Available online at www.sciencedirect.com
ScienceDirect
Bioorganic & Medicinal Chemistry Letters 18 (2008) 366–370
Bioorganic & Medicinal Chemistry Letters

Truncation and non-natural amino acid substitution studies on HTLV-I protease hexapeptidic inhibitors

Jeffrey-Tri Nguyen,[a] Meihui Zhang,[a,b] Henri-Obadja Kumada,[a] Ayako Itami,[a] Keiji Nishiyama,[a] Tooru Kimura,[a] Maosheng Cheng,[b] Yoshio Hayashi[a,c] and Yoshiaki Kiso[a,*]

[a]*Department of Medicinal Chemistry, Center for Frontier Research in Medicinal Science and 21st Century COE Program, Kyoto Pharmaceutical University, Yamashina-ku, Kyoto 607-8412, Japan*
[b]*Key Laboratory of New Drugs Design and Discovery of Liaoning Province, School of Pharmaceutical Engineering, Shenyang Pharmaceutical University, Shenyang 110016, PR China*
[c]*Department of Medicinal Chemistry, Tokyo University of Pharmacy and Life Sciences, Hachioji, Tokyo 192-0392, Japan*

Received 26 September 2007; revised 16 October 2007; accepted 18 October 2007
Available online 24 October 2007

Abstract—The culprit behind adult T-cell leukemia, myelopathy/tropical paraparesis, and a plethora of inflammatory diseases is the human T-cell leukemia virus type 1 (HTLV-I). We recently unveiled a potent hexapeptidic HTLV-I protease inhibitor, KNI-10166, composed mostly of natural amino acid residues. Herein, we report the derivation of potent tetrapeptidic inhibitor KNI-10516, possessing only non-natural amino acid residues.
© 2007 Elsevier Ltd. All rights reserved.

Adult T-cell leukemia (ATL), an aggressive malignancy of CD4 T lymphocytes, that presents as skin lesions, lymphadenopathy, and hepatosplenomegaly, was first described by Takatsuki and co-workers in Japan in 1977.[1] HTLV-I-associated myelopathy/tropical spastic paraparesis (HAM/TSP), predominantly associated with CD8 T lymphocytes in the later stage of the disease, primarily affects the spinal cord at the thoracic level causing symmetrical weakness and paralysis of the lower limbs.[2] The causative agent for ATL, HAM/TSP, and several inflammatory diseases was identified in 1980 by Gallo and co-workers as the human T-cell leukemia virus type 1 (HTLV-I).[2,3] Infecting 15–20 million people worldwide, the virus is often transmitted through breast-feeding, sexual intercourse, blood transfusion, and the sharing of contaminated injection devices, thereby allowing the virus to be endemic in the equatorial regions of Africa, Central and South America, the Caribbean, Melanesia, the Middle East, and south-western Japan, and spreading to the United States and Europe through injection drug use and sexual transmission.[2,4] Although there are therapies to alleviate the symptoms of the diseases, there is no effective treatment to eradicate HTLV-I. Infection with HTLV-I is lifelong. HTLV-I protease (PR), first identified and isolated in 1989, plays a pivotal role in HTLV-I replication.[5] Inhibition of HTLV-I PR would essentially stop viral replication and provide a possible cure to HTLV-I related diseases, thereby directly attacking at the source of the problems. The genome for HTLV-I encodes for several key proteins, namely Gag, Pro, Pol, and Env.[6] HTLV-I PR cleaves the Gag and Pol precursor polyproteins into several shorter proteins that are subsequently assembled and developed into a mature virion that ultimately becomes the cause for HTLV-I-associated diseases. Hence, our approach to the problem is to inhibit HTLV-I PR with a substrate mimic, and thereby stopping the spread of the virus.

One of the cleavage sites involved in the processing of the precursor Gag polyprotein, the matrix/capsid cleavage site, can accommodate for a specific substrate (**1**, Table 1).[7] In our previous work, based on the substrate sequence (**1**) and the concept of ''transition-state mimic'' at the P_1/P'_1 cleavage position using (2*S*,3*S*)-3-amino-2-hydroxy-4-phenylbutyric acid (allophenylnorstatine,

Keywords: Human T-cell leukemia virus; Adult T-cell leukemia; Human immunodeficiency virus; Protease inhibitor.
* Corresponding author. Tel.: +81 75 595 4635; fax: +81 75 591 9900; e-mail: kiso@mb.kyoto-phu.ac.jp

0960-894X/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2007.10.066

Table 1. HTLV-I PR substrates and peptidic inhibitors

Compounds (KNI-No.)		Structure										HTLV-I PR IC_{50}[a] (nM)	HIV-1 PR inhibition[b] (%)
			P_4	P_3	P_2	P_1	P'_1	P'_2	P'_3	P'_4	P'_5		
1	Substrate	H	Pro	Gln	Val	Leu	Pro	Val	Met	His	Pro	OH	—	—
2	10161	H	Pro	Gln	Val	Apns	Pro	Val	Met	His	OH		159	<30
3	10162	H	Pro	Gln	Val	Apns	Dmt	Val	Met	His	OH		152	58
4	10127		Ac	Gln	Val	Apns	Dmt	Val	Met	NH_2			353	96
5	10166		Ac	Ile	Ile	Apns	Dmt	Ile	Met	NH_2			88	98

[a] HTLV-I PR inhibition, IC_{50} (nM), using HTLV-I PR mutant L40I.
[b] HIV-1 PR inhibition (%) at 50 nM of the test compound.

Apns) having a hydroxymethylcarbonyl (HMC) isostere, we developed potent octapeptidic HTLV-I PR inhibitor KNI-10161 (**2**).[8] KNI-10162 (**3**), which differed by a P'_1 non-natural proline isostere, namely (*R*)-5,5-dimethyl-1,3-thiazolidine-4-carboxylic acid (Dmt), exhibited equipotency. The result of a truncation study led to a smaller hexapeptidic inhibitor, KNI-10127 (**4**), with a fair loss of activity.[9] In order to recover inhibitory activity, we performed substitution studies by building small amino acid libraries at each residue position to derive compound KNI-10166 (**5**). Considering that the in vivo efficacy of inhibitors is severely compromised by their susceptibility to proteolytic degradation and difficulty in penetrating cells, we felt a need to explore non-natural amino acid residues and further reduce the size of the inhibitor. Herein, we report the design, synthesis, and activity of several novel hexa-, penta-, and tetrapeptidic HTLV-I PR inhibitors possessing non-natural amino acid residues.

We performed a truncation study on KNI-10166 (**5**, Table 2). Compounds **6** and **7** were synthesized and their inhibitory activities against HTLV-I and human immunodeficiency virus type 1 (HIV-1) PRs were determined using inhibitor **5** as a comparative reference. Compound **6**, which lacked a P_3 Ile moiety at the N-terminal, had a drastic loss of inhibitory activity against HTLV-I PR.[10] Compound **7**, which lacked a P'_3 Met moiety at the C-terminal, retained some activity against HTLV-I PR, thereby suggesting that the P_3 residue is a higher determinant of activity than the P'_3 residue.

In our preceding work, we built small libraries with natural amino acids at each residue position to derive inhibitor **5** and noted that the S'_2, S_2, and S_3 sites best accommodated for Ile residues.[9] To avoid recognition by endopeptidases that could prematurely metabolize the compound in the body, in the current study, we opted for non-natural amino acid residue substitution and chose L-*tert*-leucine (Tle) and L-(+)-α-phenylglycine (Phg) to mimic the branched amino acid, Ile (Table 3). Among the Tle and Phg residues, the S_3 site accommodated better Phg (cf. **8** and **9**), whereas a P_2 Tle would exhibit more potent HTLV-I PR inhibitory activity (cf. **10** and **11**), and Phg is the preferred moiety at the P'_2 position (cf. **13** and **14**).

Although in our published work on HTLV-I PR inhibitors,[8,9] we designed inhibitors possessing an Apns residue with an HMC isostere at the P_1 position, based on our published HIV-1 PR inhibitor studies, in the current study, we explored Apns' diastereomer (2*R*,3*S*)-3-amino-2-hydroxy-4-phenylbutyric acid (phenylnorstatine, Pns) and did not observe an improvement in HTLV-I PR inhibition (cf. **5** and **12**).

We noted that a P'_3 Ala residue could be accommodated by the reverse transcriptase–ribonuclease H/integrase S'_3 cleavage junction,[6] and synthesized compound **15** that exhibited lower PR inhibitory activity than reference inhibitor **5**. In our previous study, we discovered that KNI-10153 and KNI-10156, each, respectively, possessing a P'_3 Phe or Gln residue, exhibited fairly potent HTLV-I PR inhibition,[9] and accordingly, we synthesized and evaluated HTLV-I PR inhibitory activity of compounds **16** and **17**, in which a corresponding P'_3 Phe or Gln residue was present. Compounds **5** and **16** exhibited similar potencies, while compounds **15** and **17** were equipotent.

From the results of the truncation study on compound **5** (Table 2), we decided to optimize the P_3–P'_1 residues and then remove the P'_2–P'_3 residues. First, we synthesized P_3–P'_1 optimized inhibitor **18**, which exhibited potent inhibitory activity against both HTLV-I and HIV-1 PRs (cf. **5**). In consideration of the vast structural differences between the P'_3 residues in compounds **5** and **15–17**, and that they exhibited minor variations in HTLV-I PR inhibitory activity (cf. **5** and **16**; **15** and **17**), we inferred that the P'_3 residue is a minor determi-

Table 2. Truncation study on KNI-10166

Compounds (KNI-No.)		Structure								HTLV-I PR IC_{50}[a] (nM)	HIV-1 PR inhibition[b] (%)
			P_3	P_2	P_1	P'_1	P'_2	P'_3			
6	10167		Ac	Ile	Apns	Dmt	Ile	Met	NH_2	>1000	89
5	10166	Ac	Ile	Ile	Apns	Dmt	Ile	Met	NH_2	88	98
7	10168	Ac	Ile	Ile	Apns	Dmt	Ile	NH_2		249	57

Shaded value denotes that a truncation study has been performed.
[a] HTLV-I PR inhibition, IC_{50} (nM), using HTLV-I PR mutant L40I.
[b] HIV-1 PR inhibition (%) at 50 nM of the test compound.

Download English Version:

https://daneshyari.com/en/article/1376941

Download Persian Version:

https://daneshyari.com/article/1376941

Daneshyari.com