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Synthesis and Conformation of Gly-Gly Dipeptides Constrained with Phenylalanine-like Aminocaproic Acid Linkers

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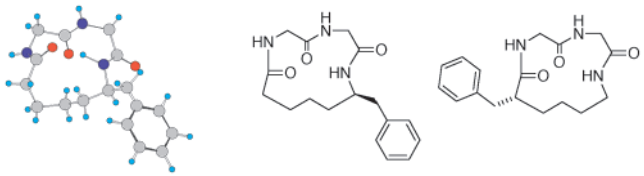
ABSTRACT



The constraint of dipeptides with linkers derived from 6-aminocaproic acid (Aca) is a useful means of constructing a β-turn peptidomimetic. The extension of this concept to the mimicry of a tripeptide entails the incorporation of a side chain moiety on either end of the Aca chain. The synthesis and conformational analysis of two exemplary compounds is discussed.

The utility of β-turn peptidomimetics as pharmacological probes and possible drug candidates has inspired a number of approaches for their design.[1] Among these, macrocyclization with a linking group has proved a fruitful approach for the synthesis of β-turn mimics, often with a good degree of conformational restriction.[2] Macrocycles composed of aminocaproic acid (Aca) and a dipeptide are known to adopt a β-turn conformation in which the amino acid residues occupy positions corresponding to the $i + 1$ and $i + 2$ positions of a natural β-turn (Figure 1).[3] Work in this laboratory has focused on the effect of Aca substituent stereochemistry on the conformation of similar macrocycles.[4] In that work, we established linker stereochemistry to have a significant effect on the type of β-turn adopted in a series of macrocycles based on the Ala-Gly motif. Despite the appealing simplicity and

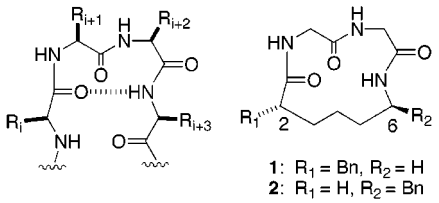


Figure 1. A generalized β-turn (left) and an Aca-constrained β-turn peptidomimetic containing Gly-Gly in the $i + 1$ and $i + 2$ positions.

(1) Reviews: (a) Farmer, P. S. In *Drug Design*; Ariëns, E. J., Ed.; Academic: New York, 1980; Vol. 10, pp 119−143. (b) Giannis, A.; Kolter, T. *Angew. Chem., Int. Ed. Engl.* **1993**, *32*, 1244−1267. (c) Olson, G. L.; Bolin, D. R.; Bonner, M. P.; Bös, M.; Cook, C. M.; Fry, D. C.; Graves, B. J.; Hatada, M.; Hill, D. E.; Kahn, M.; Madison, V. S.; Rusiecki, V. K.; Sarabu, R.; Sepinwall, J.; Vincent, G. P.; Voss, M. E. *J. Med. Chem.* **1993**, *36*, 3039−3049. (d) Gante, J. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 1699−1720. (e) Aubé, J. In *Advances in Amino Acid Mimetics and Peptidomimetics*; Abell, A., Ed.; JAI Press: Greenwich, 1997; Vol. 1, pp 193−232.

(2) For some recent examples, see: (a) Bach, A. C., II; Eyermann, C. J.; Gross, J. D.; Bower, M. J.; Harlow, R.; Weber, P. C.; DeGrado, W. F. *J. Am. Chem. Soc.* **1994**, *116*, 3207−3219. (b) Shao, H.; Wang, S. H. H.; Lee, C.-W.; Ösapay, G.; Goodman, M. *J. Org. Chem.* **1995**, *60*, 2956−2957. (c) Miller, S. J.; Grubbs, R. H. *J. Am. Chem. Soc.* **1995**, *117*, 5855−5856. (d) Shao, H.; Lee, C.-W.; Zhu, Q.; Gantzel, P.; Goodman, M. *Angew. Chem., Int. Ed. Engl.* **1996**, *35*, 90−92. (e) Bach, A. C., II.; Espina, J. R.; Jackson, S. A.; Stouten, P. F. W.; Duke, J. L.; Mousa, S. A.; DeGrado, W. F. *J. Am. Chem. Soc.* **1996**, *118*, 293−294. (f) Feng, Y.; Wang, Z.; Jin, S.; Burgess, K. *J. Am. Chem. Soc.* **1998**, *120*, 10768−10769. (g) Feng, Y.; Pattarawarapan, M.; Wang, Z.; Burgess, K. *Org. Lett.* **1999**, *1*, 121−124.

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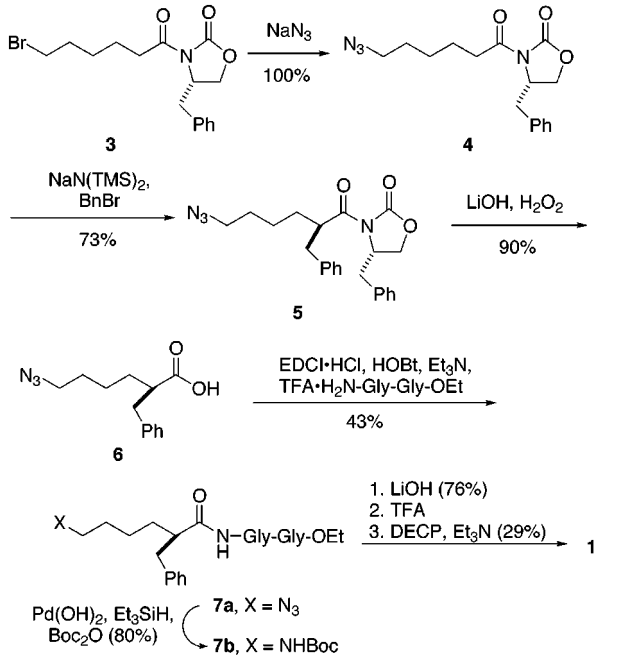
generality of this approach, it has been underutilized as a strategy in peptidomimetic design.

Often, the biological activity of a peptide is dependent on three consecutive amino acid residues, such as the well-known RGD triad.[5] Utilization of an Aca-linking strategy for the mimicry of such compounds would require the tether to mimic a third amino acid residue by incorporating an amino acid side-chain equivalent onto itself. Two obvious positions for side-chain substitution are at C-2 and C-6 of the Aca chain, as pictured in Figure 1. In this Letter, we describe the synthesis of two Gly-Gly mimics containing a monosubstituted Aca linker representing an additional phenylalanine moiety (**1** and **2**, Figure 1). The choice of Gly-Gly as the central dipeptide was made to permit examination of the conformational tendencies of the linkers in the absence of a conformationally demanding dipeptide unit. In earlier work,[4] it had been established that a 3-methyl-substituted Aca chain occupied type II β-turn space to a surprising degree as determined by CD spectroscopy. However, the position of the methyl group in that compound did not correspond to that of a side chain in a naturally occurring β-turn.

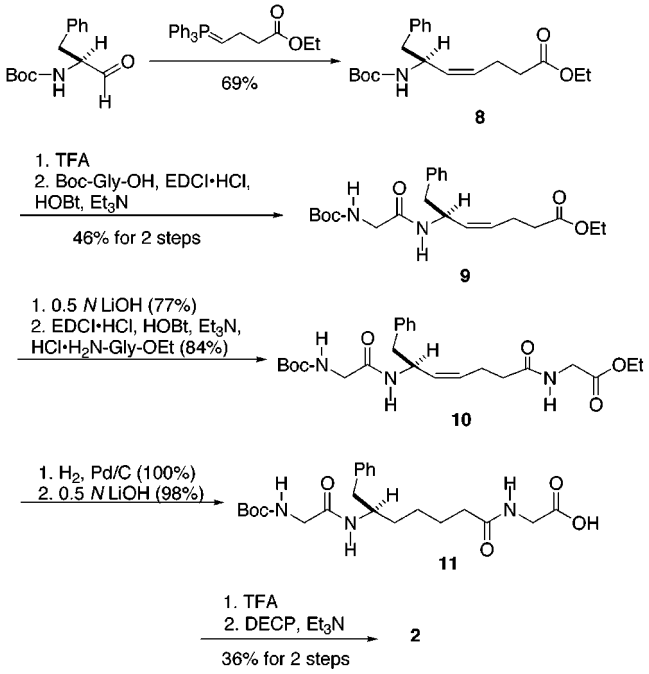
We envisioned using methods that would be widely applicable to a variety of proteinogenic side-chain types with modest modifications. While the final stages in the syntheses followed standard peptide-coupling protocols, the synthesis of the 2-benzyl-substituted linker was accomplished using an oxazolidinone-mediated asymmetric alkylation reaction as the key step (Scheme 1).[6] Thus, 6-bromohexanoyl chloride was coupled to the standard Evans chiral auxiliary to form **3**. After displacement of the bromide with NaN_3, deprotonation and alkylation with benzyl bromide afforded **5** (73%, ≥98% de). Following the removal of the chiral auxiliary,[7] the azido acid was coupled to the *N*-terminus of glycylglycine ethyl ester to afford a seco-tripeptide. The azide moiety was then directly reduced[8] to afford Boc-protected amine **7b**. This step proved convenient inasmuch as attempted direct reductions to the unprotected amine proved problematic, possibly due to catalyst poisoning by the product. Ester hydrolysis, Boc deprotection, and cyclization with diethyl cyanophosphonate (DECP) resulted in the formation of **1**. No racemization was evident by chiral HPLC examination of azide **7a** (Chiralcel OD-H column, 7% EtOH/hexane) using a racemic mixture as the standard.

Scheme 1



The synthesis of the Aca tether bearing a C-6 benzyl substituent utilized a chiral pool approach (Scheme 2). Elongation of the carbon chain of Boc-L-phenylalanal[9] was effected by Wittig reaction with [3-(ethoxycarbonyl)propyl]-

Scheme 2



(3) (a) Deslauriers, R.; Leach, S. J.; Maxfield, F. R.; Minasian, E.; McQuie, J. R.; Meinwald, Y. C.; Némethy, G.; Pottle, M. S.; Rae, I. D.; Scheraga, H. A.; Stimson, E. R.; Van Nispen, J. W. *Proc. Natl. Acad. Sci. U.S.A.* **1979**, *76*, 2512−2514. (b) Némethy, G.; McQuie, J. R.; Pottle, M. S.; Scheraga, H. A. *Macromolecules* **1981**, *14*, 975−985. (c) Deslauriers, R.; Evans, D. J.; Leach, S. J.; Meinwald, Y. C.; Minasian, E.; Némethy, G.; Rae, I. D.; Scheraga, H. A.; Somorjai, R. L.; Stimson, E. R.; Van Nispen, J. W.; Woody, R. W. *Macromolecules* **1981**, *14*, 985−996. (d) Maxfield, F. R.; Bandekar, J.; Krimm, S.; Evans, D. J.; Leach, S. J.; Némethy, G.; Scheraga, H. A. *Macromolecules* **1981**, *14*, 997−1003. (e) Bandekar, J.; Evans, D. J.; Krimm, S.; Leach, S. J.; Lee, S.; McQuie, J. R.; Minasian, E.; Némethy, G.; Pottle, M. S.; Scheraga, H. A.; Stimson, E. R.; Woody, R. W. *Int. J. Pept. Protein Res.* **1982**, *19*, 187−205. (f) Perczel, A.; Fasman, G. D. *Protein Sci.* **1992**, *1*, 378−395. (g) Perczel, A.; Hollósi, M.; Sándor, P.; Fasman, G. D. *Int. J. Pept. Protein Res.* **1993**, *41*, 223−236.

(4) Kitagawa, O.; Vander Velde, D.; Dutta, D.; Morton, M.; Takusagawa, F.; Aubé, J. *J. Am. Chem. Soc.* **1995**, *117*, 5169−5178.

(5) (a) Ojima, I.; Chakravarty, S.; Dong, Q. *Bioorg. Med. Chem.* **1995**, *3*, 337−360. (b) Samanen, J. *Ann. Rep. Med. Chem.* **1996**, *31*, 91−99.

(6) Evans, D. A. In *Asymmetric Synthesis*; Morrison, J. D., Ed.; Academic: Orlando, 1984; Vol. 3, pp 2−110.

(7) Li, Z.-H.; Bulychev, A.; Kotra, L. P.; Massiva, I.; Mobashery, S. *J. Am. Chem. Soc.* **1998**, *120*, 13003−13007.

(8) Kotsuke, H.; Ohishi, T.; Araki, T. *Tetrahedron Lett.* **1997**, *38*, 2129−2132.

triphenylphosphonium bromide[10] to give the product olefin (**8**). The six-carbon tether was incorporated into tripeptide **11** and cyclized using the standard peptide coupling techniques shown. In this example, a modest amount of racemization was encountered en route to the final targets, presumably taking place during the olefination step (Chiralcel OD-H (4% EtOH/hexane) analysis of the hydrogenated form of compound **9** indicated that this material was obtained in ca. 93% ee).

Circular dichroism (CD) spectroscopy measurements of macrocycles **1** and **2** were carried out in methanol at concentrations previously established to minimize aggregation (c = 1 mg/mL, 0.02 cm path length).[4] The CD spectrum of compound **1** resembles the standard CD curve for a βII turn as established for an Aca-containing model system (Figure 2).[3c,g] Conversely, compound **2** shows a significant

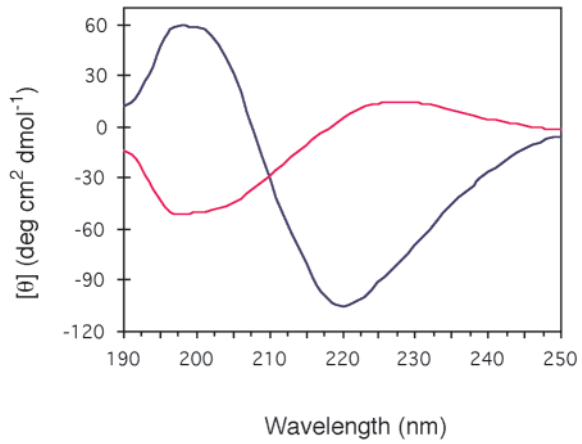


Figure 2. CD spectra of compounds **1** (blue) and **2** (red).

population of the backbone mirror-image βII′ turn as established by CD. For both **1** and **2**, NMR ROESY experiments supported our assignments based on CD spectroscopy (however, note that NMR does not readily distinguish between the enantiomeric backbones of βII and βII′ turns). Thus, both spectra showed substantial cross-peaks between one of the Gly_1 α-protons and the Gly_2 amide, as well as between the Gly_2(NH) and the Aca(NH) signals, as expected for type II or II′ β-turns. In addition, there was no evidence of an Gly_1(NH) and Gly_2(NH) interaction that would betray the presence of a type I or I′ β-turn. Some averaging of diastereotopic protons in both cases suggests that these compounds experience a degree of conformational mixing. Still, it is evident that placing a substituent on the Aca linker does not interfere with the intrinsic preference of this system for a βII/II′ conformation.

An X-ray crystallographic structure obtained from compound **2** confirmed the solution results (Figure 3; a crystal

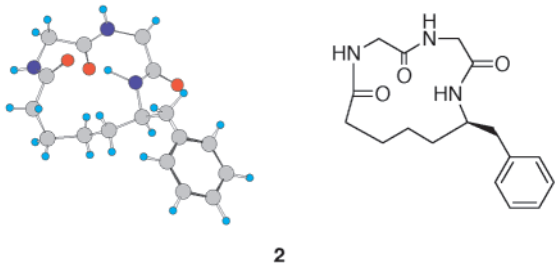


Figure 3. Ball-and-stick depiction of the X-ray crystallographic structure of compound **2**.

in the racemic space group $P2_1/a$ was crystallized from $MeOH/CH_2Cl_2$). Indeed, the observed dihedral angles are all within 26° of idealized type II′ values.[11] Interestingly, this structure also contains the "classical" β-turn H-bond between the Aca(C═O) and the Aca(NH) (O → H distance 1.892 Å). Examination of this structure shows that the benzyl substituent occupies a pseudoequatorial position in this conformation, presumably to avoid steric interactions with the remainder of the cyclic molecule.

In summary, two examples of phenylalanine-like Aca linkers have been prepared and incorporated into a cyclic peptide. Peptidomimetics containing substitution at positions 2 and 6 on the tether adopt a type II or II′ β-turn surrounding the central Gly-Gly dipeptide, respectively. We are currently extending these findings into the realm of pharmacologically relevant peptide mimicry.

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(9) Fehrentz, J.-A.; Castro, B. *Synthesis* **1983**, 676−678.
(10) Bestman, H. J.; Koschatzky, K. H.; Schätzke, W.; Süss, J.; Vostrowsky, O. *Liebigs. Ann. Chem.* **1981**, 1705−1720.
(11) Venkatachalam, C. M.; Ramachandran, G. M. *Annu. Rev. Biochem.* **1969**, *38*, 45−82.