Bioorganic & Medicinal Chemistry Letters 21 (2011) 3976–3981

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Renin inhibitors for the treatment of hypertension: Design and optimization of a novel series of tertiary alcohol-bearing piperidines

Austin Chen *, Elizabeth Cauchon, Amandine Chefson, Sarah Dolman, Yves Ducharme, Daniel Dubé, Jean-Pierre Falgueyret, Pierre-André Fournier, Sébastien Gagné, Michel Gallant, Erich Grimm, Yongxin Han, Robert Houle, Jing-Qi Huang, Gregory Hughes, Hélène Jûteau, Patrick Lacombe, Sophie Lauzon, Jean-François Lévesque, Susana Liu, Dwight MacDonald, Bruce Mackay, Dan McKay, M. David Percival, René St-Jacques, Sylvie Toulmond

Merck Frosst Centre for Therapeutic Research, 16711 Trans Canada Highway, Kirkland, Québec, Canada H9H 3L1

ARTICLE INFO

Article history:
Received 17 February 2011
Revised 3 May 2011
Accepted 4 May 2011
Available online 14 May 2011

Keywords:
Renin
Inhibitor
Anti-hypertensive

ABSTRACT

The design and optimization of a novel series of renin inhibitor is described herein. Strategically, by committing the necessary resources to the development of synthetic sequences and scaffolds that were most amenable for late stage structural diversification, even as the focus of the SAR campaign moved from one end of the molecule to another, highly potent renin inhibitors could be rapidly identified and profiled.

Despite the fact that anti-hypertensive medications with distinct mechanisms of action have become broadly available in the past two decades, hypertensive heart disease remains one of the leading causes of mortality in the developed world.[1] As a result, there continues to be a demand for the discovery of more efficacious therapeutic agents that can be used either as monotherapy or in combination with existing anti-hypertensive agents. In this regard, one attractive research strategy involves the design and synthesis of new molecules capable of inhibiting renin, an enzyme responsible for the rate-limiting conversion of angiotensinogen into angiotensin I (Fig. 1).[2] Although numerous pharmaceutical companies have embraced this strategy, to date, aliskiren is the only direct renin inhibitor that has been approved by the FDA for the treatment of mild to moderate hypertension.[3]

We have previously reported that piperidines substituted at the 4-position with an *N*-methyl pyridone (**1**, Table 1) can serve as highly potent renin inhibitors.[4] During this SAR campaign, it was observed that the pyridone functionality engages in two key stabilizing interactions with the renin enzyme: (1) formation of a H-bond between the pyridone carbonyl and indole NH of Trp45, (2) formation of a π-stack between the pyridone ring and phenol residue of Tyr83. Even though we believed these pyridone-bearing renin inhibitors possessed all the properties necessary for clinical

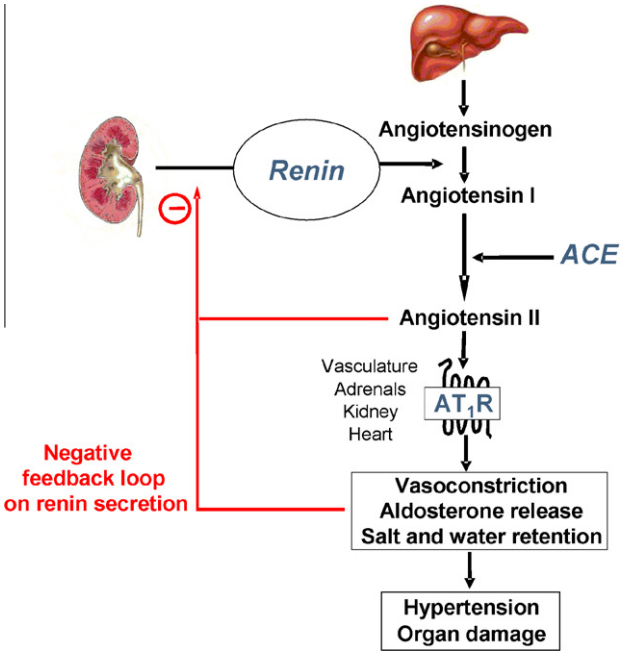
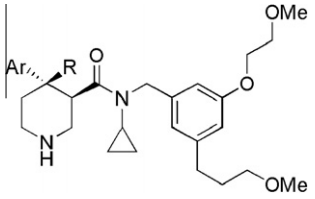


Figure 1. The renin–angiotensin–aldosterone system.

* Corresponding author. Tel.: +1 450 218 4017.
E-mail address: austin.chihyu.chen@gmail.com (A. Chen).

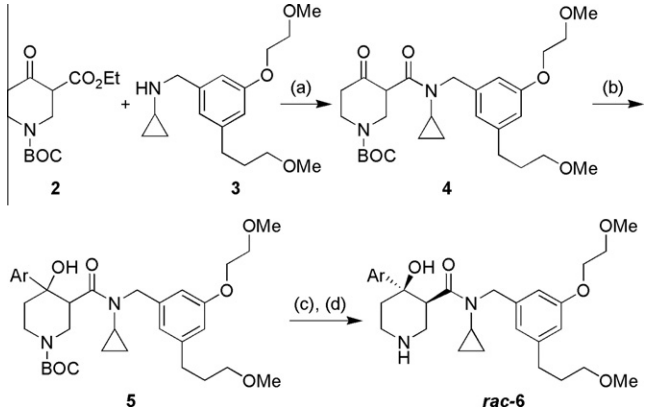
0960-894X/$ - see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2011.05.014

Table 1
SAR of select renin compounds: Northern modifications



Ar									
R	H	OH	OH	OH	OH	OH	OH	OH	OH
	1	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**
Renin buffer IC_{50}[a] (nM)	12	318	139	54	62	13	9	13	23
Renin plasma IC_{50}[a] (nM)	31	990	467	960	479	135	120	97	230

[a] Average of at least two replicates. All compounds were tested as a racemic mixture. See Ref. 8 for assay protocols.



Scheme 1. Synthesis of **6**. Reagents and conditions: (a) 0.2 equiv DMAP, 140 °C, 79%; (b) 3 equiv LiCl, 3 equiv ArMgBr, THF, rt, 1 h; (c) chromatographic resolution; (d) 30 equiv 4 M HCl in dioxane, rt, 1 h or 10 equiv $ZnBr_2$,CH_2Cl_2.

development, we were intrigued by whether the above favorable interactions could be maintained or enhanced by another suitably-functionalized aromatic plate.

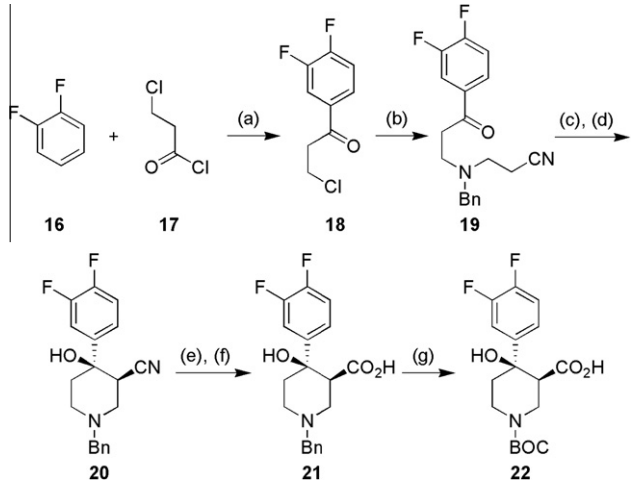
Since the chemistry developed previously for the synthesis of compound **1** necessitated the installation of the pyridone handle at an early stage, an alternative approach that would allow for end stage structural diversification at the 4-position of the piperidine ring was highly desirable. In this regard, the addition of aryl organometallics to 4-oxo-piperidine-3-carboxamides[5] developed by Bezençon et al. proved to be the most ideal solution (Scheme 1). Briefly, β-ketoester **2**[4] was first converted to β-ketoamide **4** by heating with amine **3**[6] in the presence of catalytic quantities of 4-dimethylamino-pyridine. Subsequent LiCl-mediated addition of an aryl Grignard reagent to amide **4** afforded the corresponding alcohol **5**, albeit as a ~1:1 mixture of diastereomers. Following the isolation of the desired *cis*-isomer via repeated column chromatography, the final cleavage of the BOC-protecting group could be readily accomplished either in the presence of a large excess of 4 M HCl in dioxane or with zinc(II) bromide in CH_2Cl_2.[7]

Using compound **7** with its naked benzene as a reference point, the addition of a methoxy group at the *meta*-position (i.e., **8**) to

Table 2
Key profiles of compound **15**

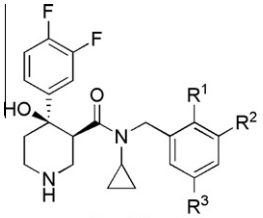
Renin IC_{50}[a,b] (nM)	Buffer		6.4
	Plasma		42
PK in SD rat (5 mpk I.V.)	*F* (%)	0.3 mpk P.O.	17
		3 mpk P.O.	29
		30 mpk P.O.	18
	Cl (mL/min/kg)		36
	$T_{1/2}$ (h)		1.5
	V_{dss} (L/kg)		5
Efficacy in dTGR (10 mpk P.O.)	Max. BP ↓ (mm Hg)		0
	Duration (h)		0
Off-target profile	hERG K_i (μM)		4.5
	CYP 3A4 Inhibition IC_{50} (μM)		0.9

[a] See Ref. 8 for assay protocols.
[b] Average of at least two replicates.

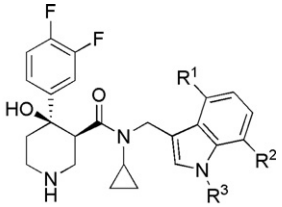


Scheme 2. Synthesis of **22**. Reagents and conditions: (a) 1.2 equiv $AlCl_3$, CS_2, 45 °C, 20 h, 95%; (b) 1 equiv $BnNH(CH_2)_2CN$, 2.5 equiv NEt_3, THF, 22 °C, 0.5 h, >99%; (c) 1.5 equiv KO^{t}Bu, THF, 30 °C, 89%; (d) chiral resolution, AD column, 7:1:1 hexanes/EtOH/*i*PrOH; (e) 2.5 equiv LiOH, 5 equiv 30% aq H_2O_2, DMSO, 45 °C; (f) KOH, ethanol, 80 °C, 10 h; (g) 0.2 equiv $Pd(OH)_2$ on carbon, 1.2 equiv di-*tert*-butyldicarbonate, 1 equiv 200 psi H_2, ethanol, 22 °C, 2 h, 70% over three steps.

Table 3
SAR of select renin inhibitors: amide modifications



23 - 25



26 - 54

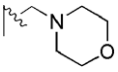
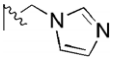
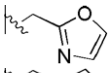
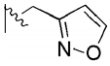
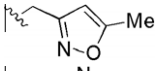
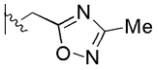
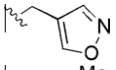
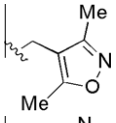
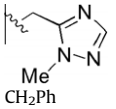
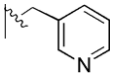
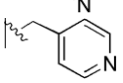
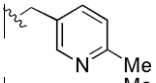
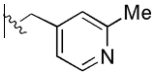
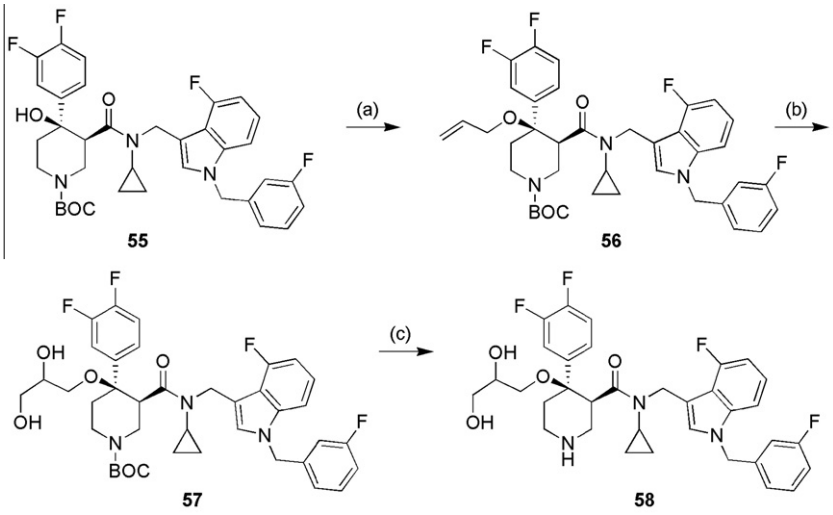
Compound	R^1	R^2	R^3	Renin potency[a]		hERG K_i (μM)	CYP 3A4 inhibition	
				Buffer (nM)	Plasma (nM)		Reversible (% activity)[b]	Time dependent (% loss)[c]
23	Cl	Cl	H	1.2	145	0.68	12	67
24	Cl	H	$(CH_2)_3OMe$	1.1	61	1.0	6	–
25	Cl	H	$(CH_2)_2OMe$	1.2	110	2.2	9	–
26	F	H	H	0.24	2.0	1.4	15	18
27	Cl	H	H	0.11	1.6	0.91	12	52
28	Br	H	H	0.17	2.1	0.46	16	61
29	F	H	$(CH_2)_3OMe$	0.07	3.4	1.3	3	–
30	F	$(CH_2)_3OMe$	H	0.10	6.2	1.1	<1	–
31	F	$(CH_2)_3OMe$	$(CH_2)_3OMe$	0.04	4.4	0.85	3	–
32	F	$(CH_2)_2CN$	$(CH_2)_3OMe$	0.26	2.5	0.60	2	–
33	F	CH_2SO_2Me	$(CH_2)_3OMe$	0.18	2.7	1.8	8	–
34	F		$(CH_2)_3OMe$	0.40	1.5	0.90	<1	–
35	F		$(CH_2)_3OMe$	0.30	2.2	1.4	<1	–
36	F	H	$CH_2(C{=}O)NHOMe$	0.45	2.2	8.4	14	14
37	F	H	$CH_2(C{=}O)N(Me)OMe$	1.0	7.4	2.1	7	–
38	F	H		0.06	0.57	1.1	8	–
39	F	H		0.15	5.2	0.90	9	–
40	F	H		0.36	11	0.88	7	–
41	F	H		0.03	0.42	1.9	8	–
42	F	H		0.38	2.8	0.62	5	–
43	F	H		14	190	0.71	5	–
44	F	H		0.76	1.5	4.4	7	–
45	F	H	CH_2Ph	0.05	1.7	0.25	10	63
46	F	H	$CH_2(2\text{-}(OCHF_2)Ph)$	1.5	330	0.24	5	–
47	F	H	$CH_2(3\text{-}(OCHF_2)Ph)$	0.10	14	0.30	6	–
48	F	H	$CH_2(4\text{-}(OCHF_2)Ph)$	0.66	130	0.16	3	–
49	F	H	$CH_2(3\text{-}FPh)$	0.04	4.8	0.25	5	–
50	F	H	$CH_2(3,4\text{-}F_2Ph)$	0.09	12	0.30	7	–
51	F	H		0.10	3.4	0.34	3	–
52	F	H		0.08	1.0	1.1	3	–

Table 3 (*continued*)

Compound	R^1	R^2	R^3	Renin potency[a]		hERG K_i (μM)	CYP 3A4 inhibition	
				Buffer (nM)	Plasma (nM)		Reversible (% activity)[b]	Time dependent (% loss)[c]
53	F	H		2.5	48	0.36	20	66
54	F	H		0.04	0.36	0.97	1	—

[a] Average of at least two replicates. See Ref. 8 for assay protocols.
[b] Calculated as a percentage of the difference in the rate of CYP3A4-mediated conversion of testosterone (250 μM) to 6-β-hydroxytestosterone in the presence of compound (10 μM in DMSO) versus blank DMSO. 50% activity corresponds to a reversible CYP 3A4 inhibition IC_{50} of 10 μM. 10% activity corresponds to a reversible CYP 3A4 inhibition IC_{50} of 2 μM.
[c] Calculated as a percentage of the difference in the rate of CYP3A4-mediated conversion of testosterone (250 μM) to 6-β-hydroxytestosterone before and after 30 min incubation period with the compound (10 μM in DMSO). A 0% loss corresponds to no measurable time-dependent CYP 3A4 inhibition.



Scheme 3. Synthesis of **58**. Reagents and conditions: (a) 1.5 equiv NaH (60% dispersion in oil), DMF, 5 equiv allyl bromide, 80 °C, 5 h, 45%; (b) 0.1 equiv $OsCl_3$, 0.05 equiv DABCO, 3 equiv K_3CO_3, 3 equiv $K_3Fe(CN)_6$, *t*-BuOH, water, rt, 15 h, 89%; (c) 10 equiv $ZnBr_2$,CH_2Cl_2, rt, 1.5 h, 55%.

Table 4
Key profiles of compounds **49** and **58**

Compound		**49**	**58**
Renin IC_{50}[a,b] (nM)	Buffer	0.04	<0.02
	Plasma	4.8	16
SD rat (3 mpk P.O., 5 mpk I.V.)	*F* (%)	37	<1
	P.O. AUC (μM*h)	0.34	0.04
	Cl (mL/min/kg)	93	16
	$T_{1/2}$ (h)	9.4	2.6
	V_{dss} (L/kg)	48	0.4
Efficacy in dTGR (3 mpk P.O.)	Max. BP ↓ (mm Hg)	15	—
	Duration (h)	48	—
hERG K_i (μM)		0.25	0.80
CYP 3A4 IC_{50} (nM)	Reversible (% activity)[c]	5	67
	Time dependent (% loss)[d]	—	31

[d]Calculated as a percentage of the difference in the rate of CYP3A4-mediated conversion of testosterone (250 μM) to 6-β-hydroxy-testosterone before and after 30 min incubation period with the compound (10 μM in DMSO). A 0% loss corresponds to no measurable time-dependent CYP 3A4 inhibition.
[a] See Ref. 8 for assay protocols.
[b] Average of at least two replicates.
[c] Calculated as a percentage of the difference in the rate of CYP3A4-mediated conversion of testosterone (250 μM) to 6-β-hydroxy-testosterone in the presence of compound (10 μM in DMSO) versus blank DMSO. 50% activity corresponds to a reversible CYP 3A4 inhibition IC_{50} of 10 μM. 10% activity corresponds to a reversible CYP 3A4 inhibition IC_{50} of 2 μM.

better engage renin enzyme's Trp45 was found to be beneficial for binding. The intrinsic renin potency could be improved further by simply replacing the methoxy group with either a chlorine (i.e., **9**) or a fluorine (i.e., **10**) atom. However, it should be noted that these substitutions also afforded compounds that were more shifted in the presence of human plasma. Regardless, since having a fluorine at the *meta*-position appeared to offer the best compromise between enzyme potency and plasma shift, this element was kept constant in the following SAR. Further addition of another small, lipophilic group at either the *para*- or the other *meta*-position improved the renin potency by another three- to seven-fold, with the 3,4-difluoro substitution pattern (i.e., **13**) being optimal. Consequently, compound **13** was resolved and the more potent enantiomer (i.e., **15**) was profiled (Table 2). Although we were encouraged by the lack of dependence of bioavailability on dose when **15** was given orally to SD rats, a common phenomenon that plagued earlier generations of renin inhibitors,[9] we were however alarmed by the compound's affinity for the hERG channel and by its ability to inhibit CYP3A4 activity. Furthermore when compound **15** was given to hypertensive double transgenic rats (dTGR) harboring both human renin and angiotensinogen,[10] we failed to observe any lowering of blood pressure.

In order to improve the renin potency further as well as address the above off-target concerns, we then decided to focus our SAR effort on the benzyl amide handle. Although the synthetic scheme

described in Scheme 1 could have been serviceable for this campaign, we again felt it was more prudent to re-design the synthesis so that the diversification step (i.e., amide coupling) could be postponed to the end. In this regard, a scalable route to access the requisite chiral hydroxyl acid **22** was developed (Scheme 2). Briefly, Friedel–Crafts acylation of 1,2-difluorobenzene (**16**) with 3-chloro-propionyl chloride (**17**) in carbon disulfide afforded ketone **18** in 95% yield. Its subsequent amination with commercially available 3-(benzylamino)propanenitrile proceeded quantitatively with triethylamine as base. In the presence of 1.5 equiv of potassium *tert*-butoxide, ketone **19** underwent intramolecular cyclization to afford the desired hydroxyl nitrile as a single diastereomer. Following chiral separation on a Chiralpak AD column, the desired enantiomer **20** was hydrolyzed to the corresponding acid **21** by a two step process: an initial oxidation to the intermediate amide with lithium peroxide followed by hydrolysis with ethanolic potassium hydroxide. Finally, a protecting group switch from benzyl amine **21** to *tert*-butyl carbamate **22** under a hydrogen atmosphere was best accomplished with Pearlman's catalyst in the presence of di-*tert*-butyl-dicarbonate and triethylamine.

With hydroxyl acid **22** in hand, all the amides shown in Table 3 were prepared from the requisite amines with HATU as the coupling agent followed by cleavage of the BOC protecting group. As expected,[4] the best amine identified for the earlier generation renin inhibitors[6] was no longer optimal for our current series. Indeed, when benzyl amine **3** was switched for *N*-(2,3-dichlorobenzyl)cyclopropanamine, a six-fold increase in intrinsic renin potency was observed (i.e., **23** vs **15**). However in the absence of a suitable tail designed to anchor the inhibitor into the renin s3 sub-pocket, compound **23** was also highly shifted by human plasma. Although the plasma shift observed could be slightly decreased when either 3-methoxypropyl (i.e., **24**) or 2-methoxyethyl (i.e., **25**) tail was re-introduced at the *meta*-position, neither compound exhibited sufficient renin plasma potency to warrant further profiling. On the other hand, a major potency breakthrough was realized when we abandoned the benzene ring (i.e., **7** ~ **15**, **23** ~ **25**) for an indole scaffold (i.e., **26** ~ **54**). Indeed, with only a halogen substituent at the 4-position of an otherwise unfunctionalized indole ring (i.e., **26** ~ **28**), sub-nanomolar renin inhibitors were obtained. Although it was not possible to differentiate between these analogues on the basis of their respective plasma renin IC_{50}, the 4-fluoro derivative **26** appeared to possess the best, albeit still unacceptable, off-target profile of the three. When compound **26** was docked into the renin active site in silico, molecular modeling revealed that the renin s3 sub-pocket can be accessed from both the 1- and 7-positions of the indole ring. Consequently, a 3-methoxylpropyl tail was installed at either of these two positions to assess whether more improvements in potency could be achieved. While compounds **29** and **30** were both more potent than their un-alkylated precursor **26** in the enzyme assay, these renin inhibitors were however more plasma shifted. Furthermore, these modifications failed to improve the off-target profile. From **29**, the addition of a second alkyl chain capped by methoxide (i.e., **31**), cyanide (i.e., **32**), sulfone (i.e., **33**), morpholine (i.e., **34**) or imidazole (**35**) also failed to deliver any tangible improvement over **26**. In contrast, the replacement of 3-methoxypropane in **29** by an *N*-methoxyacetamide (i.e., **36**) did lead to a six-fold decrease in binding affinity to the hERG channel. We have also evaluated the impact of capping compound **26**'s indole NH with either a benzyl (i.e., **45** ~ **50**) or a heteroaryl methyl (i.e., **38** ~ **44**, **51** ~ **54**) residue. Compounds capped with a five-membered heterocycle such as 1,3-oxazole (i.e., **38**) and 3-methyl-1,2,4-oxadiazole (i.e., **41**), or a 6-membered heterocycle such as 4-methyl-3-pyridine (i.e., **54**), were found to be sub-nanomolar renin inhibitors even in the presence of human plasma. However, further addition of methyl groups to 5-membered heterocycles proved to be deleterious for renin potency (i.e., **40** vs **39**, **43** vs **42**). For analogues capped with either a benzyl or a pyridyl group, *ortho*- and *para*-substitutions were also not tolerated (i.e., **46** vs **45**, **48** vs **45**, **53** vs **51**). On the other hand, small substituents at the *meta*-position (i.e., **49** and **54**) were beneficial for renin potency.

Although great strides in terms of renin potency were made and several low picomolar renin inhibitors were identified, closer examination would reveal that these gains were often achieved at the expense of the off-target profile (i.e., hERG binding, CYP3A4 inhibition or both). Consequently, in order to ascertain whether the judicious introduction of polarity could free the current series from this zero-sum quagmire, the tertiary alcohol of compound **49**[11] was functionalized as depicted in Scheme 3. Starting from its *tert*-butyl carbamate **55**, alkylation with allyl bromide was best accomplished by heating the reactants with sodium hydride in DMF. Subsequent dihydroxylation of the olefin in **56** using modified Upjohn conditions[12] afforded the desired diol **57** as a ~1:1 mixture of diastereomers. Finally, removal of the BOC protecting group with excess zinc(II) bromide delivered compound **58** uneventfully.[7]

The key characteristics of compounds **49** and **58** are summarized in Table 4. As expected, the introduction of polarity did improve the off-target profile to a more manageable level. Unfortunately, this was accompanied by a drop in plasma renin potency and more importantly, compound **58** was no longer orally bioavailable in rats. Consequently, this series was put on hold.

In summary, by committing the necessary resources to the development of synthetic sequences that were most amenable for late stage structural diversification, we were able to make a rapid 'no go' decision on this series of renin inhibitors. Although compounds suitable for further development could not be identified, several members of this series were found to exhibit dose-independent oral bioavailability in rats. Their use to elucidate the role of efflux transporters on the poor oral bioavailability shared by early generation renin inhibitors at low dose will be presented in a future manuscript.

References and notes

1. http://ucatlas.ucsc.edu/cause.php
2. Zaman, M. A.; Oparil, S.; Calhoun, D. A. *Nat. Rev. Drug Discovery* **2002**, *1*, 621.
3. Hollenberg, N. K. *Nat. Rev. Nephrology* **2010**, *6*, 49.
4. Chen, A.; Campeau, L.-C.; Cauchon, E.; Chefson, A.; Ducharme, Y.; Dubé, D.; Falgueyret, J.-P.; Fournier, P.-A.; Gagné, S.; Grimm, E.; Han, Y.; Houle, R.; Huang, J.-Q.; Lacombe, P.; Laliberté, S.; Lévesque, J.-F.; Liu, S.; MacDonald, D.; Mackay, B.; McKay, D.; Percival, M. D.; Regan, C.; Regan, H.; St-Jacques, R.; Stump, G.; Soisson, S.; Toulmond, S. *Bioorg. Med. Chem. Lett.* **2011**. doi:10.1016/j.bmcl.2011.05.013.
5. Corminboeuf, O.; Bezençon, O.; Remeň, L.; Grisotomi, C.; Richard-Bildstein, S.; Bur, D.; Prade, L.; Strickner, P.; Hess, P.; Fischli, W.; Steiner, B.; Treiber, A.; Vaughn, C. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 6291.
6. Chen, A.; Bayly, C.; Bezençon, O.; Richard-Bildstein, S.; Dubé, D.; Dubé, L.; Gagné, S.; Gallant, M.; Gaudreault, M.; Grimm, E.; Houle, R.; Lacombe, P.; Laliberté, S.; Lévesque, J. F.; Liu, S.; MacDonald, D.; Mackay, B.; Martin, D.; McKay, D.; Powell, D.; Remeň, L.; Soisson, S.; Toulmond, S. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 2204.
7. Nigam, S. C.; Mann, A.; Taddei, M.; Wermuth, C.-G. *Synth. Commun.* **1989**, *19*, 3139.
8. *Buffer assay*: Human recombinant renin (Proteos) at 100 pM was incubated in the presence or absence of renin inhibitors and 6 μM of Q-FRET substrate 9 DNP-Lys–His–Pro–Phe–His–Leu–Val–Ile–His-$_{D,L}$-Amp in 50 mM MOPS, 100 mM NaCl, pH 7.4, 0.002% Tween. The reactions take place in a Costar 384 well black plate at 37 °C for 3 h. Fluorescence was measured at times 0 and 3 h in a SpectraMax Gemini EM reader with excitation and emission filters at 328 and 388 nm, respectively. *Plasma assay*: Frozen human EDTA-plasma was rapidly thawed in warm water and centrifuged at 2900 g for 15 min at 40 °C. The supernatant was collected and recombinant human renin (Proteos) added at 1 nm nominal concentration. The plasma was transferred to Costar black 384 well plates, renin inhibitors added and the mixture pre-incubated at 37 °C for 10 min. The renin Q-FRET substrate *QXL520-Lys–His–Pro–Phe–His–Leu–Val–Ile–His–Lys-(5-FAM)* (Proteos), diluted in 3 M Tris/200 mM EDTA, pH 7.2 was added to the plasma with final concentrations of 342 mM Tris, 23 mM EDTA and 6.8 μM substrate. The plate was incubated at 37 °C for 1 h and the plate read in a SpectraMax Gemini EM reader with excitation and emission filters at 490 and 520 nm, respectively, at time 0 and 1 h.

9. In fact, the oral bioavailability of renin inhibitors such as aliskiren increases nonlinearly with dose. It has been hypothesized that because aliskiren is a good substrate of endogenous human and rat PGP transporters, this efflux mechanism would become increasingly saturated at higher doses. Drugs that exhibit this behavior could have the potential to be victims of drug–drug interactions when co-dosed with compounds known to inhibit PGP transporters.
10. Hess, P.; Clozel, M.; Clozel, J.-P. *J. Appl. Physiol.* **1996**, *81*, 1027.
11. During in silico docking experiment with compound **15**, modeling revealed that the tertiary alcohol residue points out into solvent and does not engage in any stabilizing interactions with the renin enzyme.
12. Minato, M.; Yamamoto, K.; Tsuji, J. *J. Org. Chem.* **1990**, *55*, 766.