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Discovery of *N*-benzyl-*N*′-(4-pipyridinyl)urea CCR5 antagonists as anti-HIV-1 agents (II): Modification of the acyl portion

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ABSTRACT

Modification of the acyl moiety in the CCR5 lead molecule **2** led to identification of several new classes of CCR5 antagonists. Antiviral activity and pharmacokinetic properties of the synthesized compounds were evaluated. Structure–activity relationship (SAR) derived from these studies further guided the optimization efforts, ultimately leading to the discovery of **36** with an acceptable drug-like profile.

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CCR5 targeted viral entry inhibitors represents a novel antiviral mechanism against HIV infection by preventing M-tropic virus from entering host. Blockage of CCR5 has successfully demonstrated the prevention of the viral infection.[1] The recently approved CCR5-antagonist maraviroc[2] has been shown to be an effective component of highly active antiretroviral therapy (HAART) for patients harboring predominately M-tropic virus. However, novel CCR5 antagonists with improved phamarcokinetics (PK) and safety profiles are needed to further expand the use of this class of antiretrovirals in anti-HIV therapy.[3]

In our preceding communication,[4] we reported the optimization of **1** at the amine portion, which led to identification of *N*′-[(4-cyanophenyl)methyl]-*N*-(3-fluorophenyl)-*N*-piperidinylurea as the optimal amine moiety, exemplified in **2**. This compound exhibited highly potent antiviral activity and a promising pharmacokinetic profile in rats, providing a foundation for the modification of the acyl terminal in **3** with the aims to further improve the overall profile of this class of CCR5 antagonists (Fig. 1).

The synthesis of the acyl analogs **3** was carried out as depicted in Scheme 1. Reductive amination of 1,1-dimethylethyl 4-oxo-1-piperidine carboxylate **4** with 3-fluoroaniline provided secondary aniline **5**, one of the urea precursors. Next, 4-(aminomethyl) benzonitrile **6** was reacted with excess of phosgene in the presence of pyridine, generating isocyanate **7**, which was subsequently treated with **5** to form urea **8**. Removal of Boc in **8** followed by a second reductive alkylation with aldehyde **9**[5] yielded **10**. After deprotection of the Boc, the unmasked piperidine in **10** was coupled with various acids under HATU conditions to furnish **11–36**.

All compounds were tested for antiviral activity in both Human OsteoSarcoma (HOS) ($n = 2$) and peripheral blood lymphocytes (PBL) cells ($n \geqslant 4$) against the Ba-L strain of HIV-1.[6] Compound induced cellular toxicity was also investigated using CellTiter-Glo reagent (Promega). None of the compounds induced cell killing up to 1 μM (data not shown) indicating that the observed potency

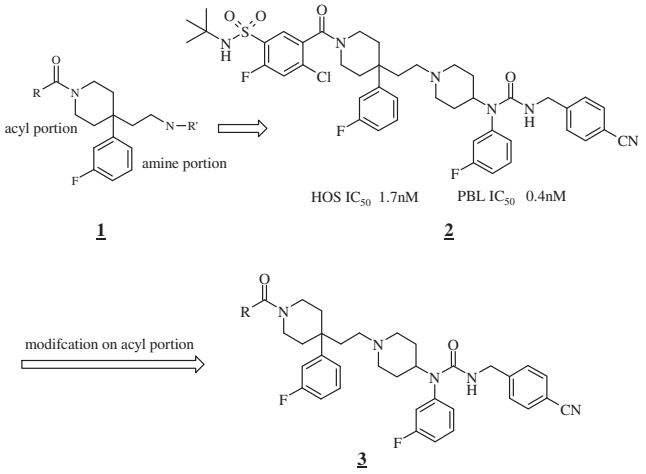

Figure 1. Evolution of the piperidine based CCR5 antagonists.

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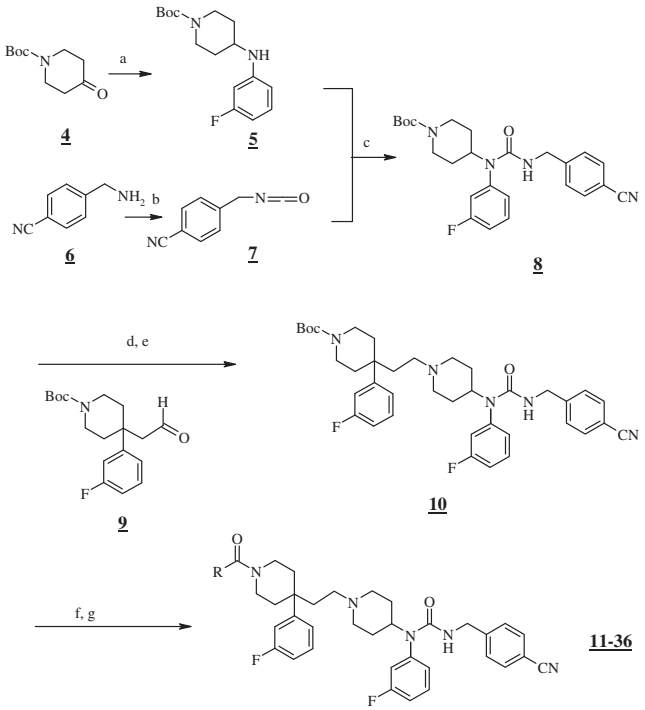


Scheme 1. Reagents and conditions: (a) 3-fluoroaniline, $NaBH(OAc)_3$, AcOH, CH_2Cl_2, rt, overnight, quant.; (b) phosgene, pyridine, CH_2Cl_2, 0 °C to rt., 3 h; (c) *i*-Pr_2EtN, CH_2Cl_2, rt., overnight, 90%; (d) HCl, rt, quant.; (e) compound **9**, $NaBH(OAc)_3$, *i*-Pr_2EtN, CH_2Cl_2, rt., 60%; (f) 4 M HCl, rt, quant.; (g) RCOOH, HATU, EtN_3, DMF, rt, 30–60%.

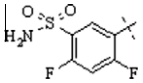
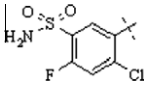
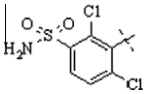
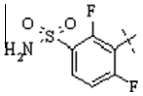
is the result of an antiviral effect and not toxicity. Pharmacokinetic properties were also evaluated in Sprague–Dawley rats at 1 mg/kg using 5% mannitol with 0.05% acetic acid in DMSO as dose vehicle. The data is presented in Tables 1–5.

The SAR of lead **2** from our preceding communication suggested that further modification of the acyl portion is warranted. To this end, *tert*-butyl based sulfonamide **11** with a different dichloro substitution pattern slightly improved PBL potency, but showed inferior bioavailability in rats (Table 1). The pyrrolidyl moiety in sulfonamides **13** and **15** consistently resulted in subnano molar potencies in PBL assays; however, their PK properties were disappointing. Hydroxylated pyrrolidine appeared even worse, increasing rat clearance ~2-folds (**12** vs **13** and **14** vs **15**). Therefore, further optimization of the sulfonamides region was required to improve pharmacokinetics.

The sulfonamides in Table 1 appeared to be highly hydrophobic as suggested by their high *c* log *P* values. To increase the polarity and also reduce the size of the sulfonamides, a series of primary sulfonamides **16–19** was synthesized and evaluated as shown in Table 2.

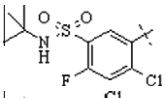
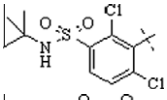
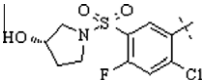
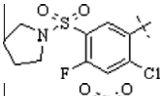
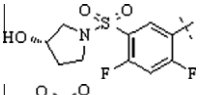
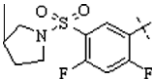
These sulfonamides maintained antiviral activity at a low single digit nM level. Interestingly, halogen substituents appeared to modulate the clearance in rats with a preference for chloro over fluoro substituents. Disappointingly, the primary sulfonamides were not bioavailable in rats presumably because of their poor permeability as suggested by their low apparent permeability coeffi-

Table 2
Antiviral potency in HOS and PBL cell assays and pharmacokinetics in rats of primary sulfonamides **16–19**

Compd #	R	HOS IC_{50} (nM)	PBL IC_{50} (nM)	Cl (mL/min/kg) (rat, 1 mg/kg)	Papp (nM/s @ pH 7.05)
16		4	5.2	31	<3
17		6.8	0.9	9.7	<3
18		5.7	2.8	16	<3
19		6.6	2	25	NA

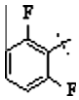
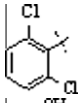
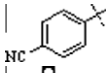
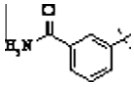
NA, not available.

Table 1
Antiviral potency in HOS and PBL cell assays and pharmacokinetics in rats of *tert*-butyl and pyrrolidyl sulfonamides **2**, **11–15**

Compd #	R	HOS IC_{50} (nM)	PBL IC_{50} (nM)	Rat PK (1 mg/kg)		c log P (day light)
				Cl (mL/min/kg)	%F	
2		0.4	1.7	18	22	7.5
11		3	0.8	9	10	7.7
12		1.5	0.5	24	8	5.4
13		1.5	0.6	9.5	9	6.5
14		1.3	NA	43	0	4.9
15		0.9	0.6	29	10	5.9

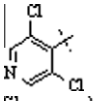
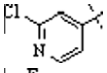
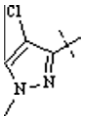
NA, not available.

Table 3
Antiviral potency in HOS and PBL cell assays and pharmacokinetics in rats of benzoic acid derivatives **20**–**24**

Compd #	R	HOS IC_{50} (nM)	PBL IC_{50} (nM)	Rat PK (1 mg/kg)		*c* log *P* (day light)	DOF IC_{50} (uM)
				Cl (mL/min/kg)	%*F*		
20		1.9	6.5	18.6	19	6	1.2
21		2.6	NA	13.3	19	7.2	1
22		6.6	32	11.3	46	6	0.8
23		13	26	8.1	23	5.2	0.4
24		3.7	NA	20	0	4.7	20

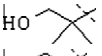
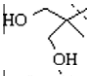
NA, not available.

Table 4
Antiviral potency in HOS and PBL cell assays and pharmacokinetics in rats of heterocyclic acyl derivatives **25**–**30**

Compd #	R	HOS IC_{50} (nM)	PBL IC_{50} (nM)	Rat PK (1 mg/kg)		
				Cl (mL/min/kg)	$t_{1/2}$ (hr)	%*F*
25		2.7	4.8	24	4.9	50
26		19	13	14.6	5.8	64
27		6.4	6.1	22.5	3.4	60
28		17	22	25	2.6	11
29		8.2	NA	13	7.2	3
30		1.1	41	124	NA	0

NA, not available.

Table 5
Antiviral potency in HOS and PBL cell assays and pharmacokinetics in rats of aliphatic acid derivatives **31**–**36**

Compd #	R	HOS IC_{50} (nM)	PBL IC_{50} (nM)	Rat PK (1 mg/kg)		
				Cl (mL/min/kg)	$t_{1/2}$ (hr)	%*F*
31		11	5.3	10	2.4	11
32		19	20	22	2.6	63
33		22	NA	26	4.4	7
34		17	22	25	2.6	11
35		6.4	49	12	4.3	28
36		8	8.1	28	4.4	53

NA, not available.

Table 6
Pharmacokinetics of **36** in dog and monkey at 1 mg/kg dose[a]

Animal	Cl (mL/min/kg)	$t_{½}$ (h)	%*F*
Dog	12	6.6	45
Monkey	30	2.2	14

[a] Dose vehicle: 5% mannitol with 0.05% acetic acid in DMSO.

Table 7
Physical properties of **2** and **36** and their oral exposures in rat and dog (AUC) at 1 mg/kg dose

Compd #	MW	*c* log *P*	PSA(Ertl)	AUC (ng hr/mL)(1 mg/kg p.o.)	
				Rat	Dog
2	849	7.5	126	205	22
36	644	4.8	100	383	515

cients (Papp) using MDCK cell line as well as high polar surface area (PSA (Ertl) = 140).

We then focused our efforts on removing the sulfonamide moiety altogether in order to further reduce the size and PSA. The di-halogen substituted benzene analogs **20** and **21** offered similar potency, and dramatically improved bioavailability as compared to the primary sulfonamides (Table 3). Small polar groups CN and OH on the benzene ring (**22** and **23**) were detrimental to inhibitory potency in PBL assays though they did increase compound bioavailability in rats. In contrast, amide in **24** retained potent HOS IC_{50}, but was not bioavailable. In addition, hERG liability measured by dofetilide binding assay (DOF IC_{50})[7] became a concern for this class of compounds (except for **24**) most likely due to the increase in lipophilicity.

Heterocyclic acyls with reduced lipophilicity were subsequently explored (Table 4). In particular, pyridine analogs **25**, **26** and **27** not only maintained single digit nM IC_{50}s without dofetilide binding concerns (data not shown), but also exhibited superior pharmacokinetic properties in rats; unfortunately, these compound were

relatively potent inhibitors of CPY 3A4 (data not shown). In addition, the five-membered heterocyles **28**, **29** and **30** failed to improve either potency or PK.

Aliphatic acyl groups were also pursued (Table 5). The *tert*-butyl derivative **31** had good antiviral activity in PBL, however, its bioavailability was not satisfactory. The mono-hydroxylated *tert*-butyl analog **32** lost antiviral potency, but, surprisingly, showed the highest %*F* among the compounds reported in this communication. Both the di-hydroxyl and the THP analogs **33** and **34** did not exhibit an improvement in either antiviral potency or DMPK properties. The *gem*-dimethyl nitrile **35** had the most promising pharmacokinetics in rats, but, unfortunately, its antiviral activity in the PBL assays was substantially decreased. The dimethyl hydroxyl **36** demonstrated the most balanced antiviral and rat PK profiles with single digit nM IC_{50}s in both HOS and PBL assays and 53% bioavailability in rats.

Additional evaluation of **36** for dofetilide binding (IC_{50} = 10 μM), CYP inhibitions and the reactive metabolite assays suggested no associated concerns. It was then advanced to dog and monkey PK studies. Encouragingly, **36** demonstrated low clearance and high bioavailability in dogs at 1 mg/kg dose. Unfortunately, its monkey PK was slightly disappointing with 14% bioavailability due to the relatively high clearance (Table 6).

In summary, several classes of acyl analogs of **2** were synthesized and evaluated in antiviral assays and animal PK models. Modulation of physiochemical properties of the molecule by reducing mass, lowering *c* log *P* and decreasing PSA (Table 7) finally led to the identification of **36**, a potent CCR5 antagonist with substantial improvement in oral exposures across animal species, particularly, in dog. This compound was progressed to further preclinical studies and the results will be published in due time.

References and notes

1. Dragic, T.; Litwin, V.; Allaway, P. G.; Martin, R. S.; Huang, Y.; Nagashima, A. K.; Cayanan, C.; Maddaon, J. P.; Koup, A. R.; Moore, P. J., et al *Nature* **1996**, *381*, 667.
2. Price, D. A.; Armour, D.; De Groot, M.; Leishman, D.; Napier, C.; Perros, M.; Stamman, B. L.; Wood, A. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 4633.
3. Kazmierski, W. M.; Gudmundsson, K. S.; Piscitelli, S. C. *Annu. Rep. Med. Chem.* **2007**, *42*, 301.
4. Duan, M.; Peckham, J.; Edelstein, M.; Ferris, R.; Kazmierski, M. W.; Wheelan, P.; Xiong, Z. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 7397.
5. Kazmierski, W. M.; Aquino, C.; Chauder, B.; Deanda, F.; Ferris, R.; Jones-Hertog, D.; Kenakin, T.; Koble, C.; Watson, C.; Wheelan, P.; Yang, H.; Youngman, M. *J. Med. Chem.* **2008**, *51*, 6538.
6. Muniz-Medina, V.; Jones, S.; Maglich, J.; Galardi, C.; Hollinggswirth, R.; Kazmierski, W.; Ferris, R.; Edelstein, M.; Chiswell, K.; Kenakin, T. *Mol. Pharmacol.* **2009**, *75*, 490.
7. (a) Singleton, D. H.; Boyd, H.; Steidl-Nichols, J. V.; Deacon, M.; Croot, M. J. D.; Price, D.; Nettleton, D. O.; Wallace, N. K.; Troutman, M.; Williams, C.; Boyd, J. G. *J. Med. Chem.* **2007**, *50*, 2931; (b) Deacon, M.; Singleton, D. H.; Szalkki, N.; Pasieczny, R.; Peacock, C.; Price, D.; Boyd, J.; Boyd, H.; Steidl-Nichols, J. V.; Williams, C. *J. Pharmacol. Toxicol. Methods* **2007**, *55*, 255.