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Communications

Synthesis of Vinyl Sulfone-tethered Proline Derivatives as Highly Selective Cathepsin S Inhibitors

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Cathepsin S (CatS), a cysteine protease belonging to the papain family, has been proposed as an attractive therapeutic target for asthma[1] and autoimmune diseases[2] due to the fact that CatS is closely implicated in the process of antigen presentation.[3,4] Indeed, it has been reported that inhibition of CatS decreases chronic inflammatory pain in a collagen-induced arthritis (CIA) model of rheumatoid arthritis.[5] Thus, there is a growing interest among medicinal chemists to identify CatS inhibitors. Morpholineurea-leucine homophenylalanine vinyl sulfone 1 (LHVS, Figure 1) has been previously identified as a potent CatS inhibitor but limited selectivity for members of the cathepsin family.[6] A number of selective CatS inhibitors have been reported as potential therapeutic agents, including cyanopeptide-, pyrrolopyrimidine-, aminopyrimidine-, cyanopyrazolidine-based inhibitors.[7,8] Although some CatS inhibitors are currently being evaluated in clinical trials,[9] potent and selective CatS inhibitors are still required. With respect to development of selective CatS inhibitors, we recently reported novel selective cathepsin S inhibitors based on a proline scaffold.[10] Following up on this work, we herein report the synthesis of a series of proline analogues with vinyl sulfone group, which is a key functional group of LHVS. We evaluated these novel compounds for their *in vitro* CatS inhibitory activity to investigate a structure-activity relationship (SAR).

The general synthetic procedure for preparing 4-sulfonyl substituted proline-derived analogues is summarized in Scheme 1. Mesylation of the commercially available proline

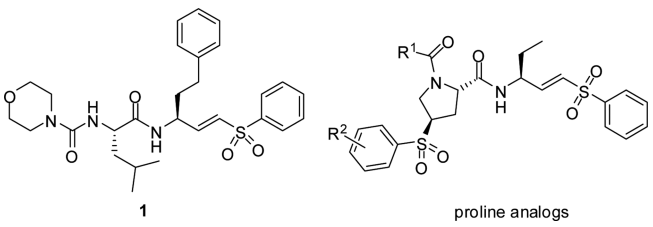
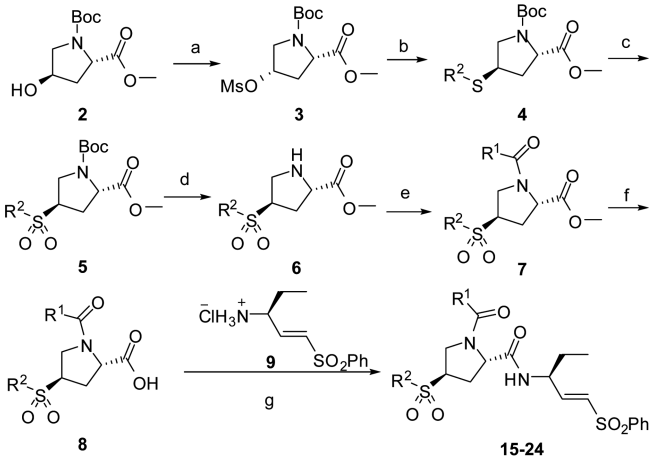
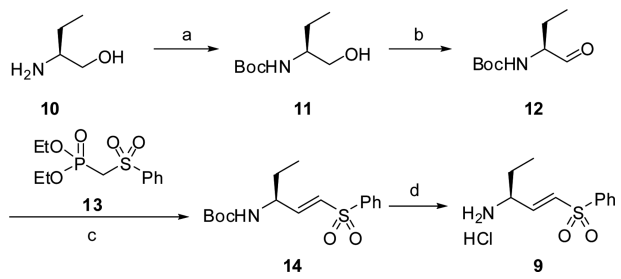


Figure 1. Structure of LHVS and design of proline-based CatS inhibitors.



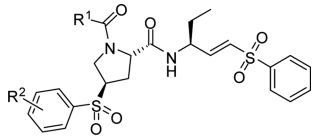
Scheme 1. (a) MsOH, DIAD, PPh_3, TEA, THF, 70 °C, 15 h; (b) R^2SH, NaH, THF, 0 °C, 30 min then, 50 °C, 4 h; (c) *m*CPBA, CH_2Cl_2, 8 h; (d) 4 N HCl in 1,4-dioxane, CH_2Cl_2, 2 h; (e) $(R^1CO)_2O$, TEA, MeOH, 2 h or R^1COCl, DIPEA, CH_2Cl_2, 2 h; (f) LiOH in H_2O/MeOH, THF, 0 °C → rt, 2 h; (g) EDCI, HOBt, DIPEA, CH_2Cl_2, rt, 5-10 h.

analogue **2** was performed under Mitsunobu condition and subsequent nucleophilic substitution by a thiol afforded the thioether **4**, which was converted to sulfone **5** by *m*CPBA oxidation. After deprotection of *tert*-Boc, acid chloride or



Scheme 2. (a) Boc_2O, DCM, rt, 2 h; (b) Dess-martin periodinane, 0 °C to rt, 1 h; (c) **13**, NaH, THF, 0 °C to rt, 3 h; (d) 4 N HCl in 1,4-dioxane, DCM, 1 h.

Table 1. Inhibitory activities of derivatives for *h*CatS



Compound	R^1	R^2	IC_{50} (nM)[12]
15	CH_3	H	2.6
16	*iso*-Pr	H	1.2
17	Cyclohexyl	H	2.6
18	1-Piperidinyl	H	2.2
19	4-Pyridinyl	H	2.0
20	Phenyl	H	1.4
21	CH_3	2-Cl	1.4
22	CH_3	3-Cl	1.9
23	CH_3	4-Cl	1.9
24	CH_3	4-phenyl	2.2
LHVS			1.9

Table 2. Inhibitory activities for Cathepsin S, B, and K

Compound	IC_{50} (nM)		
	*h*Cat S	*h*Cat B	*h*Cat K
15	2.6	> 1,000	> 1,000
16	1.2	> 1,000	> 1,000
20	1.4	> 1,000	> 1,000
LHVS	1.9	-	4.0
CA-074	-	3.0	

anhydride were coupled with an amine **6** to provide the amide **7**. Next, compound **7** was hydrolyzed and treated with vinyl sulfone amine **9** in the presence of EDCI and HOBt to yield the desired analogues (**15**-**24**). The synthetic route for compound **9** is illustrated in Scheme 2. Commercially available amino-alcohol **10** was protected with *t*-Boc group, followed by oxidation with Dess–Martin periodinane to furnish aldehyde **12** in high yield (85% over 2 steps).

Olefination of **12** with phosphonate **13**[11] gave the required vinyl sulfone **14** and Boc deprotection provided the desired fragment **9**. We attempted to modify a proline moiety to increase selectivity while retaining the vinyl sulfone, which is critical for activity of LHVS. In order to investigate SAR, the R^1 moiety was modified to various alkyl (**15** -**18**) or aryl (**19** and **20**) substituents. All of the R^1-modified analogues showed excellent IC_{50} values for human CatS (*h*Cat S) in the range of 1.2 to 2.6 nM, which was similar to LHVS (Table 1). Likewise, most vinyl sulfone analogues showed better activity than the 2-acyl benzoxazole analogues that we previously reported.[10] We next evaluated the influence of R^2-substituents on cathepsin S, for which R^1 were fixed as methyl group. Regioisomeric chlorophenyl derivatives **21**-**23** and the 4-biphenyl compound **24** exhibited inhibitory activity similar to the unsubstituted phenyl analogue **15**. Several of the potent CatS inhibitors then were screened for inhibitory activity for cathepsin B and K to determine if they were selective for CatS (Table 2). While LHVS showed potent inhibitory activity for CatK as well as CatS, the selected compounds did not inhibit CatB and CatK. CA-074 was used as a positive control for CatB.[13]

In summary, we developed a series of novel vinyl sulfone tethered proline derivatives as potent CatS inhibitors. The representative compounds showed excellent *in vitro* activity and selectivity for CatS over CatB and CatK. Thus, these compounds could serve as leads for the development of CatS inhibitors.

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