room temperature under an argon atmosphere. The mixture was then heated to 110 °C and stirred for 5 h. After cooling, the reaction mixture was concentrated to dryness. Ice water was added to the residue, and the resulting precipitate was collected and washed with water. The crude product was recrystallized from ethanol to give **30** (802 mg in two crops,

69%) as a white solid: mp 255–256 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.83 (s, 1H), 8.66 (d, J = 4.8 Hz, 1H), 8.02 (d, J = 4.8 Hz, 1H), 7.70 (d, J = 8.8 Hz, 2H), 7.63 (d, J = 8.8 Hz, 2H), 3.51 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 166.53, 163.19, 148.90, 148.08, 140.02, 134.55, 130.67, 130.59, 129.49, 127.54, 121.77; MS (FAB) m/z 304 (M^++1). Anal. Calcd for $C_{14}H_{10}N_3OSCl$: C, 55.36; H, 3.32; N, 13.83; S, 10.56; Cl, 11.67. Found: C, 55.31; H, 3.34; N, 14.06; S, 10.54; Cl, 11.64.

Compounds **31** and **34** were prepared in a similar manner.

5.1.38. 2-[(4-Chlorophenyl)(methyl)amino]-4*H*-1,3-benzothiazin-4-one (31)

Beige solid; 69%; mp 216–218 °C (from EtOH); ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.23 (d, J = 7.9 Hz, 1H), 7.42–7.72 (m, 7H), 3.50 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 168.14, 163.44, 140.60, 134.52, 133.53, 132.88, 130.94, 130.82, 129.77, 128.61, 126.60, 122.15; MS (FAB) m/z 303 (M^++1). Anal. Calcd for $C_{15}H_{11}N_2OSCl{\cdot}0.2H_2O$: C, 58.80; H, 3.75; N, 9.14; S, 10.47; Cl, 11.57. Found: C, 58.77; H, 3.58; N, 9.14; S, 10.43; Cl, 11.52.

5.1.39. 2-[(4-Chlorophenyl)(methyl)amino]-4*H*-thieno[2,3-*e*][1,3]thiazin-4-one (34)

Brown solid; 57%; mp 246–247 °C dec (from EtOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.70 (d, J = 5.6 Hz, 1H), 7.53 (d, J = 8.8 Hz, 2H), 7.30 (d, J = 8.4 Hz, 2H), 6.94 (d, J = 5.2 Hz, 1H), 3.61 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 165.42, 164.85, 140.18, 135.80, 134.16, 132.82, 130.85, 129.80, 125.92, 124.39, 41.04; MS (FAB) m/z 309 (M^++1). Anal. Calcd for $C_{13}H_9N_2OS_2Cl$: C, 50.56; H, 2.94; N, 9.07; S, 20.77; Cl, 11.48. Found: C, 50.44; H, 2.84; N, 9.00; S, 20.65; Cl, 11.24.

5.1.40. 2-[(4-Chlorophenyl)(methyl)amino]-4*H*-1,3-benzoxazin-4-one (36)

Triethylamine (1.40 ml, 10.1 mmol) was added dropwise to an ice-cooled mixture of methyl salicylate (**35**, 1.52 g, 9.99 mmol), cyanogen bromide (1.06 g, 10.0 mmol), and acetone (5 ml) over a period of 3 min. After stirring for 10 min, the mixture was warmed to room temperature and stirred for 5 min. Insoluble matter was filtered off, and a solution of 4-chloro-*N*-methylaniline (1.41 g, 9.96 mmol) in acetone (5 ml) was added dropwise to the filtrate over a period of 3 min. To this mixture was added benzoic acid (1.22 g, 9.99 mmol), and the whole was stirred overnight at room temperature. The mixture was poured into ice water, and the resulting precipitate was collected and washed with water. The crude product was washed with hot ethyl acetate/hexane to give **36** (2.03 g, 71%) as a white solid: ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.10–8.17 (m, 1H), 7.51–7.59 (m, 1H), 7.45 (d, J = 8.4 Hz, 2H), 7.35 (t, J = 7.6 Hz, 1H), 7.29 (d, J = 8.8 Hz, 2H), 6.92–7.12 (br, 1H), 3.57 (s, 3H); ^{13}C NMR (125 MHz, $CDCl_3$) δ 167.00, 157.16, 153.70, 139.96, 134.07, 133.59, 129.76, 127.85, 127.77, 125.80, 117.34, 115.60, 39.49; MS (FAB) m/z 287 (M^++1). Anal. Calcd for $C_{15}H_{11}N_2O_2Cl$: C, 62.84; H, 3.87; N, 9.77; Cl, 12.37. Found: C, 62.70; H, 3.78; N, 9.71; Cl, 12.41.

5.2. Pharmacology

5.2.1. Inhibition of kainate-induced neurotoxicity in primary rat hippocampal cultures[35]

Hippocampal cell cultures prepared from embryonic day 18–20 Wistar rats were used after 8 or 9 days' culture in vitro and treated simultaneously with test compounds and 300 μM kainate. Overall neuronal cell injury was quantitatively assessed by measuring lactate dehydrogenase (LDH) release from damaged or destroyed cells into the extracellular fluid 24 h after kainate exposure. LDH activity was measured using an LDH assay kit in a 7250 Automatic Analyzer (Hitachi). A single experiment was performed in triplicate for each compound.

5.2.2. Maximal electroshock seizure (MES) in ICR mice[26]

A group of 10 male ICR mice was used. Sixty minutes after test compounds were orally administered, electroshock stimulation (50 mA, 0.2 s, 50 Hz) was applied to each mouse via the cornea. Inhibition of the maximal electroshock seizure action was evaluated by the presence or absence of tonic extension development as an index, and the ED_{50} value was calculated by the probit method.

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