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Synthesis, SAR and evaluation of [1,4′]-bipiperidinyl-4-yl-imidazolidin-2-one derivatives as novel CCR5 antagonists

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ABSTRACT

Elaboration of our previously disclosed spiropiperidine template led to the development of a series of novel CCR5 antagonists. Results of SAR exploration and preliminary lead characterization are described.

The utility of CCR5 antagonists for the treatment of HIV[1] and for their potential use in a variety of autoimmune indications[2] remains an area of interest. We have described our earlier efforts in this area which resulted in the discovery of a novel spiropiperidine template **1**.[3] In a continuation of our program, analysis of a series of CCR5 antagonists disclosed by AnorMed[4] (represented by compound **2**) suggested replacement of the Roche spiro group with the imidazolidinone fragment from the AnorMed template. This strategy is illustrated in Figure 1, affording a novel new series represented by compound **3**.[5]

As seen in Table 1, compound **3** showed promising activity in binding and antiviral assays.[6] Assessment in commonly used in vitro pharmacokinetic assays demonstrated a number of issues which could affect bioavailability. Screening in human liver microsomes (HLM) showed medium to low metabolic stability[7] with a clearance of 31 μL/min/mg protein, which corresponds to projected in vivo clearance close to hepatic blood flow.[8] As Table 1 also shows, screening of compound **3** in Caco-2 cells[9] showed low intrinsic permeability[10] and high P-glycoprotein (P-gp)[11] mediated active efflux, additional factors which could limit in vivo exposure.

Thus, the goal was to improve antiviral activity and address metabolic and efflux issues with this template.

Our experience working with CCR5 antagonists has taught us that meeting this goal can be quite challenging. The CCR5 receptor binding pocket is largely lipophilic in nature and antagonists with high lipophilicity exhibit the best potency. Unfortunately, highly lipophilic templates frequently suffer from low metabolic stability and high clearance issues. A strategy to reduce lipophilicity and block metabolism by incorporation of polar functionalities can lead to improved metabolic stability and lower clearance. However, as these analogs become more hydrophilic, antiviral potency typically decreases and issues with permeability and efflux arise. Thus the key challenge is to balance the opposing trends of antiviral potency and permeability vs. metabolic stability and efflux.

A targeted SAR study of the lead template was thus initiated to address these issues. For convenience, the right hand side of the

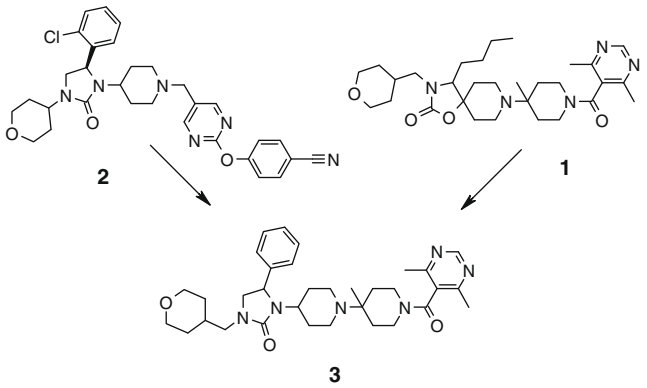

Figure 1.

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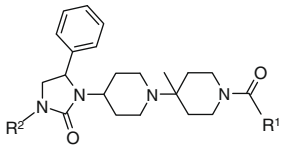
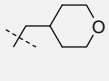
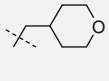
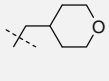
Table 1
SAR of lead compound **3**



#	R^1	R^2	R^3	Binding IC_{50}[a]	Antiviral IC_{50}[b]	HLM[c]	Caco[d] AB/ER
3			*rac*	27	47	31	0.4/33
4			(*R*)	20	32	16	0.5/29
5			(*S*)	>500	>650	ND[e]	ND[e]

[a] Competitive binding evaluated versus RANTES with IC_{50} values in nM, as mean of two experiments.
[b] Antiviral IC_{50} values in nM as mean of two experiments.
[c] Human liver microsomal intrinsic clearance (μL/min/mg protein).
[d] Permeability in Caco-2 cells AB, apical to basolateral and BA, basolateral to apical movement of test compound in 21 day cultured Caco-2 cells ($cm/s \times 10^{-6}$). ER, efflux ratio of BA to AB.
[e] Not determined.

template, containing the 2,6-dimethyl pyrimidine group is referred to as the head region and the left hand side, containing the substituted imidazolidinone is called the tail region.

As a first step, the individual enantiomers of racemate **3**, (*R*) and (*S*) isomers **4** and **5** were prepared. As can be seen in Table 1, all antiviral activity resided in the (*R*) enantiomer **4** and subsequent analogs maintained this stereochemistry.

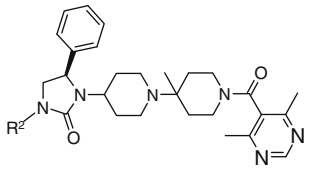
As seen in Table 2, elongation of the tail linker of analog **4** to two carbons afforded compound **6,** which demonstrated comparable antiviral activity, but at a cost of decreased metabolic stability. Conversely, shortening of the tail linker in analog **7** resulted in loss of activity, but did afford improved metabolic stability. Next, a variety of substituted piperidine analogs were prepared and characterized. Carbamate analog **8** maintained antiviral potency while sulfonamide **9** demonstrated improved activity, but both were significantly less stable. Incorporation of a polar urea functionality in compound **10** improved metabolic stability, but at a price of antiviral potency. Improved permeability and decreased efflux was observed with *trans*-4-methoxy and ethoxy cyclohexyl analogs **11** and **12,** along with excellent antiviral activity. While the higher lipophilicity for these compounds may be driving these improved permeability characteristics, it also contributes to their unfavorable metabolic stability. The corresponding *trans*-4-hydroxy-cyclohexyl analog **13** did have a favorable combination of antiviral activity and metabolic stability. Unfortunately, limited permeability and increased efflux are significant problems with this compound.

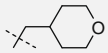
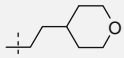
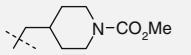
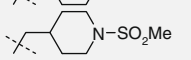
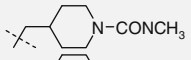
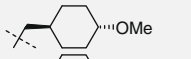
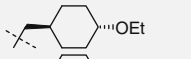
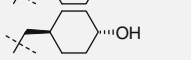
Building on the trend that a number of analogs had promising metabolic stability parameters but lacked the necessary antiviral potency, a series of compounds were prepared by introducing a cyanopyridine as a replacement for the pyrimidine head group. Our previous studies with spiropiperidine CCR5 antagonists had demonstrated that incorporation of this group had a beneficial effect on antiviral potency.

As can be seen in Table 3, replacement of the pyrimidine group in compounds **4**, **7**, **8** and **13** by the corresponding cyanopyridine head group (compounds **14**–**17**) resulted in improved antiviral potency of 4–90-fold. Even compound **18**, where the tail group had been trimmed down to a simple methyl group demonstrated good potency with an IC_{50} of 22 nM. Although inclusion of the cyanopyridine group generally gave a slight decrease in metabolic stability, analogs such as **15** had a good balance of metabolic stability and antiviral activity.

As a next step, modifications of the tail side chain group were investigated. Substitution on the phenyl ring of **4** with a variety of groups such as chloro, fluoro and cyano all resulted in a dramatic loss of antiviral activity. Similar loss of activity was observed by replacement of the phenyl group with 2-pyridyl, 4-pyran or cyclohexyl (data not shown). Recovery of good antiviral potency and reasonable metabolic stability could be achieved by combination of the pyridyl side chain and the cyanopyridine head group as seen

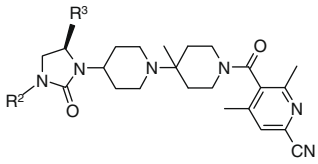
Table 2
SAR of tail substituents

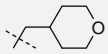
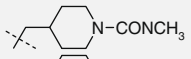
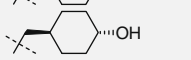


#	R^2	Binding IC_{50}[a]	Antiviral IC_{50}[a]	HLM[a]	clog *P*	Caco[a] AB/ER
4		20	32	16	1.6	0.5/29
6		8	25	96	2.1	0.8/24
7		90	>625	8	0.9	0.6/28
8		26	26	155	2.1	0.3/56
9		34	7	224	1.2	0.2/38
10		32	99	13	1.3	0.1/13
11		62	8	69	2.5	1.4/13
12		6	3	47	2.8	3/4
13		20	8	12	1.9	0.2/61

[a] Defined as in Table 1.

Table 3
SAR of analogs containing cyanopyridine head



#	R^2	R^3	Binding IC_{50}[a]	Antiviral IC_{50}[a]	HLM[a]	Caco[a] AB/ER
14		Ph	12	2	57	1/20
15		Ph	21	7	15	1/12
16		Ph	26	25	53	0.7/10
17		Ph	33	3	34	0.4/47
18	Me	Ph	24	22	44	0.6/25

[a] Defined as in Table 1.

in Table 4 (compounds **19** and **20**). Replacement of the phenyl group with n-butyl resulted in a loss of antiviral potency as seen with compounds **21** and **22**. Introduction of the cyanopyridine head group in **23** did bring back reasonable antiviral activity. The poor correlation between CCR5 binding inhibition and antiviral activity observed in analogs such as **7**, **21** and **22** is not unusual and has been previously observed.[3,12] It most likely results from differences in the allosteric modes of inhibition for chemokine versus viral binding.

A number of compounds with promising in vitro properties were further profiled in single dose pharmacokinetic experiments in rat and monkey as seen in Tables 5 and 6. In rat PK, analogs **3** and **15** (screened as the corresponding racemate) showed the most promising bioavailability with moderate clearance and half life. Subsequently, analog **4** (*R* enantiomer of **3**) and **15** were screened in monkey PK. Although compound **15** demonstrated a superior profile, permeability and efflux are still limiting factors on bioavailability.

A particular off-target issue associated with CCR5 antagonists is affinity for the hERG channel leading to a potential for cardiac arrhythmias. Screening a set of representative examples from our series in a PatchXpress clamp system[13] showed a range of hERG IC_{20} values from 0.3 to 30 μM. Analogs **3** and **15** (racemate) showed hERG IC_{20} values of 22 and 3.5 μM, respectively.

A representative synthetic sequence for the preparation of our series is outlined in Scheme 1 for the preparation of analog **4**.[14]

Deprotection of ketal **24**[15] under acidic conditions afforded ketone **25**. Debenzylation of **25** under hydrogenolysis conditions in the presence of Boc anhydride gave intermediate **26**. The imidazolidinone portion of the molecule was prepared starting from commercially available (*R*)-(−)-2-amino-2-phenyl ethanol **27**. Amine protection with Boc anhydride afforded compound **28**. Mesylation followed by treatment with sodium azide gave the corresponding azide **29**. Removal of the Boc group of **29** with trifluoroacetic acid followed by reductive amination of ketone **26** with sodium triacetoxyborohydride afforded compound **30**. Catalytic hydrogenation of azide **30** gave the corresponding amine **31**, which was subjected to reductive alkylation with 4-tetrahydropyran carboxaldehyde followed by subsequent cyclization with triphosgene to give the substituted imidazololidinone **32**. Acidic treatment of **32** to remove the Boc protecting group followed by amide formation with 4,6-dimethylpyrimidine-5-carboxylic acid afforded target compound **4**.

In summary, we have discovered a novel series of potent CCR5 small molecule antagonists, exemplified by analogs **4** and **15**, possessing a promising antiviral, pharmacokinetic and off-target profile.

Table 5
Pharmacokinetic profiles in rat[a,b]

PK	RLM[7]	C_{max} (ng/mL)	AUC (ng * h/mL)	Cl (mL/min/kg)	$T_{1/2}$ (h)	*F*%
3	14	56	189	44	1.5	27
12	89	21	49	65	0.8	10
15[c]	55	43	85	16	5.7	23
19	87	38	187	29	1	19

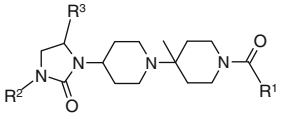
[a] Doses were 1 mg/kg IV and PO.
[b] C_{max}, AUC and *%F* was determined after oral dosing. Cl, Vd_{ss}, $t^{1/2}$ were determined from the iv dose.
[c] Racemate.

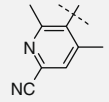
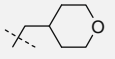
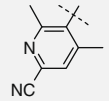
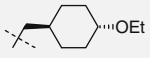
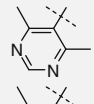
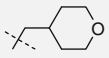
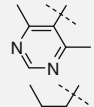
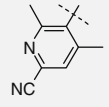
Table 6
Pharmacokinetic profiles in cynomologous monkey[a,b]

PK	kFLM[7]	C_{max} (ng/mL)	AUC (ng * h/mL)	Cl (mL/min/kg)	$T_{1/2}$ (h)	*F*%
4	65	16	55	39	1.9	14
15[c]	33	133	418	17	1.9	42

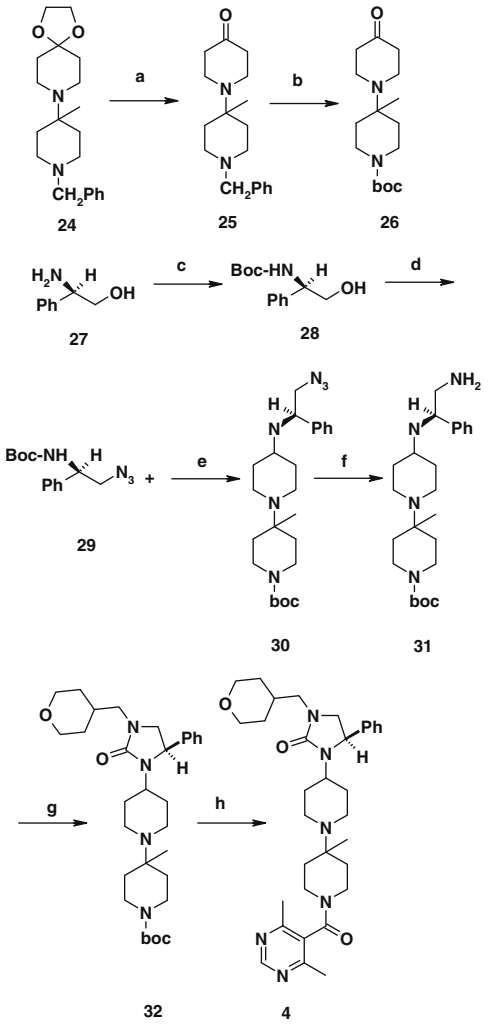
[a] Doses were 1 mg/kg IV and PO.
[b] C_{max}, AUC and *%F* was determined after oral dosing. Cl, Vd_{ss}, $t^{1/2}$ were determined from the iv dose.
[c] Racemate.

Table 4
SAR of tail side chain analogs



#	R^1	R^2	R^3	Binding IC_{50}[a]	Antiviral IC_{50}[a]	HLM[a]	Caco[a] AB/ER
19			 (R)	12	22	18	0.1/77
20			 (R)	25	9	40	0.5/30
21			*n*Bu (*rac*)	43	335	36	0.4/40
22			*n*Bu (*rac*)	84	>625	9	0.4/52
23			*n*Bu (*rac*)	65	41	20	1/14

[a] Defined as in Table 1.



Scheme 1. Reagents and conditions: (a) (i) concd HCl, MeOH, reflux; (ii) 1 N HCl, reflux 65%; (b) H_2, $Pd(OH)_2$, $(Boc)_2O$, MeOH, 87%; (c) $(Boc)_2O$, 1 M NaOH, Et_2O, 97%; (d) (i) MsCl, TEA, CH_2Cl_2; (ii) NaN_3, DMF, 60 °C, 67% for two steps; (e) (i) TFA, CH_2Cl_2; (ii) **26**, $NaBH(O_2CCH_3)_3$, $ClCH_2CH_2Cl$, 76% for two steps; (f) H_2, 5% Pd/C, MeOH, 98%; (g) (i) 4-tetrahydropyrancarboxaldehyde, $NaBH(O_2CCH_3)_3$, $ClCH_2CH_2Cl$; (ii) $(CCl_3O)_2CO$, pyridine, CH_2Cl_2, 79% for two steps; (h) (i) TFA, CH_2Cl_2, 99%; (ii) 4,6-dimethylpyrimidine-5-carboxylic acid, HOBT, EDAC, DIEA, DMF, 60%.

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5. Subsequent to our investigative efforts, related templates have been reported: Bourque, E.; Metz, M.; Baird, I. R.; Yang, W.; Bridger, G.; Skerlj, R. T. WO2008070758.
6. For binding and antiviral assay protocols see: Melville, C. R.; Rotstein, D. M. WO2008119663.
7. The thresholds above which intrinsic microsomal clearance values were considered to be high were $\geqslant$35 μl/min/mL for HLM, $\geqslant$90 μl/min/mL for RLM and $\geqslant$43 μl/min/mL for kfLM (cynomologous monkey). These scaled to in vivo clearance values $\geqslant$70% of liver blood flow using the well-stirred model. Intrinsic microsomal clearance values corresponding to $\leqslant$30% of liver blood flow ($\leqslant$6.5 μl/min/mL for HLM, $\leqslant$15 μl/min/mL for RLM and ($\leqslant$14 μl/min/mL for kFLM) were considered to be low.
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