Synthesis of 4,4-Disubstituted 2-Aminocyclopentanecarboxylic Acid Derivatives and Their Incorporation into 12-Helical \(\beta\)-Peptides

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Received August 20, 2004

ABSTRACT

An enantioselective synthetic route is reported for trans-2-aminocyclopentanecarboxylic acids (ACPC) bearing geminal side chain pairs at the 4-position. \(\beta\)-Peptides containing the 4,4-disubstituted ACPC residues adopt the 12-helical conformation, as demonstrated by 2D NMR analysis in aqueous solution. These 4,4-disubstituted ACPC residues display functional groups, including acidic and hydrogen bond donating groups, in a geometrically defined fashion, which should be useful for the design of \(\beta\)-peptides for specific applications.

Unnatural oligomers with discrete folding propensities (“foldamers”) have received much attention in recent years.\(^1\)\(^2\) Oligomers consisting of \(\beta\)-amino acids constrained with five-membered rings (Figure 1), such as trans-2-aminocyclopentane-1,2-dicarboxylic acid (ACPC) and trans-3-aminopyrrolidin-4-carboxylic acid (APC), have been shown to adopt a helical conformation defined by 12-membered-ring hydrogen bonds, \(\text{C}^=\text{O}(i) \rightarrow \text{N}^H(i + 3)\) (“12-helix”).\(^3\) A 12-helical structure can be observed with as few as six residues in aqueous solution.\(^4\) Properly designed 12-helical \(\beta\)-peptides mimic the antimicrobial activity of natural host-defense peptides.\(^5\) Biological applications of non-12-helical \(\beta\)-peptides have also been reported.\(^6\)

The synthesis of a diverse set of \(\beta\)-amino acid residues is vital for developing \(\beta\)-peptides with useful functions.\(^7\) Previously we showed that side chains can be introduced into 12-helical \(\beta\)-peptides by sulfonylation of the ring nitrogen of APC\(^8\) or by introduction of a few acyclic \(\beta\)-amino acids among ACPC and/or APC residues.\(^9\) Diversity can be achieved also by introducing side chains on the cyclopentane ring. Recently, we reported the synthesis of ACPC residues

\[\text{ACPC} \quad \text{APC} \quad \begin{array}{c} \text{R = OMe} \\ \text{R = CH}_3\text{NHBOc} \\ \text{R = }\end{array} \]

\[\begin{array}{c} \text{R = Me} \\ \text{R = Ph} \\ \text{R = CH}_3\text{O}^\text{Bu} \\ \text{R = CO}^\text{Bu} \end{array}\]

Figure 1. \(\beta\)-Amino acids with five-membered ring constraint.
went oxidative C–C bond cleavage in aqueous KMnO₄ to furnish the desired diacid 4a.¹¹ The more hydrophobic 4,4-diphenylcyclohexanone could not be cleaved using aqueous conditions. Instead, the ketone was converted to TMS enol ether 5,¹² which was oxidized to diacid 4b in organic solvents.¹³

![Scheme 1](image link)

**Scheme 1**

**Scheme 2**

Precursors for monomers 1c and 1d were prepared by the oxidation of disubstituted cyclohexenes (Scheme 2). Disubstituted cyclohexenes 6 and 7 were prepared from the Diels–Alder reaction between butadiene and methylidene malonate generated in situ from the corresponding malonate ester and formaldehyde.¹⁴ Cyclohexene 6 was reduced to diol 8 and protected as the bis-tert-butyl ether 9. Disubstituted cyclohexenes 7 and 9 were oxidized to the corresponding diacids 4c and 4d in aqueous KMnO₄.

Diacids 4a–d were converted to enantiopure aminooxysters 12a–d and 13a–d through an auxiliary-based synthesis (Scheme 3). The diacids 4a–d were first converted to the corresponding diesters 10a–d, which were cyclized to the β-keto esters 11a–d. The 4,4-disubstituted regioisomeric product was predominant in each case, presumably because of steric effects. The β-keto esters were converted to diastereomeric trans-amino esters 12a–d and 13a–d, in one-pot operations, by enamine formation with α-methylbenzylamine and subsequent reduction using NaBH₃CN.¹⁵ In each case the reduction was highly trans-selective, albeit with low selectivity between the two trans diastereomers. These isomers could be readily separated from each other and from the minor cis isomers by column chromatography, allowing access to both enantiomers of the β-amino acids.¹⁶ The stereochemistry of the diastereomeric β-amino esters was assigned by X-ray crystallography of salts derived from 12a–c.¹⁷ Saponification of the ester, hydrolysis of the auxiliary, and Fmoc protection yielded the protected β-amino acids 14a–d and ent-14a–d.¹⁹

To determine whether 4,4-disubstituted ACPC residues are compatible with the 12-helix, we prepared β-peptides 15 and...


Hexamers 15 and 16 were both suitable for two-dimensional NMR analysis (Figure 2). The proton resonances of these β-peptides were well resolved, both in CD$_3$OD and in aqueous solution. NOEs are indicated as ambiguous where resonance overlap precludes a definitive assignment (Figure 2).

Hexamer 15 displayed NOEs in methanol consistent with the 12-helical conformation. Three unambiguous C$_3$H(i) to C$_a$(i + 2) NOEs were assigned (of four possible NOEs of this type); four unambiguous C$_4$H(i) to NH(i + 2) NOEs were assigned (of five possible NOEs), and two unambiguous C$_3$H(i) to NH(i + 3) NOEs were assigned (of four possible NOEs). These observations are fully consistent with a 12-helical structure. Hexamer 15 displayed a similar set of NOEs in water. Two of four C$_3$H(i) to C$_a$(i + 2) NOEs, four of five C$_3$H(i) to NH(i + 2) NOEs, and two of four C$_3$H(i) to NH(i + 3) NOEs were unambiguously assigned.

Interestingly, a previously unobserved type of NOE was observed for hexamer 15 in water. Two NOEs from C$_3$H(i) to C$_a$(i + 3) were identified. Crystal structure data for 12-helical ACPC oligomers suggest that these two protons would be too far apart for an NOE between them to be observed. This contradiction suggests that the “12-helix” may be a family of related conformers, perhaps in rapid exchange with one another, rather than a single, rigid conformation. The introduction of side chains at particular positions on the five-membered rings of individual residues may perturb the 12-helical conformational manifold in such a way that new NOEs are detected.

Hexamer 16 also displayed NOEs consistent with 12-helical structure in both methanol and water. In methanol, one of four C$_3$H(i) to C$_a$(i + 2) NOEs, four of five C$_3$H(i) to NH(i + 3) NOEs, and four of five C$_3$H(i) to NH(i + 4) NOEs were unambiguously assigned.

(19) ent-$14c$ could be incorporated into β-peptides and deprotected with no observed decarboxylation of the β-dicarboxylic acids; see Supporting Information.

(20) For CD characterization of these and other β-peptides containing 4,4-disubstituted ACPC residues, see Supporting Information.


Reagents and conditions: (a) MeOH, benzene, H$_2$SO$_4$ or Mgl, K$_2$CO$_3$, DMF or BnBr, Cs$_2$CO$_3$, CH$_3$CN, 70–100%; (b) KO'Bu, THF, 51–70%; (c) (i) (R)-(+)-α-methylbenzylamine, AcOH, MeOH, (ii) NaBH$_3$CN, 38–41% of 12a–d, 10–19% of 13a–d; (d) LiOH, 0 °C; (e) 10% Pd/C/NH$_4$HCO$_3$, MeOH, reflux, 16 h; (f) Fmoc-OSu/NaHCO$_3$, acetone/H$_2$O, rt, 12 h, 63–76% over 3 steps.

Figure 2. NOEs between nonadjacent residues observed for hexamers containing 4,4-disubstituted ACPC residues in 9:1 H$_2$O/D$_2$O at pH 3.8. Dashed lines indicate NOEs that are ambiguous because of resonance overlap. Data were obtained on a Varian 600 MHz spectrometer. Data for 15 was acquired at 14 °C at 5–10 mM concentration. Data for 16 was acquired at 4 °C at 5–10 mM concentration.
to NH(i + 2) NOEs, and one of four CαH(i) to NH(i + 3) NOEs were unambiguously assigned. In water, the same types of NOEs were identified and unambiguously assigned: three of four CαH(i) to CαH(i + 2) NOEs, four of five CαH(i) to NH(i + 2) NOEs, and one of four CαH(i) to NH(i + 3) NOEs. Only one fewer NOE was observed in water for this peptide than in methanol, perhaps suggesting that the structural differences between the two solvents are minimal.

Acknowledgment. This research was supported by the NIH (GM56414). T.J.P was supported by a NIH postdoctoral fellowship (GM065713). E.P.E. was supported by a NDSEG fellowship that is sponsored by the Department of Defense. NMR spectrometers were purchased with partial support from NIH and NSF. CD spectra were obtained in the NSF-supported Biophysics Instrumentation Facility. Crystallographic work was performed by Dr. Ilia A. Guzei.

Supporting Information Available: General procedures, characterization data, 1H and 13C spectra of all numbered intermediates in the schemes and crystallographic data in CIF format. This material is available free of charge via the Internet at http://pubs.acs.org.

OL0483293