Anticonvulsant Activity of Piperidinol and (Dialkylamino)alkanol Esters

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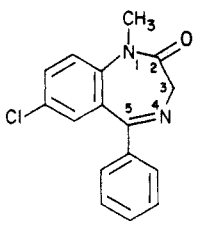
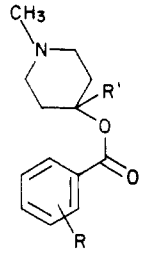
Aromatic and heterocyclic esters of 1-methyl-4-piperidinol and 1,4-dimethyl-4-piperidinol and aromatic esters of (dialkylamino)alkanols were prepared and evaluated for antiepileptic activity by the maximal electroshock seizure (MES) and subcutaneous pentylenetetrazole seizure threshold (scMet) assays and for minimal central neurotoxicity by the rotorod ataxia test. The most potent compound, namely the 2-phenylbenzoate (**57**) of 3-(diethylamino)propanol, was slightly more potent than diphenylhydantoin in the MES assay, while the 2-phenylbenzoate (**24**) of 1-methyl-4-piperidinol and the 2-phenylbenzoate (**56**) of (diethylamino)ethanol displayed activity comparable to that of diphenylhydantoin. The 2-phenethylbenzoate ester (**6**) of 1-methyl-4-piperidinol exhibited one-third the activity of diphenylhydantoin. The 2,4,5-trimethylbenzoate **40** and 2,4,6-trimethylbenzoate **41** of 1-methyl-4-pieridinol were even less potent, but did display activity in the phenobarbital–methsuximide range. Certain compounds interact with sites associated with the GABA receptor–chloride channel complex, but their potencies as anticonvulsant agents do not correlate with interaction at sites on the channel complex. Certain analogues antagonize binding of a batrachotoxin analogue to sodium channel sites, a property indicative of local anesthetic activity. There are structural similarities between 2-phenylbenzoates **57**, **56**, and **24** and diphenylhydantoin, and the latter anticonvulsant also antagonizes binding of the batrachotoxin analogue.

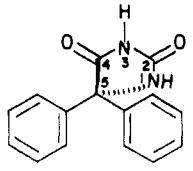
Aromatic piperidinol esters of type I can assume, due to a high degree of conformational mobility, spatial arrangements between the aromatic ring and the tertiary amine that are similar to those of the 5-phenyl group and N-1 of diazepam. In addition, certain 2-phenylbenzoates of type II show spatial similarities between the phenyl rings and the ester carbonyl with the 5-diphenyl and 4-keto groups of phenytoin. Similarities in spatial arrangements between hydrophobic and electron donating groups have been noted for phenytoin and diazepam.[1]



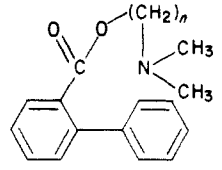
diazpam



I: R = mono-, di-, or trisubstitution; R' = H, CH_3



phenytoin



II: $n = 2.3$

Benzodiazepines exhibit activity in a variety of partial and generalized epileptic seizures, especially in status epilepticus, whereas phenytoin is effective against generalized tonic–clonic seizures (grand mal).[2]

The anticonvulsant activities of 66 esters, the majority being of type I and II, were determined by the maximal electroshock seizure (MES) and the subcutaneous pentylenetetrazole seizure (scMet) assays. Neurotoxicity was determined by the rotorod toxicity test.

Because of possible structural relationships of the present esters to benzodiazepines and/or phenytoin, a selected group of active esters were evaluated as antagonists of (i) [^{3}H]diazepam binding to ascertain possible benzodiazepine-like activity, (ii) [^{3}H]muscimol binding to ascertain possible interactions with GABA receptors, (iii) [^{35}S]tributyl bicyclophosphorothionate (TBPS) binding to ascertain possible interactions with chloride channel sites associated with GABA receptors, and (iv) [^{3}H]batrachotoxinin A benzoate binding to ascertain possible interactions with voltage-dependent sodium channels. The results suggest that interaction at sodium channel sites would be significant for the esters at in vivo doses required for anticonvulsant activity.

Results and Discussion

Initially, a group of compounds were selected from five different ester types (aromatic, heterocyclic, aralkyl, cycloalkyl, and alkyl) of 1-substituted-4-piperidinols and 1,4-disubstituted-4-piperidinols (Chart I). From the preliminary assay data, it was ascertained that anticonvulsant activity was associated mainly with one of the ester types, namely the benzoates, in which the aromatic ring is attached directly to the carbonyl function. Based on this observation, additional compounds of this type with various (dialkylamino)alkanols (Chart II) were synthesized.

The aromatic esters and the majority of the heterocyclic esters were prepared by the acid chloride method. Because of the lability of the pyrrole ring toward protic acids, the synthesis of pyrrole ester **60** was achieved by using basic conditions, namely transesterification of ethyl 3,5-dimethyl-4-acetyl-2-pyrrolecarboxylate in the presence of an excess of 1-methyl-4-piperidinol and a catalytic amount of sodium at 180 °C.

Compounds **1–66** were assayed for anticonvulsant activity (MES and scMet tests) and for neurotoxicity in mice by the National Institute of Neurological and Communicative Disorders and Stroke (NINCDS). The MES assay has predictive value for agents of potential therapeutic value in the management of grand mal epilepsy, whereas the scMet test is predictive for those likely to be effective against petit mal.[3] Twenty-two compounds exhibited activity in the primary MES screen. Eleven of these exhibited excellent activity (protection at 30 mg/kg) from which eight were selected by the NINCDS for their secondary screen (ED_{50} determination).

A moderately potent anticonvulsant of the aromatic esters was the 1-methyl-4-piperidinol 2-phenethylbenzoate

(1) Camerman, A.; Camerman, N. In *Antiepileptic Drugs: Mechanism of Action*; Glaser, G. H., Penry, J. K., Woodbury, D. M., Eds.; Raven: New York, 1980; pp 223–231.
(2) Gallagher, B. B. In *Anticonvulsants*; Vida, J. A., Ed.; Academic: New York, 1977; p 48.
(3) Woodbury, D. M. In *Experimental Models of Epilepsy*; Purpura, D. P., Penry, J. K., Tower, D., Woodbury, D. M., Walter, R., Eds.; Raven: New York, 1972; p 557.

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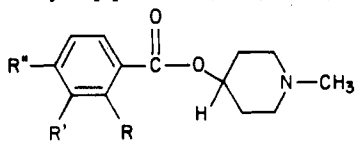
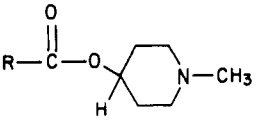
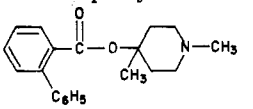
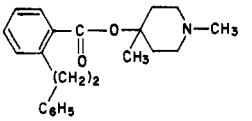
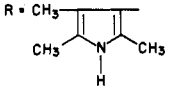
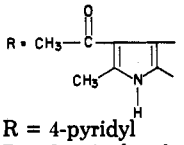
Chart I. Esters of 1-Methyl-4-piperidinol (1–51, 59–66) and 1,4-Dimethyl-4-piperidinol (52, 53)[a]



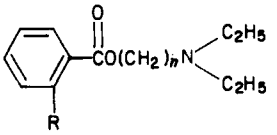


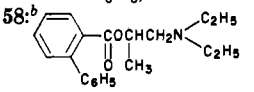
1: R = CH_3; R′, R″ = H
2: R = OCH_3; R′, R″ = H
3: R = OC_2H_5; R′, R″ = H
4: R = Cl; R′, R″ = H
5: R = OC_6H_5; R′, R″ = H
6: R = $C_6H_5CH_2CH_2$; R′, R″ = H
7: R′ = CH_3; R, R″ = H
8: R′ = OH; R, R″ = H
9: R′ = OCH_3; R, R″ = H
10: R″ = CH_3; R, R′ = H
11: R″ = OCH_3; R, R′ = H
12: R″ = OC_2H_5; R, R′ = H
13: R″ = $O(CH_2)_3CH_3$; R, R′ = H
14: R″ = F; R, R′ = H
15: R″ = Cl; R, R′ = H
16: R″ = CN; R, R′ = H
17: R, R′ = OCH_3; R″ = H
18: R, R″ = CH_3; R′ = H
19: R, R″ = OCH_3; R′ = H
20: R′, R″ = CH_3; R = H
21: R′, R″ = OCH_3; R = H
22: R, R′, R″ = OCH_3
23: R = C_2H_5; R′, R″ = H
24: R = C_6H_5; R′, R″ = H
25: R = H; R′, R″ = OC_2H_5
26: R, R′ = H; R″ = I
27: R = Br; R′, R″ = H
28: R, R′ = CH_3; R″ = H
29: R = $C_6H_5CH_2$; R′, R″ = H
30: R = NO_2; R′, R″ = H
31: R = F_3C; R′, R″ = H
32: R, R′ = H; R″ = C_6H_5
33: R = $C_6H_5C(O)$; R′, R″ = H
34: R = 4-F($C_6H_4C(O)$); R′, R″ = H
35: R = $C_6H_5OCH_2$; R′, R″ = H

36: R = 2,5-$(CH_3)_2C_6H_3$
37: R = 2,6-$(CH_3)_2C_6H_3$
38: R = 3,5-$(CH_3)_2C_6H_3$
39: R = 3,4-$(OCH_2O)C_6H_3$
40: R = 2,4,5-$(CH_3)_3C_6H_2$
41: R = 2,4,6-$(CH_3)_3C_6H_2$
42: R = $C_6H_5CH(CH_3)CH_2$
43: R = $C_6H_5CH_2$
44: R = 3,4-$(CH_3O)_2C_6H_3(CH_2)_2$
45: R = $C_6H_5(CH_2)_3$
46: R = $C_6H_5(CH_2)_2$
47: R = $(C_6H_5)_2CHCH_2$
48: R = $(C_6H_5)_2CH$
49: R = $(C_6H_5)_2C(CH_3)$
50: R = 1-naphthyl
51: R = 2-naphthyl
52:[b] 
53:[b] 
59: 
60: 
61: R = 4-pyridyl
62: R = 3-quinolynyl
63: R = CH_3CH_2
64: R = 1-adamantyl
65: R = (−)-3-pinanyl
66: R = (+)-3-pinanyl

[a] These compounds were obtained as salts as indicated in the Experimental Section and references cited therein and in Table II. [b] Complete formula is shown.

Chart II. Aromatic Esters of (Dialkylamino)alkanols[a]



54: R = $C_6H_5(CH_2)_2$; n = 2
55: R = $C_6H_5(CH_2)_2$; n = 3
56: R = C_6H_5; n = 2
57: R = C_6H_5; n = 3
58:[b] 

[a] These compounds were obtained as salts as indicated in Table II. [b] Complete formula is shown.

(6), with an ED_{50} = 25 mg/kg in the MES assay (Table I). The MES potency of 6 was comparable with that of phenobarbital, ED_{50} = 22, and approximately one-third that of phenytoin, ED_{50} = 9.5. The 1-methyl-4-piperidinol 2-phenylbenzoate (24) (ED_{50} = 12) and the (diethylamino)ethanol 2-phenylbenzoate (56) (ED_{50} = 10) displayed potencies comparable to phenytoin. The most potent compound proved to be 3-(diethylamino)propanol 2-phenylbenzoate 57, ED_{50} = 7.8. Other 2-substituted benzoates, namely 2-alkoxy (2, 3), 2-chloro (4), 2-phenoxy (5), 2-ethyl (23), 2-bromo (27), 2-nitro (30), 2-trifluoromethyl (31), 2-benzoyl (33), and 2-(*p*-fluorobenzoyl) (34) compounds were inactive in the MES assay. In general, the 3- and 4-monosubstituted benzoates 7–16 were inactive, as were the di- and trisubstituted benzoates 17–22 and 36–41. The results indicate that in aromatic esters of dialkylaminoalkanols and 1-methyl-4-piperidinol an aryl or aralkyl substituent at the 2-position of the aromatic ring promotes activity.

Several of the piperidinol esters, of the general type presented in Table I, have analgesic activity in the codeine–morphine range by the mouse hot-plate method, but in general display no physical dependence liability of the morphine type in monkeys.[4–6] In addition, these esters show little or no binding to the opiate receptor in rat brain homogenates.[5,6] Two of the more potent analgesics differ from morphine in that they decrease spontaneous motor activity of mice.[7] The site of action of such esters in the

(4) Waters, J. A. *J. Med. Chem.* 1977, *20*, 1094.
(5) Waters, J. A. *J. Med. Chem.* 1977, *20*, 1496.
(6) Waters, J. A. *J. Med. Chem.* 1978, *21*, 628. Cheng, C.-Y.; Brochmann-Hanssen, E.; Waters, J. A. *J. Med. Chem.* 1982, *25*, 145.

Table I. Anticonvulsant Activity (MES) and Inhibition of Binding of Ligands to the Voltage-Dependent Sodium Channel ([^{3}H]BTX) or to the GABA–Receptor/Chloride Channel ([^{3}H]Diazepam, [^{3}H]Muscimol, [^{35}S]TBPS)

compd	EC_{50} MES, mg/kg	IC_{50} [^{3}H]BTX, μM	% inhibn or stimuln[a] of binding at 100 μM drug		
			[^{3}H]diazepam	[^{3}H]muscimol	[^{35}S]TBPS
2	N[b]	≫10 (-3)[c]			
5	N	3.6			
6	25	5.4	50	27	-50
8	N	>10 (16)			
9	<100[d]	>10 (24)			
10	N	10.0			
15	N	>10 (31)			
21	N	>10 (10)			
24	12	~10 (46)			
27	N	>10 (21)			
29	<30	3.4			
32	<100	~10 (49)			
33	N	>10 (38)	46	21	-1
35	26	>10 (44)	37	35	-37
40	46	>10 (29)	37	57	-3
44	N	≫10 (4)			
51	<100	>10 (38)			
52	N	3.8			
53	28	3.2	42	40	-50
54	<300	3.9			
55	<30	6.0			
56	10	7.5	26	31	4
57	7.8	4.6	51	59	47
58	<30	6.8			
59	N	>10 (20)			
63	N	≫10 (0)	-14	10	-9
64	N	11.5			
65	N	3.0			
66	<300	5.0			
phenytoin	9.5	18.5			
phenobarbital	22	≫10 (5.4)			
methsuximide	76				

[a] Stimulation is indicated by minus sign before the number. [b] No anticonvulsant activity at 300 mg/kg. [c] Percent inhibitions at 10 μM drug are in parentheses; see footnote *a*. [d] In a preliminary evaluation, a mouse was protected from convulsion at the dose indicated.

central nervous system remains unclear. The highly active anticonvulsants **6** and **40** were inactive as analgesics, while the most active analgesics, **21** and **51**,[4,6] were inactive as anticonvulsants.

In the rotorod ataxia assay, a test for minimal central neurotoxicity, the 2-phenethylbenzoate **6** (TD_{50} = 50), 2-phenoxymethylbenzoate **35** (TD_{50} = 49), and 2-phenethylbenzoate **53** (TD_{50} = 50) were somewhat more toxic than phenytoin (see the Experimental Section). 2,4,5-Trimethyl **40** (TD_{50} = 118) and 2,4,6-trimethylbenzoate **41** (TD_{50} = 122) showed less neurotoxicity than phenytoin and phenobarbital, which had TD_{50} = 66 and 69, respectively. 2-Phenylbenzoate **24**, (TD_{50} = 35) and 2-phenylbenzoates of the open-chained amino alcohols (**56**, TD_{50} = 31 and **57**, TD_{50} = 20) exhibited greater neurotoxicity than the controls. It should be pointed out that neurotoxicity is generally decreased in compounds substituted at the 4-position of the aromatic ring. This is illustrated by the lower toxicity of the 2,4-dimethylbenzoate **18** and the 3,4-dimethylbenzoate **20** in comparison with that of their lower homologues, the 2-methylbenzoate **1** and the 3-methylbenzoate **7**, respectively. 4-Iodobenzoate (**26**) and 4-phenylbenzoate (**32**) also showed reduced neurotoxicity in the preliminary rotorod assay (see Experimental Section; rotorod toxicity at 30 min).

A set of compounds with activity as anticonvulsants and two inactive compounds were evaluated as inhibitors of binding of ligands to the GABA–receptor/chloride channel complex. This complex is the site of action of many compounds having convulsant or anticonvulsant activities.[8-10] All of the compounds tested that were active in the in vivo MES assay (**6**, **35**, **40**, **53**, **56**, **57**) were relatively weak antagonists of binding of [^{3}H]diazepam (Table I): IC_{50} values were estimated to be in the 100–150 μM range. The IC_{50} value for compound **57** was determined to be 167 μM. One compound inactive in the MES assay (**33**) showed a similar potency in the [^{3}H]diazepam binding assay to the MES-active compounds, while another MES-inactive compound (**63**) was inactive in the binding assay. The above compounds were also relatively weak antagonists of binding of [^{3}H]muscimol causing 20–60% inhibition at a concentration of 100 μM (Table I). The IC_{50} values for compounds **40** and **57** were 76 and 73 μM, respectively. The MES-inactive compound **63** was nearly inactive in this binding assay. Remarkably, certain of the compounds (**6**, **35**, **53**) at a concentration of 100 μM enhanced binding of [^{35}S]TBPS to sites on the chloride channel, while others either had nearly no effect (**33**, **40**, **56**, **63**) or in the case of **57** inhibited binding by 50%. This phenomenon was not investigated further, since binding to none of the sites on the GABA–receptor/chloride channel complex appeared predictive of the MES anticonvulsant activity of these esters. Indeed, it appears unlikely from these results that the anticonvulsant activity of the piperidinol and (dialkylamino)alkanol esters are due primarily or at all to interactions at sites on the GABA–chloride channel complex.

A more extensive set of active and inactive compounds were evaluated vs. binding of a batrachotoxin analogue to

(7) Harris, L. S.; Aceto, M. D.; May, E. L.; Waters, J. A.; Dewey, W. L. *Int. Congress Pharmacol. 7th (Paris)* **1978**, Abstr. 1149, 447.
(8) Weissman, B. A.; Cott, J.; Bolger, G. T.; Weber, K. H.; Horst, W. D.; Paul, S. M.; Skolnick, P. *J. Neurochem.* **1985**, *44*, 1494.
(9) Yuneda, Y.; Kiurayama, K. *Brain Res.* **1980**, *197*, 554.
(10) Supavilai, P.; Karobath, M. *Eur. J. Pharmacol.* **1983**, *91*, 145.

Table II. Synthetic Summary of 4-Piperidinol Esters

compd	synth method[a]	mp, °C	formula[b]	recrystn solvent
18	B	233–235	$C_{15}H_{21}NO_2$·HCl	acetone–EtOH
19	A	194–195	$C_{15}H_{21}NO_4$·HCl	acetone–EtOH
24	A	185–187	$C_{19}H_{21}NO_2$·HCl	acetone
28	B	208–214	$C_{15}H_{21}NO_2$·HCl	acetone–EtOH
33	A	235–237	$C_{20}H_{21}NO_3$·HCl	acetone–EtOH
34	A	238–241	$C_{20}H_{20}NO_3ClF$·HCl·0.5H_2O	acetone–EtOH
35	A	198–200	$C_{20}H_{23}NO_3$·HCl	acetone–EtOH
46	D	144–146	$C_{15}H_{21}NO_2$·HCl	acetone
47	D	150–153	$C_{21}H_{25}NO_2$·HCl	acetone
48	C	110–115	$C_{20}H_{23}NO_2$·HCl·0.75H_2O	acetone
49	D	165–167	$C_{21}H_{25}NO_2$·HCl	C_6H_6
50	D	176–194	$C_{17}H_{19}NO_2$·HCl	acetone–EtOH
51	C	200–202	$C_{17}H_{19}NO_2$·HCl	acetone–EtOH
52	D	191–192	$C_{20}H_{23}NO_2$·HCl·H_2O	acetone
53	D	152–154	$C_{22}H_{27}NO_2$·HCl	acetone–Et_2O
54	D	107–110	$C_{21}H_{27}NO_2$·HCl	acetone
55	D	104–107	$C_{22}H_{29}NO_2$·HCl	acetone–Et_2O
56	D	109–111	$C_{19}H_{23}NO_2$·HCl	acetone
57	D	120–123	$C_{20}H_{25}NO_2$·HCl	acetone
58	D	139–141	$C_{20}H_{25}NO_2$·HCl	acetone–EtOH
65	A	209–213	$C_{17}H_{29}NO_2$·HCl·0.25H_2O	acetone–EtOH
66	A	176–178 and 211–213	$C_{17}H_{29}NO_2$·HCl	acetone–EtOH

[a] The compounds were obtained by the general synthetic procedure described in ref 6: (A) the ester HCl was obtained by converting the crude HCl to the free base and subsequent treatment with ethereal HCl; (B) the ester HCl was obtained by addition of C_6H_6 to the reaction mixture; (C) the ester HCl was obtained by addition of acetone to the reaction mixture; (D) the crude ester HCl was chromatographed on silica gel. [b] Analytical results obtained within ±0.4% of the theoretical values for C, H, and N.

sodium channel sites. Anticonvulsants, such as phenytoin, do inhibit binding of the batrachotoxin analogue,[11,12] and activity at voltage-dependent sodium channels has been proposed as one factor involved in the anticonvulsant activity of phenytoin and certain other drugs (see ref 12). Phenytoin has an IC_{50} value of 18.5 μM vs. binding of [^{3}H]BTX to sites on the sodium channel (Table I). All of the active anticonvulsant compounds with ED_{50} values of 7.8–28 in the MES assay, namely **57**, **56**, **24**, **6**, **35**, and **53**, or with primary protections at 30 mg/kg in the MES assay, namely **29**, **55**, and **58**, had IC_{50} values of 3.2–7.5 μM in inhibiting [^{3}H]batrachotoxin benzoate binding to the sodium channel. IC_{50} values were not obtained for compounds **24** and **35**, but at 10 μM these compounds gave inhibition of 44% and 46% of control, respectively. All nine of the above named compounds contain some type of phenyl substitution of the 2-position of the benzoate ring, i.e., C_6H_5–, $C_6H_5CH_2$–, $C_6H_5(CH_2)_2$–, or $C_6H_5OCH_2$–; i.e., all could be construed to be able to assume structures related to that of phenytoin (see structure II). However, activity at the sodium channel, as assessed with the binding assay, is not predictive of anticonvulsant activity in the present series of compounds, since several compounds (**5**, **10**, **64**, **65**) that are inactive at 300 mg/kg in the MES assay were very potent with IC_{50} values of 3.0–11.5 μM as antagonists of [^{3}H]BTX binding. Certain compounds (**2**, **8**, **21**, **27**, **44**, **59**, **63**) inactive in the MES assay were inactive or had low activity as antagonists of [^{3}H]BTX binding. Clearly, although interactions with sites on sodium channels may play a role in the anticonvulsant properties of phenytoin, and perhaps of the present series of aromatic esters, other sites of action and/or pharmacokinetic factors also must be important to the in vivo potency and efficacy.

A major molecular feature for many anticonvulsant drugs is that they possess two hydrophobic regions and two electron-rich centers interconnected with similar spatial distances.[1] Certain of the present compounds also possess these features. The three most MES active compounds (**57**, **56**, and **24**), which have strong affinities for voltage-sensitive sodium channels, have phenyl groups attached directly to the 2-position of the benzoate ring. Drieding models show a 4.8-Å distance between the centers of the two lipophilic 5-diphenyl rings in phenytoin, while **57**, **56**, and **24** show 4.2 Å between the centers of the benzoate and 2-phenyl ring systems. In addition, the distance from the centers of the 2-phenyl groups of **57**, **56**, and **24** placed in a planar fashion to a perpendicularly positioned keto group is 3.9 Å, identical to the distance from the 5-phenyl center to the 4-keto function of phenytoin. In a more rigid system found in cyheptamide, the plane of the amide group is nearly perpendicular to each of the two fused phenyl ring planes.[13] Although the tertiary amine function in the present compounds is chemically dissimilar to the amide N-1 of phenytoin, if the methylene groups in the general structure II are staggered, the nitrogen to carbonyl oxygen distance of **57** and **56** assumes the same value as that present in the N-1 and carbonyl-4 of phenytoin, i.e., 3.6 Å. Ester groups present in the potent 2-phenylbenzoates could serve as electron-donating groups similar to that postulated for the amide grops of carbamazepine and cyheptamide,[13] or as hydrogen-bond acceptors for a hydrogen-donating receptor site postulated on the sodium channel.[12] Smythies[14] and Poupaert et al.[15] also have suggested that the C-2 carbonyl and NH-3 alternate C(=O)NH groups of phenytoin may be involved in hydrogen bonding to a putative receptor.

Experimental Section

Chemistry. Melting points were taken on a Köfler hot stage and are corrected. Analytical results obtained were within ±0.4% of theoretical values. Some of the compounds have been synthesized previously as follows: compound **59**;[4] compounds **39**, **61**, and **62**,[5] compounds **1–17**, **20–23**, **25–27**, **29–32**, **36–38**, **40–45**, **63**, and **64**.[6] New compounds, with the exception of **60**, were syn-

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(12) Willow, M.; Catterall, W. A. *Mol. Pharmacol.* **1982**, *22*, 627–635.
(13) Codding, P. W.; Lee, T. A.; Richardson, J. F. *J. Med. Chem.* **1984**, *27*, 649–654.
(14) Smythies, J. R. In Reference 1, p 207.
(15) Poupaert, D. H.; Vandervorst, D.; Guiot, P.; Moustafa, M. M. M.; Dumont, P. *J. Med. Chem.* **1984**, *27*, 76.

thesized by the general procedure described in ref 6 and are summarized in Table II.

1-Methyl-4-piperidinol 3,5-Dimethyl-4-acetylpyrrole-2-carboxylate (60). 1-Methyl-4-piperidinol (15.0 g, 0.13 mmol), contained in a large test tube with a plug of cotton at the top, was placed in a warm oil bath (75–85 °C). To the warm, magnetically stirred piperidinol was added 300 mg of finely cut pieces of metallic sodium. Stirring and heating were continued until all of the sodium had dissolved (4 h). To 3.0 g (0.014 mol) of ethyl 3,5-dimethyl-4-acetylpyrrole-2-carboxylate in a 50-mL round-bottomed flask was added the sodium piperidinolate solution described above. The magnetically stirred mixture was then heated on an oil bath maintained at 170–180 °C for 2.5 h. The flask was then fitted with a distillation sidearm and the excess piperidinol distilled (20 mm) from the reaction mixture at an oil bath temperature not greater than 200 °C. The thick, reddish brown residue was cooled, dissolved in CH_2Cl_2–MeOH and chromatographed on 90 g of silica gel. Elution with 9–12% MeOH in CH_2Cl_2 gave the desired ester **30** as a yellow solid, homogenous by TLC (silica gel GF; $CHCl_3$–MeOH 7:3). The solid was dissolved in 600 mL of ether and filtered to remove a small amount of insoluble residue. To the ethereal solution was added ethereal HCl until no more precipitation occurred. After decantation of the ether, the solid was triturated several times with fresh portions of ether and then collected by suction filtration. The yield of colorless **60** HCl salt was 2.767 g (63%). Recrystallization from acetone–EtOH gave the analytical sample: mp 256–257 °C. Anal. ($C_{15}H_{22}N_2O_3$·HCl) C, H, N.

Biochemistry. [^{3}H]Diazepam binding was ascertained with rat brain cerebral cortical membranes essentially as described.[8] Incubations were in 50 mM Tris buffer, pH 7.4, with 5 nM [^{3}H]diazepam (76 Ci/mmol) in a total volume of 1 mL with about 1000 μg of protein for 30 min at 0 °C. Nonspecific binding was determined with 10 μM clonazepam. [^{3}H]Muscimol binding was ascertained with rat brain membranes essentially as described.[9] Incubations were in 50 mM Tris–citrate buffer, pH 7.1, with 5 nM [^{3}H]muscimol (11.5 Ci/mmol) in a total volume of 1 mL with about 1000 μg of protein for 30 min at 0 °C. Nonspecific binding was determined with 100 μM GABA. [^{35}S]TBPS binding was ascertained with rat cerebral cortical membranes essentially as described.[10] Incubations were in 50 mM Tris–citrate buffer, pH 7.5, containing 200 mM NaCl with 2 nM [^{35}S]TBPS (108 Ci/mmol) for 90 min at 23 °C with about 1000 μg of protein. Nonspecific binding was determined with 10 μM picrotoxin. All of the above incubations were terminated by rapid filtration through GFB filters, followed by one 5-mL wash with buffer. The radioactivity retained on the filters was determined in hydrofluor. [^{3}H]Batrachotoxinin A benzoate binding was determined as described.[11] The results of binding assays are presented in Table I.

Pharmacology. Antiepileptic activity and neurological toxicity assays were carried out by the Antiepileptic Drug Development Program, Epilepsy Branch, National Institute of Neurological and Communicative Disorders and Stroke, National Institute of Health, Bethesda, MD. The compounds were solubilized in 0.9% sodium chloride solution and administered intraperitoneally in a volume of 0.01 mL/g of body weight to adult male Carworth Farms No. 1 mice. All compounds were tested at these dose levels (30, 100, and 300 mg/kg) at 30 min after administration. Maximal electroshock seizures (MES) were elicited with a 60-cycle alternating current of 50 mA (5–7 times that necessary to elicit minimal electroshock seizures) delivered for 0.2 s via corneal electrodes. A drop of 0.9% saline is instilled in the eye prior to the application of electrodes in order to prevent the death of the animal. Abolition of the hind limb tonic extension component of the seizure is defined as protection. Subcutaneous pentylenetetrazole seizures (scMet) were produced by 85 mg/kg of pentylenetetrazole (produces seizures in greater than 97% of mice) administered as a 0.5% solution in the posterior midline. Failure to observe even a threshold seizure (a single episode of clonic spasm of at least 5-s duration) is defined as protection. The rotorod test was used to evaluate neurotoxicity. The animal is placed on a 1-in.-diameter knurled plastic rod rotating at 6 rpm. Normal mice can remain on a rod rotating at this speed indefinitely. Neurologic toxicity is defined as the failure of the animal to remain on the rod for 1 min. Anticonvulsant quantification, i.e., the median effective dose (ED_{50}) in the MES test and the median neurotoxic dose (TD_{50}), were determined on select compounds displaying sufficient antiepileptic activity and low neurotoxicity from the above primary evaluations.

Maximal Electroshock Assay. Preliminary evaluation of the MES activity of the compounds not listed in Table I is summarized below. Protection at 30 min at a dose of 100 mg/kg was shown for compounds **18**, **48**, and **49**; protection at 300 mg/kg was shown for compounds **13** and **26**. Inactive compounds were **1**, **3**, **4**, **7**, **11**, **12**, **14**, **16**, **17**, **19**, **20**, **22**, **25**, **28**, **30**, **31**, **34**, **36–39**, **42**, **43**, **45–47**, **50**, and **60–62**. Compound **41** gave an ED_{50} of 84.5.

Rotorod Toxicity Assay. Preliminary evaluation of the rotorod toxicity (at 30 min) of the compounds is summarized below. Compounds showing no toxicity at 30, 100, and 300 mg/kg were **16**, **46**, **61**, and **63**. Compounds showing no toxicity at 30 and 100 mg/kg were **8–10**, **14**, **15**, **17**, **18**, **20**, **22**, **23**, **26**, **32**, **38**, **42–45**, **50**, **54**, **64**, and **66**. Compounds showing no toxicity at 30 mg/kg only were **1–5**, **7**, **11–13**, **19**, **21**, **23**, **25**, **27–31**, **33**, **34**, **37**, **39**, **47–49**, **51**, **58–60**, **62**, and **65**. Compounds **24**, and **55–57** were toxic at 30 mg/kg. TD_{50}'s (mg/kg) were determined on the following compounds: **6** = 50, **24** = 35, **35** = 49, **40** = 118, **41** = 122, **53** = 50, **56** = 31, and **57** = 20. The standards show TD_{50}'s as follows: phenytoin = 66, phenobarbital = 69, methsuximide = 188.

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Registry No. 1, 102698-42-0; **2**, 102698-43-1; **3**, 102698-44-2; **4**, 102698-45-3; **5**, 102698-46-4; **6**, 102698-47-5; **7**, 102698-48-6; **8**, 102698-49-7; **9**, 102698-50-0; **10**, 102698-51-1; 11, 101114-86-7; **12**, 102698-52-2; **13**, 101866-85-7; **14**, 102698-53-3; **15**, 100380-21-0; **16**, 102698-54-4; **17**, 102698-55-5; **18**, 102698-93-1; **18** (HCl salt), 102698-56-6; **19**, 102698-94-2; **19** (HCl salt), 102698-57-7; **20**, 102698-58-8; **21**, 92703-09-8; **22**, 102698-59-9; **23**, 102698-60-2; **24**, 102698-95-3; **24** (HCl salt), 80239-79-8; **25**, 102698-61-3; **26**, 102698-62-4; **27**, 102698-63-5; **28**, 102698-96-4; **28** (HCl salt), 102698-64-6; **29**, 102698-65-7; **30**, 102698-66-8; **31**, 102698-67-9; **32**, 102698-68-0; **33**, 102698-97-5; **33** (HCl salt), 102698-69-1; **34**, 102698-98-6; **34** (HCl salt), 102698-70-4; **35**, 102698-99-7; **35** (HCl salt), 102698-71-5; **36**, 102698-72-6; **37**, 1024-83-5; **38**, 92782-49-5; **39**, 66724-18-3; **40**, 102698-73-7; **41**, 102698-74-8; **42**, 102698-75-9; **43**, 99553-61-4; **44**, 102698-76-0; **45**, 102698-77-1; **46**, 102699-00-3; **46** (HCl salt), 102698-78-2; **47**, 102699-01-4; **47** (HCl salt), 102698-79-3; **48**, 97762-23-7; **48** (HCl salt), 1952-14-3; **49**, 102699-02-5; **49** (HCl salt), 102698-80-6; **50**, 102699-03-6; **50** (HCl salt), 102698-81-7; **51**, 102699-04-7; **51** (HCl salt), 102698-82-8; **52**, 102699-05-8; **52** (HCl salt), 102698-83-9; **53**, 102699-06-9; **53** (HCl salt), 102698-84-0; **54**, 102699-07-0; **54** (HCl salt), 102698-85-1; **55**, 102699-08-1; **55** (HCl salt), 102698-86-2; **56**, 102699-09-2; **56** (HCl salt), 102698-87-3; **57**, 102699-10-5; **57** (HCl salt), 102698-88-4; **58**, 102699-13-8; **58** (HCl salt), 102698-89-5; **59**, 62967-21-9; **60**, 102699-12-7; **60** (HCl salt), 102699-11-6; **61**, 64219-66-5; **62**, 102698-90-8; **63**, 80047-12-7; **64**, 102698-91-9; **65**, 102724-51-6; **65** (HCl salt), 102698-92-0; Na, 7440-23-5; Cl^-, 16887-00-6; 1-methyl-4-piperidinol, 106-52-5; ethyl 3,5-dimethyl-4-acetylpyrrole-2-carboxylate, 2386-26-7.