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STRUCTURE–ACTIVITY STUDIES ON A NOVEL SERIES OF CHOLINERGIC CHANNEL ACTIVATORS BASED ON A HETEROARYL ETHER FRAMEWORK

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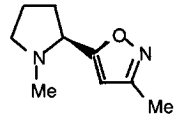
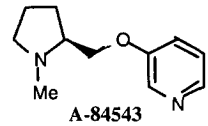
Abstract: Analogs of compound **1** with a variety of azacycles and heteroaryl groups were synthesized. These analogs exhibited K_i values ranging from 0.15 to > 10,000 nM when tested *in vitro* for cholinergic channel receptor binding activity (displacement of [^{3}H](-) cytisine from whole rat brain synaptic membranes).

Consistent losses of nicotinic receptors have been measured in the brain tissue of patients suffering from neurodegenerative disorders such as Alzheimer's disease and Parkinson's disease[1] Thus, neuronal nicotinic acetylcholine receptors (nAChRs) have been suggested in recent years as potential molecular targets for the development of agents to treat CNS disorders. New findings related to the molecular biology of this receptor class[2] and the development of novel ligands[3,4] have prompted increased interest in this therapeutic approach.

Recently, we have identified ABT-418, an isoxazole bioisostere of (*S*)-nicotine, that selectively interacts with nAChRs. [3,5–8] ABT-418 differentially activates nAChR subtypes to elicit a variety of behavioral effects, including cognitive-enhancing and anxiolytic-like activity,[3,9–11] while maintaining a reduced side-effect liability profile compared to the classical nAChR agonist, (*S*)-nicotine.[10] More recently, several members of a novel series of 3-pyridyl ether compounds, such as A-84543 (**1**), were described[12] which possess subnanomolar binding affinity for nAChRs and differentially activate specific subtypes of neuronal nAChRs. Since compound **1** exhibits high affinity for nAChRs, and was shown to have agonistic properties in the $^{86}Rb^+$ efflux functional assay in cell lines containing nAChRs, we focused our attention on structural modifications of this compound with a view of exploring the structural requirement for potent interaction with nAChRs. In this paper we describe structure-activity studies on the azacyclic and heteroaryl moieties of the pyrrolidinyl heteroaryl methyl ether based structure.



ABT-418



A-84543

1

Chemistry: The 2-substituted and 3-substituted analogs were synthesized as outlined in Scheme 1. Most of the N-methyl pyrrolidine analogs, as exemplified by the preparation of **4**, were synthesized via addition of the sodium salt of N-methyl-pyrrolidinol to the corresponding heteroaryl bromide. Pyridyl analogs were synthesized employing the Mitsunobu reaction as described previously[12] The azetidine analog **14** was prepared by reaction of the sodium salt of *N*-Boc-azetidinol with 5-bromo-pyrimidine followed by deprotection with trifluoroacetic acid. N-methylation with formic acid and formaldehyde furnished compound **15**.

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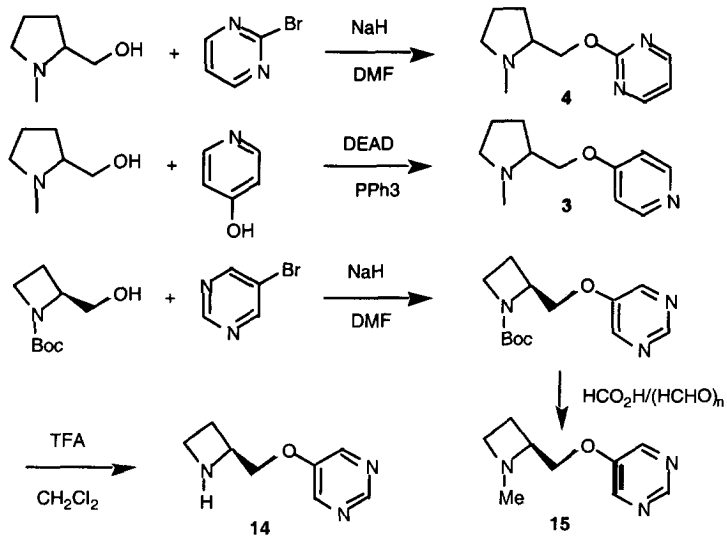


Scheme 1

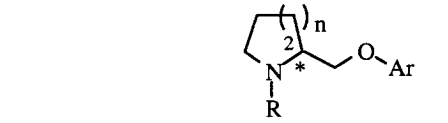
Results and Discussion: Two of the major classes of nAChRs in the brain are those labeled with high affinity by [^{3}H](-)-nicotine or [^{3}H](-) cytisine, and those labeled with high affinity by [^{125}I]α-bungarotoxin (α–BgT) [13] The distribution of high affinity nicotine binding sites in rat brain tends to coincide with the distribution of the α4β2 subunit combination in rat brain,[13,14] which is supported by immunoprecipitation experiments demonstrating that greater than 90% of the high affinity nAChR binding sites can be precipitated by antibodies raised against the α4 and β2 subunits[15] A good correlation has also been noted between the distribution of α7 mRNA and that of the high affinity binding sites for α–BgT in rodent brain.[13]

Our previous investigations have shown that A-84543 (**1**) potently interacts (K_i = 0.15 nM) with the putative α4β2 subtype of nAChRs, but has much lower affinity (K_i = 1530 nM) for the putative α7 nAChR subtype[16] Thus, in the present study, the interaction of all analogs with the putative α4β2 subtype was investigated. The ability of the more potent analogs to displace [^{125}I]α-BgT binding was also examined.

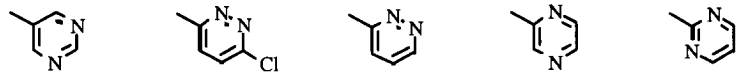
The effect of the heteroaromatic moiety on binding affinity was examined. As shown in Table 1, replacement of the 3-pyridyl fragment of potent compound **1** with a 2-pyridyl group (i.e., **2**) resulted in a 3300-fold decrease in binding affinity towards the [^{3}H]-cytisine binding site. A further reduction in binding affinity (>10,000-fold) was observed in analog **3** (K_i = 8000 nM) having a 4-pyridyl substituent. Thus, a significant portion of the binding affinity is lost upon shifting the nitrogen atom from the 3-position (numbered from ether appendage) to either the 2- or 4-position. These results suggest that the C3 nitrogen atom plays an important role in the binding activity. When an additional nitrogen atom is incorporated into the heteroaromatic ring to give 2-pyrimidinyl analog **4**, a 7-fold lower affinity than 2-pyridyl compound **2** is observed. Replacement of the 3-pyridyl group with 2-pyrazinyl moiety (i.e., compound **5**) resulted in a 145-fold decrease in binding potency. Changing the 2-pyrazinyl group to a 5-pyrimidinyl (**7**) moiety decreased the K_i value to 2.4 nM (a 16-fold decrease for the corresponding 3-pyridyl analog), while replacement of 3-pyridyl ring with a 3-pyridazinyl group (i.e., **8**) resulted in a 286-fold decrease in binding potency. In general, heteroaryl analogs containing two

nitrogen atoms exhibit lower affinity than the corresponding 3-pyridyl analogs. The deleterious effect is due to the presence of an additional nitrogen atom on the ring, which may possibly interfere with the hydrogen bond to the receptor that is important in the proposed nicotinic pharmacophores.[17,18] In addition, compounds that possess the nitrogen atom at the 3 position of the pyridine ring(numbered from ether appendage) are usually more potent than the ones without it. Based on these results, nitrogen atom appears to be important for optimal binding potency for the heteroaryl methyl ether analogs. It is also interesting to note that introduction of the electron withdrawing group such as chloro atom has only little effect on binding affinity (**8** vs **9**).

Table 1. Binding data of heteroaryl pyrrolidine methyl ether analogs



Ar :



5-pyrimidinyl | 3-chloro-6 pyridazinyl | 3-pyridazinyl | 2-pyrazinyl | 2-pyrimidinyl

Compound	Stereochemistry at C2	azacycle	Ar	[^{3}H]Cytisine Binding K_i (nM)[a]
1	*S*	1-methyl-pyrrolidinyl	3-pyridyl	0.15 ± 0.01
2	*S*	1-methyl-pyrrolidinyl	2-pyridyl	495 ± 10
3	*S*	1-methyl-pyrrolidinyl	4-pyridyl	7914 ± 26
4	*S*	1-methyl-pyrrolidinyl	2-pyrimidinyl	1223 ± 431
5	*S*	1-methyl-pyrrolidinyl	2-pyrazinyl	16.3 ± 0.7
6	*R*	1-methyl-pyrrolidinyl	2-pyrazinyl	406 ± 9
7	*S*	1-methyl-pyrrolidinyl	5-pyrimidinyl	2.39 ± 0.08
8	*S*	1-methyl-pyrrolidinyl	3-pyridazinyl	42.07 ± 1.88
9	*S*	1-methyl-pyrrolidinyl	3-chloro-6-pyridazinyl	29 ± 3
10	*R*	1-methyl-pyrrolidinyl	3-chloro-6-pyridazinyl	85.35 ± 3.05
11	*S*	1-methyl-azetidinyl	3-chloro-6-pyridazinyl	63.82 ± 2.88
12	*S*	azetidinyl	3-chloro-6-pyridazinyl	15.37 ± 0.36
13	*S*	1-methyl-azetidinyl	2-pyridazinyl	28.17 ± 0.83
14	*S*	azetidinyl	5-pyrimidyl	0.13 ± 0.02
15	*S*	1-methyl-azetidinyl	5-pyrimidyl	6.54 ± 0.34

[a] The ability of compounds to displace [^{3}H](-)-cytisine binding to whole rat brain membranes was performed as described. Values are the means ± S. E. M; n = 3–4. In all cases, the Hill co-efficient was close to unity indicative of an interaction with a single class of binding sites.

To examine whether heteroaryl analogs have the same stereochemical bias to nicotinic receptors at C2' position as we observed with **1** and its corresponding enantiomer,[12] the enantiomers of compounds **5** and **9** also

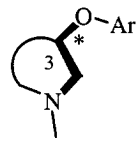
were evaluated for the affinity at receptor binding sites. Table 1 reveals that (*S*)-2-pyrazyl (**5**) and (*S*)-3-chloro-6-pyridazinyl (**9**) analogs were more potent than the corresponding (*R*)-enantiomers **6** and **10**.

The effect of the azacyclic moiety on the binding affinity followed the trend observed in the parent A-84543 **1** series.[12] (*S*)-*N*-Me azetidine compounds have lower affinity than their corresponding (*S*)-*N*-Me pyrrolidine compounds (**9** vs **11** and **7** vs **15**). With respect to N-methylation in azetidine analogs, the trends are similar to those found for the 3-pyridyl ether analogs. Thus, for the corresponding azetidines, the (*S*)-*N*-H compounds (**12** and **14**) exhibit 4- to 50-fold higher affinity than the (*S*)-*N*-Me congeners (**11** and **15**).

We are also interested in the 3-substituted compounds, as exemplified by the structure shown in Table 2. These analogs possess the key elements as defined in the 2-substituted analogs for good nAChR binding, including a basic nitrogen and a two carbon ether chain (highlighted by the bold line) between the nitrogen atom and heteroaryl moiety. As demonstrated in Table 2, the 3-substituted heteroaryl ether analogs are usually less potent than the corresponding 2-substituted analogs. In the case of 3-pyridyl compounds, the 2-substituted (**1**) and 3-substituted analogs (**17**) displayed a 25-fold difference in binding affinities. In contrast to 3-pyridyl analogs, greater differences in binding potency were observed with 3-chloro-6-pyridazinyl analogs (**9** is 172-fold more potent than **19**). The trend regarding the effect of heteroaryl moiety on binding affinities in the 3-substituted series is similar to that observed in the 2-substituted series with an order of potency 3-pyridyl > 5-pyrimidinyl > 3-chloro-6-pyridazinyl > 2-pyrazinyl > 2-pyridyl (cf. **16**, **21**, **19**, **20**, **18**).

The effect of stereochemistry on the binding affinities at the C3 position of pyrrolidine ring also was examined. In contrast to the 100-fold difference in binding potency with compound **1** and its enantiomer[12] the (*S*)-3-pyridyl analog (**17**) is only 2-fold more potent than its corresponding *R* enantiomer (**16**).

Table 2. Binding Data of Heteroaryl Azacyclic Methyl Ether Analogs



Compound	Stereochemistry at C3	azacycle	Ar	[^{3}H]Cytisine Binding K_i (nM)[a]
16	*R*	1-methyl-pyrrolidinyl	3-pyridyl	4.44 ± 0.16
17	*S*	1-methyl-pyrrolidinyl	3-pyridyl	2.67 ± 0.25
18	*R,S*	1-methyl-pyrrolidinyl	2-pyridyl	> 10,000
19	*R,S*	1-methyl-pyrrolidinyl	3-chloro-6-pyridazinyl	46.4 ± 0.5
20	*R,S*	1-methyl-pyrrolidinyl	2-pyrazinyl	359 ± 26
21	*R,S*	1-methyl-pyrrolidinyl	5-pyrimidinyl	11.7 ± 0.4
22	*R,S*	1-methyl-pyrrolidinyl	3-pyridylmethyl	11900 ± 1920

[a] The ability of compounds to displace [^{3}H](-)-cytisine binding to whole rat brain membranes was performed as described. Values are the means ± S. E. M; n = 3–4. In all cases, the Hill co-efficient was close to unity indicative of an interaction with a single class of binding sites.

We are also interested in the N1-substituted compounds, as exemplified by the structure shown in Table 3. These analogs also possess the key elements as defined in the 2-substituted analogs for good nAChR binding (highlighted by the bold line). As demonstrated in Table 3, the N1-substituted heteroaryl ether analog is usually less potent than the corresponding 2- and 3-substituted analogs when heteroaryl is 3-pyridyl, 2-pyrazinyl or 3-cloro-6-pyridazinyl moiety. In the case of the 2-pyridyl analogs, the N1-substituted analog **24** is more potent than the corresponding 2-substituted (**2**) and 3-substituted (**18**) analogs. The trend regarding the effect of heteroaryl moiety on binding affinities in the N1-substituted series is different from those observed in the 2- and 3-substituted series with an order of potency 2-pyridyl > 3-pyridyl > 2-pyrazinyl > 3-chloro-6-pyridazinyl ≥2-pyrimidinyl (cf. **24**, **23**, **26**, **27**, **25**).

Table 3. Binding Data of Heteroaryl Azacyclic Methyl Ether Analogs

Compound	azacycle	Ar	[^{3}H]Cytisine Binding K_i (nM)[a]
23	pyrrolidinyl	3-pyridyl	240 ± 30
24	pyrrolidinyl	2-pyridyl	160 ± 10
25	pyrrolidinyl	2-pyrimidinyl	1400 ± 300
26	pyrrolidinyl	2-pyrazinyl	560 ± 70
27	pyrrolidinyl	3-chloro-6-pyridazinyl	1300 ± 500

[a] The ability of compounds to displace [^{3}H](-)-cytisine binding to whole rat brain membranes was performed as described. Values are the means ± S. E. M; n = 3–4. In all cases, the Hill co-efficient was close to unity indicative of an interaction with a single class of binding sites

Four selected potent analogs, namely, **14**, **15**, **16** and **17**, were also investigated for their ability to displace [^{125}I]α-BgT binding to rat brain membranes. All of the compounds were found to display very weak affinity toward this nAChR subtype (K_i > 10,000 nM). While the binding affinity of these ligands for the putative α7 subtype is much lower than that found for the putative α4β2 subtype, it is important to point out that acetylcholine, presumably the endogenous α7 ligand, also displaces ^{125}I[α]-BgT binding to rat brain with relatively weak affinity.

In conclusion, we have shown that the heteroaryl group and azacycles moiety of heteroaryl azacyclic methyl ether analogs, as exemplified by **1**, have a profound effect on the binding potency. Among the analogs synthesized, the preferred heteroaryl ring is 5-pyrimidyl group. With regard to azacycles, *N*-H-azetidines are the optimial azacyclic moiety for the preparation of potent ligands . In addition, the stereochemistry at either C2 or C3 position play an important role in determining the binding affinities in both 2- and 3-substituted series.

REFERENCE AND NOTES

1. Nordberg, A. *J. Reprod. Fert. Suppl.* **1993**, *46*, 145-154.

2. Sargent, P. B. *Annual Review of Neuroscience.* **1993**, *16*, 403-443.

3. Arneric, S. P.; Sullivan, J. P.; Briggs, C. A.; Donnelly-Roberts, D.; Anderson, D. J.; Raszkiewicz, J. L.; Hughes, M. L.; Cadman, E. D.; Adams, P.; Garvey, D. S.; Wasicak, J. T.; Williams, M. *J. Pharmacol. Exp. Therap.* **1994**, *270*, 310-318.

4. Meyer, E. M.; de Fiebre, C. M.; Hunter, B. E.; Simpkins, C. E.; Frauworth, N.; de Fiebre, N. E. C. *Drug Development Research.* **1994**, *31*, 127-134.

5. 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*, 1055-1059.

6. Garvey, D. S.; Wasicak, J. T.; Elliott, R. L.; Lebold, S. A.; Hettinger, A.-M.; Carrera, G. M.; Lin, N.-H.; He, Y.; Holladay, M. W.; Anderson, D. J.; Cadman, E. D.; Raszkiewicz, J. L.; Sullivan, J. P.; Arneric, S. P. *J. Med. Chem.* **1994**, *37*, 4455-4463.

7. Lin, N.-H.; He, Y.; Anderson, D. J.; Wasicak, J. T.; Kasson, R.; Sweeny, D.; Sullivan, J. P.. *Bioorg. Med. Chem. Lett* **1994**, *4*, 2389-2394.

8. Lin, N.-H.; He, Y.; Arneric, S. P.; Sullivan, J. P. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 1141-1146.

9. Arneric, S. P.; Anderson, D. J.; Bannon, A. W.; Briggs, C. A.; Buccafusco, J. J.; Brioni, J. D.; Decker, M. W.; Donnelly-Roberts, D.; Gopalakrishnan, M.; Holladay, M. W.; Kyncl, J.; Marsh, K. C.; Pauly, J.; Radek, R. J.; Rodrigues, D.; Sullivan, J. P. *CNS Drug Rev.* **1995**, *1*, 1-26.

10. 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. T.; Garvey, D. S.; Williams, M.; Arneric, S. P. *J. Pharmacol. Exp. Therap.* **1994**, *270*, 319-328.

11. Brioni, J. D.; Kim, D. J. B.; Brodie, M. S.; Decker, M. W.; Arneric, S. P. *Psychopharmacology.* **1995**, *119*, 368-375.

12. Abreo, M. A.; Lin, N.-H.; Garvey, D. S.; Gunn, D. E.; Hettinger, A.-M.; Wasicak, J. T.; Pavlik, P. A.; Martin, Y. C.; Donnelly-Roberts, D. L.; Anderson, D. J.; Sullivan, J. P.; Williams, M.; Arneric, S. P.; Holladay, M. W. *J. Med. Chem.* **1996**, *39*, 817-825.

13. Wada, E.; Wada, K.; Boulter, J.; Deneris, E.; Heinemann, S.; Patrick, J.; Swanson, L. W. *J. Comp. Neurol.* **1989**, *284*, 314-335.

14. Flores, C. M.; Rogers, S. W.; Pabreza, L. A.; Wolfe, B. B.; Kellar, K. J. *Mol. Pharmacol.* **1991**, *41*, 31-37.

15. Seguela, P.; Wadiche, J.; Dineley-Miller, K.; Dani, J. A.; Patrick, J. W. *J. Neurosci.* **1993**, *13*, 596-604.

16. Unpublished result.

17. Beers, W. H.; Reich, E. *Nature.* **1970**, *228*, 917-922.

18. Sheridan, R. P.; Nilakantan, R.; Dixon, J. S.; Venkataraghavan, R. *J. Med. Chem.* **1986**, *29*, 899-906.