

Synthesis of a new hydroxyethylene dipeptide isostere PheΨ [CH₂CH(OH)]Phe as HIV-1 protease inhibitor

Huai Gu CHEN¹, Tomi K SAWYER¹, Peter G M WUTS² (¹Upjohn Laboratories, ² Process Research and Development, The Upjohn Company, Kalamazoo MI 49001, USA)

ABSTRACT A new type of pseudodipeptide isostere exemplified by PheΨ[CH₂CH(OH)]Phe was synthesized from phenylalanine. The HIV protease inhibitory activity (IC₅₀) of Noa-His-PheΨ[CH₂CH(OH)]Phe-Ile-Amp was 0.8 pmol·L⁻¹.

KEY WORDS dipeptides; HIV protease inhibitors; structure-activity relationship

Dipeptide isosteres⁽¹⁾ provide powerful synthetic modifications in the design of pseudopeptide or peptidomimetic analogs of biologically important peptide hormones, neurotransmitters, growth factors and a plethora of peptide substrates for proteases, and other post-translationally active enzymes. In the field of protease inhibitor discovery, the impact of dipeptide isosteres has been witnessed over the past decade by the design of nonhydrolyzable replacements of the scissile dipeptide moiety of naturally-occurring substrates. In the specific case of aspartyl protease inhibitor design. It is well recognized the hydroxyalkyl replacements of the so-called P₁-P_{1'} amide may lead towards the development of remarkably potent, peptide-base inhibitors⁽²⁾. Of such a series of dipeptide isosteres we were motivated by the surprising paucity of compounds of the generic structure (4), a structural isomer of homolog of the peptide isosteres^(3,4) (1), (2), (3) illustrated in Fig 1. In this report was described the synthesis of dipeptide isosteres of structural class

XaaΨ[CH₂CH(OH)]Yaa (4) based upon facile, stereocontrolled methodology which takes advantage of readily available amino acids and Evans chiral aldol condensation to control the stereochemistry. This strategy may lead towards the design and preparation of a number of dipeptide isosteres of the generic structure (4) utilizing inexpensive starting materials and only a few synthetic steps

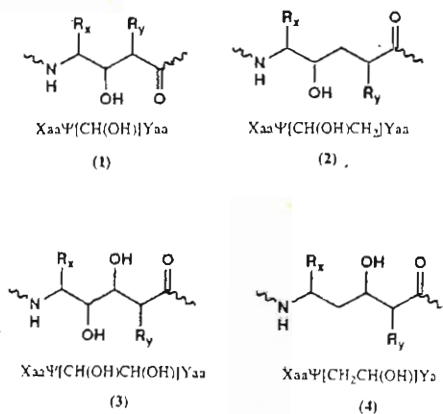


Fig 1. Representative hydroxyalkyl-modified dipeptide isosteres.

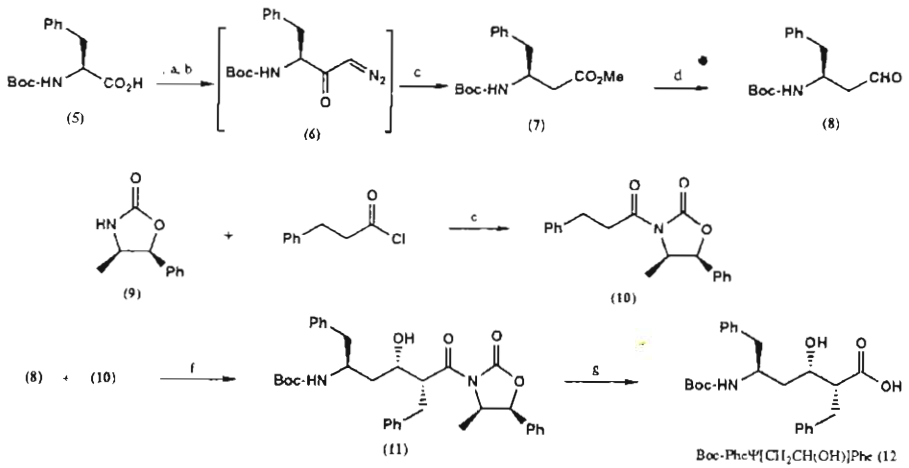
The synthesis of a typical example, Boc-PheΨ[CH₂CH(OH)]Phe (12), is illustrated in Scheme I. the required Boc-β-amino ester (7) was prepared by homologation of Boc-Phe-OH (5) employing the Amdt-Eistert type reaction⁽⁵⁾. The conversion of Boc-L-Phe-OH (5) to the homo ester (7) was accomplished by the treatment of the acid (5) with N-methylmorpholine (NMM) (1.05 equiv) and isobutyl chloroformate (1.1 equiv) in DME at -20 °C. The white precipitate was filtered after 15 min and the filtrate was treated with diazomethane

Received 1993-09-28

Accepted 1993-11-18

¹ Now in: Parke-Davis Pharmaceutical Research Division, Warner-Lambert Co. 2800 Plymouth Road, Ann Arbor MI 48105, USA.

Scheme I

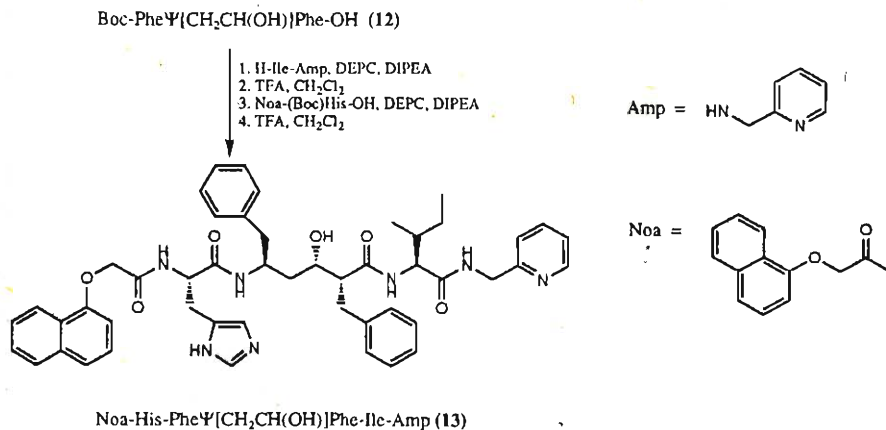


a. ClCO_2iBu , NMM, DME, -20°C b. CH_2N_2 , Et_2O , 0°C c. PhCO_2Ag , Et_3N , MeOH, 87%
 f. Bu_2BOTf , iPr_2NEt , CH_2Cl_2 , 82% g. LiOH , H_2O_2 , THF- H_2O , 0°C , 76%
 d. DIBAL, PhMe, -78°C , 92% e. $n\text{BuLi}$, -78°C

(1.5 equiv) ethereal solution at 0°C to afford the diazoketone (6). The crude (6) was decomposed with PhCO_2Ag (0.2 equiv) and Et_3N (3.8 equiv) in MeOH in a Wolff type rearrangement yielding the homo ester (7), after flash chromatography (silica gel, hexane: AcOEt/3:1), 87% yield, mp $53-54^\circ\text{C}$, $[\alpha]_{\text{D}} = -8^\circ\text{C}$ ($c=0.99$, CHCl_3). The key aldehyde (8) was prepared by the reduction of ester (7) with diisobutylaluminum hydride (2.0 equiv) in dry toluene at -78°C . The crude product was flash chromatographed (silica gel, hexane: AcOEt/4:1) to give the amino aldehyde (8), 92% yield, mp $89-90^\circ\text{C}$, $[\alpha]_{\text{D}} = -20^\circ\text{C}$ ($c=1.01$, CHCl_3). The deprotonation of (4R, 5S)-4-methyl-5-phenyl-2-oxazolidinone (9) with $n\text{BuLi}$ at -78°C in dry THF, followed by treatment with 3-phenylpropanoyl chloride gave acyloxazolidinone (10), 95% yield, mp $95-96^\circ\text{C}$, $[\alpha]_{\text{D}} = +35^\circ\text{C}$ ($c=0.99$, CHCl_3). The stereoselective aldol condensation⁽⁶⁾ was accomplished by treatment of the acyloxazolidinone (10) with Bu_2BOTf (1.2 equiv) at 0°C , followed by slow addition of DIPEA (1.3 equiv). After 1 h at 0°C , the mixture was cooled to -78°C

and aldehyde (8) was added. After another 30 min at -30°C and 2 h at 0°C , the reaction was quenched with pH 7 aqueous buffer. MeOH at 0°C . Standard work-up and flash chromatography (silica gel, hexane: AcOEt/3:1) gave the desired (2R, 3S, 5S)-isostere (11), 82% yield, mp $63-65^\circ\text{C}$, $[\alpha]_{\text{D}} = +19^\circ\text{C}$ ($c=0.99$, CHCl_3). HPLC (Analytical) HPLC was performed on Beckman HPLC instrument, with a 4.6×250 mm, C-18, Vydac 218TO54 column, $\text{H}_2\text{O}/\text{CH}_3\text{CN}/90:10$, flow rate $1.5 \text{ mL} \cdot \text{min}^{-1}$, to give a single peak, $t_{\text{R}} = 15.32$ min) analysis showed a $>96\%$ D (diastereomeric excess) of the stereoselective aldol condensation. Hydrolysis of the chiral auxiliary of (11) with a solution of $\text{LiOH} \cdot \text{H}_2\text{O}$ (2.0 equiv) in H_2O_2 (8 equiv) gave the final product (2R, 3S, 5S)-5-[(*tert*-butoxycarbonyl)amino]-3-hydroxy-6-phenyl-2-phenylmethyl-1-hexanoic acid (12), after flash chromatography (silica gel, hexane: AcOEt:AcOH/10:50:5), 76% yield, mp $168-169^\circ\text{C}$ $[\alpha]_{\text{D}} = -26^\circ\text{C}$ ($c=1.00$, CHCl_3). The overall yield of the synthesis was 42%. This hydroxyethylene dipeptide isostere (12) was then elaborated to give the peptidomimetic (13).

Scheme II



using standard solution-phase peptide synthesis (Scheme II).

The HIV protease inhibitory activity (IC_{50}) of compound (13) was determined to be about $800 \text{ nmol} \cdot \text{L}^{-1}$ (7). Relative to the previously reported peptidomimetic inhibitor Noa-His-PheΨ[CH(OH)(CH(OH))]Phe-Ile-Amp ($K_i = 19 \text{ nmol} \cdot \text{L}^{-1}$) (4), the potency of compound (13) was decreased to about 1/40. The structure-activity relationship of a series of Ψ[CH₂CH(OH)] modified peptidomimetic inhibitors of HIV protease will be published soon. In summary, the above hydroxyethylene modified dipetide isostere exemplifies a novel synthon which may provide versatile application in the field of peptide-base drug design.

ACKNOWLEDGMENT We thank Dr A G Tomasselli, Dr R L Henrikson, and J O Hui for the IC_{50} determination of compound (13).

REFERENCES

1 Hruby VJ, Al-Obeidi F, Kazmierski W. Emerging approaches in the molecular design of receptor-selective peptide ligands: conformational, topographical and dynamic

considerations.

J Biochem 1990; **268**: 249-62.

- 2 Williams RM. Design and synthesis of biologically active peptide mimics. In: Williams WV, Weiner DB, editors. The development and utilization of biologically active peptides; vol 1. Lancaster (PA); Technomic Publ Co, 1993; 187-215.
- 3 Wuts PGM, Ritter AR, Pruitt LE. Synthesis of the hydroxyethylene isostere of Lcu-Val. J Org Chem 1992; **57**: 6696-700.
- 4 Thastrivongs S, Tomasselli AG, Moon JB, Hui J, McQuade TJ, Turner SR, Strohbach JW, Howe WJ, Tarpley WG, Henrikson RL. Inhibitors of the protease from human immunodeficiency virus; design and modeling of a compound containing a dihydroxyethylene isostere insert with high binding affinity and effective antiviral activity. J Med Chem 1991; **34**: 2344-56.
- 5 Mendre C, Rodriguez M, Laur J, Aumelas A, Martinez J. Peptide and pseudopeptide analogues of cholecystokinin. Chemical modification of the Met²⁸-Gly²⁹ region. Tetrahedron 1988; **44**: 4415-30.
- 6 Evans DA, Mathre DJ. Asymmetric synthesis of the enkephalinase inhibitor thirphan. J Org Chem 1985; **50**: 1830-5.
- 7 Tomasselli AG, Hui JO, Sawyer TK, Staples DJ, Banow C, Reardon IM, Howe WJ, DeCamp DL, Craik CS, Henrikson RL. Specificity and inhibition of proteases from human immunodeficiency viruses 1 and 2. J Biol Chem 1990; **265**: 14675-83.