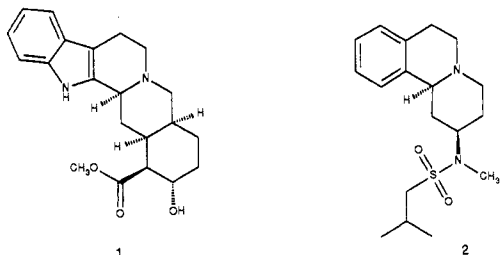


Communications to the Editor

(8 α ,12 α ,13 α)-5,8,8a,9,10,11,12,12a,13,13a-Decahydro-3-methoxy-12-(methylsulfonyl)-6H-isoquino[2,1-g][1,6]naphthyridine, a Potent and Highly Selective α_2 -Adrenoceptor Antagonist¹

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



octahydro-2H-benzo[b]furo[2,3-a]quinolizine⁵⁻⁷ and berbaine^{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 (8 α ,12 α ,13 α)-5,8,8a,9,10,11,12,12a,13,13a-decahydro-3-methoxy-12-(methylsulfonyl)-6H-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 5¹³ (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 8 α ,12 α ,13 α 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.

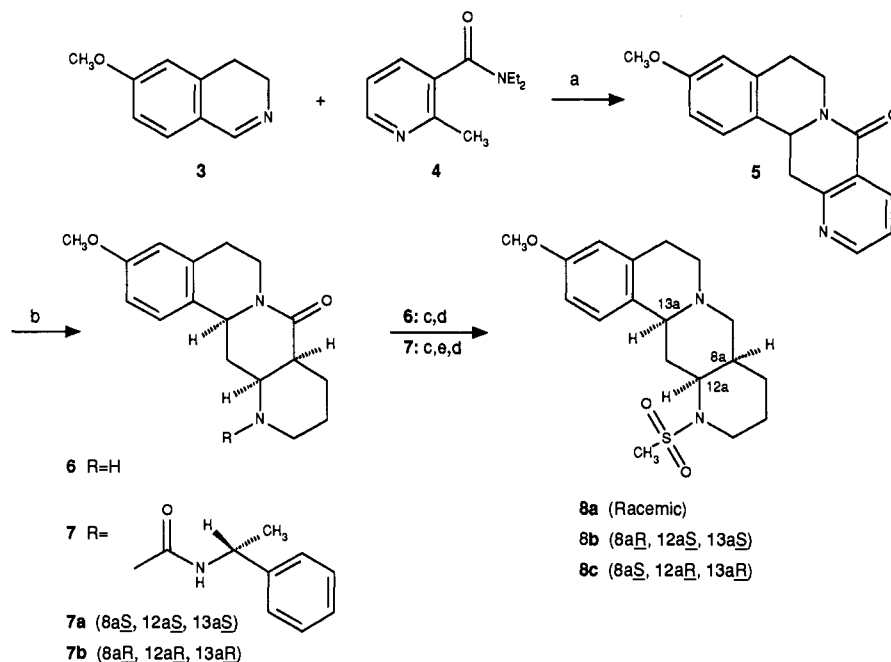
Resolution of 8a was effected by a variation of the optically active (1-phenylethyl)urea method.¹⁶ Thus, treatment of the racemate 6 with (+)-(*R*)-1-phenylethyl isocyanate afforded the diastereomeric ureas 7a and 7b, which were separated by medium-pressure chromatography. The diastereomeric purities of 7a and 7b so obtained were determined to be >98% by ¹H 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; [α]_D +13°, *c* 0.3, MeOH) and 8c (HCl salt, mp 256–258 °C; [α]_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 [³H]prazosin and [³H]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*

- (1) Contribution No. 780 from the Institute of Organic Chemistry.
- (2) Starke, K.; Borowski, E.; Endo, T. *Eur. J. Pharmacol.* 1975, 34, 385.
- (3) Starke, K.; Tanaka, T. *Eur. J. Pharmacol.* 1980, 63, 191.
- (4) (a) Lattimer, N.; McAdams, R. P.; Rhodes, K. F.; Sharma, S.; Turner, S. J.; Waterfall, J. F. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1984, 327, 312. (b) Ward, T. J.; White, J. F.; Lattimer, N.; Rhodes, K. F.; Sharma, S.; Waterfall, J. F. *J. Med. Chem.* 1988, 31, 1421.
- (5) Huff, J. R.; Anderson, P. S.; Baldwin, J. J.; Clineschmidt, B. V.; Guare, J. P.; Lotti, V. J.; Pettibone, D. J.; Randall, W. C.; Vacca, J. P. *J. Med. Chem.* 1985, 28, 1759.
- (6) Pettibone, D. J.; Clineschmidt, B. V.; Lotti, V. J.; Martin, G. E.; Huff, J. R.; Randall, W. C.; Vacca, J.; Baldwin, J. J. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1986, 333, 110.
- (7) Pettibone, D. J.; Clineschmidt, B. V.; Lotti, V. J.; Baldwin, J. J.; Huff, J. R.; Randall, W. C.; Vacca, J.; Young, S. D. *Naunyn-Schmiedeberg's Arch. Pharmacol.* 1987, 336, 169.
- (8) Vizi, E. S.; Harsing, L. G.; Gaal, J.; Kapocsi, J.; Bernath, S.; Somogyi, G. T. *J. Pharmacol. Exp. Ther.* 1986, 238, 701.
- (9) Vizi, E. S.; Toth, L.; Somogyi, G. T.; Szabo, L.; Harsing, L. G.; Szantay, C. *J. Med. Chem.* 1987, 30, 1355.
- (10) Chapleo, C. B.; Meyers, P. L.; Butler, R. C. M.; Doxey, J. C.; Roach, A. G.; Smith, C. F. C. *J. Med. Chem.* 1983, 26, 823.
- (11) Clark, R. D.; Michel, A. D.; Whiting, R. L. In *Progress in Medicinal Chemistry*; Ellis, G. P., West, G. B., Eds.; Elsevier: Amsterdam, 1986; Vol. 23, p 1.

- (12) Baumgarten, P.; Dornow, A.; *Ber. Dtsch. Chem. Ges. B* 1939, 72, 563.
- (13) Satisfactory elemental analyses ($\pm 0.4\%$ 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 8 $\alpha\beta$,12 $\alpha\beta$,13 $\alpha\alpha$ (30%) and 8 $\alpha\beta$,12 $\alpha\alpha$,13 $\alpha\alpha$ (5%) diastereomers were also formed and were separated from 6 chromatographically.
- (16) Schönerberger, 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.
- (19) Drew, G. M. *Br. J. Pharmacol.* 1978, 64, 293.

Scheme I^a

^a (a) LDA, THF, -40 °C. (b) H₂, HOAc, Rh-Al₂O₃. (c) LAH, THF. (d) MsCl, CH₂Cl₂, Et₃N. (e) NaOBu, *n*-BuOH.

Table I. Radioligand Binding and Functional Results

compd	ligand binding, pK _i ^a			functional antagonism, pA ₂ ^a		
	[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 [³H]yohimbine minus pK_i [³H]prazosin. ^c Reversal of phenylephrine-induced contraction. ^d Reversal of inhibitory effect of UK-14304 on contractile response to field stimulation. ^e Antilog of pA₂ (UK-14304) minus pA₂ (phenylephrine).

assays are given in Table I. Reversal of clonidine-induced mydriasis in the rat was used as an *in vivo* assay for central α₂-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 α₂-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 α₂-adrenoceptor antagonists of the octahydro-2*H*-benzo[*b*]furo[2,3-*a*]quinolizine class.⁵⁻⁷

Functional results obtained in the rat aorta (α₁) and guinea pig ileum (α₂) confirmed the potent and selective α₂-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 α₂-adrenoceptor antagonist reported to date. Furthermore, the α₂/α₁ selectivity of 8b appears to be unprecedented.²¹

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

compd	IC ₅₀ , ^a μg/kg		ratio oral/ <i>iv</i>
	<i>iv</i>	oral	
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 α₂-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

(20) Berridge, T. L.; Gadie, B.; Roach, A. G.; Tulloch, I. F. *Br. J. Pharmacol.* 1983, 78, 507.

(21) For example, L-657,743 has been reported to have a pK_i of 9.3 in ligand binding studies using both [³H]rauwolscine and [³H]clonidine with a selectivity ratio of 240 (ref 7). The berbaine CH-38083 had a pK_i of 8.7 ([³H]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.

Acknowledgment. We thank Dr. Henri Pinhas and Isabelle Jullien, Syntex Research France, for the chiral HPLC determinations and Dr. Frederick Hollander, University of California, Berkeley, for the X-ray crystallographic data. We are grateful to Lani Russell for preparing the manuscript.

(23) Gillespie, J. S. In *Adrenergic Activators and Inhibitors*, Part 1; Szekeres, L., Ed.; Springer-Verlag: Berlin, 1980.

(24) The B_{\max} is the number of fmoles of [³H]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.

† Syntex Research, Palo Alto.

‡ Syntex Research, Edinburgh.

Robin D. Clark,*† David B. Repke†
Andrew T. Kilpatrick,† Christine M. Brown†
Alison C. MacKinnon,† Ruth U. Clague†
Michael Spedding*†

*Institute of Organic Chemistry
Syntex Research
Palo Alto, California 94304
and Department of Pharmacology
Syntex Research Center
Edinburgh, Scotland EH 14 4AP
Received February 13, 1989*

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 Schally¹ 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 an 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 atrophy⁷ 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

(2) Hurley, D. M.; Brian, R.; Outch, K. *N. Engl. J. Med.* **1984**, *310*, 1069.

(3) (a) Labrie, F.; Belanger, A.; Cusan, L.; Sequin, G.; Pelletier, G.; Kelly, P. A.; Reeves, J. J.; Lefebure, F. A.; Lemay, A.; Gourdeau, Y.; Raymond, J. P. *J. Androl.* **1980**, *1*, 209. (b) Johnson, E. S.; Gendrich, R. L.; White, W. F. *Fertil. Steril.* **1976**, *27*, 853.

(4) (a) Rivier, C.; Vale, W.; Rivier, J. *J. Med. Chem.* **1983**, *26*, 1545. (b) DeSombre, E. R.; Johnson, E. S.; White, W. F. *Cancer Res.* **1976**, *36*, 3830.

(5) (a) van Steenbrugge, G. J.; Ultee-van Gessel, A. M.; Groen, M.; de Jong, F. H.; Schroeder, F. H. *Life Sci.* **1987**, *40*, 1335. (b) Garnick, M. B.; Glode, L. M. *N. Engl. J. Med.* **1984**, *311*, 1281.

(6) Manoni, A.; Santen, R.; Harvey, H.; Lipton, A.; Max, D. *Endocr. Rev.* **1986**, *7*, 89.

(7) Arimura, A.; Sato, H.; Kumasaka, T.; Worobec, R. B.; Debeljuk, L.; Dunn, J. D.; Schally, A. V. *Endocrinology* **1973**, *93*, 1092.

(8) Arimura, A.; Debeljuk, L.; Schally, A. V. *Endocrinology* **1974**, *95*, 323.

(9) Trachtenberg, J.; Pont, A. *Lancet* **1984**, *2*, 433.

(10) Bhasin, S.; Sikka, S.; Fielder, T.; Sod-Moriah, U.; Levine, H. B.; Swerdloff, R. S.; Rajfer, J. *Endocrinology* **1986**, *118*, 1229.

(1) Schally, A. V.; Arimura, A.; Baba, Y.; Naiy, R. M. G.; Matsuo, H.; Redding, T. W.; Debeljuk, L.; White, W. F. *Biochem. Biophys. Res. Commun.* **1971**, *43*, 393.