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Synthesis, Reactions, and Pharmacological Screening of Heterocyclic Derivatives Using Nicotinic Acid as a Natural Synthon

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Summary. A series of substituted pyridine derivatives were prepared from 3-acetylpyridine, which was prepared from the corresponding nicotinic acid as a natural starting material. Reaction of 3-acetylpyridine with indole-3-carboxaldehyde afforded the corresponding 3-β-(3-indolyl)-acryloylpyridine, which was reacted with hydroxylamine hydrochloride in pyridine or acetic acid in the presence of sodium acetate to afford 3-β-(3-indolyl)acryloylpyridine oxime and oxazole derivatives. The oxime was treated with ethyl isothiocyanate or toluene-3,5-diisocyanate in refluxing dioxane to give the corresponding ethyl thiosemicarbazide and 3,5-bissemicarbazide derivative. 3-β-(3-Indolyl)acryloylpyridine was condensed with malononitrile in refluxing ethanol in the presence of piperidine as a catalyst to give cyanoaminopyrane, while it was condensed with ethyl cyanoacetate or malononitrile in the presence of ammonium acetate to yield cyanopyridone and cyanoaminopyridine derivatives. Cyclization of acryloylpyridine with *o*-phenylenediamine in refluxing butanol led to the formation of the corresponding benzodiazipine *via* the intermediate **A**. Finally, cycloaddition reaction of acryloylpyridine with thiourea yielded thioxopyrimidine, which was treated with chloroacetic acid to yield thiazolopyrimidine. An arylmethylene derivative was prepared by reacting thiazolopyrimidine with indole-3-carboxaldehyde or by reacting thioxopyrimidine with indole-3-carboxaldehyde and chloroacetic acid in one step. The pharmacological screening showed that many of these obtained compounds have good analgesic and anticonvulsant activities comparable to Valdecoxib® and Carbamazepine® as reference drugs.

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Keywords. Nicotinic acid; Indole-3-carboxaldehyde; Thioxopyrimidine; Analgesics; Anticonvulsants.

Introduction

In previous work we reported that certain substituted pyridines and their chiral macrocyclic derivatives have antidepressant, antimicrobial, anticancer, analgesic, and anticonvulsant activities [1–7]. Nicotinic acid and its derivatives constitute an important class of naturally occurring compounds. Minute amounts of nicotinic acid occur in all living cells, liver, yeast, and corn [8] and it exhibits a wide spectrum of biological activities [9, 10]. It has been reported that substituted indole derivatives are anti-inflammatory [11, 12] and anticancer agents [13–15]. Additionally, some indole derivatives are used as antimicrobial agents, in particular, their 3-substituted derivatives [16–18]. Recently, some new substituted pyridine, pyrane, and pyrimidine derivatives have been synthesized, which exhibit analgetic, anti-inflammatory, antiparkinsonian, and androgenic-anabolic activities [19, 20]. On the other hand, semicarbazide, thiosemicarbazide, and macrocyclic pyridine derivatives show promising biological activities [21–23]. In view of these observations and as continuation of our previous work on pyridine chemistry, we synthesized some new compounds containing pyridine and indole moieties, and tested their selected pharmacological activities.

Results and Discussion

Synthesis

Nicotinic acid (pyridine-3-carboxylic acid) is a naturally occurring carboxylic acid that is found mainly in bacterial endospores [24]. In our previous work we have reported the synthesis and a preliminary biological activity screening of several pyridine derivatives based on 3-β-indolylacryloylpyridine (**4**), which was prepared as starting material from the corresponding nicotinic acid (**1**) *via* its ethyl ester (**2**) and the corresponding 3-acetylpyridine (**3**) according to literature methods [21] (Scheme 1). Condensation of **4** with hydroxylamine hydrochloride in pyridine afforded the corresponding 3-β-(3-indolyl)-acryloylpyridine oxime (**5**) (Scheme 1). The latter was cyclized with refluxing acetic anhydride to the oxazole **6**, which was also prepared directly from **4** by reaction with hydroxylamine hydrochloride in the presence of anhydrous sodium acetate in refluxing acetic acid. Oxime **5** was reacted with ethyl isothiocyanate in refluxing dioxane to afford the ethyl thiosemicarbazide **7**. Additionally, **5** was treated with toluene-3,5-diisocyanate to give the bissemicarbazide derivative **8** (Scheme 1).

Cyanoaminopyrane derivative **9** was prepared by condensation of the α,β-unsaturated ketone **4** with malononitrile in the presence of piperidine as a catalyst in refluxing ethanol. On the other hand, **4** was reacted with ethylcyanoacetate or malononitrile in the presence of ammonium acetate to give the corresponding cyanopyridone **10** and cyanoaminopyridine **11**. A one-step synthesis of **10** could be achieved by condensation of **3** with 1-cyano-1-carboethoxy-2-(3-indolyl)ethylene or ethyl cyanoacetate and indole-3-carboxaldehyde in the presence of ammonium acetate (Scheme 2). Similarly, a one-step synthesis of **11** could be achieved by condensation of **3** with 1,1-dicyano-2-(3-indolyl)ethylene or malononitrile and indole-3-carboxaldehyde in the presence of ammonium acetate (Scheme 2).

Several publications [25–27] have reported that the reaction of 1,2-diamines with α,β-unsaturated ketones gave different products depending on the experimental conditions. In the present work, **4** was reacted with 1,2-phenylenediamine in refluxing butanol without catalyst affording the corresponding diazepine derivative **12** (Scheme 3). Formation of **12** is believed to take place by the addition of the diamine on the ethylenic double bond of **4** followed by interamolecular condensation of the amino group with the carbonyl function affording **12** as the final product (Scheme 2).

The reaction of **4** with thiourea in the presence of ethanolic potassium hydroxide yielded the corresponding thioxopyrimidine derivative **13**, which was condensed with chloroacetic acid in a mixture of acetic acid/acetic anhydride in the presence of anhydrous sodium acetate to yield the thiazolopyrimidine derivative **14**. Thiazolopyrimidine **14** was treated with indole-3-carboxaldehyde in the presence of anhydrous sodium acetate in a mixture of glacial acetic acid/acetic anhydride yielding the corresponding arylmethylene derivative **15**. However, **15** was also prepared directly from the thioxopyrimidine **13** by reacting it with chloroacetic acid, indole-3-carboxaldehyde, and anhydrous sodium acetate in refluxing acetic acid/acetic anhydride mixture (Scheme 3).

Pharmacological Screening

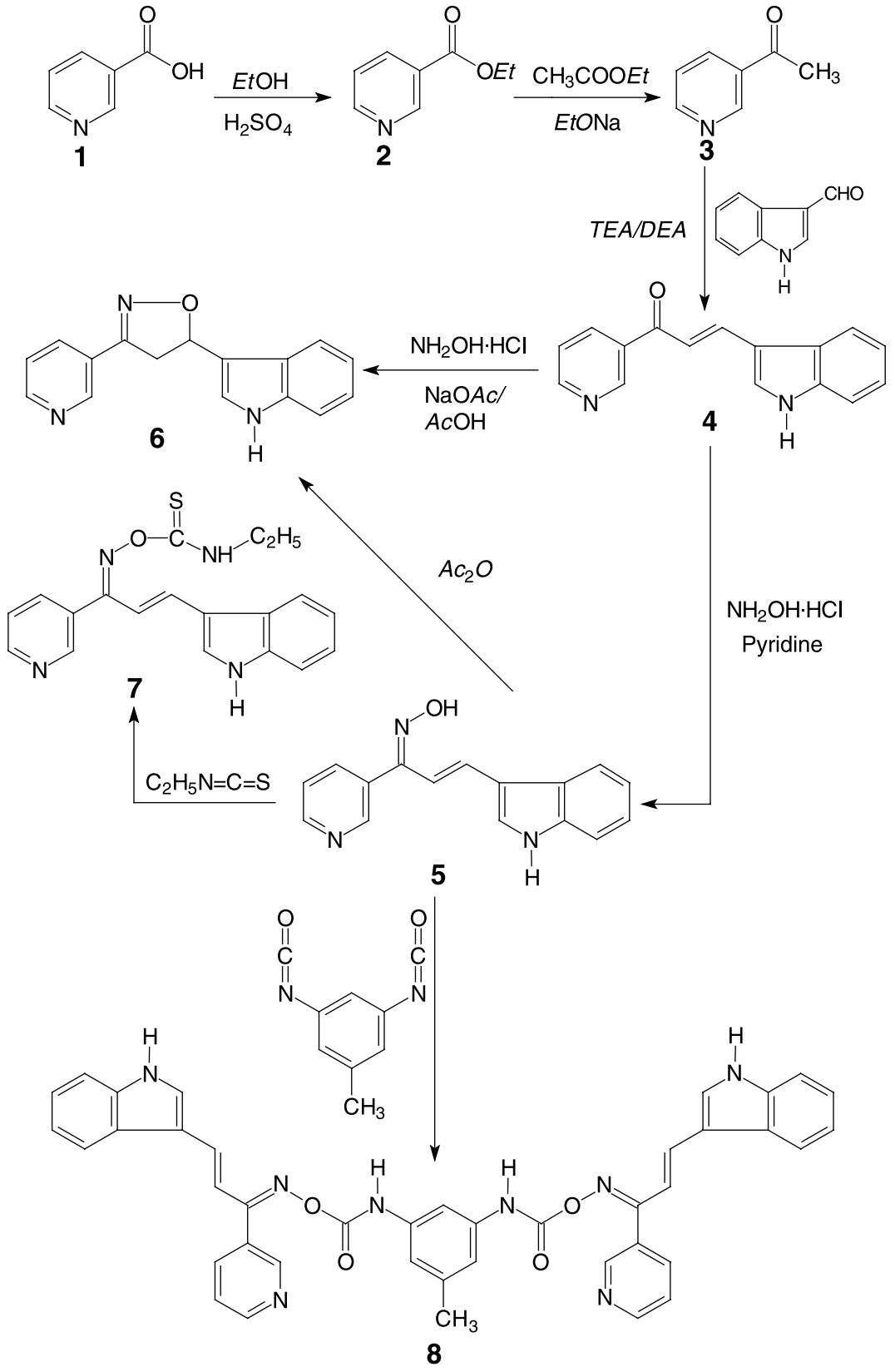
Analgesic and anticonvulsant activity was tested despite of their different biological, yet neurological receptors. Ten representative compounds, namely **5–13**, and **15** were studied with respect to analgesic and anticonvulsant activities.

Analgesic Activity

All tested compounds exhibited analgesic activity in a hot plate assay (Table 1). The most potent ones are **12** and **15**, which displayed higher activity than the standard drug Valdecoxib® by nearly 130–160% (compound **12** showed the most pronounced effect). Also, the analgesic activities of **5–11**, and **13** approached those of Valdecoxib®, and showed 61–93% activity as compared with Valdecoxib® activity (Table 1).

Anticonvulsant Activity

Antagonism against yohimbine-induced seizures in mice is considered to be a predictive model of potential anticonvulsant and *GABA*-mimetics [28]. Compounds **7–9** are devoid of anticonvulsant activity in the yohimbine-induced clonic seizures assay, in which they provide no protection against yohimbine-induced clonic seizures. Compounds **5**, **6**, and **10** showed interesting anticonvulsant activity.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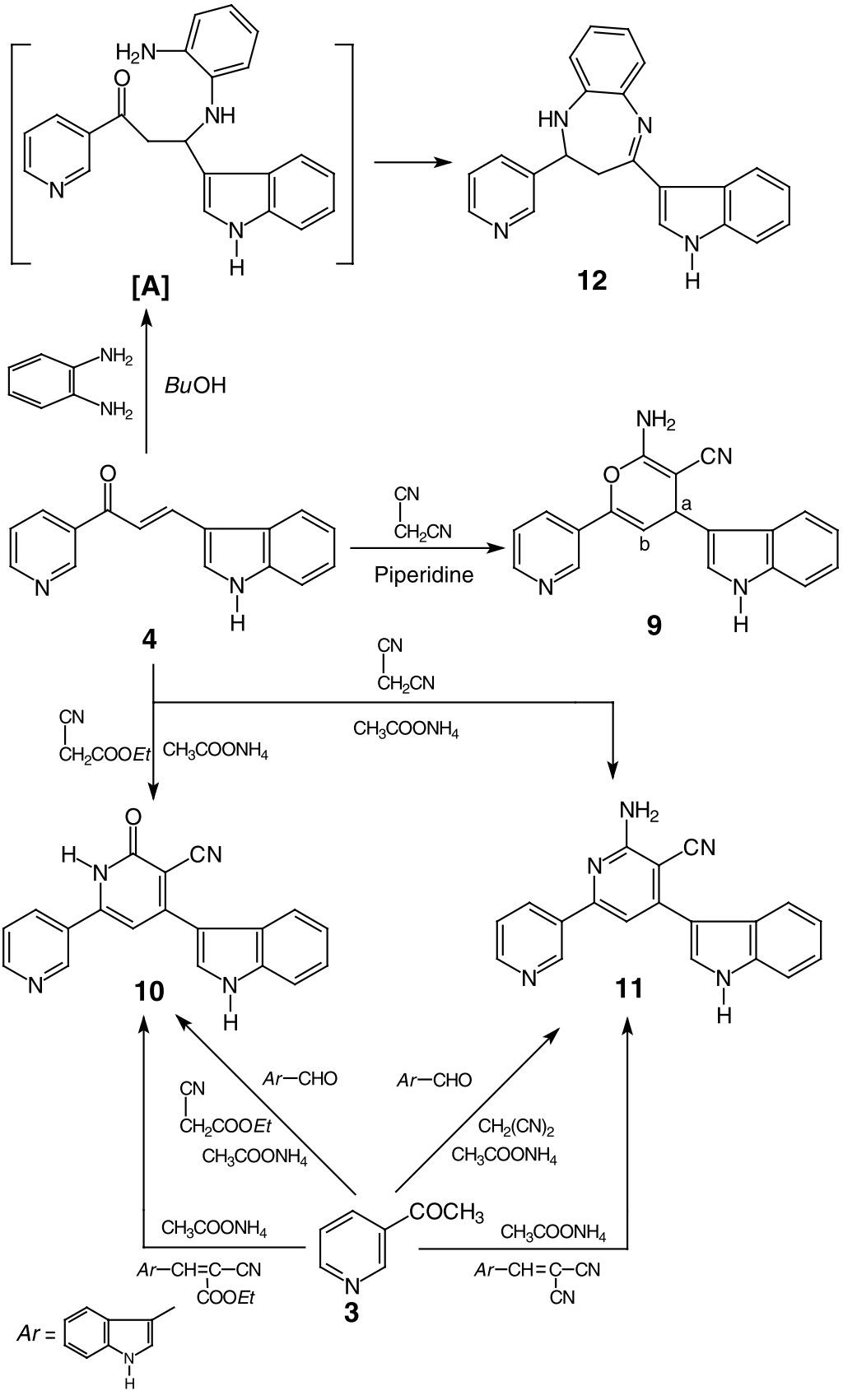
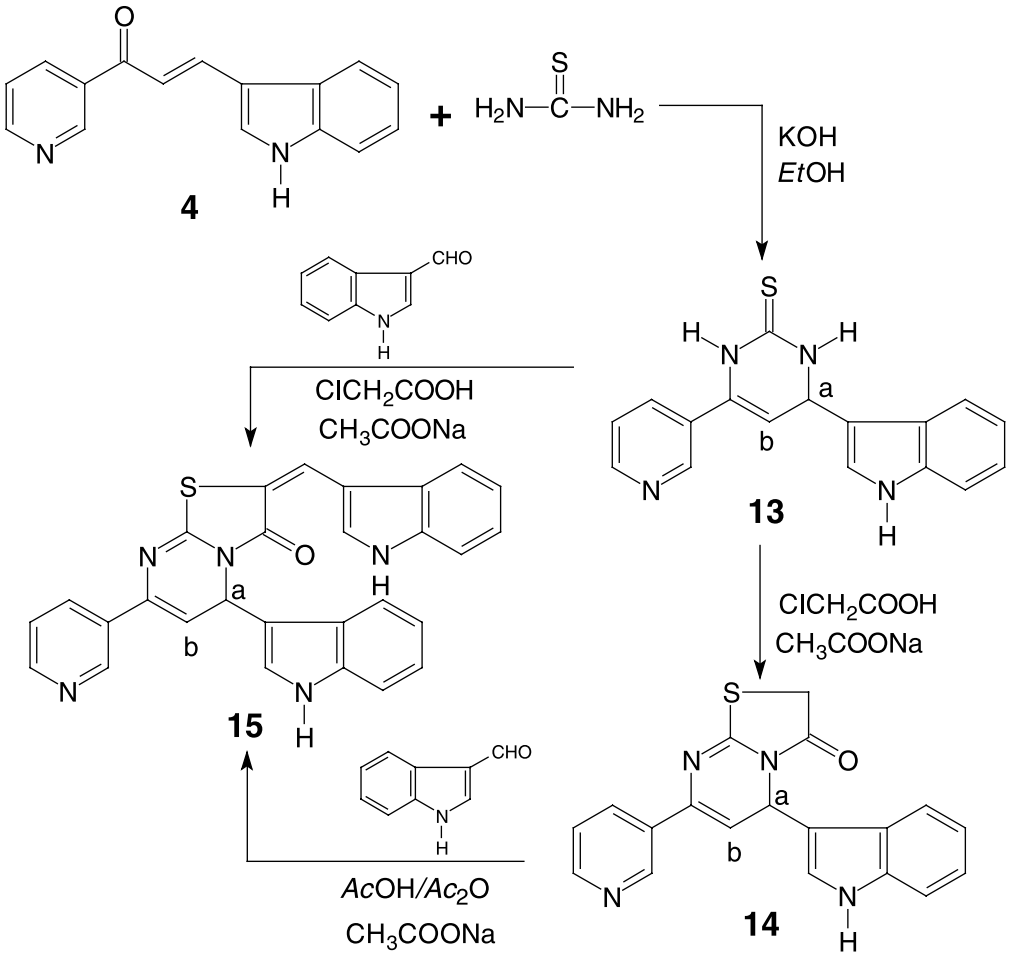
H2SO4
CH3COOEt
EtONa
TEA/DEA
NH2OH·HCl
NaOAc/
AcOH
Ac2O
NH2OH·HCl
Pyridine
C2H5N=C=S
1
2
3
4
5
6
7
8

Scheme 1



Scheme 2



Scheme 3

Table 1. Analgesic activities of selected compounds in a hot plate assay

Compound	Analgesic activity related to Valdecoxib® after:						
	10 min ± SE	20 min ± SE	30 min ± SE	45 min ± SE	60 min ± SE	90 min ± SE	120 min ± SE
Valdecoxib®	1.00 ± 0.010	1.00 ± 0.010	1.00 ± 0.010	1.00 ± 0.010	1.00 ± 0.010	1.00 ± 0.010	1.00 ± 0.010
5	0.89 ± 0.011	0.89 ± 0.011	0.89 ± 0.011	0.91 ± 0.017	0.92 ± 0.016	0.93 ± 0.015	0.90 ± 0.017
6	0.65 ± 0.012	0.63 ± 0.012	0.89 ± 0.012	0.88 ± 0.016	0.88 ± 0.021	0.89 ± 0.017	0.88 ± 0.018
7	0.78 ± 0.012	0.86 ± 0.014	0.85 ± 0.012	0.87 ± 0.015	0.87 ± 0.018	0.84 ± 0.012	0.84 ± 0.019
8	0.61 ± 0.013	0.74 ± 0.012	0.79 ± 0.001	0.81 ± 0.015	0.86 ± 0.016	0.85 ± 0.016	0.84 ± 0.035
9	0.83 ± 0.014	0.90 ± 0.015	0.93 ± 0.017	0.95 ± 0.021	0.95 ± 0.032	0.94 ± 0.018	0.94 ± 0.026
10	0.61 ± 0.013	0.65 ± 0.011	0.75 ± 0.012	0.76 ± 0.018	0.77 ± 0.011	0.78 ± 0.011	0.77 ± 0.013
11	0.92 ± 0.011	0.92 ± 0.009	0.93 ± 0.016	0.88 ± 0.019	0.83 ± 0.021	0.79 ± 0.016	0.65 ± 0.012
12	1.30 ± 0.180	1.43 ± 0.16	1.44 ± 0.130	1.45 ± 0.200	1.41 ± 0.320	1.42 ± 0.290	1.40 ± 0.280
13	0.63 ± 0.010	0.64 ± 0.017	0.73 ± 0.013	0.73 ± 0.018	0.74 ± 0.019	0.75 ± 0.016	0.78 ± 0.013
15	0.98 ± 0.013	0.97 ± 0.015	1.41 ± 0.140	1.56 ± 0.210	1.55 ± 0.350	1.60 ± 0.340	1.43 ± 0.450

Their relative potencies compared to the standard drug Carbamazepine® are 0.68, 0.96, and 0.75. Compounds **11–13**, and **15** are more potent than Carbamazepine® with relative potencies of 2.30, 2.54, 2.02, and 1.84 (Table 2). The ED_{50} value was estimated *via* determining the dose which protected 50% of the tested animals against the convulsant induced by yohimbine.

Table 2. Anticonvulsant activities of selected compounds (as ED_{50} values) needed to antagonize yohimbine-induced clonic seizure and compared to the anticonvulsant activity of Carbamazepine®

Compound	$ED_{50}/mg \cdot kg^{-1} \pm SE$	Relative potency compared to Carbamazepine® ± SE
Control	0	0
Carbamazepine®	29 ± 0.31	1.00 ± 0.011
5	53 ± 0.41	0.68 ± 0.010
6	30 ± 0.32	0.96 ± 0.008
7	No protection	No protection
8	No protection	No protection
9	No protection	No protection
10	34 ± 0.35	0.75 ± 0.008
11	12 ± 0.10	2.30 ± 0.018
12	11 ± 0.112	2.54 ± 0.020
13	14 ± 0.12	2.02 ± 0.024
15	16 ± 0.123	1.84 ± 0.019

Experimental

All melting points were taken on an Electrothermal IA 9000 series digital melting point apparatus. Elemental analytical data (in accord with the calculated values) were obtained from the microanalytical unit, Cairo University, Cairo, Egypt. The IR spectra (*KBr*) were recorded on a Pye Unicam SP-1000 spectrophotometer. The ^{1}H NMR spectra were recorded at 270 MHz on a Varian EM-360 Spectrometer using *TMS* as an internal standard at the Central Services Laboratory, Centre of Molecular and Macromolecular Studies, Polish Academy of Science, Poland. The mass spectra were performed using a VG 2AB-3F spectrometer (70 eV). All reactions were followed by TLC (silica gel, aluminum sheets 60 F_{254}, Merck).

3-[β-(3-Indolyl)acryloyl]pyridine (**4**, $C_{16}H_{12}N_2O$)
A mixture of 0.12 g **3** (1 mmol) and 0.145 g indole-3-carboxaldehyde (1 mmol) together with 3 cm^3 of a mixture of *TEA*/*DEA* (1:1) in 30 cm^3 ethanol was refluxed for 4 h. The reaction mixture was evaporated under reduced pressure to dryness, the obtained residue was solidified with ether. The separated solid was filtered off, dried (under vacuum) and crystallized to afford 0.21 g (86%) **4**. Mp 240–242°C (*Ac*OH/H_2O); IR (film): $\bar{\nu} = 3340$–3260 (NH), 1679 (C=O), 1607 (C=C) cm^{-1}; ^{1}H NMR (*DMSO*-d_6): $\delta = 6.50$ (d, $J = 14.60$ Hz, CH-olefinic), 6.90 (d, $J = 14.65$ Hz, CH-olefinic), 7.0–7.40 (m, Ar–H), 7.80–8.25 (m, pyrid-4,5,6), 8.60 (s, pyrid-2), 9.50 (s, NH-indole, exchangeable with D_2O) ppm; MS (EI, 70 eV): $m/z = 248$ [M$^+$, 45], 106 [100, base peak].

3-[β-(3-Indolyl)acryloyl]pyridine oxime (**5**, $C_{16}H_{13}N_3O$)
A mixture of 0.248 g **4** (1 mmol) and ~0.1 g of $NH_2OH \cdot HCl$ (1 mmol) in 30 cm^3 dry pyridine was refluxed for 6 h. The reaction mixture was cooled, poured into ice-water, and neutralized with 1 *N* HCl. The obtained solid was collected by filtration, dried (under vacuum) and crystallized to give 0.20 g (82%) **5**. Mp 225–227°C (*Ac*OH/H_2O); IR (film): $\bar{\nu} = 3540$–3260 (OH, NH), 1670 (C=N), 1605 (C=C) cm^{-1}; ^{1}H NMR (*DMSO*-d_6): $\delta = 2.45$ (s, OH, exchangeable with D_2O), 6.35 (d, $J = 14.55$ Hz, CH-olefinic), 6.80 (d, $J = 14.60$ Hz, CH-olefinic), 6.95–7.55 (m, indole-H), 7.85–8.35 (m, pyrid-4,5,6), 8.68 (s, pyrid-2), 9.65 (s, NH-indole, exchangeable with D_2O) ppm; MS (EI, 70 eV): $m/z = 263$ [M$^+$, 100, base peak].

2-(Pyridin-3-yl)-4-(indol-3-yl)oxazole (**6**, $C_{16}H_{13}N_3O$)
Method A: A solution of 0.263 g **5** (1 mmol) in 50 cm^3 acetic anhydride was refluxed for 10 h. After cooling, the reaction mixture was poured into ice-water, the obtained solid was filtered off, washed with water, dried (under vacuum), and crystallized to give 0.20 g (76%) **6**. Mp 232–234°C (*Et*OH); IR (KBr): $\bar{\nu} = 3340$–3240 (NH), 1665 (C=N), 1608–1600 (C=C) cm^{-1}; ^{1}H NMR (*DMSO*-d_6): $\delta = 1.6$–1.9 (m, CH_2-oxazole), 4.2 (m, CH-oxazole), 6.82–7.35 (m, indole-H), 7.80–8.30 (m, pyrid-4,5,6), 8.62 (s, pyrid-2), 9.45 (s, NH-indole, exchangeable with D_2O) ppm; ^{13}C NMR (*DMSO*-d_6): $\delta = 42.10$, 77.80, 155.10 (oxazole-C), 110.10, 116.5, 118.0, 119.10, 121.2, 121.9, 126.5, 135.3 (indole-C), 122.90, 125.30, 136.20, 150.5, 151.0 (pyridine-C) ppm; MS (EI, 70 eV): $m/z = 263$ [M$^+$, 15], 147 [100, base peak].

Method B: A mixture of 0.248 g **4** (1 mmol), ~0.1 g $NH_2OH \cdot HCl$ (1 mmol), and 0.082 g anhydrous sodium acetate (1 mmol) in 30 cm^3 glacial acetic acid was refluxed for 6 h. The reaction mixture was cooled, poured into ice-water, the obtained solid was collected by filtration, washed with water, dried (under vacuum), and crystallized to give 0.17 g (66%) **6** as identified by mp and TLC in comparison with authentic sample.

3-[β-(3-Indolyl)acryloylethylthiosemicarbazide]pyridine (**7**, $C_{19}H_{18}N_4OS$)
A mixture of 0.263 g **5** (1 mmol) and 0.087 g ethyl isothiocyanate (1 mmol) in 50 cm^3 dry dioxane containing 2 cm^3 of *TEA* was heated under reflux for 10 h. The solvent was evaporated under reduced pressure, the obtained residue was solidified with *n*-hexane. The obtained solid was filtered off, washed with diethyl ether, dried (under vacuum), and crystallized to give 0.24 g (68%) **7**. Mp 189–191°C (dioxane/benzene); IR (film): $\bar{\nu} = 3336$–3250 (NH), 1668 (C=N), 1225 (C=S) cm^{-1}; ^{1}H NMR (*DMSO*-d_6): $\delta = 0.8$ (t, 3H, CH_3), 2.40 (d, CH_2), 4.10–4.30 (bs, NH-CS, exchangeable with D_2O), 6.20 (d, $J = 14.59$ Hz, CH-olefinic), 6.50 (d, $J = 14.62$ Hz, CH-olefinic), 6.90–7.25 (m, Ar–H), 7.85–8.40 (m, pyrid-4,5,6), 8.75 (s, pyrid-2), 9.44 (s, NH-indole, exchangeable with D_2O) ppm; ^{13}C NMR (*DMSO*-d_6): $\delta = 17.60$ (CH_3), 34.60 (CH_2), 109.75, 116.25, 117.85, 119.05, 121.25, 122.05, 126.35, 135.45 (indole-C), 123.0, 125.15, 136.40, 150.55, 150.95 (pyridine-*C*), 137.15, 141.75 (CH=CH), 156.65 (C=S), 162.90 (C=N) ppm; MS (EI, 70 eV): $m/z = 350$ [M$^+$, 32], 246 [100, base peak].

3,5-Bis{3-[β-(3-indolyl)acryloylsemicarbazide]pyridine} toluene (**8**, $C_{41}H_{32}N_8O_4$)
A mixture of 0.526 g **5** (2 mmol) and 0.174 g toluene-3,5-diisocyanate (1 mmol) in 50 cm^3 dry dioxane containing 2 cm^3 *TEA* was refluxed for 12 h. The solvent was evaporated

under reduced pressure and the oily product was triturated with *n*-hexane and petroleum ether (40–60°C). The obtained solid was filtered off, dried (under vacuum), and crystallized to give 0.45 g (65%) **8**. Mp 196–198°C (*Et*OH); IR (film): $\bar{\nu} = 3460–3290$ (NH), 1710–1695 (C=O), 1685–1675 (C=N) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 1.20$ (s, CH_3), 6.35 (d, $J = 14.60$ Hz, CH-olefinic), 6.70 (d, $J = 14.65$ Hz, CH-olefinic), 6.85–7.20 (m, Ar–H), 7.80–8.50 (m, 2 × pyrid-4,5,6), 8.65 (s, NHCO, exchangeable with D_2O), 8.76 (s, pyrid-2), 9.56 (s, NH-indole, exchangeable with D_2O) ppm; ^{13}C NMR (*DMSO*-d_6): $\delta = 19.65$ (CH_3), 104.75, 117.18, 134.80, 136.70 (Ph-C), 109.85, 116.35, 118.05, 119.0, 121.24, 122.10, 126.30, 135.35 (indole-C), 122.95, 125.28, 136.42, 150.49, 151.05 (pyridine-*C*), 137.18, 142.65 (CH=CH), 152.80 (C=O), 163.10 (C=N) ppm; MS (EI, 70 eV): m/z (%) = 700 [M^+, 5], 176 [100, base peak].

2-Amino-4-(3-indolyl)-6-(3-pyridyl)-pyran-3-carbonitrile (**9**, $C_{19}H_{14}N_4O$)

Method A: A mixture of 0.248 g **4** (1 mmol), 0.066 g malononitrile (1 mmol), and a few drops of piperidine in 100 cm^3 ethanol was stirred at room temperature for 2 h. The solvent was concentrated under reduced pressure; the formed solid was filtered off, dried (under vacuum) and crystallized to give 0.26 g (84%) **9**. Mp 210–212°C (*Ac*OH); IR (film): $\bar{\nu} = 3560–3390$ (NH, NH_2), 2218 (C≡N), 1675 (C=N) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 3.76$ (bs, NH_2, exchangeable with D_2O), 4.48 (s, H_a-pyrane), 6.95–7.35 (m, Ar–H + H_b-pyrane), 7.86–8.55 (m, pyrid-4,5,6), 8.74 (s, pyrid-2), 9.60 (s, NH-indole, exchangeable with D_2O) ppm; MS (EI, 70 eV): $m/z = 314$ [M^+, 25], 198 [100, base peak].

Method B: A mixture of 0.248 g **4** (1 mmol), 0.066 g malononitrile (1 mmol), and 2 g anhydrous sodium acetate in 25 cm^3 glacial acetic acid was refluxed for 4 h. The reaction mixture was poured onto crushed ice, the obtained precipitate was collected by filtration, dried (under vacuum), and crystallized to give 0.20 g (65%) **9** as identified by mp and R_f-values in comparison with authentic samples previously obtained by Method A.

*Synthesis of Cyanopyridone **10** and Cyanoaminopyridine **11***

Method A: A mixture of 0.248 g **4** (1 mmol), ethyl cyanoacetate or malononitrile (1 mmol), and 0.6 g ammonium acetate (8 mmol) in 30 cm^3 ethanol was refluxed for 4 h. The reaction mixture was evaporated under reduced pressure and the obtained residue was triturated with diethyl ether. The formed solid was collected by filtration, washed with ether, dried (under vacuum), and crystallized to give 0.26 g (85%) **10** and 0.29 g (92%) **11**.

Method B: A mixture of 0.12 g **3** (1 mmol), 1-cyano-1-ethyl ester-2-(3-indolyl)-ethylene or 1,1-dicyano-2-(3-indolyl)ethylene (1 mmol) and 0.6 g ammonium acetate (8 mmol) in 30 cm^3 ethanol was refluxed for 6 h. The reaction mixture was concentrated and left to cool. The formed solid was filtered off and crystallized to give 0.23 g (75%) **10** and 0.17 g (55%) **11**.

Method C: A mixture of 0.12 g **3** (1 mmol), 0.145 g indole-3-carboxaldehyde (1 mmol), and ethyl cyanoacetate or malononitrile (1 mmol) in the presence of 0.6 g ammonium acetate (8 mmol) in 30 cm^3 ethanol was refluxed for 5 h. The solvent was evaporated under reduced pressure and the obtained residue was triturated with *n*-hexane. The obtained solid was filtered off, washed with *n*-hexane, and crystallized to give 0.21 g (68%) **10** and 0.20 g (65%) **11**. The obtained products from methods B and C were identified by their mps, mixed mps, and R_f values on TLC by comparison with authentic samples from method A.

3-[3′-Cyano-2′-oxo-4′-(3″-indolyl)-6′-pyridyl]pyridine (**10**, $C_{19}H_{12}N_4O$)

Mp 148–149°C (*Ac*OH/H_2O); IR (film): $\bar{\nu} = 3427–3350$ (NH), 2217 (C≡N), 1708 (C=O) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 6.98–7.45$ (m, Ar–H), 7.65–8.35 (m, pyrid-4,5,6), 8.45 (s, 1H, pyrid-2), 8.90 (s, NH-pyridone, exchangeable with D_2O), 9.56 (s, NH-indole, exchangeable with D_2O) ppm; ^{13}C NMR (*DMSO*-d_6): $\delta = 114.90$ (CN), 106.40, 110.80, 118.05, 119.15, 121.24, 125.10, 128.30, 134.85 (indole-C), 123.30, 125.40, 136.22, 149.96, 151.15 (pyridine-*C*), 109.80, 120.15, 155.30, 163.80 (pyridone-C), 158.65 (C=O) ppm; MS (EI, 70 eV): $m/z = 312$ [M^+, 12], 196 [100, base peak].

3-[2′-Amino-3′-cyano-4′-(3″-indolyl)-6′-pyridyl]pyridine (**11**, $C_{19}H_{13}N_5$)

Mp 178–179°C (*Et*OH); IR (film): $\bar{\nu} = 3560–3380$ (NH, NH_2), 2221 (C≡N), 1770 (C=N) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 3.65$ (s, NH_2, exchangeable with D_2O), 7.05–7.60 (m, Ar–H), 7.80–8.30 (m, pyrid-4,5,6), 8.60 (s, pyrid-2), 9.48 (s, NH-indole, exchangeable with D_2O) ppm; MS (EI, 70 eV): m/z (%) = 311 [M^+, 100, base peak].

*Cyclocondensation of α,β-Unsaturated Ketone **4** with o-Phenylenediamine*

2-(Pyridin-3-yl)-4-(indol-3-yl)benzo[b]diazipene (**12**, $C_{22}H_{18}N_4$)

A mixture of 0.248 g **4** (1 mmol) and 0.11 g *o*-phenylenediamine (1 mmol) in 30 cm^3 dry *n*-butanol was heated under reflux for 18 h. The solvent was evaporated under reduced pressure till dryness, the oily product was triturated with petroleum ether (40–60°C), and the separated solid was filtered off, washed with petroleum ether (40–60°C), dried (under vacuum), and crystallized to give 0.22 g (66%) **12**. Mp 155–156°C (*Et*OH); IR (film): $\bar{\nu} = 3380–3342$ (NH), 1610 (C=N) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 3.20$ (d, CH_2-diazepine), 4.10 (m, CH-diazepine), 6.84–7.65 (m, Ar–H), 7.78–8.35 (m, pyrid-4,5,6), 8.66 (s, pyrid-2), 9.62 (s, NH-indole, exchangeable with D_2O), 11.20 (s, NH-diazepine, exchangeable with D_2O) ppm; ^{13}C NMR (*DMSO*-d_6): $\delta = 42.20$ (CH_2), 58.45 (CH), 110.42, 111.60, 119.05, 120.05, 121.50, 125.35, 128.38, 134.50 (indole-C), 122.85, 126.55, 136.40, 149.65, 150.90 (pyridine-*C*), 114.80, 118.50, 122.90, 128.0, 133.85, 144.15 (Ph-C), 161.05 (C=N) ppm; MS (EI, 70 eV): $m/z =$ 338 [M^+, 10] and at 312 [100, base peak].

6-(Pyridin-3-yl)-1,2,3,4-tetrahydro-2-thioxo-4-(indol-3-yl)pyrimidene (**13**, $C_{17}H_{14}N_4S$)

To a mixture of 0.248 g **4** (1 mmol) and 0.2 g KOH in 1 cm^3 water in 30 cm^3 ethanol, 0.15 g thiourea (2 mmol) were added.

The reaction mixture was heated under reflux for 5 h. The reaction mixture was concentrated under reduced pressure and then poured into ice water. The solid formed was filtered off and crystallized to give 0.23 g (76%) **13**. Mp 155–156°C (*Et*OH/H_2O); IR (film): $\bar{\nu} = 3440–3320$ (NH), 1210 (C=S) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 5.10$ (d, H-a, pyrimidine), 6.80–7.50 (m, indole-H + H-b pyrimidine), 7.85–8.25 (m, pyrid-4,5,6), 8.40 and 8.55 (2s, NH-pyrimidine, exchangeable with D_2O), 8.68 (s, pyrid-2), 9.55 (s, NH-indole, exchangeable with D_2O) ppm; MS (EI, 70 eV): $m/z = 306$ [M^+, 55], 190 [100, base peak].

7-(Pyridin-3-yl)-5-(indol-3-yl)-2,3-dihydro-5H-thiazolo[3,2-a]pyrimidine (**14**, $C_{19}H_{14}N_4OS$)
A mixture of 0.306 g **13** (1 mmol) and 0.094 g chloroacetic acid (1 mmol) in 60 cm^3 of a mixture of glacial acetic acid and acetic anhydride (1:1) in the presence of 3 g anhydrous sodium acetate was refluxed for 6 h. The reaction mixture was cooled and poured onto cold water with stirring, the solid formed was filtered off, and crystallized to give 0.25 g (72%) **14**. Mp 135–136°C (*Me*OH/H_2O); IR (film): $\bar{\nu} = 3360–3320$ (NH), 1735 (C=O) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 3.75$ (s, CH_2-thiazole), 5.65 (d, H-a, for pyrimidine ring), 6.88–7.46 (m, indole-H + H-b pyrimidine ring), 7.78–8.45 (m, pyrid-4,5,6), 8.64 (s, pyrid-2), 9.66 (s, NH-indole, exchangeable with D_2O) ppm; ^{13}C NMR (*DMSO*-d_6): $\delta = 29.86$ (CH_2), 44.38, 119.65, 141.15, 162.10 (pyrimidine-C), 110.05, 111.50, 118.15, 119.18, 121.30, 122.90, 126.10, 135.85 (indole-C), 122.95, 126.40, 130.22, 149.65, 150.86 (pyridine-*C*), 169.76 (C=O) ppm; MS (EI, 70 eV): $m/z = 346$ [M^+, 35], 230 [100, base peak].

7-(Pyridin-3-yl)-2-(indolyl methylene)-5-(indol-3-yl)-2,3-dihydro-5-thiazolo[3,2-a]pyrimidine (**15**, $C_{28}H_{19}N_5OS$)
Method A: A mixture of 0.306 g **13** (1 mmol), 0.094 g chloroacetic acid (1 mmol), 1.5 g anhydrous sodium acetate in 60 cm^3 of a mixture of glacial acetic acid and acetic anhydride (1:1), and 0.145 g indole-3-carboxaldehyde (1 mmol) was refluxed for 6 h. The reaction mixture was cooled and poured onto ice-water, the obtained solid was collected by filtration, and crystallized to give 0.35 g (74%) **15**. Mp 207–209°C (*Et*OH); **15**. IR (film): $\bar{\nu} = 3365–3315$ (NH), 1706 (C=O) cm^{-1}; ^1H NMR (*DMSO*-d_6): $\delta = 5.70$ (d, H-a, pyrimidine ring), 6.90–7.60 (m, indole-H + H-b pyrimidine ring + 1H benzylic proton), 7.75–8.50 (m, pyrid-4,5,6), 8.62 (s, pyrid-2), 9.60 and 9.68 (2s, NH-indole, exchangeable with D_2O) ppm; MS (EI, 70 eV): $m/z = 473$ [M^+, 100, base peak].

Method B: A mixture of 0.346 g **14** (1 mmol) and 0.145 g indole-3-carboxaldehyde (1 mmol) in 40 cm^3 of a mixture of acetic acid/acetic anhydride (3:1) was refluxed for 5 h, allowed to cool, and then poured onto water. The solid formed was collected by filtration and crystallized to yield 0.29 g (62%) **15**. The obtained product from method B was identified by its mp, mixed mp, and R_f value on TLC by comparison with the authentic sample from method A.

Pharmacology Screening
All animals were obtained from Animal House Colony, Research Institute of Ophthalmology, Giza, Egypt.

Analgesic Activity
Sixty *Webster* mice of both sexes weighting 20–25 g were divided into 10 groups. One group was kept as control (received saline), the second group received vehicle (Gum acacia), and the third one received Valdecoxib® as a reference drug, whereas the other groups received the test compounds (SC administration). Mice were dropped gently in a dry glass beaker of 1 dm^3 capacity maintained at 55–55.5°C. Normal reaction time in seconds for all animals was determined at time intervals of 10, 20, 30, 45, 60, 90, and 120 min. This is the interval extending from the instant the mouse reaches the hot beaker till the animals lick their feet or jump out of the beaker (dose 5 mg/kg) [29]. The relative potencies to Valdecoxib® were determined (Table 1).

Anticonvulsant Activity
Male *Webster* mice (20–30 g) were individually placed in a clear plastic cylinder and the tested compounds were administrated intraperitoneally (5 mg/kg) 30 min prior to a dose of 45 mg/kg of yohimbine-HCl. The animals were observed for the onset and number of clonic seizure [30] (Table 2).

Determination of ED_{50}
ED_{50} values of the compounds with 95% confidence limit were calculated for the antagonism of yohimbine-induced clonic by the method of *Austen* and *Brocklehurst* [31].

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