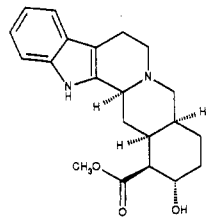
Communications to the Editor

(8aα,12aα,13aα)-5,8,8a,9,10,11,12,12a,13,13a-Decahydro-3-methoxy-12-(methylsulfonyl)-6*H*-isoquino[2,1-*g*][1,6]naphthyridine, a Potent and Highly Selective α_2-Adrenoceptor Antagonist[1]

Sir:

A number of selective α_2-adrenoceptor antagonists have been prepared on the basis of the lead structure rauwolscine (1), which, along with its isomer yohimbine, was one of the first compounds found to display a reasonable degree of selectivity for antagonism of the α_2-adrenoceptor vs the α_1-adrenoceptor.[2,3] These include the benzoquinolizine WY 26703 (**2**)[4] and several recently reported



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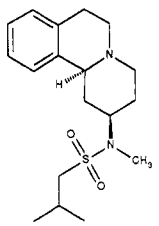

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octahydro-2*H*-benzo[*b*]furo[2,3-*a*]quinolizine[5-7] and berbane[8,9] derivatives. In addition, idazoxan (2-(1,4-benzodioxan-2-yl)-2-imidazoline) and related compounds, which are structurally unrelated to rauwolscine, have been shown to be selective α_2-adrenoceptor antagonists.[10,11] Such agents are of current interest for their role in elucidating the pharmacological and physiological significance of α_2-adrenoceptors and for their potential clinical utility for the treatment of a number of diseases including depression.[11] We now wish to describe the synthesis and preliminary pharmacological evaluation of (8aα,12aα,13aα)-5,8,8a,9,10,11,12,12a,13,13a-decahydro-3-methoxy-12-(methylsulfonyl)-6*H*-isoquino[2,1-*g*][1,6]-naphthyridine (**8a**), a structural hybrid between rauwolscine and WY 26703. This tetracyclic compound, which has been resolved to afford the active enantiomer **8b**, is a remarkably selective and potent α_2-adrenoceptor antagonist.

The preparation of the racemate **8a** and the enantiomers **8b** and **8c** is outlined in Scheme I. Condensation of 6-methoxy-3,4-dihydroisoquinoline (**3**) with *N*,*N*-diethyl-2-methylnicotinamide (**4**)[12] in the presence of lithium diisopropylamide afforded the key intermediate tetracyclic naphthyridine **5**[13] (mp 115–116 °C) in 78% yield. A full account on this type of annelation procedure has recently been published.[14] Catalytic hydrogenation of **5** gave the 8aα,12aα,13aα diastereomer **6** (mp 118–119 °C) as the major product in 60% yield.[15] Reduction of **6** with lithium aluminum hydride followed by mesylation afforded **8a** in 75% overall yield for the two steps: HCl salt, mp 256–258 °C.

Resolution of **8a** was effected by a variation of the optically active (1-phenylethyl)urea method.[16] Thus, treatment of the racemate **6** with (+)-(*R*)-1-phenylethyl isocyanate afforded the diastereomeric ureas **7a** and **7b**, which were separated by medium-pressure chromatography. The diastereomeric purities of **7a** and **7b** so obtained were determined to be >98% by ^{1}H NMR analysis. Lithium aluminum hydride reduced the lactam of **7a** and **7b** without affecting the urea functionality. The chiral auxiliary urea was subsequently removed with sodium butoxide in 1-butanol at reflux and mesylation of the derived amines afforded the enantiomers **8b** (HCl salt, mp 256–258 °C; $[\alpha]_D$ +13°, *c* 0.3, MeOH) and **8c** (HCl salt, mp 256–258 °C; $[\alpha]_D$ −13°, *c* 0.4, MeOH). The enantiomeric purities of **8b** and **8c** were determined to be >99% by chiral HPLC analysis.[17]

The relative and absolute configurations assigned to **8a–c** were based on X-ray crystallographic analysis of urea **7a**, which was found to have absolute configuration 8a*S*,12a*S*,13a*S* relative to the known 1′*R* auxiliary group. Thus, the absolute configuration of the derived **8b** is as shown in Scheme I (8a*R*, 12a*S*, 13a*S*) and corresponds to the absolute stereochemistry of rauwolscine (1).

The relative affinities of **8a–c** for α-adrenoceptors were determined by measurement of radioligand displacement from rat cerebral cortical membrane binding sites using [^{3}H]prazosin and [^{3}H]yohimbine to label α_1 and α_2-adrenoceptors, respectively.[11] The ability of test compounds to inhibit phenylephrine-induced contraction of the rabbit aorta was determined as a measure of functional α_1-adrenoceptor antagonism.[18] Functional α_2-adrenoceptor antagonism was determined by reversal of the inhibitory effects of UK-14304 on the contractile response to field stimulation of guinea pig ileum.[19] Results of these in vitro

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(12) Baumgarten, P.; Dornow, A.; *Ber. Dtsch. Chem. Ges. B* **1939**, *72*, 563.
(13) Satisfactory elemental analyses (±0.4% for C, H, N) were obtained for all compounds reported in this paper.
(14) Clark, R. D.; Jahangir, *J. Org. Chem.* **1987**, *52*, 5378.
(15) The 8aβ,12aβ,13aα (30%) and 8aβ,12aα,13aα (5%) diastereomers were also formed and were separated from **6** chromatographically.
(16) Schönenberger, B.; Brossi, A. *Helv. Chim. Acta* **1986**, *69*, 1486.
(17) LKB, Enatiopac, α_1-acid glycoprotein column.
(18) Ruffolo, R. R.; Waddell, J. E. *Br. J. Pharmacol.* **1982**, *77*, 169.
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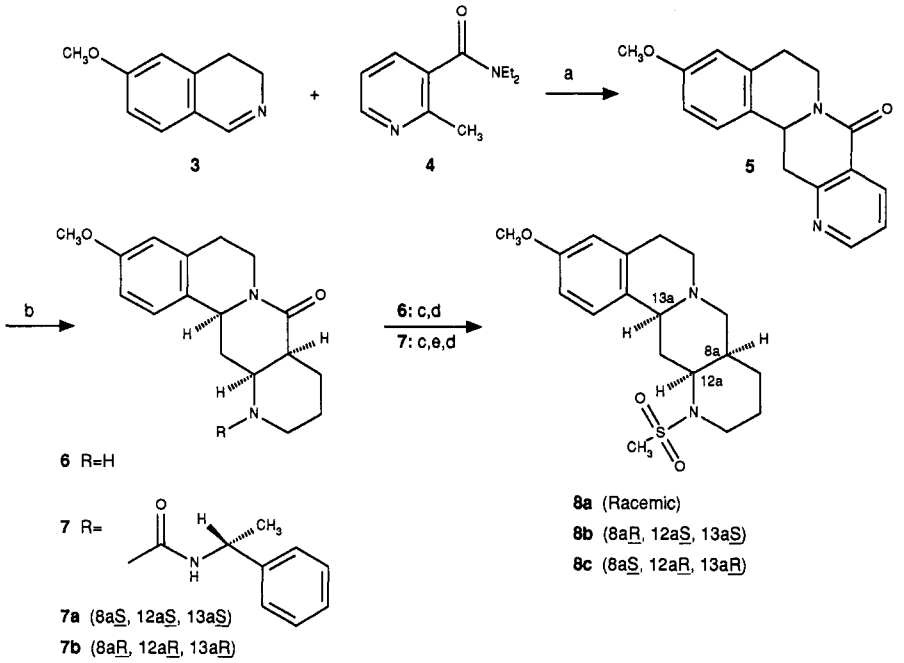
Scheme I[a]



[a] (a) LDA, THF, −40 °C. (b) H_2, HOAc, Rh–Al_2O_3. (c) LAH, THF. (d) MsCl, CH_2Cl_2, Et_3N. (e) NaOBu, *n*-BuOH.

Table I. Radioligand Binding and Functional Results

	ligand binding, pK_i[a]			functional antagonism, pA_2[a]		
compd	[^{3}H]prazosin (α_1)	[^{3}H]yohimbine (α_2)	selectivity[b]	rabbit aorta (α_1)[c]	guinea pig ileum (α_2)[d]	selectivity[e]
8a	4.99 ± 0.10	9.18 ± 0.12	15000	<6	9.7 ± 0.04	>4000
8b	5.29 ± 0.10	9.45 ± 0.16	15000	6.1	9.7 ± 0.10	4000
8c	<5	6.32 ± 0.08	>50	5.0	6.5 ± 0.10	32
idazoxan	6.10 ± 0.08	7.96 ± 0.04	72	NT	8.4 ± 0.03	
yohimbine	6.40 ± 0.03	7.90 ± 0.03	32	6.1	7.5 ± 0.10	25

[a] Values are means for at least three separate determinations. ± indicates mean standard error or where not indicated, mean standard errors were <0.2 log unit. [b] Antilog of pK_i [^{3}H]yohimbine minus pK_i [^{3}H]prazosin. [c] Reversal of phenylephrine-induced contraction. [d] Reversal of inhibitory effect of UK-14304 on contractile response to field stimulation. [e] Antilog of pA_2 (UK-14304) minus pA_2 (phenylephrine).

assays are given in Table I. Reversal of clonidine-induced mydriasis in the rat was used as an in vivo assay for central α_2-adrenoceptor antagonism.[20] The results of this assay, using both iv and oral routes of administration to assess bioavailability, are presented in Table II.

The results of the ligand binding assays in Table I indicate that the racemate **8a** is a high-affinity, remarkably selective α_2-adrenoceptor ligand. Furthermore, the activity of the racemate resides almost exclusively in the 8a*R*, 12a*S*, 13a*S* isomer **8b**, which has over 1000 times the affinity of enantiomer **8c**. This type of enantioselective affinity has been previously described for several α_2-adrenoceptor antagonists of the octahydro-2*H*-benzo[*b*]furo[2,3-*a*]-quinolizine class.[5-7]

Functional results obtained in the rat aorta (α_1) and guinea pig ileum (α_2) confirmed the potent and selective α_2-adrenoceptor antagonist properties of **8a** and **8b** (Table I).

On the basis of the data in Table I and on comparative literature data, it would appear that **8b** is at least as potent as any α_2-adrenoceptor antagonist reported to date. Furthermore, the α_2/α_1 selectivity of **8b** appears to be unprecedented.[21]

Table II. Reversal of Clonidine-Induced Mydriasis in the Rat

	IC_{50},[a] μg/kg		ratio
compd	iv	oral	oral/iv
8a	13	210	16.2
8b	6.5	95	14.6
8c	>10000	>10000	
idazoxan	17	1200	71
yohimbine	185	2800	15

[a] Dose required to reduce pupillary diameter to 50% of maximum induced by clonidine administration. Measurement of pupillary diameter was made 10 min after dosing with test compound. Values are means of five determinations and standard deviations were less than 10%.

Results in Table II indicate that **8b** is a potent antagonist of the centrally active α_2-adrenoceptor agonist clonidine in vivo with reasonable oral bioavailability relative to the standards idazoxan and yohimbine. It has also been determined that in addition to its higher potency, **8b** displays a longer duration of action compared to idazoxan upon oral administration in the mydriasis assay.[22] Thus, **8b** has been characterized as a potent, orally active, central

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(21) For example, L-657,743 has been reported to have a pK_i of 9.3 in ligand binding studies using both [^{3}H]rauwolscine and [^{3}H]clonidine with a selectivity ratio of 240 (ref 7). The berbane CH-38083 had a pK_i of 8.7 ([^{3}H]idazoxan binding) with a selectivity ratio of 1368 (ref 8). Direct comparative data on these agents and **8b** are not yet available.

(22) Duration was determined by measuring the dose ratio of clonidine administered at 1, 2, 4, and 6 h after oral dosing with test compound. The dose ratio is defined as the multiple of the control dose of clonidine required to induce a 50% increase in pupillary diameter. For **8b** (3 mg/kg po), clonidine dose ratios of 70, 53, 18, and 6 were measured at 1, 2, 4, and 6 h, respectively. For idazoxan (30 mg/kg po), the corresponding clonidine dose ratios were 40, 10, 2, and 2 at the same points. A dose ratio of 2 was considered threshold.

α_2-adrenoceptor antagonist in the rat.

A decrease in the number of cortical β-adrenoceptors (down-regulation) has been associated with a number of chronic antidepressant therapies, including treatment with antidepressants of the tricyclic, monoamine oxidase inhibitor, and atypical classes, as well as with electroconvulsive therapy.[11] This β-adrenoceptor down-regulation can be ascribed to increased levels of norepinephrine in the brain. Since prejunctional α_2-adrenoceptors mediate norepinephrine levels through a negative-feedback mechanism,[23] antagonism of α_2-adrenoceptors should increase norepinephrine levels and also result in β-adrenoceptor down-regulation and possibly a concomitant antidepressant effect. It was therefore of interest that administration of **8b** at 0.5 mg/kg once daily for 14 days in the rat was found to significantly reduce the number of cortical β-adrenoceptors to a B_{max} of 57 ± 5 fmol/mg ($p < 0.05$) relative to a control B_{max} of 72 ± 3 fmol/mg.[24] On the basis of this finding and on its potency and selectivity as an α_2-adrenoceptor antagonist, **8b** should be an important agent for the pharmacological evaluation of the α_2-adrenoceptor and may have clinical application, e.g. for the treatment of depression.

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(24) The B_{max} is the number of fmoles of [^{3}H]dihydroalprenolol which binds per mg of protein. A B_{max} of 50 was determined for desipramine after 14 days of once daily dosing at 7.5 mg/kg ip.

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LH-RH Antagonists: Design and Synthesis of a Novel Series of Peptidomimetics

Sir:

Luteinizing hormone releasing hormone (LH-RH) also known as gonadotropin releasing hormone (GnRH) was isolated and characterized by Schally[1] and co-workers in 1971. LH-RH is produced in the hypothalamus and controls secretion of luteinizing hormone (LH) and follicle stimulating hormone (FSH) from the anterior pituitary. Various LH-RH agonists have been shown to produce an initial stimulation of gonadotropin release from the anterior pituitary, which results in ovulation in women with amenorrhea and testosterone secretion, together with spermatogenesis in hypogonadotropic men.[2] However, chronic administration of LH-RH agonists results in a paradoxical inhibition of the pituitary–gonadal axis characterized by a decrease of the levels of sex steroids and the atrophy of accessory sex organs.[3,4] This discovery of gonadal steroid suppression produced by an LH-RH agonist led to the successful development of clinical therapeutic agents such as Lupron (TAP Pharmaceuticals) for use in the treatment of hormone-dependent breast and prostatic carcinoma.[5]

Currently available LH-RH superagonists are polypeptides (containing 9–10 amino acid residues) and less than 1% bioavailable when administered orally. The very principle by which the superagonists induce the hypophysis to release large amount of LH during the first 2–3 weeks of desensitization period also produces undesirable side effects.[6] One attractive alternative to this problem has been to design and antagonist which, in principle, should attain the same goal through the direct blockade of LH release. It has been shown that rabbits which produced an antibody to LH-RH developed gonadal antropy[7] and that administration of rabbit anti-LH-RH serum to normally cycling rats prevented the preovulatory surge of LH and FSH as well as blocked ovulation.[8] We decided to try to develop an orally active, preferably nonpeptidic, antagonist of LH-RH, which we envisioned should competitively block the pituitary receptors and lead to the suppression of gonadal steroids.

It has been reported that an antifungal drug called ketoconazole (Nizoral, Janssen Pharmaceutica, Beerse, Belgium), when given orally to patients in a dosage of 200–1200 mg daily produces a dose-dependent suppression of serum testosterone and leads to the remission of prostate cancer.[9] The mechanism leading to the suppression of serum testosterone by ketoconazole was studied in great detail by Bhasin et al.[10] and it was concluded that the suppression of testosterone biosynthesis was primarily due to the inhibition of a number of enzymes in the biosynthetic pathway. During our search for a nonpeptidic lead in the LH-RH program, we became extremely interested in this antifungal compound and focused our attention primarily on its involvement at the pituitary level. To our surprise, we found that ketoconazole exhibits weak but competitive binding affinity for the pituitary LH-RH receptor. In this communication, we wish to report our

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