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Glycosylated Peptoids as Prototypical HIV-1 Protease Inhibitors

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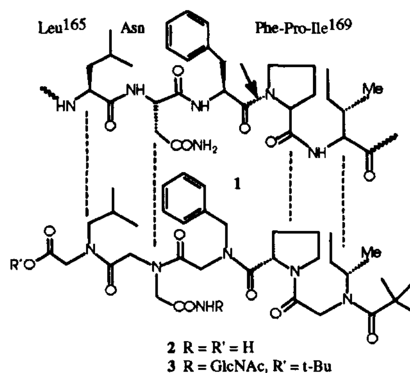
Abstract: Using a blockwise approach, N-substituted oligoglycine (NSGs) peptoids bearing N-acetylglucosamine residues in different position of the side chain were efficiently synthesized by a reiterative strategy involving mono N-alkylation and bromoacetylation. © 1997 Elsevier Science Ltd.

Inhibition of HIV-1 (human immunodeficiency virus type 1) aspartyl protease (PR) is an active area of current therapeutic investigations.[1] This enzyme cleaves the viral *gal-pol* polyprotein and is therefore essential for the maturation of fully infectious virions. A large number of more or less classical peptidomimetics based on amino acid sequences derived from physiological substrates have been shown to be potent protease inhibitors and few candidates are in clinical trials.[2] As an extension to these analogs, the syntheses of an interesting family of novel peptidomimetics based on N-substituted oligoglycines (peptoids) readily amenable to combinatorial library synthesis sound appealing.[3] Some derivatives of this class of peptide analogs have been shown to have nanomolar activity against opioid receptors.[4]

This note describes the synthesis of peptoid and glycosylated peptoid analogs derived from one of the physiological HIV-1 PR substrate sequence Leu^{165}-Ile^{169}. Few of the structural similarities between the natural substrate and the peptoid analogs are illustrated in Scheme 1. The glycosylated version of the peptoids has been further proposed on the basis that several glycosylated peptide drugs were shown to have higher bioavailability, increased absorption, and attenuated *in vivo* clearance.[5]

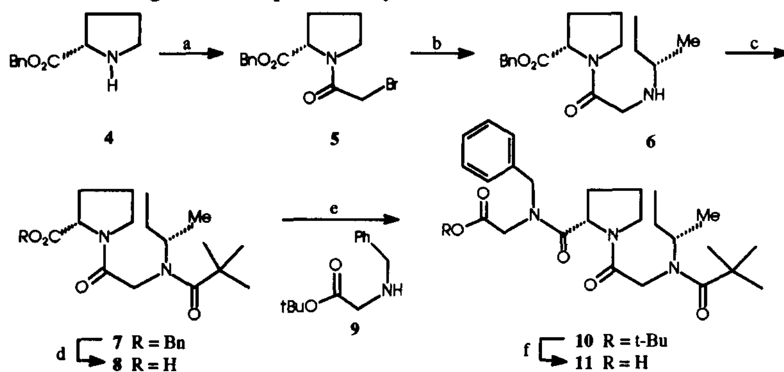


Scheme 1. Schematic representation illustrating structural similarities between the amino acid sequence of an HIV-1 protease substrate for (1) and its peptoid (2) and glycosylated peptoid (3) mimetics.

The natural peptide substrate chosen as lead has been selected from the *gal-pol* polyprotein sequence Leu^{165}-Asn-Phe-Pro-Ile^{169} in which the Phe-Pro constituted the scissile linkage.[1] In this communication, we wish to report the synthesis and biological data on analogous peptidomimetic versions derived from the corresponding N-substituted oligoglycine (NSGs) peptoids. Because of the numerous reports relating the pharmacological advantages conferred to natural peptide drugs onto which carbohydrates have been

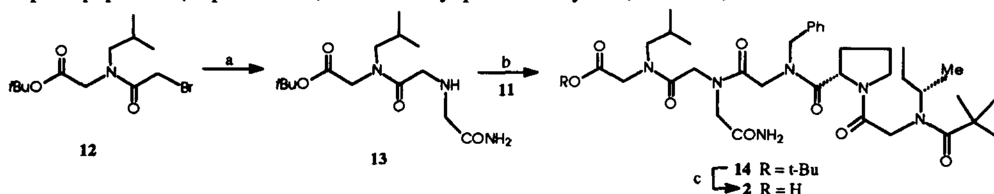
covalently linked,[5] the target peptoids have also been synthesized taking this extra advantage into consideration. Using the peptoid terminology defined by Simon *et al.*,[3a] our target sequences could be designed as the non-glycosylated NLeu-NAsn-NPhe-NPro-NIle (**2**); the N-"linked" NLeu-NAsn(GlcNAc)-NPhe-NPro-NIle (**3**), and NAsn(GlcNAc)-NLeu-NAsn-NPhe-NPro-NIle (**23**).

The synthesis was based on a blockwise approach using strategies previously outlined for N- and O-linked glycosylated peptoids from this group.[6,7] Thus, L-proline benzyl ester (**4**) was initially bromoacetylated ($BrCH_2COCl$, Py, CH_2Cl_2) to give **5** in 94% yield.[8] As previously observed with N-substituted oligoglycines bearing secondary amide linkages,[6] the ^{1}H-NMR spectra of **5** indicated the presence of two amide rotamers in a 1:1 ratio. Compound **5** was then treated with (S)-*sec*-butylamine in the presence of diisopropylethylamine (DIPEA, 0°C) to give the monoalkylated amine **6** in 68% yield. The new amide rotamer ratio (1:3.7) of **6** was determined from the integration of the methyl signals (δ 0.76 and 0.82 ppm, 2t, J = 7.4 Hz). Compound **6** was acylated with pivaloyl chloride [$(CH_3)_3COCl$, Py, CH_2Cl_2] to provide **7** in 93% yield. Removal of the benzyl group under hydrogenolysis over 10% Pd-C in methanol gave acid **8** in quantitative yield (Scheme 2). Coupling acid **8** with the known secondary amine **9**[6a] in the presence of dicyclohexylcarbodiimide (DCC, HOBt, CH_2Cl_2) afforded **10** in 87% yield. Saponification of the *tert*-butyl ester with 0.1 M KOH in EtOH provided the desired acid fragment **11** in quantitative yield.

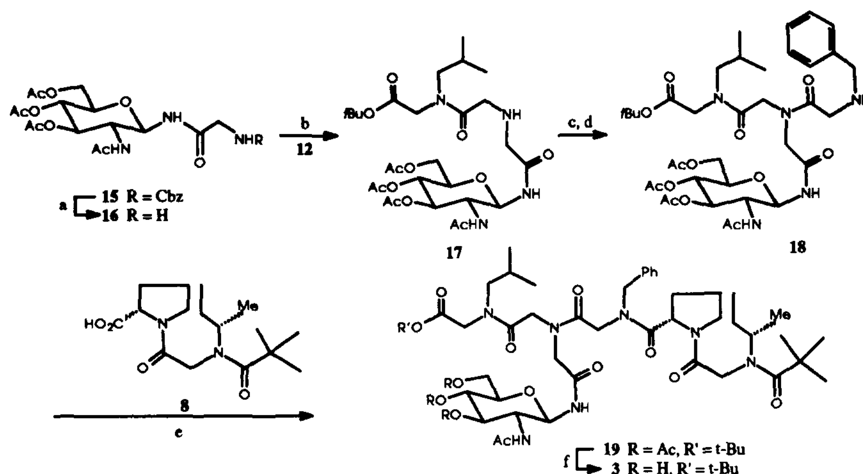


Scheme 2. a) $(BrCH_2CO)_2O$, C_5H_5N, CH_2Cl_2, 0°C, 1 h, 94%; b) (S)-*sec*-butylamine, DIPEA, CH_3CN, 0°C, 30 min, 68%; c) Me_3CCOCl, DIPEA, CH_2Cl_2, 0°C, 1 h, 93%; d) H_2, 10% Pd-C, MeOH, rt, 1 h, quant; e) DCC, HOBT, CH_2Cl_2, rt, 1 h, 87%; f) 0.1 M KOH, EtOH, rt, 12 h, then H^+ resin, quant.

In order to complete the synthesis of the non-glycosylated pentapeptoid **2** from tripeptoid acid **11**, the required dipeptoid block **13** was prepared in 86% yield along with 10% N,N-dialkylated product by N-alkylating glycinamide hydrochloride with known fragment **12**[6b] (DIPEA, CH_3CN/H_2O, 1:1, 0°C). The ^{1}H NMR spectra of **13** indicated the two amide rotamers to be present in a 2:1 ratio (δ 0.84 and 0.89 ppm, 2d, J = 6.6 Hz, $CHMe_2$; 1.41 and 1.42 , 2s, 9H, CO_2t-Bu). The final amide bond formation between amine **13** and acid **11** (DCC, HOBt, CH_2Cl_2, rt, 3 h) provided **14** in 84% yield (mp 90 - 92°C, α_D 4.2°, c 1.09, $CHCl_3$). Hydrolysis from **14** with 0.1 M KOH in EtOH at room temp followed by neutralization with IR-120 (H^+) resin gave pentapeptoid **2** (m.p 95-97 °C) in essentially quantitative yield (Scheme 3).

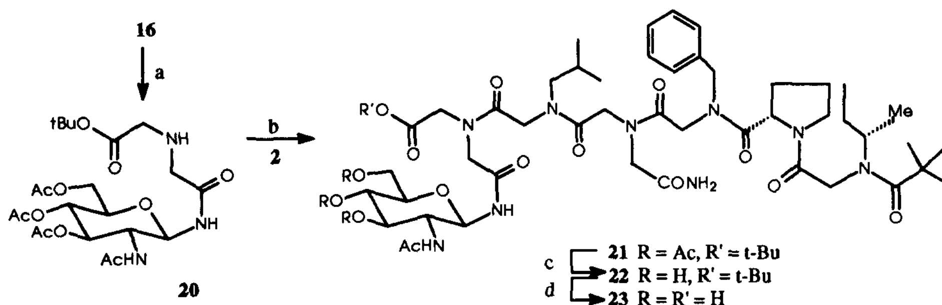


Scheme 3. a) Glycine amide HCl, DIPEA, CH_3CN/H_2O (1:1), 86%; b) DCC, HOBT, rt, 3 h, 84%; c) 0.1 M KOH, EtOH, 12 h, rt, then H^+ resin.



Scheme 4. a) H_2, 10% Pd-C, MeOH, 1 h, quant; b) DIPEA, CH_3CN, 0°C, 1 h, 70%; c) $(BrCH_2CO)_2O$, DIPEA, CH_2Cl_2, 0°C, 30 min, 90%; d) $PhCH_2NH_2$, DIPEA, CH_3CN, 0°C to rt, 1 h, 76%; e) DCC, HOBT, CH_2Cl_2, rt, 3 h, 81%; f) NaOMe, MeOH, pH = 8.5, 1 h then H^+ resin, quant.

The N-"asparagine"-linked version of the above pentapeptoid **2**, carrying an N-acetylglucosamine (GlcNAc) residue in the middle of the chain was synthesized starting from the known glycopeptide **15**.[6a] Hydrogenolysis of the Cbz-group of **15** (H_2, 10% Pd-C, MeOH, 1 h) gave **16** quantitatively (Scheme 4). N-Alkylation of **16** with **12** gave **17** (70%) which was then successively treated with bromocetic anhydride (DIPEA, CH_2Cl_2, 0°C , 30 min, 90%) and benzylamine (DIPEA, CH_3CN, 0°C to rt, 1 h) to afford secondary amine **18** in 76% yield. Amine **18** was then treated with acid **8** (DCC, HOBT, CH_2Cl_2) as above to give glycopeptoid **19** in 81% yield (mp 120-122°C; α_D 8.9°, c 1.0, $CHCl_3$). Zemplén deacetylation of **19** (NaOMe, MeOH, pH = 8.5, 1 h) gave **3** in 94% yield (mp 134-135°C).



Scheme 5. a) $BrCH_2CO_2t$-Bu, DIPEA, CH_3CN, 0°C, 1 h, 79%; b) DCC, HOBT, CH_2Cl_2, rt, 3 h, 84%; c) NaOMe, MeOH, pH = 8.5, 1 h, H^+ resin, quant; d) 0.1 M KOH, EtOH, 12 h, rt then H^+ resin, quant.

Since the N-acetylglucosamine moiety was near the susceptible hydrolytic cleavage site (Scheme 1), we also planned to install it at a more remote position to avoid possible hindrance factor. Therefore, mono N-alkylation of amine **16** with *tert*-butyl bromoacetate (DIPEA, CH_3CN, 0°C, 1 h) gave **20** in 79% yield (Scheme 5). Coupling of secondary amine **20** with acid **2** (DCC, HOBT, CH_2Cl_2, rt, 1 h) afforded glycosylated peptoid **21** in 84% yield after silica gel column chromatography (mp 145-147 °C, α_D 4.83°, c 1.07, $CHCl_3$). Compound **21** was treated under Zemplén conditions (NaOMe, MeOH, pH 8.5 rt, 1 h) to

provide **22** quantitatively (mp 128-131 °C). The ^{1}H-NMR spectra of **22** showed the two rotameric pivaloyl amide signals at δ 1.34 and 1.36 ppm in a 1:1 ratio while the characteristic singlets attributed to the *tert*-butyl ester in the vicinity of the secondary amide bond appeared at δ 1.45 and 1.46 ppm also as a 1:1 ratio. Finally, complete deprotection of **22** with 0.1 M KOH in EtOH (12 h, rt) followed by neutralization with H^+ resin gave fully deprotected glycohexapeptoid **23** in quantitative yield (mp 138-140°C). The analogs showed only low inhibitory activity in standard HIV-1 protease assays (**3** showed ~10% inhibition at 250μM).

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References and notes

1. (a) Yarchoan, R.; Mitsuya, H.; Broder, S. *TiPS* **1993**, *14*, 196; (b) Huff, J. R. *J. Med. Chem.* **1991**, *34*, 2305.
2. (a) Tomasselli, A. G.; Howe, W. J.; Sawyer, T. K.; Wlodawer, A.; Heinrikson, R. L. *Chim. Oggi* **1991**, *9*, 6; (b) Wlodawer A.; Erickson, J. W. *Ann. Rev. Biochem.* **1993**, *62*, 543; (c) De Clercq, E. *J. Med. Chem.* **1995**, *38*, 2491.
3. (a) Simon, R.J.; Kania, R.S.; Zuckermann, R.N.; Huebner, V.D.; Jewell, D.A.; Banville, S.; Ng, S.; Wang, L.; Rosenberg, S.; Marlowe, C.K.; Spellmeyer, D.C.; Tan, R.; Frankel, A.D.; Santi, D.V.; Cohen, F.E.; Bartlett, P.A. *Proc. Natl. Acad. Sci. USA* **1992**, *89*, 9367-9371; (b) Zuckermann, R.N.; Kerr, J.M.; Kent, S.B.H.; Moos, W.H. *J. Am. Chem. Soc.* **1992**, *114*, 10646-10647.
4. Zuckermann, R. N.; Martin, E. J.; Spellmeyer, D. C.; Stauber, G. B.; Shoemaker, K. R.; Kerr, J. M.; Figliozzi, G. M.; Goff, D. A.; Siani, M. A.; Simon, R. J.; Banville, S. C.; Brown, E. G.; Wang, L.; Richter, L. S.; Moos, W. H. *J. Med. Chem.* **1994**, *37*, 2678.
5. (a) Polt, R.; Porecca, F.; Szabò, L.; Hruby, V. J. *Glycoconj. J.* **1993**, *10*, 261; (b) Fisher, J. F.; Harrison, A. W.; Bundy, G. L.; Wilkinson, K. F.; Rush, B. D.; Ruwart, M. J. *J. Med. Chem.* **1991**, *34*, 3140; (c) Varga-Defterdarovic, L.; Horvat, S.; Chung, N. N.; Schiller, P. W. *Int. J. Peptide Protein Res.* **1992**, *39*, 12; (d) Kihlberg, J.; Åhman, J.; Walse, B.; Drakenberg, T.; Nilsson, A.; Söderberg-Ahlm, C.; Bengtsson, B.; Olsson, H. *J. Med. Chem.* **1995**, *38*, 161.
6. (a) Saha, U.K.; Roy, R. *Tetrahedron Lett.* **1995**, *36*, 3635-3638; (b) Saha, U.K.; Roy, R. *J. Chem. Soc., Chem. Commun.* **1995**, 2571-2573; (c) Saha, U.K.; Roy, R. *Chem. Commun.* **1996**, 210-202.
7. (a) Kim, J.M.; Roy, R. *Carbohydr. Lett.* **1996**, *1*, 465-468; (b) Kim, J.M.; Roy, R. *Carbohydr. Res.* **1997**, *298*, 173; (c) Kim, J.M.; Roy, R. *Tetrahedron Lett.* **1997**, *38*, 3487-3490.
8. All new compounds showed satisfactory spectral and elemental/mass analysis. Selected spectroscopic data are as follow: **7** (rotamers) : ^{1}H NMR ($CDCl_3$), δ (ppm): 0.81 (m, 3H), 1.07, 1.09 (2d, 3H, J=6.4 Hz, CH_3), 1.16, 1.20 (2s, 9H, CMe_3), 1.45 (m, 2H, CH_2), 1.7-2.2 (m, 4H, 2 x CH_2), 3.6 (m, 3H), 4.1 (m, 2H), 4.6 (m, 1H), 4.9, 5.1 (ABq, CH_2Ph), FAB-MS (m/z): 403 (M^+ + 1, 15.3%); **13** (rotamer 1:2) ^{1}H NMR ($CDCl_3$): 0.84, 0.89 (2d, 6H, J=6.6 Hz, CH*Me*$_2$), 1.41, 1.42 (2s, 9H, CMe_3), 1.70-1.90 (m, 1H, *CH*Me_2), 2.41 (s, 1H, NH), 2.98, 3.17 (2d, 2H, J=7.5 Hz, N*CH*$_2$CMe_2), 3.25, 3.26 (2s, 2H), 3.31, 3.46 (2s, 2H), 3.79, 3.91 (2s, 2H, N*CH*$_2$COO*t*-Bu), 5.90, 7.20 (2bs, 2H, $CONH_2$); CI-MS (m/z): 302 (M^+ + 1, 84%); **14** ^{1}H NMR ($CDCl_3$): 0.8-1.0 (m, 9H, 3 x CH_3), 1.1-1.35 (m, 12H, CH_3 and $COCMe_3$), 1.37-1.6 (m, 11H, COO*t*Bu and CH_2), 1.7-2.4 (m, 5H), 3.0-3.2 (m, 4H, 2 x CH_2), 3.60-5.4 (m, 14H), 7.15-7.4 (m, 5H); HRMS-FAB: calcd for $C_{39}H_{62}N_6O_8$ is 742.4629, found 743.4707 (M + 1); **2** m.p 95-97°C, FAB-MS (m/z): 819 (M^+ + Cs, 1.0%); **18** ^{1}H NMR ($CDCl_3$): 0.8 (m, 6H, CH$\underline{Me}_2$), 1.35 (bs, 9H, CMe_3), 1.72-1.93 (6s, 4 x Ac), 2.35 (bs, 1H, NH), 2.85-3.3 (m, 4H), 3.5-4.3 (m, 12H), 4,85-5.2(m, 2H, H-3, H-4), 5.35 (t, 1H, J=9.7 Hz), 6.1, 6.25, 6.55 (3d, 1H, J=9.1 Hz), 8.35, 8.5, 9.2 (3d, 1H, J=8.7, NH); HRMS-FAB calcd for $C_{53}H_{81}N_7O_{16}$ 1071.5739, found 1071.5682; **3** FAB-MS (m/z): 1079 (M^+ + Cs, 1.2%), 947 (M^+ + 1, 30%); **20** ^{1}H NMR ($CDCl_3$): 1.42 (s, 9H, CMe_3), 1.88 (s, 3H, NAc), 2.0, 2.02, 2.05 (3s, 9H, 3 x OAc), 3.1-3.3 (m, 4H, 2xCH_2), 3.7-3.8 (m, 1H), 4.0-4.3 (m, 3H), 5.0-5.15 (m, 3H), 6.07 (d, 1H, J=8.7 Hz), 8.13 (d, 1H, J=9.4 Hz); CI-MS (m/z): 518 (M^+, 50%); **21** ^{1}H NMR ($CDCl_3$): 0.8-1.05 (m, 9H, 3xCH_3), 1.1-1.35 (m, 12H, $COCMe_3$, CH_3), 1.4-1.6 (m 11H, CH_2, COO^{t}Bu), 1.8-2.4 (m, 17H, 4 x Ac, 2 x CH_2, $\underline{CH}Me_2$), 3.0-3.3 (m, 2H), 3.6-4.4 (m, 25H), 4.9-5.2 (m, 3H), 7.1-7.4 (m, 5H); HRMS-FAB calcd mass for $C_{57}H_{87}N_9O_{18}$ 1185.6169, found 1186.6247 (M + 1); **23** ^{1}H NMR (D_2O): 0.8-1.02 (m, 9H, 3xCH_3), 1.05-1.4 (m, 14H), 1.57-1.65 (m, 2H), 1.80-2.15 (m, 7H), 3.03-3.25 (m, 2H), 3.5- 3.95 (m, 9H), 4.0-4.65 (m, 11H), 4.95-5.2 (m, 2H), 7.25-7.5 (m, 5H); FAB - MS (m/z): 1136.3 (M^+ + Cs, 1.5%), 1004.4 (M^+, 8.9%).

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