α-Helix Mimetics

DOI: 10.1002/ange.200803778

Controlling Curvature in a Family of Oligoamide α-Helix Mimetics**

*Ishu Saraogi, Christopher D. Incarvito, and Andrew D. Hamilton**

The structure of the α-helix was first proposed by Pauling and Corey in 1951 and was subsequently found to be a common motif in protein secondary structure.[1] As more structural data on proteins became available, it was realized that the idealized α-helix and β-sheet models proposed by Pauling are often distorted. In optimizing their packing interactions, a large percentage of α-helices curve or kink,[2,3] (Figure 1) and β-sheets often deviate from planarity.[4] Kinks arising from proline, for example, have been found to be conserved across many species and may have a functional role.[5]

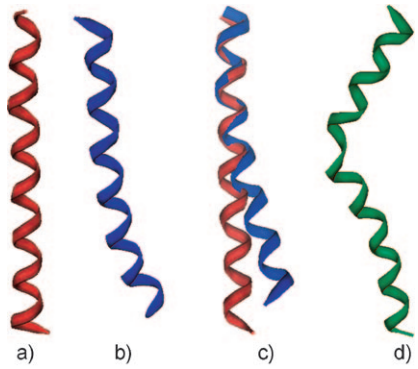
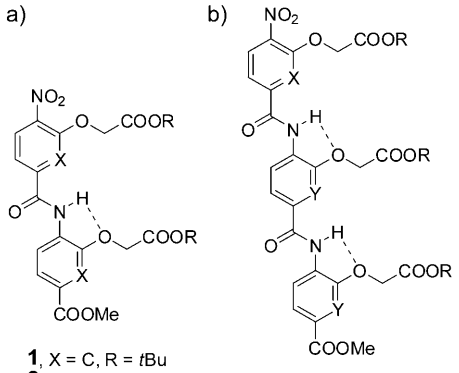


Figure 1. a) A straight helix and b) a curved helix in granulyte colony-stimulating factor (PDB ID: 1BGC); c) the two helices in (a) and (b) superimposed to highlight the curvature in the helix in (b); d) A kinked helix in lysin (PDB ID: 1LIS); adapted from Kumar et al.[6]

A survey of the protein data bank has shown that the α-helix backbone can be classified as linear, curved, or kinked, with a majority of α-helical domains showing a smooth curvature with a radius of less than 100 Å.[3,6–8] These deviations from the idealized linear form result from distortions in peptide-bond geometry and, in the case of amphipathic helices, from variations in the hydrogen-bonding patterns in the hydrophobic core versus the hydrophilic exterior.[8,9] Curvature has a significant bearing on the packing interactions, dipole moments, and spectroscopic properties of α-helices.[3,8] For example, the NMR spectra of curved helices show a periodic shift in the amide proton resonance that has been attributed to the varying degrees of magnetic anisotropy of the peptide carbonyl group.[10] Although such deviations in each residue are small and the overall backbone geometry is maintained, the cumulative effects along the helix make them significant. In seeking to mimic the structure and function of α-helices in synthetic scaffolds, it will be important to similarly reproduce different degrees of curvature.

The synthetic mimicry, using non-natural oligomers, of protein secondary structure and function has attracted considerable interest over the last two decades.[11] A recent advance in this area is the characterization of higher-order assemblies of these structures.[12] We have reported synthetic α-helix analogues[13] that mimic the recognition properties of residues at positions i, $i+3$ (and/or $i+4$), and $i+7$ of the helix and successfully disrupt key protein–protein interactions.[14–16]

One such family of scaffolds is based on an aryl amino acid oligomer in which intramolecular hydrogen bonds maintain the integrity of the structure (Scheme 1). When the aryl ring is



Scheme 1. Oligoamides used in this study: a) dimers and b) trimers.

pyridine, a curvature is seen and the direction of side-chain projection is affected.[15,16] This curvature in the solid-state structure appears to come from the geometrical restraints imposed by the hydrogen bond from the pyridine nitrogen atom to the amide -NH group. We reasoned that removing the hydrogen bond and introducing additional steric crowding through the use of a benzene ring in place of the pyridine ring would cause a straightening of the molecule and a decrease in the curvature of the backbone. In this way we would have a simple means of controlling the curvature.

A systematic search for arylcarboxamide-based structures in the Cambridge Structural Database (CSD)[17] revealed that

[*] I. Saraogi, Dr. C. D. Incarvito, Prof. A. D. Hamilton
Department of Chemistry, Yale University
225 Prospect Street, P.O. Box 208107
New Haven, CT 06520-8107 (USA)
Fax: (+1) 203-432-6144
E-mail: andrew.hamilton@yale.edu

[**] We thank the National Institutes of Health (GM69850) for financial support of this work.

Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/anie.200803778.

the hydrogen-bonding characteristics of the molecule had a significant effect on the angle at which the two aryl rings are inclined to each other (Figure 2). The degree of inclination

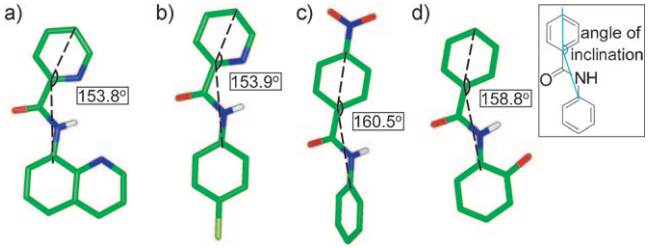


Figure 2. Some representative structures from the CSD showing different angles of inclination of the aryl rings; CSD Refcode: a) WOYVAH, b) GEPQIC, c) ACALAR, and d) SAQKEA; non-NH hydrogen atoms have been omitted for clarity; red O, blue N, green C, white H, light green Cl; the angle of inclination as defined in this study is shown as inset.

was defined as the angle at which the carbon atom of the aryl ring bound to the amide -NH group is inclined to the line connecting carbon atoms 1 and 4 of the preceding aryl ring (Figure 2, inset). Molecules with a *N*-phenylpicolinamide substructure have a relatively smaller angle of inclination presumably because of greater distortion of the pyridyl–amide bond angle to accommodate the hydrogen bond (Figure 2a,b). In *N*-phenylbenzamides, where no intramolecular hydrogen bonding is possible, the two rings can rotate more freely about the C(=O)–aryl and N(H)–aryl bonds. In this case, the angle of the tilt is larger (Figure 2c). This is also true when an *o*-hydroxy group is present, as in our proposed molecules (Figure 2d). The larger angle of inclination compared to the phenylpicolinamides implies that longer oligomers of *N*-phenyl-*p*-benzamide will be straighter, that is, they will have a smaller overall backbone curvature.[18]

Based on this information from the CSD, we first synthesized the dimeric arylcarboxamide analogues of these mimetics containing benzene (**1**) and pyridine (**2**) groups (Scheme 1a). The syntheses of these and their longer homologues (see below) were straightforward and involved an iterative acyl chloride coupling and nitro-group reduction protocol of the appropriate pyridine and benzene monomers (see the Supporting Information). Support for the presence of the second hydrogen bond to the pyridine ring comes from NMR studies showing the amide proton in **2**, which participates in bifurcated hydrogen bonding, to be shifted significantly downfield compared to that in **1**, which forms only a single hydrogen bond to the alkoxy group (Table 1).

Table 1: NMR shifts of the amide proton in molecules **1**–**6**.

Compound	δ [ppm]	Compound	δ_1 [ppm]	δ_2 [ppm]
1[a]	9.64	**3**[a]	9.45	9.71
2[b]	10.22	**4**[16][b]	10.12	10.25
		5[a]	8.85	10.36
		6[a]	9.46	10.57

NMR solvents used:[a] $CDCl_3$; [b] $[D_6]DMSO$.

A superimposition of the X-ray crystal structures of the oligobenzamide dimer **1** (red) and oligopyridylamide dimer **2** (blue) shows the diminished curvature in **1** (Figure 3). The

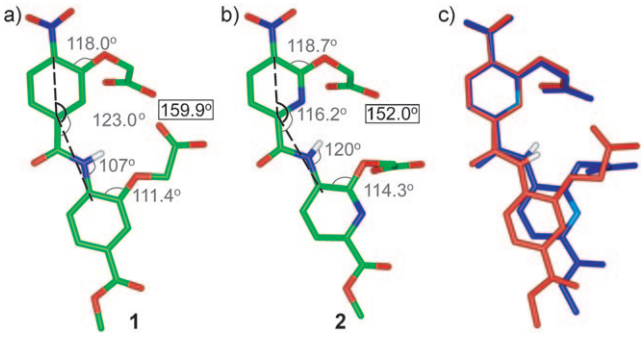


Figure 3. X-ray crystal structures of **1** (a) and **2** (b) showing the relevant angles; the two structures have been superimposed (c) to show the difference in curvature (red **1**, blue **2**). The nitrogen atoms in the pyridine rings are in light blue. Non-NH hydrogen atoms and *t*Bu ester groups in **1** have been omitted for clarity; red O, blue N, green C, white H.

two pyridine rings in **2** are tilted at an angle of 152.0° to each other, compared to 159.9° in **1**. As a consequence of the hydrogen bonding, the geometry of the amide bond is more distorted in **1** as evidenced by the H-N-C_{aryl} bond angle, which is significantly smaller (107°) than the ideal amide bond angle of 120°. The corresponding angle in the pyridine analogue is 120°. The N-C-C(=O) bond angle in **2** (116.2°) is much smaller than that in the benzene counterpart (123.0°) to allow for the bifurcated hydrogen bonds to form. A similar trend is also seen in the C-C-$O_{sidechain}$ angles, which are smaller in cases where hydrogen bonding takes place. The additive effect of these amide bond distortions leads to a significantly greater curvature in **2** relative to **1**.

The effects of the hydrogen bonding and consequently the curvature should be more pronounced in trimers **3**–**6** (Scheme 1b) because of an accumulation of these effects. In analogy with the above dimers, the oligobenzamide compound **3** is expected to be less curved than the oligopyridylamide compound **4**. Molecules **5** and **6**, with mixed oligopyridylamide and oligobenzamide backbones, should lie in between the two extremes. This hypothesis was supported by molecular modeling and energy minimization studies of trimers **3**–**6** using the consistent value force fields (CVFF) within InsightII (Figure 4).

An iterative strategy similar to that for the dimers was used to synthesize molecules **3**, **4**,[16] **5**, and **6** (see the Supporting Information). As with the dimers, the peaks in the NMR spectra for both the amide protons in **4**, but only one proton in both **5** and **6**, appear at lower field strengths compared to the amide protons in **3** (Table 1).

The X-ray crystal structures of **3**, **4**,[16] **5**, and **6** (Figure 5) confirmed the predicted trend in backbone curvature and the bond angles followed a trend similar to the dimers. The N-C-C(=O) bond angle in the case of benzene rings was found to be approximately 124° while the same angle was much smaller (≈115–117°) in the case of pyridine rings where bifurcated

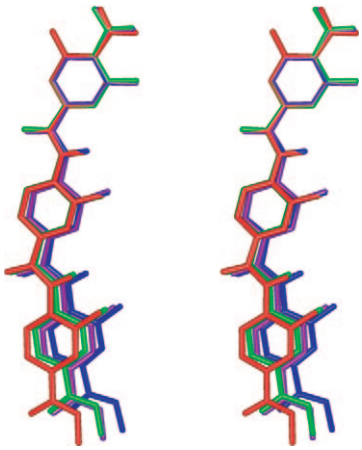


Figure 4. Superimposed stereoview representation of the acid derivatives of trimers **3**–**6** obtained from modeling in InsightII (red **3,** blue **4**, green **5**, purple **6**). Only the oxygen atom of the sidechains has been shown for clarity.

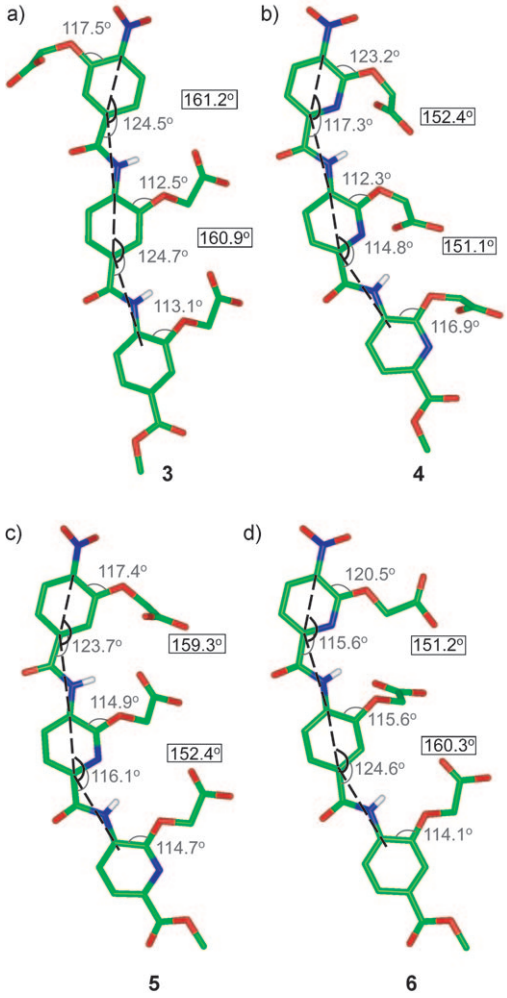


Figure 5. X-ray crystal structures of a) **3**, b) **4**[16] c) **5**, and d) **6** showing the relevant angles; non-NH hydrogen atoms and *t*Bu ester groups in **3**, **5**, and **6** have been omitted for clarity; red O, blue N, green C, white H.

hydrogen bonding occurs. In **3**, the angles at which the aromatic rings are tilted to each other are 161.2° and 160.9°, while in **4** the same angles are 152.4° and 151.1°. In **5**, where the top half of the molecule closely resembles **3**, this angle is 159.3° while the bottom half is similar to **4** with an angle of 152.4°. As expected, these numbers are reversed for **6** with values of 151.2° and 160.3° respectively. Thus, molecules **5** and **6** have an overall curvature that lies between that of **3**, which is least curved, and that of **4**, which is most curved.

In conclusion, we have shown that our oligoarylamide α-helix mimetics adopt a curved backbone structure in analogy to the majority of α-helices found in protein crystal structures. The curvature can be tuned by modifying the hydrogen-bonding characteristics of these simple compounds through the choice of appropriate building blocks, that is, by switching from phenyl to pyridine rings. This approach adds to the repertoire of tools available for the effective mimicry of protein secondary structures by choosing a scaffold that most closely mimics the α-helix of interest.

Received: August 1, 2008
Revised: September 17, 2008
Published online: November 6, 2008

Keywords: curvature · helical structures · hydrogen bonds · oligoamides · proteomimetics

[1] L. Pauling, R. B. Corey, H. R. Branson, *Proc. Natl. Acad. Sci. USA* **1951**, *37*, 205–211.
[2] C. Chothia, *Annu. Rev. Biochem.* **1984**, *53*, 537–572.
[3] D. J. Barlow, J. M. Thornton, *J. Mol. Biol.* **1988**, *201*, 601–619.
[4] B. K. Ho, P. M. G. Curmi, *J. Mol. Biol.* **2002**, *317*, 291–308.
[5] F. S. Cordes, J. N. Bright, M. S. P. Sansom, *J. Mol. Biol.* **2002**, *323*, 951–960.
[6] S. Kumar, M. Bansal, *Biophys. J.* **1996**, *71*, 1574–1586.
[7] S. Kumar, M. Bansal, *Biophys. J.* **1998**, *75*, 1935–1944; V. Geetha, *Int. J. Biol. Macromol.* **1996**, *19*, 81–89.
[8] T. Blundell, D. Barlow, N. Borkakoti, J. Thornton, *Nature* **1983**, *306*, 281–283.
[9] P. Chakrabarti, M. Bernard, D. C. Rees, *Biopolymers* **1986**, *25*, 1087–1093.
[10] I. D. Kuntz, P. A. Kosen, E. C. Craig, *J. Am. Chem. Soc.* **1991**, *113*, 1406–1408; N. E. Zhou, B. Y. Zhu, B. D. Sykes, R. S. Hodges, *J. Am. Chem. Soc.* **1992**, *114*, 4320–4326; F. J. Blanco, J. Herranz, C. Gonzalez, M. A. Jimenez, M. Rico, J. Santoro, J. L. Nieto, *J. Am. Chem. Soc.* **1992**, *114*, 9676–9677.
[11] *Foldamers: Structure, Properties and Applications* (Eds.: S. Hecht, I. Huc), Wiley-VCH, Weinheim, **2007**; D. Seebach, D. F. Hook, A. Glattli, *Biopolymers* **2006**, *84*, 23–37; S. H. Gellman, *Acc. Chem. Res.* **1998**, *31*, 173–180; D. J. Hill, M. J. Mio, R. B. Prince, T. S. Hughes, J. S. Moore, *Chem. Rev.* **2001**, *101*, 3893–4011; C. M. Goodman, S. Choi, S. Shandler, W. F. DeGrado, *Nat. Chem. Biol.* **2007**, *3*, 252–262; I. Huc, *Eur. J. Org. Chem.* **2004**, 17–29; J. Garner, M. M. Harding, *Org. Biomol. Chem.* **2007**, *5*, 3577–3585; Y. Hamuro, S. J. Geib, A. D. Hamilton, *J. Am. Chem. Soc.* **1997**, *119*, 10587–10593; V. Berl, I. Huc, R. G. Khoury, M. J. Krische, J. M. Lehn, *Nature* **2000**, *407*, 720–723; A. R. Sanford, B. Gong, *Curr. Org. Chem.* **2003**, *7*, 1649–1659; S. Chatterjee, R. S. Roy, P. Balaram, *J. R. Soc. Interface* **2007**, *4*, 587–606, and references therein.
[12] K. Huang, C. W. Wu, T. J. Sanborn, J. A. Patch, K. Kirshenbaum, R. N. Zuckermann, A. E. Barron, I. Radhakrishnan, *J. Am. Chem. Soc.* **2006**, *128*, 1733–1738; E. J. Petersson, A. Schepartz,

J. Am. Chem. Soc. **2008**, *130*, 821 – 823; W. S. Horne, J. L. Price, J. L. Keck, S. H. Gellman, *J. Am. Chem. Soc.* **2007**, *129*, 4178 – 4180; G. V. M. Sharma, P. Jayaprakash, K. Narsimulu, A. R. Sankar, K. R. Reddy, P. R. Krishna, A. C. Kunwar, *Angew. Chem.* **2006**, *118*, 3010 – 3013; *Angew. Chem. Int. Ed.* **2006**, *45*, 2944 – 2947; Q. Gan, C. Y. Bao, B. Kauffmann, A. Grelard, J. F. Xiang, S. H. Liu, I. Huc, H. Jiang, *Angew. Chem.* **2008**, *120*, 1739 – 1742; *Angew. Chem. Int. Ed.* **2008**, *47*, 1715 – 1718.

[13] H. Yin, A. D. Hamilton, *Angew. Chem.* **2005**, *117*, 4200 – 4235; *Angew. Chem. Int. Ed.* **2005**, *44*, 4130 – 4163; J. M. Davis, L. K. Tsou, A. D. Hamilton, *Chem. Soc. Rev.* **2007**, *36*, 326 – 334.

[14] J. Becerril, A. D. Hamilton, *Angew. Chem.* **2007**, *119*, 4555 – 4557; *Angew. Chem. Int. Ed.* **2007**, *46*, 4471 – 4473; J. M. Rodriguez, A. D. Hamilton, *Angew. Chem.* **2007**, *119*, 8768 – 8771; *Angew. Chem. Int. Ed.* **2007**, *46*, 8614 – 8617; H. Yin, G. I. Lee, H. S. Park, G. A. Payne, J. M. Rodriguez, S. M. Sebti, A. D. Hamilton, *Angew. Chem.* **2005**, *117*, 2764 – 2767; *Angew. Chem. Int. Ed.* **2005**, *44*, 2704 – 2707; J. T. Ernst, O. Kutzki, A. K. Debnath, S. Jiang, H. Lu, A. D. Hamilton, *Angew. Chem.* **2002**, *114*, 288 – 291; *Angew. Chem. Int. Ed.* **2002**, *41*, 278 – 281; H. Yin, G. I. Lee, K. A. Sedey, J. M. Rodriguez, H. G. Wang, S. M. Sebti, A. D. Hamilton, *J. Am. Chem. Soc.* **2005**, *127*, 5463 – 5468; O. Kutzki, H. S. Park, J. T. Ernst, B. P. Orner, H. Yin, A. D. Hamilton, *J. Am. Chem. Soc.* **2002**, *124*, 11838 – 11839; L. Moisan, S. Odermatt, N. Gombosuren, A. Carella, J. Rebek, *Eur. J. Org. Chem.* **2008**, 1673 – 1676; P. Maity, B. Konig, *Org. Lett.* **2008**, *10*, 1473 – 1476.

[15] J. T. Ernst, J. Becerril, H. S. Park, H. Yin, A. D. Hamilton, *Angew. Chem.* **2003**, *115*, 553 – 557; *Angew. Chem. Int. Ed.* **2003**, *42*, 535 – 539.

[16] L. A. Estroff, C. D. Incarvito, A. D. Hamilton, *J. Am. Chem. Soc.* **2004**, *126*, 2 – 3.

[17] F. H. Allen, *Acta Crystallogr. Sect. B* **2002**, *58*, 380 – 388.

[18] While this work was in progress, two other examples of phenylcarboxamide oligomers were reported: J. M. Ahn, S. Y. Han, *Tetrahedron Lett.* **2007**, *48*, 3543 – 3547; J. Plante, F. Campbell, B. Malkova, C. Kilner, S. L. Warriner, A. J. Wilson, *Org. Biomol. Chem.* **2008**, *6*, 138 – 146.

[19] CCDC 696873 (**1**),697087 (**2**), 697088 (**3**), 697089 (**5**),and 697090 (**6**) contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

