Pergamon

Bioorganic & Medicinal Chemistry Letters, Vol. 5, No. 9, pp. 991-996, 1995

0960-894X/95 $9.50+0.00

0960-894X(95)00154-9

PHENYL PYRROLIDINE ANALOGUES AS POTENT NICOTINIC ACETYLCHOLINE RECEPTOR (nAChR) LIGANDS

Richard L. Elliott,* Keith B. Ryther, David J. Anderson, Joanna L. Raszkiewicz§, Jeffrey E. Campbell, James P. Sullivan, and David S. Garvey.‡

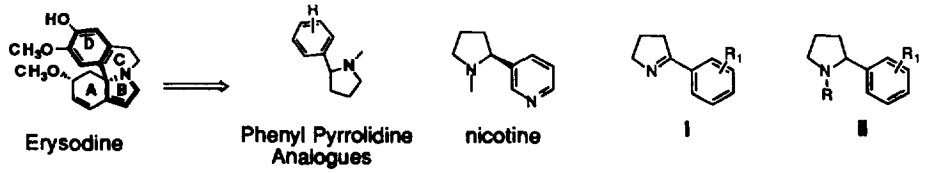
Neuroscience Research, Pharmaceutical Products Division, Abbott Laboratories, One Abbott Park Road, Abbott Park, IL 60064-3500.

Abstract: The synthesis and SAR of a series of 2-phenyl pyrrolidines as neuronal nAChR ligands are described. Substitution on the aryl ring had a dramatic effect on receptor binding affinity, with K_i values ranging from 46 nM to >10,000 nM. Analogues **8**, **9**, and **14** were the most potent ligands evaluated, having K_i values of 68 nM, 75 nM, and 46 nM; respectively.

The development of safe and efficacious agents for the treatment of Alzheimer's disease (AD) is one of the major medical challenges of this century, with over 4 million cases in the U.S. alone,[1] and the number expected to rise to nine million by the year 2040 at a cost estimated over 58 billion dollars per annum.[2] However, the search for agents to prevent, cure, or ameliorate the symptoms of AD is complicated by many factors, including the slow progression of the disease, the lack of good diagnostic tools for early identification of the disease, the heterogeneity of the disease, and the lack of appropriate animal models which mimic the neurodegenerative processes which occur in AD.

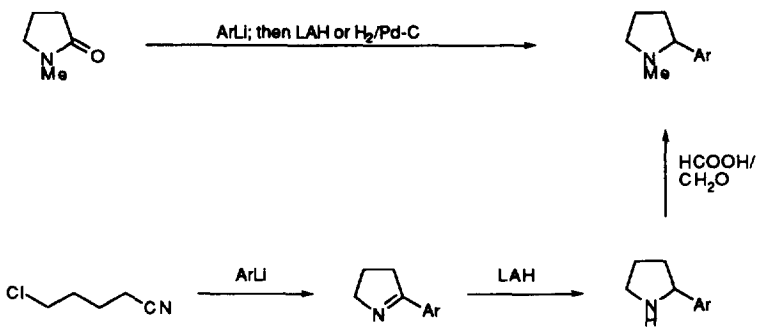
Recent studies have demonstrated that nicotine and other nicotinic agonists are potent cognition enhancers in a number of behavioral paradigms in both rodents[3-9] and non-human primates.[10, 11] Furthermore, nicotine has been shown to be efficacious in improving the cognitive performance of AD patients.[12-14] Unfortunately, nicotine's cognitive-enhancing properties are accompanied by a number of undesirable properties, including addictive liability as well as cardiovascular and gastrointestinal effects.[15] The discovery of novel and selective nAChR ligands may allow for the development of nicotinic-based therapeutic agents devoid of the these undesirable side-effects.[16,17] Toward this end, we recently reported[18-21] on novel 2-pyrrolidinyl isoxazoles which are potent and selective nAChR ligands with cognition-enhancing properties.

Our continuing search for novel nAChR agents led us to examine the structures of other known, potent nAChR ligands. We became interested in the group of *Erythrina* alkaloids, many of which bind potently to the nicotinic receptor.[22a-g] In particular, the tetracyclic structure of the aromatic class of *erythrina* alkaloids, as exemplified by erysodine (K_i = 5 nM)[22g], possess key elements as defined in the Beers-Reich pharmacophore model for good nAChR binding,[23] including a basic nitrogen and a number of oxygen atoms to serve as hydrogen-bond acceptors. However, it was unclear whether the oxygenated phenyl ring (**D**-ring) or the oxygenated **A**-ring was fulfilling this role. Conceptually, removal of the **A**- and **C**-rings and reduction of the pyrroline **B**-ring affords an oxygenated phenyl pyrrolidine structure, which can formally be considered as des-aza analogue of nicotine. We report herein the synthesis and nAChR binding properties of these phenyl pyrroline (**I**) and phenyl pyrrolidine (**II**) analogues.



Methods: The phenyl pyrrolidine (racemic) and pyrroline analogues were synthesized as outlined in Scheme 1. Most of the *N*-methyl compounds were synthesized via addition of the appropriately substituted aryllithium reagent (commercially available or synthesized via metal-halogen exchange from the corresponding aryl bromide or iodide) to *N*-methylpyrrolidinone, followed by *in situ* reduction of the resultant aminal using either $LiAlH_4$ (LAH) or H_2/Pd-C.[24] The pyrroline compounds were synthesized by addition of the aryllithium to 4-chlorobutyronitrile;[25,26] subsequent LAH reduction of the pyrrolines afforded the corresponding *N*-H pyrrolidine analogues. All final compounds were fully characterized by ^{1}H-NMR, ms, and elemental analysis.

Scheme 1.



The compounds were tested for nAChR binding using [^{3}H]-(-)-cytisine (selective for the $\alpha_4\beta_2$ nAChR subtype) following a modified procedure of Pabreza et al,[27] using a whole rat brain preparation. [^{125}I]-α-bungarotoxin (α-Bgt, selective for the α_7-homomeric nAChR subtype) binding was determined in membranes prepared from whole rat brain using a modification of the method of Marks et al.[28]

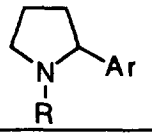
Results and Discussion

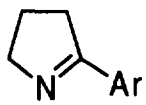
In general, aromatic substitution on the phenyl pyrrolidine had a dramatic effect on binding affinity for the neuronal nACh receptor, with binding affinities of the 22 analogues synthesized ranging from 46 nM to >10,000 nM. Although the *N*-unsubstituted phenyl pyrrolidine analogue **1** had very low affinity for the receptor (K_i = 8696 nM) the *N*-Me analogue **2** had substantially higher affinity (K_i = 450 nM) at the brain nAChR. The binding affinity of compound **2** has been previously published K_i values of 860 nM[29] and 3500 nM[30] using [^{3}H]-cytisine and [^{3}H]-nicotine, respectively.

The high K_i values for compounds **3** and **10** reveals that substitution at the *ortho*-position of the phenyl ring is not well tolerated. *Meta*-substitution tended to have minimal effect on the binding affinity, although the K_i was

slightly improved in the case of the *meta*-chloro analogue **6**. The 3-methoxy analogue **11** bound with a K_i value of 597 nM, compared to the reported value of 1200 nM.[29] In contrast to the *meta*-substituted analogues, *para*-substitution caused large variations in the K_i values, which ranged from under 100 nM in several cases to over 1000 nM. Within the halogen-substituted series, the highest binding affinity was found for those analogues having either a chlorine or bromine atom at the *para* -position, although di-substitution at the *meta* -and *para*-positions was also tolerated. The 3,4-methylenedioxy analogue **14** had the highest potency (K_i = 46 nM) of all the analogues tested; surprisingly the closely related oxygenated analogues **11-13**, **15**, and **16** had much poorer binding affinity. As with the secondary amine analogue **1**, the phenyl pyrroline analogues **21** and **22** had very low affinity for the nACh receptor. No further pyrroline analogues were synthesized in view of their low activity.

Table 1. nAChR Binding of Phenyl Pyrrolidine Analogues.



compd[a]	R	Ar	KI (nM)[b]
1	H	phenyl	8696 ± 354
2	Me	phenyl	450 ± 56
3	Me	2-methylphenyl	9000 ± 625
4	Me	3-methylphenyl	719 ± 167
5	Me	4-methylphenyl	1333 ± 360
6	Me	3-chlorophenyl	227 ± 92
7	Me	4-chlorophenyl	164 ± 51
8	Me	3,4-dichlorophenyl	68 ± 8
9	Me	4-bromophenyl	75 ± 6
10	Me	2-methoxyphenyl	>10,000 (3)
11	Me	3-methoxyphenyl	597 ± 23
12	Me	4-methoxyphenyl	707 ± 80
13	Me	3,4-dimethoxyphenyl	9,700 ± 976
14	Me	3,4-methylenedioxyphenyl	46 ± 13
15	Me	3,4-ethylenedioxyphenyl	1220 ± 238
16	Me	3-methoxy-4-hydroxyphenyl	8250 ± 1108
17	Me	3-fluorophenyl	399 ± 23
18	Me	4-fluorophenyl	207 ± 21
19	Me	3,4-difluorophenyl	420 ± 28
20	Me	4-trifluoromethylphenyl	931 ± 76
			
21	----	4-chlorophenyl	>10,000 (3)
22	----	3-fluorophenyl	>10,000 (3)

[a] All final compounds were fully characterized by ^{1}H-NMR and ms, and had elemental analyses within ±0.4% of theoretical values. b The compounds were tested for nAChR binding in a whole rat brain preparation using [^{3}H]-(-)-cytisine, following a modification of the procedure of Pabreza et al.[27] Values are the means ± S.E.M.; n = 3-4.

In addition to the binding experiments using [^{3}H]-cytisine, which has been shown to bind with high affinity to the $\alpha4\beta2$ nAChR subtype, a major nAChR subtype accounting for greater than 90% of the high affinity [^{3}H]-nicotine binding sites in the CNS,[31,32] two of the most potent analogues (**7** and **14**) were also tested using [^{125}I]-α-bungarotoxin as the radioligand, which binds with high affinity to a putative $\alpha7$-homomeric binding site, a second major nAChR found in the CNS.[33,34] Both analogues displayed low affinity for this binding site, having K_i values greater than 10,000 nM.

As mentioned above, analogues **2** and **11** have been previously synthesized and tested by others as nAChR agents. Glennon et al.[29] reported that the phenyl analogue and *m*-methoxyphenyl analogue had much reduced affinity for the nACh receptor (*vis a vis* nicotine), and hence concluded that the pyridyl nitrogen is important for good binding. However, it appears that a significant portion of the binding affinity lost upon removing the pyridyl nitrogen can be recovered with an appropriately substituted phenyl ring. Although the binding affinity of these nAChR ligands is significantly lower than nicotine (K_i = 1.15 nM), they still bind better than one might predict considering the importance traditionally imputed to the pyridyl nitrogen for hydrogen-bonding in the classical Beers-Reich[23] pharmacophore model. Analogues possessing heteroatoms attached to the phenyl moiety, such as compound **14**, may be able to participate in hydrogen bonding interactions, although it is surprising that analogues **11**, **13**, **15**, and **16**, which also possess hydrogen bond acceptors, bind poorly. Compound **16** possesses the same 3-methoxy-4-hydroxyphenyl aromatic ring as found in Erysodine, yet has very low affinity for the nAChR receptor. The reasons why only the 3,4-methylenedioxy phenyl moiety, and none of the other closely related oxygenated phenyl rings, substituted for the 3-pyridyl ring of nicotine are not known.

Interestingly, the *para*-halogenated analogues **7**, **8**, and **9**, which are unable to participate in any significant hydrogen-bonding interactions, show significant binding affinity. Although the exact nature of the interaction of the substituted phenyl ring with the receptor is not known, binding interactions such as π-π interactions and/or hydrophobic interactions may contribute to the binding of these analogues. In any case, it is clear that the substitution pattern on the phenyl moiety of these phenylpyrrolidine analogues is important and can significantly modulate the binding affinity for the nicotinic receptor.

In conclusion, substituted phenyl rings have been demonstrated to serve as novel bioisosteric replacements for the 3-pyridyl ring of nicotine, with the 3,4-methylenedioxy, 3,4-dichloro, and 4-bromo analogues demonstrating the highest binding affinity for the nACh receptor. Although still 10-20 fold less potent than nicotine, these analogues represent novel non-heteroaromatic containing ligands which should serve as leads towards developing even more potent nAChR ligands.

* Author to whom correspondence should be addressed: Richard L. Elliott, Ph.D., Anti-infective Research Area, Dept. 47N, Abbott Park, IL 60064-3500.

§ Present Address: Natural Product Services, 1510 University Parkway, Salt Lake City, Utah 84115.

‡ Present Address: NitroMed Research Laboratories, 801 Albany Street, Boston, MA 02118.

REFERENCES

1) Schehr, R. S. *Bio/Technology* **1994**, *12*, 140.

2) Max, W. *Neurology* **1993**, *43 (8)*, S6.

3) Brioni, J. D.; Arneric', S. A. *Behavioral and Neural Biology* **1993**, *59*, 57.

4) Decker, M. W.; Majchrzak, M. J.; Anderson, D. J. *Brain Research* **1992**, *572*, 281.

5) Levin, E. D. *Psychopharmacology* **1992**, *108*, 417.

6) Bovet, D.; Bovet-Nitti, F.; Olivero, A. *Psychopharmacol.* **1966**, *10*, 1.

7) Evangelista, A. M.; Gattoni, R. C.; Izquierdo, I. *Pharmacology* **1970**, *3*, 91.

8) Haroutunian, V.; Barnes, E.; Davies, K. L. *Psychopharmacol.* **1985**, *87*, 266.

9) Gray, J. A.; Mitchell, S. N.; Joseph, M. H.; Grigoryan, G.; Dawe, S.; Hodges, H. *Drug Dev Res.* **1994**, *31*, 3.

10) Elrod, K.; Buccafusco, J. J.; Jackson, W. J. *Life Sciences* **1988**, *43*, 277.

11) Buccafusco, J.; Jackson, W. *Neurobiology of Aging* **1991**, *12*, 233.

12) Sahakian, B.; Jones, G.; Levy, R., Gray; J.; Warburton, D. *British Journal of Psychiatry* **1989**, *154*, 797.

13) Jones, G. M. M.; Sahakian, B. J.; Levy, R.; Warburton, D. M.; Gray, J. A. *Psychopharmacology* **1992**, *108*, 485.

14) Newhouse, P. A.; Sunderland, T.; Tariot, P. N.; Blumhardt, C. L.; Weingartner, H.; Mellow, A.; Murphy, D. L. *Psychopharmacology* **1988**, *95*, 171.

15) Benowitz, N. L. *New England J. Med.*, **1988**, *319*, 1318.

16) Arneric´, S. P.; Williams, M. *Int. Acad. Biomed. Drug Res.* **1993**, *7*, 30.

17) Williams, M.; Sullivan, J. P.; Arneric´, S. P. *Drug News and Perspectives* **1994**, *7*, 205.

18) Garvey, D. S.; Wasicak, J. T.; Decker, M. W.; Brioni, J. D.; Buckley, M. J.; Sullivan, J. P.; Carrera, G. M.; Holladay, M. W.; Arneric´, S. P.; Williams, M. *J. Med. Chem.* **1994**, *37*, 1055.

19) Arneric´, S. P.; Sullivan, J. P.; Briggs, C. A; Donnelly-Roberts, D.; Anderson, D. J.; Raszkiewicz, J. L.; Hughes, M.; Cadman, E. D.; Adams, P.; Garvey, D. S.; Wasicak, J.; Williams, M. *J. Pharmacol. Exp. Therap.* **1994**, *270 (1)*, 310.

20) Decker, M. W.; Brioni, J. D.; Sullivan, J. P.; Buckley, M. J.; Radek, R. J.; Raszkiewicz, J. L.; Kang, C. H.; Kim, D. J. B.; Giardina, W. J.; Wasicak, J.; Garvey, D. S.; Williams, M.; Arneric´, S. P. *J. Pharm. Exp. Ther.* **1994**, *270 (1)*, 319.

21) Lin, N.-H.; He, Y.; Anderson, D. J.; Wasicak, J. T.; Kasson, R.; Sweeny, D.; Sullivan, J. P. *Bioorg. Med. Chem. Lett.* **1994**, 4(20), 2389.

22) a) Dyke, S. F.; Quessy, S. N. *The Alkaloids. Chemistry and Physiology;* Rodrigo, R. G. A., Ed.; Academic Press, Inc.: New York, 1981; pp 1-98; b) Lehman, A. J. *J. Pharmacol. Exp. Ther.* **1937**, *60*, 69; c) Luetje, C. W.; Patrick, J. W. *Soc. Neurosci. Abstr.* **1989**, *15*, 677; d) Megirian, D.; Leary, D. E.; Slater, I. H. *J. Pharmacol. Exp. Ther.* **1955**, *113*, 212; e) Williams, M.; Robinson, J. L. *J. Neurosci.* **1984**, *4*, 2906. f) Anderson, D. J.; Arneric´, S. P.; Swanson, S.; Raszkiewicz, J. L.; Sullivan. J. P. *Soc. Neurosci. Abstr.* **1993**, *19*, 1536, Abstr. # 631.15; g) Decker, M. W.; Anderson, D. J.; Brioni, J. D.; Donnelly-Roberts, D. L.; Kang, C. H.; O'Neill, A. B.; Piattoni-Kaplan, M.; Swanson, S.; Sullivan, J. P. *European Journal of Pharmacology*, in press.

23) Beers, W. H.; Reich, E. *Nature (London)* **1970**, *228*, 917.

24) Hwang, Y. C.; Chu, M.; Fowler, F. W. *J. Org. Chem.* **1985**, *50*, 3885.

25) Craig, L. C.; Bulbrook, H.; Hixon, R. M. *J. Amer.Chem. Soc.* **1931**, *53*, 1831.

26) Starr, D. F.; Bulbrook, H.; Hixon, R. M. *J. Amer. Chem. Soc.* **1932**, *54*, 3971.

27) Pabreza, L. A.; Dhawan, S.; Kellar, K. J. *Mol. Pharmacol.* **1990**, *39*, 9.

28) Marks, M. J.; Stitzel, J. A.; Romm, E.; Wehner, J. M.; Collins, A. C. *J. Pharm. Exp. Ther.* **1986**,*30*, 427.

29) Glennon, R. A.; Maarouf, A.; Fahmy, S.; Martin, B.; Fan, F.; Yousif, M.; Shafik, R. M.; Dukat, M. *Med. Chem. Res.* **1993**, *2*, 546.

30) Caldwell, W. S.; and Lippiello, P. M. US Patent Number 5,248,690 assigned to R.J. Reynolds Tobacco Company. Sept. 28, 1993.

31) Flores, C. M.; Rogers, S. W.; Pabreza, L. A.; Wolfe, B.; Kellar, K. J. *Mol. Pharmacol* . **1992**, *41*, 31.

32) Whiting, P.; Schoepfer, R.; Lindstrom, J.; Priestly, T. *Mol. Pharmacol.* **1992**, *40*, 463.

33) Clarke, P. B. S.; Schwartz, R. D.; Paul, S. M.; Pert, C. B. ; Pert, A. *J. Neurosci.* **1985**, *5*, 1307.

34) Seguela, P.; Wadiche, J.; Dineley-Miller, K.; Dani, J. A.; Patrick, J. W. *J. Neurosci.* **1993**, *13*, 596.

(*Received in USA* 6 *March* 1995; *accepted* 28 *March* 1995)