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Pre-clinical characterization of aryloxypyridine amides as histamine H_3 receptor antagonists: Identification of candidates for clinical development

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ABSTRACT

The pre-clinical characterization of novel aryloxypyridine amides that are histamine H_3 receptor antagonists is described. These compounds are high affinity histamine H_3 ligands that penetrate the CNS and occupy the histamine H_3 receptor in rat brain. Several compounds were extensively profiled pre-clinically leading to the identification of two compounds suitable for nomination as development candidates.

There have been many reports over the past few years demonstrating that histamine H_3 receptors play a pivotal role in regulation of the sleep-wake cycle and that histamine H_3 antagonists should have therapeutic utility for the treatment of a variety of conditions, including ADHD, narcolepsy and potentially Alzheimer's disease, among other conditions. In fact, several pharmaceutical companies have reported initiation of clinical trials for these indications and numerous reviews covering the topic have appeared in the recent literature.[1–6]

Some of the structures of histamine H_3 antagonists that are in clinical trials, or that were in the clinic at one time, are shown in Figure 1. These include the Bioprojet compound BF2.649 (**1**, tiprolisant), a benzoazepine from GSK (**2**) and the aminopyrrolidine **3** (A-331440) from Abbott. Tiprolisant is reported to be in Phase II trials for the treatment of narcolepsy, epilepsy and Parkinson's disease and compound **2** is also in Phase II trials for narcolepsy.[2–6]

As part of our efforts to discover novel histamine H_3 antagonists we now report on a series of high affinity aryloxypyridine amides that have drug-like properties and pre-clinical profiles amenable for the selection of potential clinical candidates.

Over the past several years we have reported on a number of histamine H_3 antagonist chemotypes including propyloxypiperidines,[7] 2-aryloxymethyl-morpholines,[8] and (4-aminobutyn-1-yl)benzylamines[9] as represented by structures **4–6**, among others (Fig. 2). We have also described our efforts towards the identification of benzyl amine-based compounds, such as **7,** that are dual histamine H_3 antagonists/serotonin transporter (SERT) inhibitors with in vivo efficacy at both targets.[10] During the course of these latter studies, we prepared numerous pyridine-linked compounds as represented by **8** (Fig. 3).[11]

We immediately recognized that compound **8** and related analogs represented a possible new chemotype for optimization of histamine H_3 affinity as **8** had moderate affinity for the H_3 receptor and no affinity for SERT.

Also, based on our work in the benzyl amine series, we realized that these compounds were likely not optimized for H_3 affinity. For instance, in the benzyl amine series diazepane is preferred over

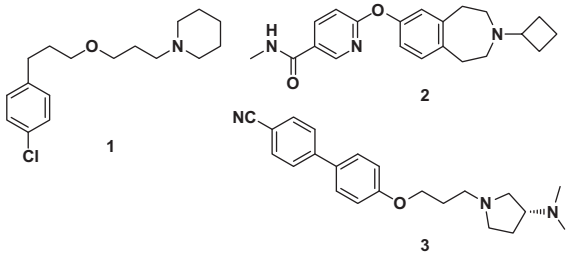

Figure 1. Literature histamine H_3 antagonists.

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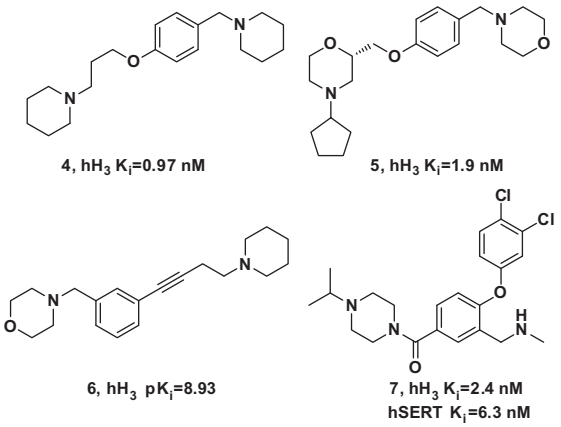


Figure 2. Literature histamine H_3 antagonists and H_3/SERT ligands.

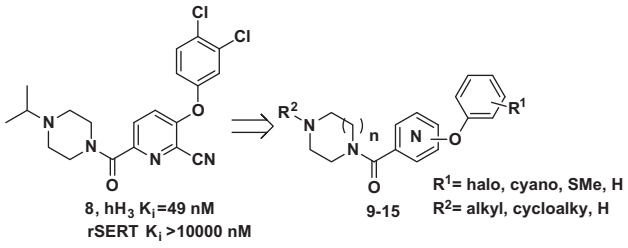
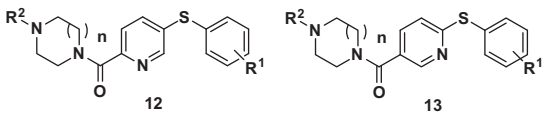


Figure 3. H_3/SERT ligands and novel histamine H_3 ligands.

piperazine.[10] In addition, the cyano substituent may not be required for H_3 affinity since both the cyano and benzyl amine substituents are tolerated. Thus, we proposed several series of isomeric aryloxy-pyridine amides (**9–15**) as potential histamine H_3 antagonists. These compounds utilize the amide that was preferred for H_3 affinity in **7** and lack the cyano moiety in **8** while retaining the pyridine linker present in **8**. We now report our progress towards the identification of compounds from these series that are suitable for progression into early development.

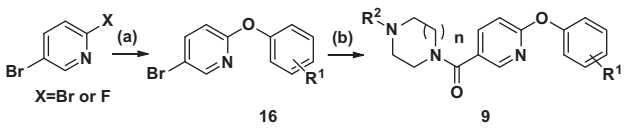
The procedures used to prepare the desired compounds are shown in Schemes 1–3. Compounds **9** (Scheme 1) were easily prepared by addition of a phenol to either 5-bromo-2-fluoro-pyridine or 2,5-dibromopyridine which produced the 5-bromo-2-aryloxy-pyridines **16** in acceptable yields. The 5-bromo-2-aryloxypyridines **16** were then converted directly to compounds **9** via an aminocarbonylation reaction.[11,12]

Compounds **10** and **11** were prepared as shown in Scheme 2 utilizing a simple amino acid coupling followed by addition of a phenol to the resulting 5- or 6-bromopyridine-2-carboxylic acid amide. The phenylsulfanylpyridines **12** and **13** were made using the same methods, substituting the appropriate benzenethiol for the phenol.

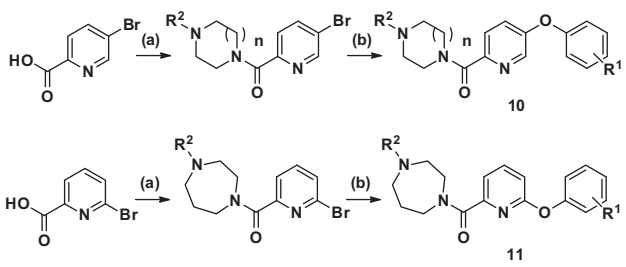


Scheme 3 details the synthesis of the 5-phenoxy-nicotinamides **14** and the 4-phenoxy-pyridine-2-carboxylic acid amides **15**. The procedures are similar to those described previously.

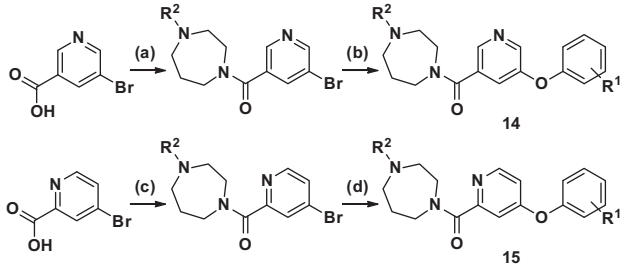
Tables 1 and 2 show representative human histamine H_3 binding affinities and pA_2's for the compounds that were prepared in this study.[13] These data clearly show that diazepane (**9n–9ab**) is preferred over piperazine (**9a**–9**j**) and that cyclic or branched alkyl groups are preferred (i.e., **9k–9m** vs **9n** and **9q**) as substituents on the diazepane. Substituents on the aryloxy phenyl ring are generally tolerated, however the position of the aryloxy ring relative to the amide, and the position of nitrogen in the pyridine ring do have a significant effect on H_3 affinity. It appears that compounds **11** have substantially decreased affinity as compared to compounds **9–10,** and **12–14**.



Scheme 1. Synthesis of compounds **9**. Reagents and conditions: (a) phenol, K_2CO_3, DMF, 90 °C, 18 h, 90–97%; (b) amine, THF, DBU, Hermann's catalyst[12], *t*-Bu_3PHBF_4, $Mo(CO)_6$, 125 °C, microwave, 6 min, ~50%.



Scheme 2. Synthesis of compounds **10–11**. Reagents and conditions: (a) amine, EDCI, HOBt, $(i\text{-Pr})_2$NEt, DCM, 23 °C, 18 h, 25–90%; (b) phenol, Cs_2CO_3, DMA, 120 °C, 18 h, 23–60%.



Scheme 3. Synthesis of compounds **14–15**. Reagents and conditions: (a) oxalyl chloride, DMF, DCM, 23 °C, 1 h, then amine, Et_3N, 18 h, 26–58%; (b) phenol, Cs_2CO_3, DMA, 200 °C, 2 h, microwave, 63–91%; (c) amine-HCl, bromotripyrro-lidinophosphonium hexaflurophosphate, $(i\text{-Pr})_2$NEt, DCM, 18 h, 26-58%; (d) phenol, Cs_2CO_3, DMA, 200 °C, 3 h, microwave, 63–91%.

Several compounds were chosen for an abbreviated in vivo screen based on their affinity for the H_3 receptor and/or preliminary microsomal stability data. Typically, the compounds were screened for affinity at the rat histamine H_3 receptor[13] and for stability in the presence of human and rat liver microsomes either prior to, or in some cases concurrent with, dosing in a rat.

Our strategy was designed to very quickly identify compounds with acceptable brain exposures following oral administration. In order to do this, compounds were dosed po in rats then plasma and brain concentrations, as well as ex vivo receptor occupancy, were obtained at 1, 3 and 6 h post dose. This allowed for a determination of relative exposure in the plasma following a po dose, thus giving a crude estimate of *%F*. The results of these studies are shown in Table 3. Some, but not all, of the compounds have lower affinity for the rat histamine H_3 receptor than the human receptor and typically the compounds are less stable in rat liver microsomes than in human liver microsomes. However, the rat brain and plasma concentrations (shown at 1 h) do not necessarily correlate with microsomal stability in rat. Even so, these experiments allowed us to identify compounds that efficiently penetrate the CNS, occupy

Table 1
Binding and functional data for the human H_3 receptor for compounds **9–10** and **12–13**

Compds	R^1	R^2	n	Human H_3 K_i (nM)[a]	Human H_3 pA_2[b]
9a	3,4-Cl	*i*-Pr	1	78 ± 28	
9b	4-MeS	*i*-Pr	1	230 ± 23	
9c	H	*i*-Pr	1	260 ± 76	
9d	3-CN	*i*-Pr	1	110 ± 39	
9e	4-F	*i*-Pr	1	79 ± 10	
9f	4-F	*c*-Pr	1	430 ± 86	
9g	4-Cl	*i*-Pr	1	73 ± 9	
9h	3-Cl	*i*-Pr	1	100 ± 48	
9i	2-Cl	*i*-Pr	1	31 ± 8	
9j	2-Cl	*c*-Pent	1	40 ± 10	
9k	4-F	H	2	9000	
9l	4-F	Me	2	860 ± 210	
9m	4-F	Et	2	34 ± 3	
9n	4-F	*c*-Pr	2	25 ± 3	8.80
9o	3-F	*c*-Pr	2	16 ± 6	
9p	3-F	*c*-Bu	2	4.1 ± 0.8	9.40
9q	4-F	*c*-Bu	2	1.4 ± 0.1	9.42 ± 0.10
9r	3-CN	*c*-Pr	2	12 ± 8	8.53
9s	3-CN	*c*-Bu	2	1.8 ± 0.7	9.39 ± 0.10
9t	H	*c*-Pr	2	10 ± 5	
9u	H	*c*-Bu	2	4.4 ± 0.5	9.57
9v	3,4-Cl	*i*-Pr	2	18 ± 5	
9w	3,4-Cl	*c*-Bu	2	7.6 ± 2.7	9.26
9x	3-Cl	*c*-Pr	2	17 ± 4	
9y	3-Cl	*c*-Bu	2	1.0 ± 0.4	9.06
9z	4-Cl	*c*-Pr	2	13 ± 5	
9aa	4-Cl	*c*-Bu	2	59 ± 4	9.22
9ab	2-Cl	*c*-Bu	2	17 ± 3	
10a	3,4-Cl	*i*-Pr	2	1.5 ± 0.3	9.51
10b	3,4-Cl	*c*-Pr	2	10 ± 2	
10c	4-Cl	*c*-Bu	2	5.5 ± 3.5	8.61
10d	3-Cl	*c*-Bu	2	4.7 ± 6.6	8.72
10e	4-F	*c*-Bu	2	2.9 ± 0.9	8.85 ± 0.55
10f	3-F	*c*-Bu	2	5.5 ± 1.0	
10g	2-F	*c*-Bu	2	4.3 ± 3.7	9.27
12a	H	*c*-Bu	2	3.7 ± 1	
12b	4-F	*c*-Bu	2	4.6 ± 0.7	
13a	H	*c*-Bu	2	2.3 ± 0.2	9.48
13b	4-F	*c*-Bu	2	29 ± 8	9.48

[a] K_i's are the mean of at least three experiments in triplicate. K_i ± SD is reported.
[b] The result of a single experiment, unless SD is shown, then it is the mean of three experiments.

Table 2
Binding and functional data for the human H_3 receptor for compounds **11** and **14–15**

Compds	R^1	R^2	Human H_3 K_i (nM)[a]	Human H_3 pA_2[b]
11a	3,4-Cl	*i*-Pr	430 ± 130	
11b	3,4-Cl	*c*-Pr	1040 ± 180	
11c	3,4-Cl	*c*-Bu	160 ± 55	
14a	H	*c*-Bu	11 ± 10	
14b	4-F	*c*-Bu	17 ± 17	8.87
15a	H	*c*-Bu	41 ± 3	
15b	4-F	*c*-Bu	5.7 ± 3	

[a] K_i's are the means of at least three experiments in triplicate. K_i ± SD is reported.
[b] The result of a single experiment.

the histamine H_3 receptor in a rat brain, and that are relatively stable to microsomes, thus allowing us to choose compounds for more thorough in vivo evaluation.

The more interesting compounds shown in Table 3 were then evaluated in more detailed rat PK studies. The results of these experiments are shown in Table 4.

Typically, the compounds have rather high clearance in rat and moderate to high volumes of distribution. Compounds **9q**, **9s** and **9aa** have high bioavailability in rat.

Balancing microsomal stability as well as rat plasma and brain concentrations and receptor occupancy, several compounds were chosen for further profiling. These included **9q**, **9s** and **10e**. Canine

Table 3
Binding and functional data for the rat H_3 receptor, RLM and HLM stability, and in vivo data for select compounds **9–10**, **12**

Compds	r.b. H_3 K_i[a] (nM)	Rat H_3 pA_2[b]	RLM[c] (%)	HLM[c] (%)	R.O.[d]	Concn[e]
9p	21 ± 2	8.38	86	99	88	1.96/0.36
9q	23 ± 8	8.45	63	87	94	2.65/1.48
9s	11 ± 9	8.02	87	99	87	0.96/6.02
9u	15 (n = 2)	8.43	90	99	n.d.	0.30/0.21
9w	10 (n = 1)	8.16	4	30	100	4.79/1.52
9x	75 (n = 2)	7.94	41	70	n.d.	0.30/0.31
9y	15 ± 4	8.30	58	98	96	1.72/0.63
9aa	23 ± 9	8.40	63	95	95	2.62/4.47
10e	24 (n = 2)	7.58	56	95	87	2.37/1.02
12b	37 (n = 1)		7	82	n.d.	0.12/0.03

[a] Except where indicated values are the means of at least three experiments in triplicate. r.b. is rat brain. K_i ± SD is reported.
[b] The result of a single experiment.
[c] Percent remaining after 30 min in rat liver microsomes (RLM) or human liver microsomes (HLM).
[d] Receptor occupancy (R.O.) at 1 h after a 10 mpk po dose.
[e] Brain/plasma concentrations (μM) at 1 h after a 10 mpk po dose.

Table 4
Pre-clinical PK data for select compounds **9** and **10e**[a]

Compds	Cl (mL/min/kg)	V_{ss} (L/kg)	$t_{1/2}$ (h)	%*F*
Rat				
9q	49	3.5	1.2	50
9s	41	1.4	0.7	89
9p	118	4.2	0.8	11
9y	104	5.0	1.0	19
9aa	36	3.0	1.3	62
10e	80	4.5	1.0	23
Canine				
9q	35	7.2	2.6	28
9s	21	1.6	0.9	29
10e	35	3.6	1.9	14
Mouse				
9q	94	8.7	1.1	28
9s	97	4.4	0.8	21

[a] For rat (Sprague–Dawley) and mouse (Balb-C), the compounds were dosed 1 mg/kg i.v. and 10 mg/kg po, for dog (beagle) the doses were 1 mg/kg i.v. and 5 mg/kg po.

PK for these compounds are shown in Table 4. All three compounds have high clearance and moderate bioavailability in canine. In vitro cardiovascular safety assessment (Table 5) revealed that although none of the compounds bound efficiently to the hERG channel, as measured by displacement of astemizole, the compounds tested variably had significant activity in a functional patch clamp assay. Based on the patch clamp data **9aa**, **9y**, and **10e** were not pursued further.

All of the compounds shown in Table 5 were also screened in a commercial panel of 50 receptor, ion channel and transporter assays (CEREP, www.cerep.com) and none had any significant affinity (at 1 μM) for any of the targets that were screened.

After evaluating all the data we had at this time compounds **9q** and **9s** were chosen for additional pre-clinical evaluation.

Table 5
In vitro CV safety assessment for select compounds **9** and **10e**

Compds	hERG binding IC_{50} (%) (μM/%inhib@10 μM)	Patch clamp IC_{50} (μM)
9q	>10/0	3.5
9s	>10/23	>10
9p	>10/0	5.4
9y	>10/28	1.6
9aa	>10/23	1.4
10e	8.4/53	0.9

Table 6
Permeability, solubility and physical properties of **9q** and **9s**

Compds	**9q**	**9s**
Caco-2 A to B	78×10^{-6} cm/s	77×10^{-6} cm/s
Caco-2 B to A	36×10^{-6} cm/s	25×10^{-6} cm/s
Solubility		
SGF[a,b]	>5 mg/mL	>1 mg/mL
Water[b]	>5 mg/mL, pH 3.82	>100 mg/mL, pH 5.49
MW	369.44	376.46
pK_a[c]	8.15	8.01(±0.04)
Log $D_{7.4}$[c]	2.08	1.38

[a] For **9q** aqueous HCl at pH 1, for **9s** 0.2% NaCl in 0.1 N HCl.
[b] For the HCl salt.
[c] Measured by Analiza (www.analiza.com).

Both compounds were found to be permeable, did not show the potential for efflux (Caco-2 data, Table 6) and both were sufficiently soluble in simulated gastric fluid and in water as the HCl salts. Neither compound showed any CYP inhibition (in vitro microsome data for 3A4, 2D6 and 2C9 at 10 μM) and both compounds met pre-clinical developability criteria with regards to physical properties including log $D_{7.4}$ and molecular weight. It is notable that the brain to plasma ratios (Table 2) for **9q** and **9s** are quite different (1.8 vs 0.16). This difference is possibly attributed to differences in V_{ss}, (3.5 L/kg vs 1.4 L/kg in rat, Table 4) and correlates with log $D_{7.4}$ (2.08 vs 1.38).

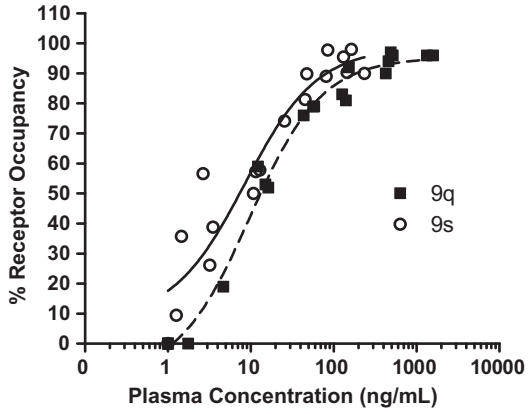


Figure 4. Ex vivo H_3 receptor occupancy with **9q** or **9s** in rat striatum: dose dependency after oral administration (t = 30 min for **9q** or 60 min for **9s**). Results are presented as% receptor occupancy versus vehicle treated rats (each data point represents an individual animal). Ex vivo autoradiography was performed as previously described using [^{3}H]-R-α-methylhistamine.[13]

Figure 4 shows the plasma drug concentration-response curves for receptor occupancy in a rat for **9q** and **9s**.[13] These data demonstrate that the plateau for maximal receptor occupancy (~90%) for **9q** was reached at 3 mg/kg po corresponding to a plasma concentration of 150 ng/mL. Likewise, **9s** had maximal receptor occu-

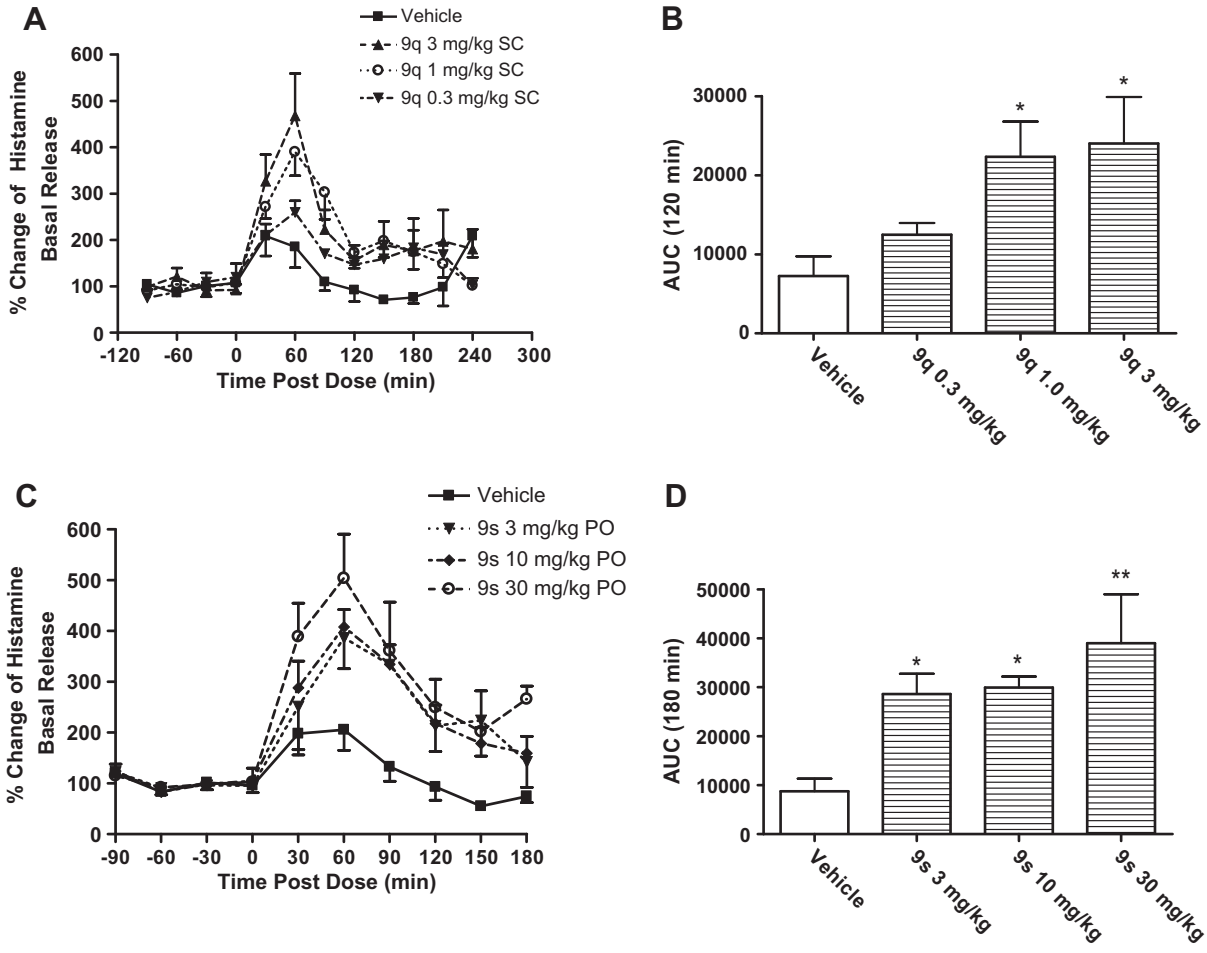


Figure 5. (A and B) Effects of **9q** (0.3, 1.0 and 3.0 mg/kg s.c.) and **9s** (3, 10 and 30 mg/kg po) on in vivo extracellular levels of histamine in the frontal cortex of freely moving rats (AP = −3.2 mm, ML = 0.8 mm, V = −5.0 mm from bregma and dura). All rats were administered the drugs after measuring stable baseline. Histamine levels were measured using high performance liquid chromatography with mass spectroscopy (HPLC MS/MS).[14] Data represents the means ± SEM, n = 4–6 animals. (A and C) Area under the curve values of the data presented in panel B and D. p^* < 0.05, $^{**}p$ < 0.01 by one way ANOVA and Newman–Keuls multiple comparison post hoc test versus vehicle treated rats.

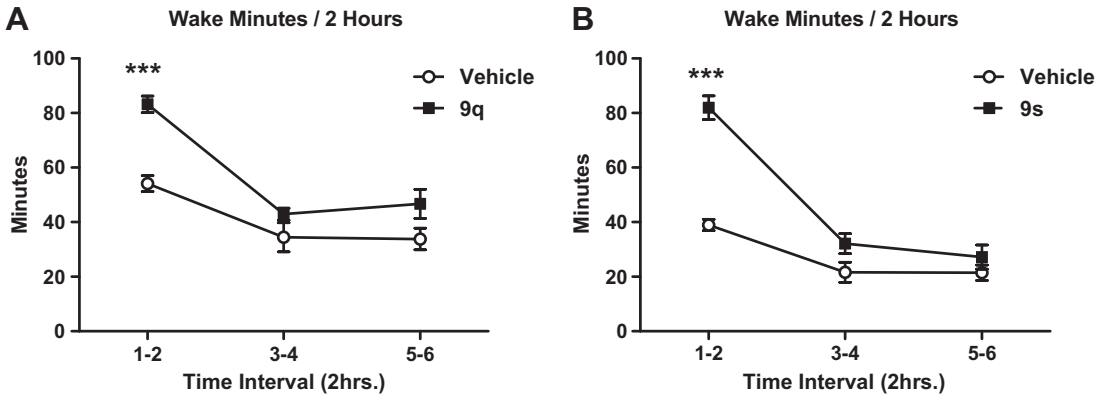


Figure 6. (A and B) Effects of **9q** (10 mg/kg s.c., $n = 7$) and **9s** (30 mg/kg s.c., $n = 6$) on wake duration (min., mean ± SEM) during the first 6 h following the administration at the beginning of the light phase. ***$p < 0.01$ v. vehicle as determined by two way ANOVA followed by Bonferroni *post hoc* analysis.

pancy (again, ~90%) at 1 mg/kg po corresponding to a plasma concentration of 160 ng/mL. It is interesting to note that, because of the differences in brain to plasma ratios, this calculation implies that much lower brain concentrations are required for occupancy with **9s** as compared to **9q**. This may be explained in part by the differences in plasma protein binding for the two compounds. Compound **9s** has a free fraction of 79% in rat whereas compound **9q** has a free fraction of only 15% in rat.

Considering that both compounds have 10-fold lower affinity for the rat receptor as compared to the human receptor (pA_2's, Tables 1 and 3), the expected plasma concentration required for human receptor 90% occupancy is estimated at ~15 ng/mL, assuming similar brain to plasma ratios in rat and human. Next, microdialysis experiments were performed on **9q** and **9s** in rat.

Figure 5 shows the increase in histamine release in the rat frontal cortex following s.c. administration of **9q** and following po administration of **9s** versus time. The concentrations of **9q** obtained in a similar experiment at 3 mpk s.c. in the brain and plasma were 0.956 μM and 0.257 μM, respectively and the concentration of **9s** in the rat brain and plasma at 10 mpk is listed on Table 3. Both compounds are linear with dose. Both compounds significantly increased the levels of histamine in the frontal cortex, a result which is consistent with antagonism of the histamine H_3 receptor in rat. Finally, EEG studies were also performed on **9q** and **9s** in rat in order to confirm wake promoting effects.[9] Figure 6 shows the total wake duration for each compound. Both compounds showed a significant increase in wake duration following s.c. administration of doses that resulted in maximal receptor occupancy.

In conclusion, we have designed and prepared a series of aryloxypyridine amides and demonstrated that select members of this series are high affinity histamine H_3 antagonists. We have also demonstrated that these compounds are highly selective for the histamine H_3 receptor and that several compounds readily penetrate the rat brain and occupy the histamine H_3 receptor following oral administration. Finally, compounds **9q** and **9s** occupy the H_3 receptor at low plasma concentrations, increase histamine release in rat frontal cortex and promote wake in rat. Because these compounds have higher affinity for the human H_3 receptor, it is expected that around 10-fold lower plasma concentrations will be required in human brain H_3 receptor occupancy compared to rat. We have also demonstrated that compounds **9q** and **9s** have physical properties consistent with good absorption and distribution and we have demonstrated acceptable PK in three pre-clinical species for both compounds. Based on this work we identified **9q** and **9s** as potential clinical candidates. Additional toleration studies and baboon PET studies are ongoing in order to support future human clinical trials.

References and notes

1. Gemkow, M. J.; Davenport, A. J.; Harich, S.; Ellenbroek, B. A.; Cesura, A.; Hallett, D. *Drug Discovery Today* **2009**, *14*, 509.
2. Esbenshade, T. A.; Browman, K. E.; Bitner, R. S.; Strakhova, M.; Cowart, M. D.; Brioni, J. D. *Br. J. Pharmacol.* **2008**, *154*(6), 1166.
3. Sander, K.; Kottke, T.; Stark, H. *Biol. Pharm. Bull.* **2008**, *31*(12), 2163.
4. Stocking, E. M.; Letavic, M. A. *Curr. Top. Med. Chem.* **2008**, *8*(11), 988.
5. Letavic, M. A.; Barbier, A. J.; Dvorak, C. A.; Carruthers, N. I. *Prog. Med. Chem.* **2006**, *44*, 181.
6. Celanire, S.; Wijtmans, M.; Talaga, P.; Leurs, R.; de Esch, I. J. P. *Drug Discovery Today* **2005**, *10*, 1613.
7. Apodaca, R.; Dvorak, C. A.; Xiao, W.; Barbier, A. J.; Boggs, J. D.; Wilson, S. J.; Lovenberg, T. W.; Carruthers, N. I. *J. Med. Chem.* **2003**, *46*, 3938.
8. Letavic, M. A.; Keith, J. M.; Ly, K. S.; Bonaventure, P.; Feinstein, M. A.; Lord, B.; Miller, K. L.; Motley, S. T.; Nepomuceno, D.; Sutton, S. W.; Carruthers, N. I. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 5796.
9. Dvorak, C. A.; Apodaca, R.; Xiao, W.; Jablonowski, J. A.; Bonaventure, P.; Dugovic, C.; Shelton, J.; Lord, B.; Miller, K.; Dvorak, L. K.; Lovenberg, T. W.; Carruthers, N. I. *Eur. J. Med. Chem.* **2009**, *44*, 4098.
10. Ly, K. S.; Letavic, M. A.; Keith, J. M.; Miller, J. M.; Stocking, E. M.; Barbier, A. J.; Bonaventure, P.; Lord, B.; Jiang, X.; Boggs, J. D.; Dvorak, L.; Miller, K. L.; Nepomuceno, D.; Wilson, S. J.; Carruthers, N. I. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 39.
11. Keith, J. M.; Letavic, M. A.; Ly, K. S.: Mani, N.; Mills, J.; Villani, F.; Zhong, H. M. U.S. 20070281923.
12. Letavic, M. A.; Ly, K. S. *Tetrahedron Lett.* **2007**, *48*, 2339.
13. The affinity of test compounds for the human recombinant H_3 receptor was determined as described in Ref. 8. Functional data (human pA_2 data) and all rat data was determined as described in Barbier, A. J.; Berridge, C.; Dugovic, C.; Laposky, A. D.; Wilson, S. J.; Boggs, J.; Aluisio, L.; Lord, B.; Mazur, C.; Pudiak, C. M.; Langlois, X.; Xiao, W.; Apodaca, R.; Carruthers, N. I.; Lovenberg, T. W. *Br. J. Pharmacol.* **2004**, *143*, 649. Under the assay conditions used (no constitutive histamine H_3 activity) we are unable to distinguish between antagonists versus inverse agonists.
14. A Supelco Discovery HS F5, 2.1 × 100 mm, 3 μm column was used with a mobile phase containing a 0.1% HCO_2H in H_2O (solvent A) and 0.1% HCO_2H in H_3CCN (solvent B). The mobile phase was held on the B:A ratio of 100:0 for 1.5 min. The linear gradient was changed over the next 0.5 min to B:A ratio of 5:95 and held for 2.5 min, then returned to the starting conditions. The total run time was 6.5 min and flow rate was 0.6 mL/min (Shimadzu LC-10AD VP with SCL-10A VP system controller). Tandem mass spectrometric (MS/MS) detection was carried out on a PE Sciex API4000 in the positive ion mode (ESI) by multiple reaction monitoring (MH^+/daughter was 112.09 95.1 *m/z*). The concentration for each sample was calculated from the peak area of the chromatographic signal and the slope from the corresponding standard curve.