dichloropyrimidine and 1,2,3,4-tetrahydro-1-naphthylamine hydrochloride by the method of preparation of **59** with ethyl acetate–hexanes (1:1) as the flash chromatography elution solvent to give 1.46 g (20%) of **60**, mp 221–222 °C. Anal. ($C_{14}H_{14}N_3Cl$) C, H, N.

(B) Benzodiazepine Binding Assay. Male Sprague-Dawley rats weighing between 110 and 220 g were decapitated, and the brains were rapidly removed and chilled in ice-cold 0.9% NaCl. The cerebellum and pons medulla were removed, and the rest of the brain was homogenized in 20 volumes of 0.32 M sucrose in a Potter–Elvehjem homogenizer fitted with a Teflon pestle with a clearance of 0.25 mm. The homogenate was centrifuged at 1000*g* for 10 min at 4 °C. The supernatant was decanted and centrifuged at 30000*g* for 15 min. The pellet was frozen at a −60 °C for later use.

The frozen pellet was resuspended at the time of use in 20 volumes of Tris-HCl buffer, pH 7.5. A 0.5-mL aliquot of this membrane suspension was incubated with 1.5×10^{-9} M [^{3}H]-diazepam and enough Tris-HCl buffer (pH 7.5) to give a final volume of 2.0 mL for 30 min at 4 °C. Ten milliliters of ice-cold buffer was added to each tube, and the samples were filtered through Whatman GF/C glass filters. The filters were washed with an additional 10 mL of the ice-cold buffer, removed, placed in scintillation vials together with 10.0 mL of Bray's solution, and counted in a Packard Tri-Carb liquid scintillation counter, Model 3320.

Nonspecific [^{3}H]diazepam binding was determined from parallel samples containing 3×10^{-6} M cold diazepam. Specific binding is defined as the difference between total and nonspecific binding and was ~90% of the total binding. This procedure is a modification of the techniques of Braestrup and Squires.[2]

(C) Pharmacology. Ovariectomized Long-Evans (Charles River) rats, deprived of food 22 h/day, worked for food pellets in daily 1-h sessions on a modified Geller–Seifter conflict schedule.[14,15] The operant chambers (Coulbourn Instruments) were located in sound-attenuating enclosures and were equipped with a lever manipulandum, a cue light above the lever, a house light, a pellet dispenser delivering 45-mg pellets (BioServ, Inc.) to a lighted feeder bin, and a water spout. Response-contingent footshock was delivered to a grid floor from a Coulbourn shock generator during the conflict portion of the schedule. Environmental control and data acquisition were performed by a Data General Nova 3/12 minicomputer via an Interact interface (BRS/LVE).

Food reinforcement was delivered on a mult VI 2-min/CRF (food + shock) schedule consisting of four 12-min periods of variable interval reinforcement during which a lever press produced a food pellet every 2 min on the average, alternating with four 3-min periods of continuous reinforcement, signaled by a cue light, during which every lever press produced a pellet and a footshock. Shock level was 0.00 mA for the first response in each conflict period and was increased by 0.05 mA for each successive response in the period.

In general, compounds were administered by oral gavage (po) in a 0.5% methylcellulose suspension 60 min prior to the operant session. Several compounds were also tested following intraperitoneal administration. Results on days of drug administration are expressed as a percent change in conflict responses from the preceding day and represent the mean and SEM of at least four rats.

Acknowledgment. We acknowledge the technical assistance of F. L. M. Tang and A. Russell in performing the receptor binding assays. Some NMR spectra were provided by Dr. B. S. Hurlbert and his staff. We acknowledge the excellent technical assistance of Mrs. Alice Melton. We thank T. Cozart, J. Appleton, and D. Tabon for assistance in preparation of the manuscript and Mr. Allen Jones for proofreading the manuscript.

Synthesis and Structure–Activity Relationship of Substituted Tetrahydro- and Hexahydro-1,2-benzisothiazol-3-one 1,1-Dioxides and Thiadiazinones: Potential Anxiolytic Agents

Magid Abou-Gharbia,* John A. Moyer, Usha Patel, Michael Webb, Guy Schiehser, Terrance Andree, and J. Thomas Haskins

Medicinal Chemistry and Experimental Therapeutics Divisions, Wyeth-Ayerst Research, CN-8000, Princeton, New Jersey 08543-8000. *Received August 25, 1988*

Several novel substituted tetrahydro- and hexahydro-1,2-benzisothiazol-3-one 1,1-dioxides and thiadiazinones were prepared and examined in a series of in vitro and in vivo tests to determine their pharmacological profile. Most compounds were orally active in blocking the conditioned avoidance response (CAR) but did not antagonize apomorphine-induced stereotyped behavior. Several compounds demonstrated moderate to high affinity for the 5-HT_{1A} receptor binding site, with compounds **37** and **38** containing 2-pyrimidinylpiperazinyl and [3-(trifluoromethyl)phenyl]piperazinyl moieties and compound **47** containing the 2-pyrazinylpiperazinyl moiety displaying the highest affinity (K_i values of 10, 4, and 9 nM, respectively). Compound **37**, 3-[4-[4-(2-pyrimidinyl)-1-piperazinyl]butyl]hexahydro-4,7-etheno-1*H*-cyclobut[*f*]-1,2-benzisothiazol-3(2*H*)-one 1,1-dioxide, buspirone, and ipsapirone showed similarities in their neurochemical and behavioral profiles. They were similar in potency in blocking CAR with AB_{50} values of 39, 32, and 42 mg/kg, respectively. They also demonstrated high affinity and selectivity for the 5-HT_{1A} receptor site (K_i = 10 nM) and exhibited partial agonist/antagonist activity in the serotonin syndrome test. In addition, compound **37** inhibited apomorphine-induced climbing behavior much more potently (ED_{50} of 3.4 mg/kg) than stereotyped behavior (ED_{50} of 32.2 mg/kg) and will be evaluated further. Structure–activity relationships within this series of compounds are discussed.

The discovery of buspirone as a non-benzodiazepine anxiolytic agent revolutionized the drug therapy of anxiety and has led to the synthesis of several compounds possessing high 5-HT_{1A} affinity, many of which are under development as anxiolytic agents.[1,2] In addition to treatment of anxiety, 5-HT_{1A} partial agonists such as gepirone (1) are now being examined for their mixed activity as anxiolytic–antidepressant agents.[3] The therapeutic potential of the 5-HT_{1A} agonists in the treatment of multi-CNS disorders was recently extended to the development of compounds that may have antipsychotic and

(1) Rowan, M. J.; Anwyl, R. *Eur. J. Pharmacol.* **1986**, *132*, 93.
(2) Hirose, A.; Kato, T.; Shimizu.; Nakamura, M. N.; Katsube, J. *Jpn. J. Pharmacol.* **1986**, *40*, 114.
(3) McMillen, B. A.; Scott, S. M.; Williams, H. L.; Sanghera, M. K. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1987**, *335*, 454.

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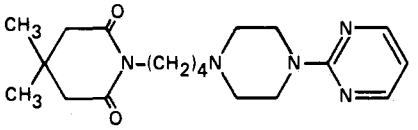
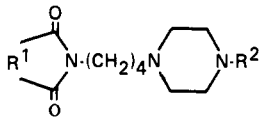
anxiolytic activity.[4,5] Our interest in the development of non-benzodiazepine anxiolytic agents that exhibit weak or moderate activity at the D_2 receptor binding sites and display high affinity for 5-HT_{1A} receptor sites has recently led to the synthesis of several polycyclic aryl- and heteroarylpiperazinyl imides **2**.[6] One of these, Wy-47,846 [2-[4-[4-(2-pyrimidinyl)-1-piperazinyl]butyl]hexahydro-4,7-etheno-1*H*-cyclobut[*f*]isoindole-1,3-(2*H*)-dione (**3**)], demonstrated high affinity at the 5-HT_{1A} receptor binding site and was 3 times more potent than buspirone in inhibiting the conditioned avoidance response (CAR).[6] Buspirone and compound **3** inhibited serotonergic neuronal activity in the dorsal raphe nucleus, having ID_{50} values of 0.03 and 0.07 mg/kg iv, respectively.[7] Buspirone, however, is not completely selective for the 5-HT system as it possesses a much more potent dopaminergic component than **3**.



1



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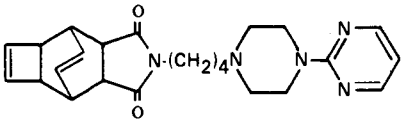
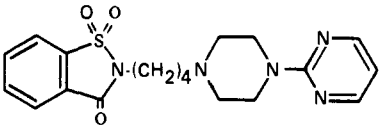


3



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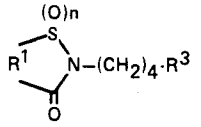


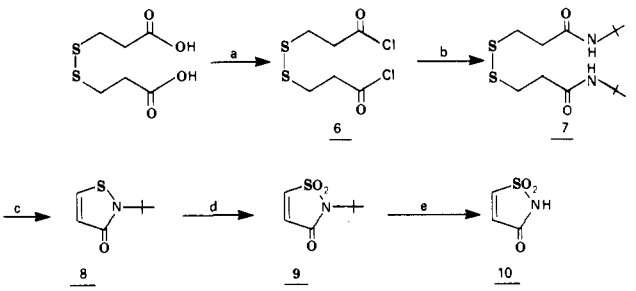
5

As an extension of our earlier investigations,[6] we examined the effect of substituting the imide carbonyl functionality in **2** with the nonclassical isosteric sulfonyl group that exists in the anxiolytic agent ipsapirone (**4**), to determine its effect on the receptor binding affinities and behavioral profile of these isothiazolone derivatives.

Our previous structure–activity relationship studies on a series of polycyclic imides[6] revealed that lipophilic heteroarylpiperazine derivatives have high affinity for 5-HT_{1A} receptor sites and weak affinity for D_2 sites. These compounds also possessed a lower potential for extrapyramidal side effect (EPS) liability than that exhibited by less lipophilic compounds. Therefore, it was anticipated that isothiazolone 1,1-dioxides with appropriate R^1 and bearing appropriate R^2 substituents might demonstrate a favorable receptor binding profile and possess reasonably potent activity in the CAR test with lower potential for side effects.

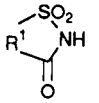
This paper describes the synthesis and behavioral and biochemical profile of a novel series of polycyclic tetrahydro- and hexahydro-1,2-benzisothiazolone 1,1-dioxides and thiadiazinones (**5**).

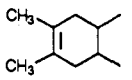
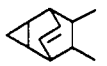
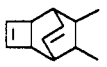
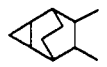
Scheme I[a]



[a] (a) $SOCl_2$; (b) H_2N-*t*-Bu, OH^-; (c) SO_2Cl_2; (d) *m*-CPBA; (e) CF_3COOH, Δ.

Table I. 2-Unsubstituted 1,2-Benzisothiazol-3-one 1,1-Dioxides



no.	R^1	mp, °C	% yield	formula
11		185–187	30	$C_8H_9NSO_3$
12		197–199	45	$C_9H_{11}NSO_3$
13		108–110	92	$C_9H_{13}NSO_3$
14		172–180	35	$C_{10}H_{11}NSO_3$
15		207–213	29	$C_{11}H_{11}NSO_3$
16		125–128	53	$C_8H_{11}NSO_3$
17		140–144	45	$C_{10}H_{13}NSO_3$
20		234–236	38	$C_{17}H_9NSO_3$

Chemistry

Literature procedures were adapted for the synthesis of the monocycle 2-*tert*-butylisothiazolin-3-one 1,1-dioxide (**10**)[8,9] in five steps as illustrated in Scheme I. 3,3′-Dithiobis(propionic acid) was converted to the corresponding diacid chloride **6** in 85% yield by refluxing in thionyl chloride. The reaction of **6** with *tert*-butylamine afforded the corresponding bis(*tert*-butylamide) derivative **7** in 90% yield, which upon refluxing with sulfuryl chloride afforded

(4) Hibert, M.; Gittos, D.; Middlemiss, D. *International Symposium on Medicinal Chemistry*; West Berlin, 1986; Abstract IX, 68.

(5) "Umespirone", a diazabicyclic imide derivative being developed as an antipsychotic agent which may have anxiolytic activity because of its high 5-HT_{1A} affinity and moderate dopaminergic effect (Int. Conf. Behav. Pharmacol. 5-HT, Amsterdam, 1987).

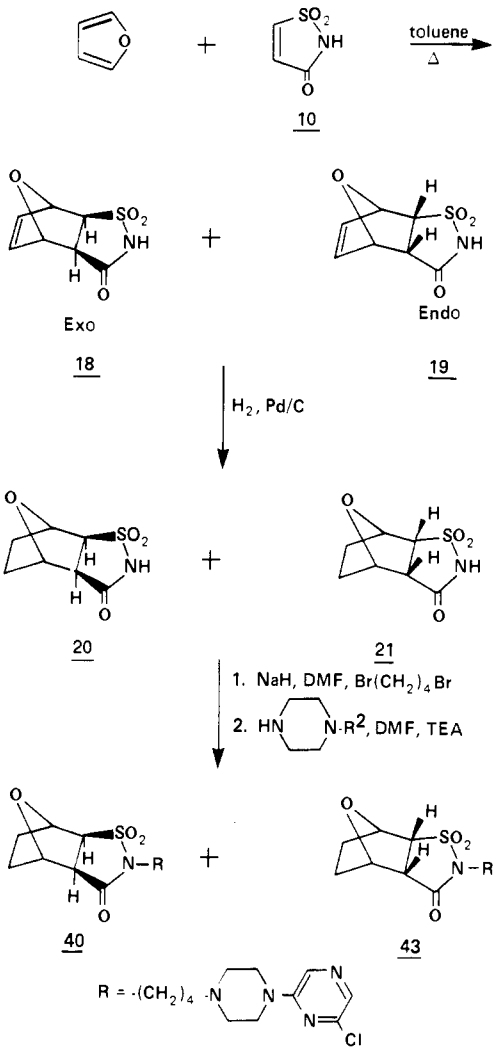
(6) Abou-Gharbia, M.; Patel, U.; Webb, M.; Moyer, J.; Andree, T.; Muth, E. *J. Med. Chem.* 1988, *31*, 1382.

(7) Haskins, J. T.; Jones, D. E.; Sacher, M. D.; Muth, E. A. *Neuroscience* 1987, *22* (Suppl.), 1051 p.

(8) Lewis, S. N.; Miller, G. A.; Hausmann, M.; Szamborski, E. C. *J. Heterocycl. Chem.* 1971, *8*, 591.

(9) Lewis, S. N.; Miller, G. A.; Hansmann, M.; Szamborski, E. C. *J. Heterocycl. Chem.* 1971, *8*, 581.

Scheme II

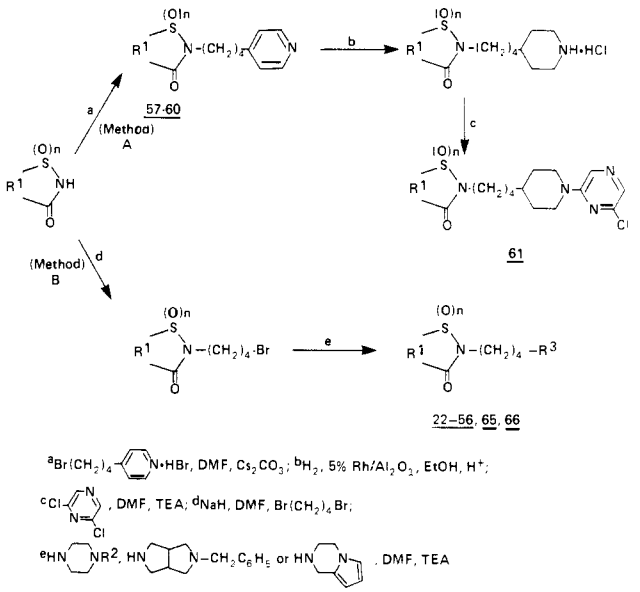


2-*tert*-butyl-4-isothiazolin-3-one (**8**) in 57% yield. The 1,1-dioxide derivative **9** was prepared in 45% yield via *m*-chloroperoxybenzoic acid oxidation of **8**. Deprotection of **9** was effected by refluxing **9** in trifluoroacetic acid for 2 h to afford **10** in 65% yield.

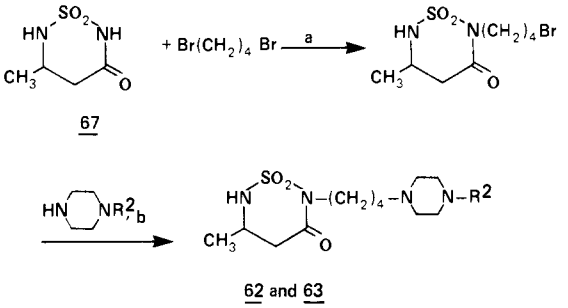
It has been reported that isothiazolin-3-one 1-oxide and its 1,1-dioxide analogues **10** undergo a Diels–Alder cycloaddition reaction with various dienes[10] while isothiazolin-3-one (**8**), the unoxidized version of **10**, being a poor dienophile, does not react with these dienes. The reaction of **10** with cyclopentadiene, 1,3-cyclohexadiene, 2,3-dimethyl-1,3-butadiene, 1,3-cycloheptadiene, and 1,3,5,7-cyclooctatetraene in refluxing toluene afforded the 2-unsubstituted 1,2-benzisothiazolin-3-one 1,1-dioxides (**11–15**) in reasonable yields (Table I). From the spectroscopic data and on the basis of Diels–Alder mechanistic considerations, the kinetically controlled endo cycloadducts **11–15** were predominantly formed. In contrast, the reaction of **10** with furan in refluxing benzene for 2 h afforded a 2:1 mixture of the unstable exo:endo adducts **18** and **19**, which were hydrogenated over Pd/C for 0.5 h in dry THF to afford the corresponding 2-unsubstitued hexahydroepoxy-1,2-benzisothiazolin-3-one 1,1-dioxides **20** and **21**, respectively. Saturated intermediates **16** and **17** were prepared via hydrogenation of **11** and **14** in a similar fashion.

The general procedure utilized in preparing 2-substituted benzisothiazolin-2-one 1,1-dioxides **2–54** (Table II) is shown in Scheme II. The synthesis of compounds **55–61**, **65**, and **66** (Table III) is illustrated in Scheme III (methods A and B). The appropriate 2-unsubstituted benzisothiazolones were reacted with 1,4-dibromobutane in DMF in the presence of sodium hydride to afford a bromobutyl intermediate, which was subsequently reacted either with the appropriately substituted aryl- or heteroarylpiperazines to give compounds **22–55**, **65**, and **66** or with the substituted octahydropyrrolopyrrole[11] to afford compound **56** (method B, Table III). Yields of all compounds in Table II were generally low, and no attempts were made to optimize yields. Scheme III also illustrates the general procedure for the synthesis of compounds in Table III, in which the 2-unsubstituted 1,2-benzisothiazolones were reacted with 4-pyridinylbutyl bromide hydrobromide[12] in DMF in the presence of cesium carbonate to afford compounds **57–60** (method A). Hydrogenation of **58** in ethanol in the presence of 5% Rh/Al_2O_3 followed by alkylation of the piperidinyl intermediate with 2,6-dichloropyrazine afforded **61** in an overall yield of 40%.

Scheme III[a]



Scheme IV[a]



[a] (a) DMF, NaH; (b) DMF, TEA.

Thiadiazinones **62** and **63** were prepared by reacting 5,6-dihydro-5-methyl-2*H*-1,2,6-thiadiazin-3(4*H*)-one 1,1-dioxide (**67**)[13] with 1,4-dibromobutane to afford the corresponding bromobutyl intermediate, which was subsequently reacted with the appropriate heteroarylpiperazine (Scheme IV). Unlike **62** and **63**, the spirothiazodiazone

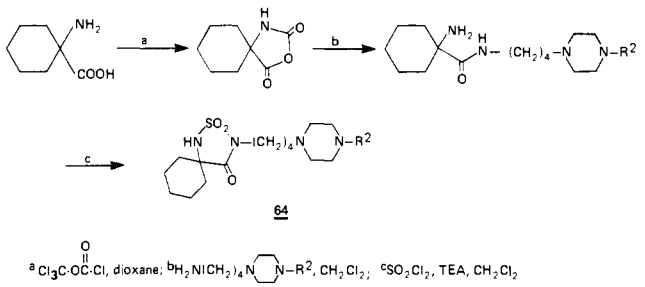
(10) Weiler, E. D.; Brennan, J. *J. Heterocycl. Chem.* **1978**, *15*, 1299.

(11) Loftus, P.; Wong, J. J. *J. Heterocycl. Chem.* **1983**, *20*, 321.

(12) Mioque, M.; Gautier, J. A. C. R. *C. R. Hebd. Seances Acad. Sci.* **1961**, *252*, 1299.

(13) Die, R.; Diez, J.; Garcia-Munoz, G.; Madronero, R.; Stud, M. *J. Heterocycl. Chem.* **1972**, *9*, 973.

Scheme V[a]



64

[a] $Cl_3C{-}O{-}C{-}Cl$, dioxane; [b] $H_2NCH_2)_4N$ $N{-}R^2$, CH_2Cl_2; [c] SO_2Cl_2, TEA, CH_2Cl_2

64 was prepared from the commercially available 1-amino-1-cyclohexanecarboxylic acid in a three-step synthesis, illustrated in Scheme V. 1-Amino-1-cyclohexanecarboxylic acid was reacted with trichloromethyl chloroformate (TCF) to afford the spirooxazolidinedione intermediate, which was subsequently reacted with the appropriate heteroarylpiperazine to afford an aminoacetanilide. Reaction with sulfuryl chloride yielded 64 in an overall yield of 23%.

Biological Results and Discussions

The affinity of these compounds for various central neuroreceptors was assessed by their ability to displace various receptor radioligands in vitro. All compounds were tested for their in vitro dopamine-2 (D_2) and serotonin-1 (5-HT_{1A}) receptor binding affinities. This was determined by measuring the ability of these agents to inhibit [^{3}H]-spiperone binding in limbic structures or to inhibit [^{3}H]-8-OH-DPAT binding in hippocampus. To further assess possible dopaminergic liability, compounds were tested for their ability to antagonize apomorphine-(APO-)induced behavior. Compounds were also tested for possible antipsychotic activity or non-benzodiazepine anxiolytic activity by measuring their abilities to block the response of rats trained to avoid electrical shock in shelf-jump and/or discrete trial conditioned avoidance response (CAR) tests after ip and/or oral administration.[14,15]

Compounds were considered to be inactive if ED_{50} values were greater than 60 mg/kg in the APO-induced stereotyped behavior test (high-dose procedure reported earlier)[16] and if AB_{50} values were greater than 40 mg/kg in the CAR paradigm. Biological data for all synthesized compounds and reference compounds buspirone and ipsapirone are shown in Tables II and III.

It is generally accepted that conflict behavior models, widely used to assess preclinical anxiolytic activity, are specific in determining activity of compounds acting at the benzodiazepine–GABA receptor complex and are less sensitive for non-benzodiazepine anxiolytic agents. Selected compounds were examined for their potential anxiolytic activity in serotonin syndrome tests.[17] This procedure assesses in vivo 5-HT_{1A} agonist/antagonist activity. Furthermore, the side effect potential of these compounds was examined in rotorod/ethanol interaction studies.[18,19]

Structure–Activity Relationships. We previously reported structure–activity relationships for the polycyclic aryl- and heteroarylpiperazinyl imide series.[6] In that series, four structural parameters dramatically affected receptor binding affinities and the behavioral profile of these compounds. These include the effects of polycycloalkyl modifications, heteroarylpiperazinyl variations, incorporation of a sulfonyl group into the polycycloalkyl moiety, and alteration of the piperazinyl moiety. Like polycyclic aryl- and heteroarylpiperazinyl imides, increasing the lipophilicity of the 1,2-benzisothiazol-3-one 1,1-dioxides enhanced their affinity for the 5-HT_{1A} receptor binding site. The effect of lipophilicity of these compounds, expressed as their octanol–water partition coefficients[20] (log p), on 5-HT_{1A} receptor affinity is shown in Table IV. Compound **37** was the most lipophilic analogue and demonstrated a high affinity for the 5-HT_{1A} receptor binding site, similar to that of ipsapirone (K_i = 10 nM). Although a correlation between lipophilicity and 5-HT_{1A} receptor affinity cannot be made from only four analogues, our findings are in agreement with our earlier investigation with polycyclic imides.[6]

Contrary to our previous findings with polycyclic imides,[6] compounds containing a pyrimidinyl moiety demonstrated the lowest affinity at D_2 receptor binding sites. Compound **39**, containing a pyrimidinyl moiety, was inactive at the D_2 site, whereas compound **46**, containing an *m*-chlorophenyl group in place of the pyrimidinyl moiety, displayed moderate affinity for D_2 receptor sites, producing 55% inhibition at 1 μM. Like polycyclic imides, incorporation of a 6-chloropyrazinyl moiety into the 1,2-benzisothiazol-3-one 1,1-dioxide and the thiadiazinone 1,1-dioxide skeleton produced or enhanced oral activity in the CAR paradigm. Compounds **29**, **39**, and **62** containing the 2-pyrimidinyl moiety were orally inactive in the CAR test, but oral activity was introduced by incorporating a 6-chloro-2-pyrazinyl moiety into their structure (**30**, **40**, and **63**). Incorporation of the serotonin mimetic substituted arylpiperazines into the 1,2-benzisothiazol-3-one 1,1-dioxides resulted in an enhancement of the affinity of these compounds for D_2 and 5-HT_{1A} receptor sites with little or no change in potency in inhibiting CAR, with the exception of compound **45**. For example, compound **38**, containing the [*m*-(trifluoromethyl)phenyl]piperazinyl moiety, was the most potent 5-HT_{1A} ligand with good D_2 potency as well. However, 5-HT_{1A} receptor selectivity was only observed with compound **45**, which demonstrated very low D_2 affinity and showed high affinity for the 5-HT_{1A} receptor binding sites (K_i = 22 nM). Replacement of the piperazinyl moiety with an octahydropyrrolyl or a piperidinyl moiety afforded compounds **56** and **61**, respectively. Both showed diminished in vitro receptor activity and diminished potency in the CAR test as well.

Apomorphine-Induced Stereotyped and Climbing Behavior. Most compounds were inactive in antagonizing high-dose APO-induced stereotyped behavior at doses up to 60 mg/kg ip, with the exception of compounds **33**, **37–40**, and **45** (Table V). Of these compounds, **37** was found to inhibit APO-induced climbing much more potently (ED_{50} of 3.4 mg/kg) than stereotyped behavior (ED_{50} of 32.2 mg/kg). The climbing behavior induced by apomorphine has been reported to be more closely related to mesolimbic dopaminergic function, as opposed to striatal function.[15] The ratio of antagonism of stereotyped behavior to antagonism of climbing behavior produced by **37** was higher than that determined for the antipsychotic agent clozapine

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(17) Smith, L. M.; Peroutka, S. J. *Biochem. Behav.* **1986**, *24*, 1513.
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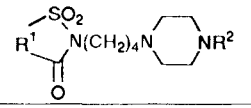
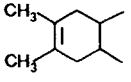
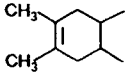
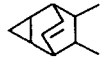
(19) Malick, J. B.; Patel, J. B.; Salama, A. J.; Meiners, B. A.; Giles, R. E.; Goldberg, M. E. *Drug. Dev. Res.* **1983**, *4*, 61.
(20) McCall, J. M. *J. Med. Chem.* **1975**, *6*, 549.

Table II. 2-Substituted 1,2-Benzisothiazol-3-one 1,1-Dioxide

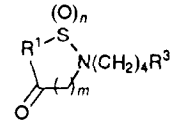


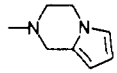
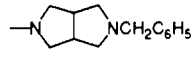
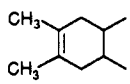
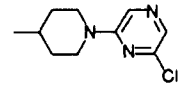
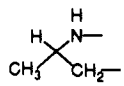
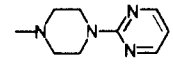
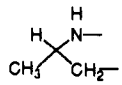
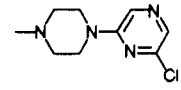
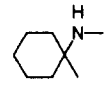
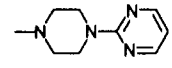
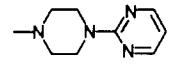
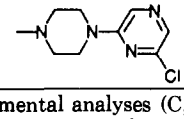
no.	R^1	R^2	mp, °C	formula[a]	inhibn of accumbens D_2 binding [K_i (95% CI)[b] or % inhibn at 1 μM]	inhibn of 5-HT_{1A} binding[c] [K_i (94% CI) or % inhibn at 1 μM]	inhibn of CAR [AB_{50}, mg/kg (95% CI)]
22		2-pyrimidinyl	257–259	$C_{21}H_{29}N_5SO_3$[d]·2HCl[e]	2000 nM (850–11 500 nM)		40[w]
23		6-chloro-2-pyrazinyl	263–267	$C_{21}H_{28}ClN_5SO_3$·HCl	57% at 3 μM		40[x]
24		2-pyrimidinyl	220–222	$C_{20}H_{27}N_5SO_3$·2HCl[f]	1100 nM (no CI)	434 nM (245–736)	45 (no CI)[y]
25		6-chloro-2-pyrazinyl	235–237	$C_{20}H_{26}ClN_5SO_3$·2HCl	1%		40[w]
26		6-chloro-3-pyridazinyl	247–249	$C_{20}H_{26}ClN_5SO_3$·2HCl[f]	3%		>40[z]
27		2-pyrimidinyl	160–162	$C_{21}H_{31}ClN_5SO_3$[g]·2HCl[f]	21%		40[x]
28		6-chloro-2-pyrazinyl	158–160	$C_{21}H_{30}ClN_5SO_3$[i]·2HCl	66% at 10 μM		40[x]
29		2-pyrimidinyl	214–215	$C_{20}H_{29}N_5SO_3$·2HCl[f]	36%	53 nM (48–59)	40[x]
30		6-chloro-2-pyrazinyl	240–242	$C_{20}H_{28}ClN_5SO_3$[j]·HCl	32%	51%	45 (39–53)[y]
31		3-$CF_3C_6H_4$	199–201	$C_{23}H_{28}F_3N_3SO_3$·2HCl	85	15 nM (12–17)	51 (no CI)[y]
32		3-$CF_3C_6H_4$	225–227	$C_{23}H_{30}FN_3SO_3$[i]·2HCl	97%	16 nM (12–20)	43 (32–71)[y]
33		3-ClC_6H_5	226–229	$C_{22}H_{30}ClN_3SO_3$[k]·2HCl	98%	45 nM (25–84)	41 (32–61)[y]
34		2-pyrimidinyl	231–234	$C_{22}H_{29}N_5SO_3$·2HCl	1407 nM (no CI)		40[x]
35		6-chloro-2-pyrazinyl	255–257	$C_{22}H_{28}ClN_5SO_3$[m]·HCl	1031 nM (no CI)	14 nM (12–17)	40[x]
36		2-pyrimidinyl	250–251	$C_{22}H_{31}N_5SO_3$[i]·HCl	70%	21 nM (16–27)	40[x]
37		2-pyrimidinyl	224–235	$C_{23}H_{29}N_5SO_3$[n]·2HCl	136 nM (116–159)	10 nM (9–12)	39 (26–143)[y]

38		3-$CF_3C_6H_4$	244–245	$C_{25}H_{32}F_3N_3SO_3$·HCl	100%	4 nM (3–5)	40[x]
39		2-pyrimidinyl	239–241	$C_{19}H_{27}N_5SO_4$[h]·2HCl	0%		>40[x]
40		6-chloro-2-pyrazinyl	231–235	$C_{19}H_{26}ClN_5SO_4$·HCl	36%	550 nM (391–841)	34 (no CI)[y]
41		3-chloro-2-pyrazinyl	67–93	$C_{19}H_{27}ClN_5SO_4$[o]·HCl[e]	3300 nM (no CI)		48 (37–73)[y]
42		2-pyrazinyl	186–193	$C_{19}H_{27}N_5SO_4$[o]·2HCl[f]	0%	25%	48 (37–73)[y]
43		6-chloro-2-pyrazinyl	268–277	$C_{19}H_{26}ClN_5SO_4$·HCl	10%	54%	40[x]
44[aa]		2-pyrazinyl	186–193	$C_{19}H_{27}N_5SO_4$·2HCl[f]	0%	25%	48 (37–73)[y]
45		3-$CF_3C_6H_4$	205–206	$C_{22}H_{28}F_3N_3SO_4$·2HCl	0%	22 nM (15–31)	40[w]
46		3-ClC_6H_4	200–205	$C_{21}H_{28}ClN_3SO_4$[p]·2HCl[q]	55%	34 nM (27–42)	40[w]
47		2-pyrazinyl	225–228	$C_{19}H_{23}N_5SO_3$[r]·2HCl	43%	9 nM (6–13)	40[w]
48		3-chloro-2-pyrazinyl	229–231	$C_{19}H_{22}ClN_5SO_3$·HCl[f]	493 nM (375–661 nM)		40[x]
49		6-chloro-2-pyrazinyl	246–249	$C_{19}H_{22}ClN_5SO_3$[s]·HCl	6%		40[w]
50			247–249	$C_{17}H_{23}N_7SO_3$·HCl	14%	52%	40[x]
51		2-pyrimidinyl	224–225	$C_{17}H_{21}N_5S_2O_3$·2HCl			>20[z]
52		6-chloro-2-pyrazinyl	255–256	$C_{17}H_{20}ClN_5S_2O_3$[f]·HCl			>40[z]
53		3-chloro-2-pyrazinyl	243–245	$C_{17}H_{20}ClN_5S_2O_3$[u]·HCl[v]			>40[z]
54		6-chloro-2-pyridazinyl	247–249	$C_{17}H_{20}ClN_5S_2O_3$·2HCl[y]			>40[z]
buspirone					119 nM (no CI)	10 nM (6–15)	32 (no CI)[y]
ipsapirone (TVXQ-7821)						10 nM (6–14)	42 (22–180)

[a] All compounds had elemental analyses (C, H, N) within ±0.4% of the theoretical values, unless otherwise specified. [b] 95% CI indicates values for 95% confidence interval. [c] Rat hippocampal tissue. [d] Calcd: C, 48.27; N, 13.41. Found: C, 48.7; N, 12.97. [e] Hydrate. [f] Hemihydrate. [g] H: calcd, 6.79; found, 6.23. [h] Sesquihydrate. [i] Calcd: C, 46.62; H, 5.96; N, 12.95. Found: C, 46.14; H, 5.43; N, 14.83. [j] N: calcd, 7.52; found, 7.11. [k] Calcd: H, 6.10; N, 8.00. Found: H, 5.67; N, 7.44. [l] N: calcd, 14.53; found, 14.02. [m] Calcd: C, 52.27; N, 13.25. Found: C, 51.7; N, 12.81. [n] C: calcd, 46.15; found, 45.58. [o] Calcd: H, 5.73; N, 13.72. Found: H, 4.81; N, 13.11. [p] Calcd: C, 44.8; H, 6.09; N, 7.46. Found: C, 45.53; H, 5.59; N, 8.28. [q] Dihydrate. [r] H: calcd, 5.31; found, 4.81. [s] N: calcd, 14.83; found, 14.06. [t] Calcd: 42.67; found, 43.09. [u] Calcd: C, 38.9; N, 13.37. Found: C, 38.2; N, 13.85. [v] $2^1/_2$ H_2O. [w] Active at single dose in discrete trial CAR, p.o. [x] Active at single dose in shelf-jump CAR, i.p. [y] Active in discrete trial CAR, p.o. [z] Indicates insignificant activity at a single dose in shelf-jump CAR, ip. [aa] Note: compound 44 is identical with 42.

Table III. 2-Substituted Tetrahydro- and Hexahydro-1,2-benzisothiazolones and Thiadiazinones



no.	R^1	n	m	R^3	mp, °C	formula[a]	% inhibn of accumbens D_2 binding at 1 μM	% inhibn of 5-HT_{1A} binding[c] at 1 μM	inhibn of CAR [AB_{50}, mg/kg (95% CI)[b]]
55		2	0		216–218	$C_{18}H_{21}N_3SO_3 \cdot HCl$	0		>40[h]
56		2	0		248–249	$C_{24}H_{31}N_3SO_3 \cdot 2HCl$	8		>40[h]
57		2	0	4-pyridinyl	211–213	$C_{17}H_{20}N_2SO_3 \cdot HCl$	1		40[i]
58		2	0	4-pyridinyl	250–252	$C_{18}H_{22}N_2SO_3$[d] $\cdot HCl$	0	35	>40[h]
59		2	0	4-pyridinyl	218–220	$C_{16}H_{22}N_2SO_4 \cdot HCl$[e]	3	0	40[i]
60		2	0	4-pyridinyl	152–154	$C_{18}H_{24}N_2SO_3$[g] $\cdot HCl$	56	8	>40[h]
61		2	0		160–162	$C_{22}H_{31}ClN_4SO_3 \cdot HCl$[f]	0	7	>40[h]
62		2	0		115–117	$C_{16}H_{30}N_6SO_3 \cdot 2HCl$[f]			40[i]
63		2	0		107–109	$C_{16}H_{25}ClN_6SO_3 \cdot 2HCl$[e]	4	61	32 (no CI)[j]
64		2	0		211–214	$C_{19}H_{30}N_6SO_3 \cdot 2HCl$[e]			40
65		1	0		220–221	$C_{19}H_{23}N_5SO_2 \cdot 2HCl$[e]	3	43	40[h]
66		1	0		222–224	$C_{19}H_{22}ClN_5SO_2 \cdot HCl$[e]	0	44	40[i]

[a] All compounds have elemental analyses (C, H, N) within ±0.4% of the theoretical values, unless other specified. [b] 95% CI indicates valles for 95% confidence interval. [c] Rat hippocampal tissue. [d] C: calcd, 56.47; found, 56.05. [e] Hemihydrate. [f] Hydrate. [g] N: calcd, 17.76; found, 18.32. [h] Indicates insignificant activity at single dose in shelf-jump CAR, ip. [i] Active at single dose in shelf-jump CAR, ip. [j] Active in discrete trials CAR, po. [k] Active at single dose in discrete trial CAR, po.

(9.57 vs 5.77), an antipsychotic agent apparently devoid of extrapyramidal side effect (EPS) liability.[21] This result suggests a low potential for extrapyramidal side effects for compound **37**.

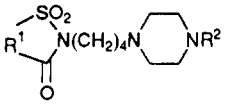
Compounds 37 and 40. Because of its dopaminergic component, buspirone was initially evaluated in the clinic as an antipsychotic agent.[22] Shortly thereafter, anxiolytic properties were discovered, and studies pursuing its mechanism of action shifted to the potential involvement of 5-HT_{1A} receptors in anxiolytic activity. We felt that compounds which demonstrated antipsychotic activity in the CAR paradigm and those with 5-HT_{1A} selectivity, i.e., those similar to buspirone in profile, should be further examined as potential anxiolytic agents. Accordingly, compounds **37** and **40** were selected for further preclinical profiling as summarized in Table VI. Both of these compounds demonstrated oral activity in the CAR testing similar to that of buspirone and ipsapirone, with AB_{50} values of 39 and 34 mg/kg. respectively. Unlike **40**, **37** displayed high affinity for 5-HT_{1A} receptor sites (K_i = 10 nM vs K_i = 550 nM) and moderate affinity for D_2 receptor sites (K_i = 136 nM). Both compounds lacked activity at the cholinergic and benzodiazepine receptor binding sites and demonstrated weak activity at the adrenergic receptor sites.

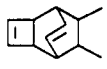
Compound **37** exhibited partial agonist/antagonist activity similar to that of buspirone and ipsapirone in the serotonin syndrome (Table VII), while **40** produced only a few components of the syndrome (muscle relaxation and hind limb abduction). Furthermore, in rotorod tests, both **37** and **40** lacked sedative and ethanol interaction activity effects, results that are similar to those for buspirone. Because of its favorable profile of activity, **37** was selected for further preclinical evaluation.

In conclusion, a series of substituted tetrahydro- and hexahydro-1,2-benzisothiazol-3-one 1,1-dioxides and thiadiazinones were synthesized. Many of these compounds

(21) Costall, B.; Naylor, R. J.; Nohvia, V. *Eur. J. Pharmacol.* **1978**, *50*, 39.
(22) Taylor, D. P.; Riblet, L. A.; Stanton, H. C.; Eison, A. S.; Eison, M. S.; Temple, D. L. *Pharmacol. Biochem. Behav* **1982**, *17*, 25.

Table IV. Effect of Lipophilicity of 2-Substituted Benzisothiazolone 1,1-Dioxides on the Affinity for the 5-HT_{1A} Receptor Binding Sites



no.	R^1	R^2	log p	inhibn of 5-HT_{1A} binding [K_i, nM (95% CI), or % inhibn at 1 μM]
42			1.14	25%
40			1.64	550 nM (391–841)
29			1.72	53 nM (84–59)
37			1.92	10 nM (9–12)

possessed high affinity for the 5-HT_{1A} receptor sites. One compound, **37**, was found to share with buspirone and ipsapirone a similar favorable neurochemical and behavioral profile and will be evaluated as a potential anxiolytic agent.

Experimental Section

Melting points were determined on a Thomas-Hoover apparatus and are uncorrected. Spectra were recorded for all compounds and were consistent with assigned structures. ^{1}H NMR spectra were recorded on Varian XL-300 and XL-100 instruments. Mass spectra were recorded with a Kratos MS-25 mass spectrometer. IR spectra were recorded with a Perkin-Elmer 299 infrared spectrophotometer. Elemental analyses were performed with a Perkin-Elmer Model 240 elemental analyzer by the analytical section of our laboratories, and analyses were within ±0.4% of the theoretical values, unless otherwise specified. Thin-layer chromatography was performed on silica gel plates (Merck).

3-Isothiazolone 1,1-Dioxide (10). A stirred solution of 2-*tert*-butyl-3-isothiazolone 1,1-dioxide (7)[8,9] (3.7 g, 0.02 mol) in 50 mL of trifluoroacetic acid was refluxed for 2 h. The solution was cooled and trifluoroacetic acid was removed under reduced pressure. The remaining oil crystallized upon trituration with a 1:1 ethyl acetate–ether mixture. The solid was filtered and dried to afford 1.7 g (65%) of **10**; mp 118–120 °C. Anal. ($C_3H_3NS_3$) C, H, N.

Cycloaddition Reaction of 3-Isothiazolone 1,1-Dioxide with Furan. Preparation of 20. A mixture of 3-isothiazolone 1,1-dioxide (5.2 g, 0.04 mol) and 5 mL of furan in toluene was refluxed for 2 h. An additional 20 mL of furan was added in 5-mL portions over 2 h. After cooling, the separated solid was filtered and dried to afford 7.5 g (93% yield) of the exo cycloadduct 3aα,4β,7β,7aα-tetrahydro-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-dioxide (**18**); mp 180–183 °C. The exoadduct (10 g, 0.04 mol) was dissolved in dry THF (100 mL) and was hydrogenated over 2 g of 10% Pd/C for 0.5 h. The catalyst was filtered and the solvent was evaporated under reduced pressure to afford the corresponding reduced exo cycloadduct 3aα,4β,7β,7aα-hexahydro-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-dioxide (**20**) in quantitative yield: mp 234–236 °C; ^{1}H NMR (DMSO-d_6) δ 1.65 (m, 4 H), 3.65 (d, J = 7 Hz, 1 H), 4.10 (d, J = 7 Hz, 1 H), 4.85 (s, 1 H), 5.10 (s, 1 H), and 12.5 (br, 1 H, exchangeable). Anal. ($C_7H_9NSO_4$) C, H, N.

In a like manner compounds **11–17** were prepared via the Diels–Alder reaction of 3-isothiazolone 1,1-dioxide with the appropriate diene.

General Procedure for the Preparation of Compounds in Table II. 3aα,4β,7β,7aα-Hexahydro-2-[4-[4-(6-chloro-2-pyrazinyl)-1-piperazinyl]butyl]-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-Dioxide (40). To a solution of the exo adduct **20** (3 g, 0.01 mol) in 50 mL of DMF was added sodium hydride (0.6 g, 0.03 mol), and the reaction mixture was stirred at 60 °C for 1 h. This solution was added to a stirred solution of 1,4-dibromobutane (6.4 g, 0.03 mol) in 50 mL of DMF, and stirring was continued at room temperature for 48 h. The DMF was then removed under reduced pressure, and the remaining semisolid was suspended in water and extracted with methylene chloride (3 × 100 mL). The combined organic extracts were washed with water, dried over anhydrous Na_2SO_4, and evaporated under reduced pressure to afford 3 g of the crude 3aα,4β,7β,7aα-hexahydro-2-(4-bromobutyl)-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-dioxide. The title compound was prepared by dissolving the above bromobutyl intermediate (3.59 g, 0.01 mol) in 150 mL of DMF, and the stirred solution was treated with triethylamine (8.6 g, 0.08 mol) and 1-(6-chloro-2-pyrazinyl)piperazine (4.9 g, 0.02 mol). After stirring for 24 h at room temperature, triethylamine (8.6 g, 0.08 mol) was added and stirring was continued for an additional 18 h. The mixture was partitioned between methylene chloride (3 × 100 mL) and water. The combined organic extracts were dried over anhydrous Na_2SO_4 and evaporated under reduced pressure to give an oil. Trituration with ethyl acetate gave a solid, which was subjected to preparative HPLC (silica gel; ethyl acetate). The fractions containing the title compound (R_f = 0.15, ethyl acetate) were combined and evaporated and the residue crystallized from ethyl acetate to give 1.1 g (22% yield) of **40** as the free base: mp 147–149 °C; ^{1}H NMR (DMSO-d_6) δ 2.46 (m, 2 H), 1.54–1.78 (m, 6 H), 2.32 (t, J = 6 Hz, 2 H), 2.43 (m, 4 H), 3.48 (t, J = 6 Hz, 2 H), 3.57 (m, 4 H), 3.73 (d, J = 8 Hz, 1 H), 4.22 (d, J = 8 Hz, 1 H), 4.85 (s, 1 H), 5.09 (s, 1 H), 7.88 (d, 1 H), and 8.31 (s, 1 H). An analytical sample was prepared by conversion to the hydrochloride salt; mp 231–235 °C. Anal. ($C_{22}H_{28}ClN_5SO_3 \cdot HCl$) C, H, N.

Compounds 22–56 were similarly prepared from the corresponding 1,2-benzisothiazol-3-one 1,1-dioxide and the appropriately substituted aryl- or heteroarylpiperazine or octahydropyrrolopyrrole.[10]

3aα,4α,7α,7aα-Hexahydro-2-[4-[4-(6-chloro-2-pyrazinyl)-1-piperazinyl]butyl]-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-Dioxide (43). The title compound was prepared following a similar procedure used for the preparation of **40** with the exception that a 2:1 exo:endo mixture of **20** and **21** (formed from the Diels–Alder reaction of furan and 3-isothiazolone 1,1-dioxide in benzene) was used instead of **20**. The title compound was separated by preparative HPLC to afford the free base: ^{1}H NMR (DMSO-d_6) δ 1.2–1.85 (m, 8 H), 2.35 (t, J = 6 Hz, 2 H), 2.44 (m, 4 H), 3.51 (t, J = 6 Hz, 2 H), 3.57 (m, 4 H), 4.07 (dd, J = 12, 6

Table V. Effect of 2-Substituted Benzisothiazolone 1,1-Dioxides and Ipsapirone on Apomorphine Antagonism[a]

compd	ED_{50}, mg/kg, ip (95% CI)[b] stereotyped behavior	ED_{50}, mg/kg, ip (95% CI)[b] climbing behavior	ratio ED_{50}(stereotyped behavior)/ ED_{50}(climbing behavior)
33	34 (no CI)	16 (1.29–27.21)	2.13
37	32.2 (no CI)	3.38 (0.97–11.7)	9.53
38	inactive	29 (no CI)	
39	34 (no CI)	16 (1.29–27.21)	2.13
40	inactive	27 (7–101)	
45	44.36 (37.28–52.78)	11.49 (no CI)	3.86
ipsapirone	inactive	26 (3–220)	
clozapine[c]	26.26 (22–31)	4.55 (1–23)	5.2

[a] Low-dose APO-induced behavior. [b] 95% confidence intervals. [c] Antipsychotic agent devoid of EPS liability.

Table VI. Receptor Binding Affinities[a] of Compounds 37, 40, and Buspirone

binding site	37	40	buspirone
5-HT_{1A}	10 nM (9–17 nM)	550 nM (391–841 nM)	10 nM (6–15 nM)
D_2	136 nM (116–159)	36%	119 (M (no CI)
Bz	0%	0%	0%
QNB		0%	
α_1	54%	6%	33%
α_2		21%	
β		8%	0%

[a] All data reported as percent inhibition at 1 μM of drug or K_i (95% confidence interval).

Table VII. Activity of Compounds 37 and 40 in the Serotonin Syndrome Test

compd (dose, mg/kg, ip)	N	elicit syndrome mean total score (±SEM)	antagonize syndrome mean total score (±SEM)
MeO-DMT (3.0)	94	14.0 (0.4)	
37 (17.0)	8	3.9 (0.4)	5.1 (0.3)[a]
40 (17.0)	8	2.1 (0.4)[b]	12.3 (0.9)
buspirone (10.0)	8	2.9 (0.4)	6.1 (0.6)[a]
ipsapirone (10.0)	8	2.4 (0.4)	8.4 (0.8)[a]

[a] Statistically different from saline control by Student's t test, $P < 0.001$. [b] Not true agonist activity; muscle relaxation and hind limb abduction.

Hz, 1 H), 4.41 (dd, J = 12, 5 Hz, 1 H), 5.05 (m, 2 H), 7.89 (s, 1 H), and 8.32 (s, 1 H). It was converted to the hydrochloride salt; mp 268–277 °C. Anal. ($C_{19}H_{26}ClN_5SO_4$·HCl) C, H, N.

3a,4,7,7a-Tetrahydro-2-[4-(4-pyridinyl)butyl]-4,7-methano-1,2-benzisothiazol-3(2*H*)-one 1,1-Dioxide (57). To a stirred solution of 4a,4,7,7a-tetrahydro-4,7-methano-1,2-benzisothiazol-3(2*H*)-one (11) (2 g, 0.01 mol) in 50 mL of dimethylformamide were added cesium carbonate (3.0 g, 0.011 mol) and 4-pyridinylbutyl bromide hydrobromide (3.2 g, 0.011 mol). The reaction mixture was stirred at room temperature for 48 h, dimethylformamide was evaporated under reduced pressure, and the residue was extracted with methylene chloride (3 × 200 mL). The methylene chloride extracts were collected, washed with water, dried over anhydrous Na_2SO_4, and evaporated under reduced pressure. The semisolid residue was subjected to preparative HPLC separation using ethyl acetate as the eluent. Evaporation of the solvent from the desired fractions (TLC R_f = 0.20, ethyl acetate) afforded 1.6 g (48% yield) of the title compound, which was converted to the hydrochloride salt by dissolving the free base in ethanol and adding ethanol saturated with hydrogen chloride, mp 211–213 °C. Anal. ($C_{17}H_{20}N_2SO_3$·HCl) C, H, N.

Compounds 58–60 were prepared similarly from the appropriate 1,2-benzisothiazol-3-one 1,1-dioxide and 4-pyridinylbutyl bromide hydrobromide.

Hexahydro-2-[4-[1-(6-chloro-2-pyrazinyl)-4-piperidinyl]butyl]-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-Dioxide (61). To an ethanolic solution of the hydrochloride salt of 58 (2.0 g, 0.004 mol) in 50 mL of ethanol under nitrogen were added 0.4 g of 5% rhodium over aluminum oxide and 1 mL of glacial acetic acid. The reaction mixture was hydrogenated at room temperature in a Parr shaker with hydrogen (50 psi) for 3 h and filtered, and the solvent was removed under reduced pressure. The remaining oil was dissolved in 50 mL of dimethylformamide, and to that solution were added 0.5 g of cesium carbonate and 2,6-dichloropyrazine (0.8 g, 0.006 mol). The reaction mixture was stirred at room temperature for 48 h and worked up as above for compound 40 to afford the title compound, which was converted to the hydrochloride salt; mp 160–162 °C. Anal. ($C_{22}H_{31}ClN_4SO_3$·HCl) C, H, N.

2-[4-[4-(6-Chloro-2-pyrazinyl)-1-piperazinyl]butyl]-5,6-dihydro-5-methyl-4*H*-1,2,6-thiadiazin-3(4*H*)-one 1,1-Dioxide (63). To a stirred solution of 5,6-dihydro-5-methyl-2*H*-1,2,6-thiadiazin-3(4H)-one 1,1-dioxide[13] (2.5 g, 0.015 mol) in 70 mL of dimethylformamide was added sodium hydride (0.73 g, 0.03 mol). The resulting clear solution was added dropwise to a stirred solution of 1,4-dibromobutane (4.3 g, 0.02 mol) in 40 mL of dimethylformamide. The reaction mixture was stirred overnight, the solvent was removed under vacuum, and the residue was partitioned between methylene chloride and water. The combined methylene chloride extracts were combined, washed with brine, and dried over anhydrous Na_2SO_4. Filtration and removal of the solvent in vacuo afforded 4 g (88% yield) of 2-(4-bromobutyl)-5,6-dihydro-5-methyl-2*H*-1,2,6-thiadiazin-3(4*H*)-one 1,1-dioxide as a yellow oil.

A stirred solution of 2-(4-bromobutyl)-5,6-dihydro-5-methyl-2*H*-1,2,6-thiadiazin-3(4*H*)-one 1,1-dioxide (2.0 g, 0.006 mol) in 50 mL of dimethylformamide was treated with triethylamine (4 mL) and 1-(6-chloro-2-pyrazinyl)piperazine hydrochloride (1.41 g, 0.006 mol). The reaction mixture was stirred overnight, DMF was removed under high vacuum, and the residue partitioned between water and methylene chloride. The methylene chloride extracts were combined, washed with brine, and dried over anhydrous Na_2SO_4. Filtration and evaporation of the solvent gave the crude free base of 63. Preparative HPLC [silica gel; ethyl acetate–methylene chloride (9:1)] gave the title compound, which was converted to the dihydrochloride salt; mp 107–109 °C. Anal. ($C_{16}H_{25}ClN_6SO_3$·2 HCl) C, H, N.

Compound 62 was prepared following the above procedure for the preparation of 63 with the exception that 1-(2-pyrimidinyl)piperazine dihydrochloride was used instead of 1-(6-chloro-2-pyrazinyl)piperazine hydrochloride.

3-[4-[4-(2-Pyrimidinyl)-1-piperazinyl]butyl]-2-thia-3-diazaspiro[4.5]decan-4-one 2,2-Dioxide (64). To a stirred suspension of 1-amino-1-cyclohexanecarboxylic acid (92.5 g, 0.017 mol) in 70 mL of dioxane was added 6 mL of trichloromethyl chloroformate (TCF), and stirring was continued for 4 h at 55 °C or until a clear solution was obtained (reaction time from 4 to 6 h). Dioxane was evaporated under reduced pressure to afford spiro[4,5]decan-2,4'-oxazolidine-2,5'-dione. This compound was dissolved in 50 mL of methylene chloride, treated with 1-(4-aminobutyl)-4-(2-pyrimidinyl)piperazine (4.7 g, 0.02 mol), and maintained at reflux for 3 h. The reaction mixture was diluted with methylene chloride, washed sequentially with water and brine, and evaporated under reduced pressure to afford 2 g of 1-amino-*N*-[4-[4-(2-pyrimidinyl)-1-piperazinyl]butyl]cyclohexanecarboxamide. The title compound was prepared by dissolving 2 g of the acetanilide in 50 mL of methylene chloride, and to the stirred solution were added 2 mL of sulfuryl chloride and 4 mL of triethylamine. Stirring was continued at room temperature for 24 h, and the reaction mixture was washed with water, dried, and removed of solvent under reduced pressure. The free base was separated via preparative HPLC [silica gel; ethyl acetate–methylene chloride (9:1)] and was converted to the dihydrochloride salt; mp 211–214 °C. Anal. ($C_{19}H_{30}N_6SO_3$·2 HCl) C, H, N.

Antagonism of Apomorphine-Induced Behaviors. For the high-dose procedure, see reference 16. Antagonism of low-dose apomorphine-induced stereotyped and climbing behavior tests were conducted according to an adaptation of the methods of Costall et al.[21] and Puech et al.[23] as described earlier.[6]

Shelf-Jump Conditioned Avoidance. Shelf-jump conditioned avoidance tests were conducted according to the method of Herman et al.[24] as previously described.[6]

Discrete Trial Conditioned Avoidance. Discrete trial conditioned avoidance tests were conducted in male CD rats (Charles River) maintained at approximately 400–450-g body weight. Rats trained previously were placed in plexiglass experimental chambers equipped with a response lever, house light, and sonalert. A steel grid floor was wired for presentation of electric shock. Each trial consisted of a 15-s warning tone (conditioned stimulus), continuing for an additional 15 s accompanied by electric shock (unconditioned stimulus). The rat could terminate a trial at any point by depressing the response lever. A response during the initial 15-s warning tone ended the trial before shock delivery and was considered an avoidance response, while a response occurring during shock delivery was an escape response. Trials were

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presented on a variable-interval schedule of 2 min. The session consisted of sixty trials. Animals were run two to three times weekly with control sessions always preceding a drug run, and with at least 1 day intervening. Compounds were administered ip or po at a pretreatment time of 30 min to a minimum of five rats at each dose level (20 or 40 mg/kg) or over a range of doses. The following experimental parameters were recorded by computer: (1) the number of intertrial interval responses, (2) the number of avoidance responses, (3) the number of escape responses, and (4) the number of trials in which no response occurred. These data were used to calculate the percent difference from control values previously determined. For active compounds, response counts were summed over all subjects at a given dose. The number of trials in which rats failed to exhibit an avoidance response (avoidance block, AB) was determined at each dose. This number was expressed as a percentage of the total trials. Control performance was arbitrarily set at 100% for avoidance responding, and the dose calculated to produce a 50% block in avoidance responding (AB_{50}) was obtained from a dose effect regression line fitted by the method of least squares.

Receptor Binding Assays. D_2 and 5-HT_{1A} receptor binding assays were performed as previously described.[6] Binding assays to other receptor sites were performed as described by Muth et al.[25]

Serotonin Syndrome and Rotorod Tests. In vivo serotonin syndrome and rotorod tests were performed according to an adaptation of the methods of Smith and Peroutka[17] and Malick et al.[19]

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Registry No. **9**, 119639-24-6; **10**, 26479-40-3; **11**, 119717-60-1; **12**, 69729-80-2; **13**, 69729-75-5; **14**, 112978-09-3; **15**, 112977-69-2; **16**, 119717-61-2; **17**, 112978-13-9; **18**, 113033-94-6; **19**, 112978-14-0; **20**, 119639-25-7; **21**, 119717-62-3; **22**, 119717-63-4; **22**·2HCl, 119785-05-6; **23**, 119717-64-5; **23**·HCl, 119785-06-7; **24**, 119718-82-0; **24**·HCl, 119785-07-8; **25**, 119717-65-6; **25**·2HCl, 119785-08-9; **26**, 119639-26-8; **26**·2HCl, 119717-69-0; **27**, 119639-27-9; **27**·2HCl, 119639-50-8; **28**, 119639-28-0; **28**·2HCl, 119639-51-9; **29**, 119717-66-7; **29**·2HCl, 119785-09-0; **30**, 119717-67-8; **30**·HCl, 119785-10-3; **31**, 119639-29-1; **31**·2HCl, 119717-70-3; **32**, 119639-30-4; **32**·2HCl, 119717-71-4; **33**, 119639-31-5; **33**·2HCl, 119717-72-5; **34**, 112977-76-1; **34**·2HCl, 112977-93-2; **35**, 112977-77-2; **35**·HCl, 112977-94-3; **36**, 112977-85-2; **36**·HCl, 112978-03-7; **37**, 112977-87-4; **37**·2HCl, 112978-04-8; **38**, 119639-32-6; **38**·HCl, 119639-52-0; **39**, 119639-33-7; **39**·2HCl, 119717-73-6; **40**, 113033-90-2; **40**·HCl, 113083-05-9; **41**, 119639-34-8; **41**·HCl, 119717-74-7; **42**, 119717-68-9; **42**·2HCl, 119785-11-4; **43**, 112977-70-5; **43**·HCl, 113033-91-3; **45**, 119639-35-9; **45**·2HCl, 119717-75-8; **46**, 119639-36-0; **46**·2HCl, 119717-76-9; **47**, 119639-37-1; **47**·2HCl, 119639-53-1; **48**, 112977-80-7; **48**·HCl, 112977-97-6; **49**, 112977-78-3; **49**·HCl, 112977-95-4; **50**, 119639-38-2; **50**·HCl, 119639-54-2; **51**, 119639-39-3; **51**·2HCl, 119639-55-3; **52**, 112977-79-4; **52**·HCl, 112977-96-5; **53**, 119656-10-9; **53**·HCl, 112977-98-7; **54**, 119639-40-6; **54**·2HCl, 119656-12-1; **55**, 119639-41-7; **55**·HCl, 119639-56-4; **56**, 119656-11-0; **56**·2HCl, 119656-13-2; **57**, 119639-42-8; **57**·HCl, 119717-77-0; **58**, 119639-43-9; **58**·HCl, 119717-78-1; **59**, 119639-44-0; **59**·HCl, 119639-44-0; **60**, 119639-45-1; **60**·HCl, 119639-57-5; **61**, 119639-46-2; **61**·HCl, 119639-58-6; **62**, 114895-53-3; **62**·2HCl, 114895-57-7; **63**, 114895-51-1; **63**·2HCl, 114895-58-8; **64**, 114895-52-2; **64**·2HCl, 114895-56-6; **65**, 119639-47-3; **65**·2HCl, 119639-59-7; **66**, 119639-48-4; **66**·HCl, 119639-60-0; **67**, 36935-58-7; furan, 110-00-9; cyclopentadiene, 542-92-7; 1,3-cyclohexadiene, 592-57-4; 2,3-dimethyl-1,3-butadiene, 513-81-5; 1,3-cycloheptadiene, 4054-38-0; 1,3,5,7-cyclooctatetraene, 629-20-9; 1,4-dibromobutane, 110-52-1; (3aα,4β,7β,7aα)-hexahydro-2-(4-bromobutyl)-4,7-epoxy-1,2-benzisothiazol-3(2*H*)-one 1,1-dioxide, 119717-59-8; 1-(6-chloro-2-pyrazinyl)piperazine, 64022-27-1; 1-(2-pyrimidinyl)piperazine, 20980-22-7; 1-(6-chloro-3-pyridizinyl)piperazine, 56392-83-7; 1-(3-trifluoromethylphenyl)-piperazine, 15532-75-9; 1-(3-chlorophenyl)piperazine, 6640-24-0; 1-(2-pyrazinyl)piperazine, 34803-68-4; 1-(3-chloro-2-pyrazinyl)-piperazine, 85386-99-8; 1-(1-methyl-5-tetraazolyl)piperazine, 119639-49-5; 1,2,3,4-tetrahydropyrrole[1,2-*a*]pyrazine, 71257-38-0; 1-benzylhexahydropyrrolo[3,4-*c*]pyrrole, 86732-22-1; 4-pyridinylbutyl bromide hydrobromide, 107266-09-1; 2,6-dichloropyrazine, 4774-14-5; 1,2-benzisothiazol-3(2*H*)-one 1,1-dioxide, 81-07-2; thieno[3,4-*d*]isothiazol-3(2*H*)-one 1,1-dioxide, 59337-79-0; 1-amino-1-cyclohexane carboxylic acid, 2756-85-6; 1,2-benzisothiazol-3(2*H*)-one 1-oxide, 14599-38-3; 1-(4-aminobutyl)-4-(2-pyrimidinyl)piperazine, 33386-20-8; spiro[4,5]decan-2,4′-oxazolidine-2,5′-dione, 3253-43-8; 2-(4-bromobutyl)-5,6-dihydro-5-methyl-2*H*-1,2,6-thiadiazin-3(4*H*)-one 1,1-dioxide, 114895-54-5; 2-amino-*N*-[4-[4-(2-pyrimidinyl)-1-piperazinyl]butyl]cyclohexanecarboxamide, 119639-61-1.

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Synthesis and in Vitro Aldose Reductase Inhibitory Activity of Compounds Containing an *N*-Acylglycine Moiety

Jack DeRuiter,* Blake E. Swearingen, Vinay Wandrekar, and Charles A. Mayfield

Division of Medicinal Chemistry, Department of Pharmacal Sciences, School of Pharmacy, Auburn University, Auburn University, Alabama 36849. *Received September 6, 1988*

A number of *N*-benzoylglycines (**6**), *N*-acetyl-*N*-phenylglycines (**7**), *N*-benzoyl-*N*-phenylglycines (**8**), and tricyclic *N*-acetic acids (**9–12**) were synthesized as analogues of the *N*-acylglycine-containing aldose reductase inhibitors alrestatin and 2-oxoquinoline-1-acetic acid. Derivatives of **6**, which represent ring-simplified analogues of alrestatin, are very weak inhibitors of aldose reductase obtained from rat lens, producing 50% inhibition only at concentrations exceeding 100 μM. Compounds of series **7** were designed as ring-opened analogues of the 2-oxoquinolines. While these derivatives are more potent than compounds of series **6** (IC_{50}s of 6–80 μM), they are less active than the corresponding 2-oxoquinolines. Analogues of series **8** were designed as hybrid structures of both alrestatin and the 2-oxoquinoline-1-acetic acids. These compounds are substantially more potent than compounds of series **6** and **7** and display inhibitory activities comparable to or greater than alrestatin or the 2-oxoquinolines (IC_{50}s of 0.1–10 μM). Of the rigid analogues of **8**, the most potent derivative is benzoxindole (**12**) with an IC_{50} of 0.67 μM, suggesting that fusion of the two aromatic rings of **8** in a coplanar conformation may optimize affinity for aldose reductase in this series.

Over the past decade, a number of structurally diverse compounds have been reported to inhibit the enzyme aldose reductase and therefore possess potential utility for the prevention of some pathologies of chronic diabetes.[1-3]

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