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ANALOGS OF UK 14,304: STRUCTURAL FEATURES RESPONSIBLE FOR α_2 ADRENOCEPTOR ACTIVITY

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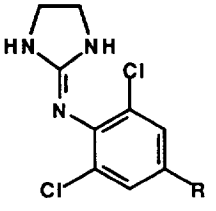
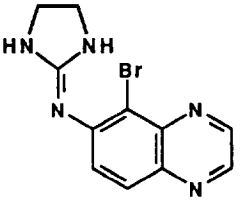
Abstract: Factors influencing the potency of UK 14,304 analogs including conformational preorganization and arrangement of hydrogen bond acceptors on the aromatic core are described The previously reported importance of twist of the iminoimidazoline ring relative to the core for enhanced α_2 activity is supported.

Agents stimulating α_2 adrenoceptors have been demonstrated to mediate a variety of physiologic functions. These functions include reduction of blood pressure, sedation, and inhibition of fluid secretion.[1] We have been interested in designing novel α_2 adrenoceptor agonists to reduce elevated intraocular pressure (IOP). This is a condition often associated with glaucoma.[2] This class of agents was first used to reduce IOP in 1966 when

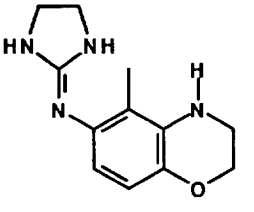
Figure I



R = H; Clonidine
R = NH_2; *p*-Aminoclonidine



UK 14,304



AGN 193080

Makabe demonstrated that clonidine, a potent α_2 adrenoceptor agonist, lowered IOP in man.[3] Two drawbacks limit the general application of clonidine for the treatment of elevated IOP. The first drawback is the ability of this agent to cross the blood-brain barrier that leads to centrally mediated side effects such as sedation. Polar agents including *p*-aminoclonidine have been introduced as CNS limited agents.[4] Clonidine's second drawback is its activity at α_1 receptors. Clonidine, like *p*-aminoclonidine, proved to be on the order of one hundred fold selective for α_2 versus α_1 adrenoceptors under our assay conditions. Enhanced α_2 versus α_1 selectivity would be a distinct

advantage for this class of agents to mitigate α_1 mediated effects including ocular vasoconstriction and mydriasis.[5] UK 14,304 is an agent with enhanced α_2 versus α_1 selectivity. This agent has proved to be 790 fold selective for the α_2 receptor versus the α_1 receptor under our assay conditions and has been used successfully to treat ocular hypertension.[6]

Our efforts to understand the features responsible for the activity of this class of compounds are detailed herein. The structure-activity relationships presented reveal the importance of conformational preorganization of both the imidazoline ring and the substituents appended to the *meta* and *para* positions. Hydrogen bonding and steric effects also influence α_2 activity. This study led to the design of AGN 193080, a potent and peripherally acting α_2 agonist.[7]

Agent synthesis proved straightforward. Appropriate aromatic amines were assembled as key intermediates. The clonidine/UK 14,304 pharmacophore was installed by treatment of the amine with imidazoline-2-sulfonic acid in either acetonitrile or *i*-butanol in the presence of triethylamine at reflux.[8] Preparation of the *N*-, *N'*-dimethyl-*N''*-arylguanidine, entry 4, was accomplished by converting 6-amino-5-bromoquinoxaline into the isothiocyanate with thiophosgene. Treatment of this intermediate with methylamine afforded the thiourea. This material was treated first with methyliodide and then with methylamine to afford the desired guanidine. The guanidine, entry 5, was obtained upon treatment of the aromatic amine with cyanamide. AGN 193080, shown as entry 12 in the table, incorporates polar functionality to minimize CNS penetration.

The agents were evaluated for their ability to discriminate between α_2 and α_1 adrenoceptors by determining binding affinities (K_i) for CHO-C10 (α_2 adrenoceptors)[9] and human cerebral cortex (α_1 adrenoceptors)[10] membrane preparations. Potent and selective agents were assessed for functional efficacy (EC_{50}) through their ability to inhibit the contractile response of rabbit *vas deferens* mounted between platinum electrodes and field stimulated.[11] Cumulative concentration-response curves in 0.25 log units were obtained for each agent. The reduction in electrically evoked peak height by the agents was measured and expressed as a percentage of the pre-treatment peak height. The EC_{50} was determined as the concentration that produced a 50% reduction in peak height. Results of the *in vitro* evaluation of the agents are presented in Table I.

Comparison of the binding affinities for UK 14,304 and entry 2 suggests that the substitution of an electron withdrawing subunit for an electron releasing subunit of comparable size has little effect on the activity of the agents. These subunits have been shown to induce a twist of the imidazoline relative to the quinoxaline nucleus.[12] These two agents are more potent at the α_2 receptor than the unsubstituted case shown in entry 3. The blocking subunit increases potency at the α_2 receptor to a greater extent than at the α_1 receptor.

The imidazoline ring of UK 14,304 is preorganized[13] relative to the agent shown in entry 4. Scission of the bond between the two carbons of the imidazoline ring causes a three order of magnitude reduction in α_2 potency. Removal of the methyl groups, entry 5, affords a two order of magnitude recovery in potency relative to the conformationally deconstrained case, entry 4. The lipophilic two carbon bridge of the imidazoline ring found in UK 14,304 does, however, afford a seventeen fold increase in binding affinity relative to entry 5.

Entry 6, a simplified agent retained α_2 activity. Enhanced activity was observed upon introduction of hydrogen bond acceptors in the *meta* and *para* positions relative to the pharmacophore, entry 7. This is the same arrangement of hydrogen bond acceptors found in norepinephrine, the endogenous ligand for α_2 receptors and demonstrates the importance of hydrogen bond acceptors *meta* and *para* positions relative to the iminoimidazoline moiety. The most potent agent in the series, entry 8, is realized upon combination of hydrogen bond acceptors at the *meta* and *para* positions relative to the iminoimidazoline moiety and a methyl group at the *ortho* position to induce twist of the imidazoline relative to the aromatic core. This arrangement also enhanced activity at α_1 receptors. Activity at α_1 receptors offers the potential of untoward side effects including a hypertensive response. The α_2/α_1 selectivity, however, remained high (500 fold). Comparison of entries 8 and 9 highlights the importance of preorganization in the right hand side of the molecule. An order of magnitude enhancement in activity is observed upon formation of the benzodioxane aromatic core versus the conformationally mobile analog, entry 9. We synthesized entry 10 to examine the effect of moving the methyl moiety from the aromatic core onto the nitrogen bridge between the aromatic subunit and the imidazoline subunit. Dexmedetomidine is a potent α_2 agonist possessing a methyl on the bridge between the aromatic core and the heterocyclic subunit suggesting that this should be an allowed modification. The agent shown in entry 10 proved dramatically less active than either the unsubstituted agent (entry 7)[14] or the C-5 methylated agent (entry 8). Modeling studies suggested that the agent was flat.[15]

Entry 11, a previously described agent incorporating amino functionality at the *meta* and *para* positions relative to the pharmacophore, was 900 fold selective for the α_2 receptor versus the α_1 receptor. This highly selective agent proved oxidatively unstable. Entry 12, AGN 193080, formed by replacement of the *para* amino with an alkylated oxygen atom for enhanced oxidative stability, was a potent and efficacious α_2 agonist. The agent had a 1.2nM binding affinity for the α_2 receptor and proved to be a potent, full agonist in the functional system. This agent was 390-fold selective for the α_2 receptor versus the α_1 receptor. While not as selective as UK 14,304, this agent had enhanced selectivity compared to clonidine.

Preliminary results from the *in vivo* evaluation of AGN 193080 have been presented and suggest that this agent does not cross the blood brain barrier.[7a] These studies suggest that AGN 193080 is efficacious for the reduction of intraocular pressure when applied topically in rabbit. AGN 193080 does not lower blood pressure when administered topically or intravenously to either rabbit or cynomologus monkeys. In sharp contrast, intracisternal injection in rabbit demonstrates that the agent dramatically reduces blood pressure when applied directly to the CNS reflecting enhanced potency at the receptor relative to clonidine. The agent has been demonstrated to be over 100 fold less sedating than clonidine in the rat activity model of sedation.

Many of the agents described herein have proved to be oxidatively stable in contrast to other peripherally acting agents including AGN 190851, AGN 192172[12], and *p*-aminoclonidine. A recent study has suggested that *p*-aminoclonidine can induce an allergic response.[16] This may be related to its hydroquinone-like structure. Agents possessing this class of subunit including amodiaquine are known to induce an allergic response.[17] Enhanced chemical stability may offer distinct advantages for reduced sensitization upon chronic administration. Full details of our studies with these agents will be reported.

Table I

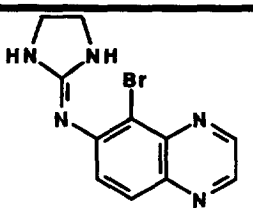
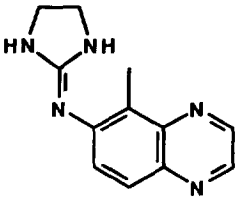
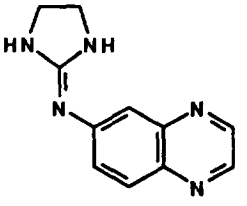
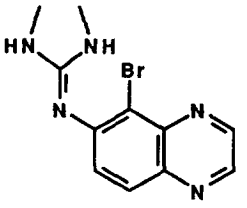
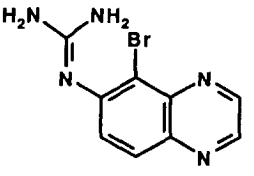
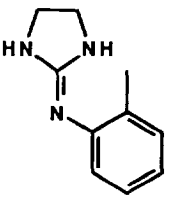
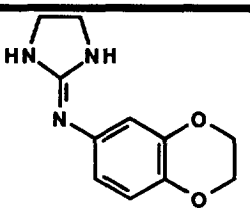
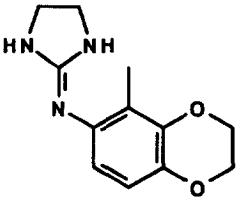
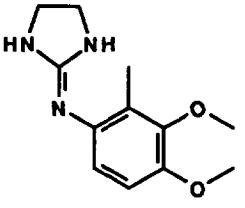
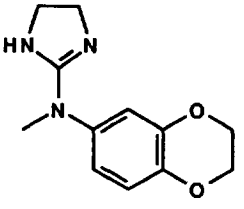
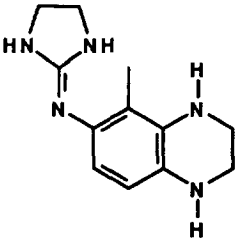
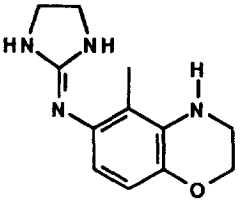
Entry	Agent	K_i (nM)[a]		EC_{50}
		α_1	α_2	α_2
1 (UK 14,304)		1,900 ± 100	2.4 ± 0.2	1.0 ± 0.1
2		1,400 ±100	1.3 ± 0.1	0.3 ± 0.3
3		11,000 ±1,000	41 ± 10	not tested
4		10,000 ± 1,000	5,300 ± 1,000	not tested
5		17,000 ± 3,000	41 ± 9.0	not tested
6		2,500 ± 150	7.8 ± 1.5	not tested

Table I con't.

Entry	Agent	K_i (nM)[a]		EC_{50}
		α_1	α_2	α_2
7		2,400 ± 500	3.1 ± 0.1	not tested
8		130 ± 4.5	0.25 ± 0.06	0.35 ± 0.08
9		1,500 ± 260	5.2 ± 1.3	not tested
10		30,000 ± 2,700	180 ± 30	not tested
11 (AGN 192172)		8,900 ± 1,000	9.6 ± 1.2	3.5 ± 1.7
12 (AGN 193080)		470 ± 21	1.2 ± 0.2	1.2 ± 0.1

References and Notes

1. (a) Maze, M.; Tranquilli, W. *Anesthesiology* **1991**, *74*, 581. (b) Hieble, J. P.; Ruffolo, Jr., R. R. In *Progress in Basic and Clinical Pharmacology: α-Adrenoceptors: Molecular Biology, Biochemistry, and Pharmacology*; Ruffolo, Jr., R. R. Ed.; S. Karger AG: Basel, 1991; Vol. 8, pp 180-220.

2. Shields, M.B. *Textbook of Glaucoma*; Williams & Wilkins: Baltimore, 1992; 3rd Ed.

3. Makabe, R. *Dtsch. Med. Wochenschr.* **1966**, *91*, 1686.

4. (a) Hernandez y Hernandez, H.; Cervantes, R.; Frati, A.; Hurtado, R.; McDonald, T. O.; DeSousa, B. *J. Toxicol. - Cut. & Ocular Toxicol.* **1983**, *2*, 99. (b) Rouot, B.; LeClerc, G. *Bull. Soc. Chim. Fr.* **1979**, 520.

5. (a) Hodapp, E.; Kolker, A. E.; Kass, M. A.; Goldberg, I.; Becker, B.; Gordon, M. *Arch. Ophthalmol.* **1981**, *99*, 1208. (b) Innemee, H. C.; de Jonge, A.; van Marel, J. C. A.; Timmermans, P. B. M. W. M.; van Zwieten, P. A. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1981**, *316*, 294.

6. Burke, J. A.; Potter, D. E. *Curr. Eye Res.* **1986**, *5*, 665.

7. (a) Munk, S. A.; Harcourt.; Burke, J.; Lai, R.; Roberts, D.; Small, D.; Gluchowski, C.; Manlapaz, C.; Padillo, E.; Kharlamb, A.; Runde, E.; Wheeler, L.; Garst, M. "Synthesis and Biological Evaluation of AGN 193080. A Potent and Selective Ocular Antihypertensive Agent." MEDI 193, 209th American Chemical Society National Meeting, Anaheim, CA, April, 1995. (b) Gluchowski, C. G. U.S. Patent 5 091 528, 1992. *Chem. Abstr.* **1993**, *118*, 234070.

8. (a) Maryanoff, C. A. ; Stanizione, R. C.; Plampin, J. N.; Mills, J. E. *J. Org. Chem.* **1986**, *51*, 1882. (b) Gluchowski, C.; Wong, H.; Garst, M. E. *ORGN 16*, 199th American Chemical Society Meeting, Boston MA, 1990.

9. Bylund, D. B.; Blaxall, H. S.; Iverson, L. J.; Caron, M. G.; Lefkowitz, R. J.; Lomasney, J. W. *Mol. Pharmacol.* **1992**, *42*, 1.

10. Greengrass, P.; Bremner, R. *Eur. J. Pharmacol.* **1979**, *55*, 323.

11. Brown, C. M.; McGrath, J. C.; Summers, R. J. *Br. J. Pharmacol.* **1979**, *66*, 553.

12. Munk, S. A.; Gluchowski, C.; Dolby, L.; Wong, H.; Burke, J.; Kharlamb, A.; Manlapaz, C.; Padillo, E.; Rodgers, D.; Ohta, B.; Wheeler, L.; Garst, M. *Bioorg. Med. Chem. Lett.* **1994**, *4*, 459.

13. (a) for an excellent overview see: Cram, D. J.; *Science* **1988**, *240*, 760 and references cited therein. (b) Dietrich, B.; Lehn, L.-M.; Suavage, J.-P.; *Tetrahedron Lett.* **1969**, *10*, 2885. (c) Pedersen, C. J. *J. Am. Chem. Soc.* **1967**, *89*, 2495.

14. (a) Chapleo C. B.; Doxey, J. C.; Frank, L. W.; Myers, P. L.; Roach, A. G.; Smith, C. F. C.; Virdee, N. K. *Eur. J. Med. Chem.* **1983**, *91*, 123. (b) Chapleo C. B.; Butler, R. C. M.; England, D. C.; Myers, P. L.; Roach, A. G.; Smith, C. F. C.; Stillings, M. R.; Tulloch, I. F. *J. Med. Chem.* **1989**, *32*, 1627.

15. The modeling study was conducted using a CAChe (Tektronics) modeling system. Minimum energy geometries were determined using the standard MM2 and optimized using the AM1 force field parameters contained in CAChe. This study suggested that the imidazoline pharmacophore and the aromatic core of entry 10 and the corresponding desmethyl agent (reported by Chapleo, *et al.* ref.14) were superimposable. This is in contrast to the twist of the imidazoline subunit relative to the aromatic core seen in the 5-methyl case (ref. 12)

16. Butler, P.; Mannschreck, M.; Lin, S.; Hwang, I.; Alvarado, J. *Arch. Ophthalmol.* **1995**, *113*, 293.

17. Sanner, M. A.; Higgins, T. J. In *Annual Reports in Medicinal Chemistry* Bristol, J., Ed., Acadamic Press: San Diego, 1991; Vol. 26, pp 181-190.

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