ORGANIC LETTERS

2002
Vol. 4, No. 26
4663−4666

Hydrogen Bond-Stabilized Helix Formation of a *m*-Phenylene Ethynylene Oligomer

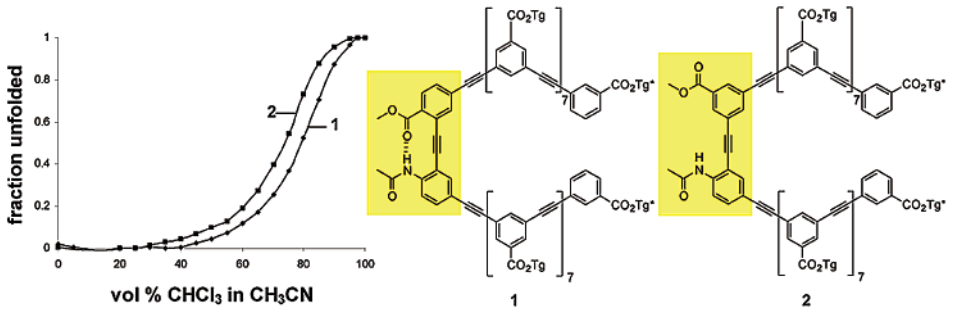
Jennifer M. Cary and Jeffrey S. Moore*

Departments of Chemistry and Materials Science & Engineering, 600 South Mathews Avenue, The University of Illinois at Urbana−Champaign, Urbana, Illinois 61801

moore@scs.uiuc.edu

Received October 14, 2002

ABSTRACT



Incorporation of a single hydrogen bonded β-turn mimic in the backbone of a *m*-phenylene ethynylene oligomer is shown to affect the thermodynamic properties of the folding reaction. Oligomers 1 and 2 both undergo solvophobic helix formation, but hydrogen bonded oligomer 1 was found to form a more stable helix with a higher tolerance to solvent denaturation than isomeric, non-hydrogen bonded oligomer 2.

Biomolecules rely on precise combinations of noncovalent interactions to interconvert between various folded and unfolded states in response to ligand binding or changes in their environment.[1] Foldamer research focuses on designing synthetic macromolecules that replicate characteristics of their natural counterparts.[2] These synthetic molecules use noncovalent interactions to drive the folding reaction, so they are able to reversibly change conformation in response to environmental stimuli. In supramolecular systems, the use of combinations of noncovalent interactions presents many challenges[3] but offers the potential for increased control. Here we show how the combination of hydrogen bonding and π-stacking interactions can be used to shift the conformational transition of a *m*-phenylene ethynylene foldamer.

One class of foldamers that has attracted recent interest is single-stranded oligomers that mimic proteins in their ability to undergo cooperative transitions between random and helical conformations.[4−6] Phenylene ethynylene (PE) oligomers **3** previously studied by our group have been found to exhibit a reversible transition from random to helical conformations in response to changes in solvent quality.[7,8] The equilibrium constant for this folding reaction (eq 1)

$$\text{random conformation} \overset{K_{eq}}{\rightleftharpoons} \text{helix} \qquad (1)$$

represents the ratio between folded and unfolded oligomers

(1) Anfinsen, C. B. *Science* **1973**, *181*, 223−230.

(2) (a) Hill, D. J. Mio, M. J.; Prince, R. B.; Hughes, T. S.; Moore, J. S. *Chem. Rev.* **2001**, *101*, 3893−4011. (b) Gellman, S. H. *Acc. Chem. Res.* **1998**, *31*, 173−180. (c) Stigers, K. D.; Soth, M. J.; Nowick, J. S. *Curr. Opin. Chem. Biol.* **1999**, *3*, 714−723. (d) Archer, E. A.; Gong, H.; Krische, M. J. *Tetrahedron* **2001**, *57*, 1139−1159.

(3) Hirschberg, J. H. K. K.; Brunsveld, L.; Ramzi, A.; Vekemans, J. A. J. M.; Sijbesma, R. P.; Meijer, E. W. *Nature* **2000**, *407*, 167−170.

10.1021/ol0270982 CCC: $22.00
Published on Web 12/07/2002

and is related to the thermodynamic parameters s and σ from the helix coil model[9] as shown in eq 2.

$$K_{eq} = \sigma s^{n - n_o} \quad (2)$$

In this equation, s accounts for the enthalpic gain from monomer−monomer interactions; n_o is equal to the number of monomers required to form one turn of the helix, and σ accounts for the entropic cost of sacrificing free rotation between monomers to form the first turn of the helix. The free energy change for the helix nucleation process (ΔG_{nuc}) is given by $-RT\ln(\sigma)$, while the free energy change for helix propagation (ΔG_{prop}) is given by $-RT(n - n_o)\ln(s)$.

Previous work in our group has demonstrated that the thermodynamics of the folding reaction for oligomer **3** can be altered through variations in chain length (n),[5] side chain polarity,[4g] and hydrophobic packing of the helical cavity.[10] However, because only aromatic stacking interactions are utilized for stabilizing the helical conformation, control over the folding properties of oligomers having a homogeneous backbone is limited. By using a combination of noncovalent interactions, we demonstrate here an increased ability to manipulate the equilibrium constant of the folding reaction.

On the basis of the value of ΔG_{nuc} for oligomers **3**, we calculate the free energy change associated with locking each acetylene bond in the folded conformation to be approximately 0.9 kcal/mol.[11] From this we predict that the incorporation of a β-turn mimic having a hydrogen bond bridging adjacent phenyl units would restrict rotation about one acetylene bond. This would lower the value of ΔG_{nuc} and increase the equilibrium constant for the folding reaction.

Diphenylacetylene is known to function as a β-turn unit in artificial β-sheet **4**,[12] so we adapted the structure to give **5** as our model of a hydrogen bonded PE monomer. We were able to obtain crystals of **5**, and the X-ray structure revealed that the molecule adopts the desired β-turn conformation with an N_{donor}−$O_{acceptor}$ hydrogen bond length of 3.10 Å (Figure 1). NMR spectroscopy showed that the β-turn conformation

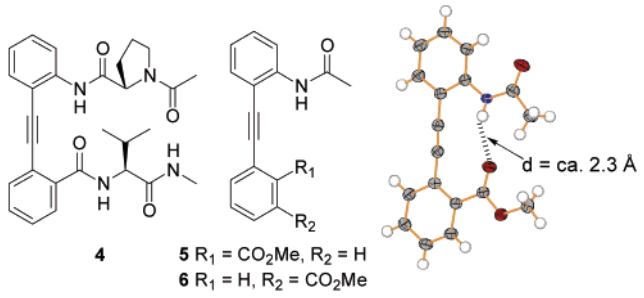
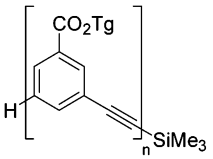


Figure 1. Model compound **5** and control isomer **6**. X-ray structure of **5** shows the hydrogen bonding interaction between adjacent units.

was retained in solution, as the amide N−H proton of **5** was shifted 1.42 ppm in chloroform and 0.97 ppm in acetonitrile relative to the amide N−H proton of control compound **6**.



3, n>10

We concluded that **5** was a suitable β-turn mimic for use in PE oligomers, while isomer **6** served as a control having similar ring electronics but a disengaged hydrogen bond. Next, we synthesized oligomers incorporating either the β-turn mimic or the control unit (Scheme 1). Synthesis of the oligomers began with acylation of 2-bromo-4-nitroaniline[13] by acetic anhydride in the presence of catalytic copper(II) trifluoromethanesulfonate.[14] Reduction of the nitroarene, followed by protection of the resulting aniline as a bis-diethyltriazine and subsequent Pd-catalyzed cross-coupling with trimethylsilylacetylene, gave amide monomer **7**. Esterification of 2-iodo-4-nitrobenzoic acid[15] gave the methyl ester. Reduction of this second nitroarene, followed by protection of the resulting aniline as a bis-diethyltriazine, yielded methyl ester monomer **8**. Coupling of monomer **7** with either **8** or **9**[5] followed by deprotection of the bis-diethyltriazine groups gave the corresponding iodides. Finally, Pd-catalyzed cross-coupling with trimethylsilylacetylene yielded β-turn monomers **10** and **11**. Coupling of either monomer **10** or **11** with 2 equiv of chiral octamer iodide **12** gave the desired octadecamers **1** and **2**.

To compare the folding properties of octadecamers **1** and **2**, UV absorption spectra of the two isomers were obtained in a series of solvent mixtures ranging from pure acetonitrile to pure chloroform. In acetonitrile, the PE backbone is poorly

(4) Examples of single-stranded oligomers that undergo cooperative conformational transitions include: (a) Kirshenbaum, K.; Barron, A. E.; Goldsmith, R. A.; Armand, P.; Bradley, E. K.; Truong, K. T. V.; Dill, K. A.; Cohen, F. E.; Zuckermann, R. N. *Proc. Natl. Acad. Sci. U.S.A.* **1998**, *95*, 4303−4308. (b) Martinez de Ilarduya, A.; Alemán, C.; García-Alvarez, M.; López-Carrasquero, F.; Muñoz-Guerra, S. *Macromolecules* **1999**, *32*, 3257−3263. (c) Cheng, J.; Deming, T. J. *Macromolecules* **2001**, *34*, 5169−5174. (d) Wu, C. W.; Sanborn, T. J.; Huang, K.; Zuckermann, R. N.; Barron, A. E. *J. Am. Chem. Soc.* **2001**, *123*, 6778−6784. (e) Fernández-Santín, J. M.; Muñoz-Guerra, S.; Rodríguez-Galán, A.; Aymamí, J.; Lloveras, J.; Subirana, J. A. *Macromolecules* **1987**, *20*, 62−68. (f) Prince, R. B.; Moore, J. S.; Brunsveld, L.; Meijer, E. W. *Chem. Eur. J.* **2001**, *7*, 4150−4154. (g) Brunsveld, L.; Prince, R. B.; Meijer, E. W.; Moore, J. S. *Org. Lett.* **2000**, *2*, 1525−1528. (h) Gin, M. S.; Moore, J. S. *Org. Lett.* **2000**, *2*, 135−138. (i) Petitjean, A.; Cuccia, L. A.; Lehn, J.-M.; Nierengarten, H.; Schmutz, M. *Angew. Chem., Int. Ed.* **2002**, *41*, 1195−1198.

(5) Prince, R. B.; Saven, J. G.; Wolynes, P. G.; Moore, J. S. *J. Am. Chem. Soc*. **1999**, *121*, 3114−3121.

(6) Nelson, J. C.; Saven, J. G.; Moore, J. S.; Wolynes, P. G. *Science* **1997**, *277*, 1793−1796.

(7) Hill, D. J., Moore, J. S. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 5053−5057.

(8) Defined according to classic polymer chemistry: Flory, P. J. *Principles of Polymer Chemistry*; Cornell University Press: Ithaca, NY, 1953.

(9) Zimm, B. H.; Bragg, J. K. *J. Chem. Phys.* **1959**, *31*, 526−535.

(10) Prince, R. B. Ph.D. Thesis, University of Illinois at Urbana−Champaign, Urbana, 2000.

(11) See supplemental information

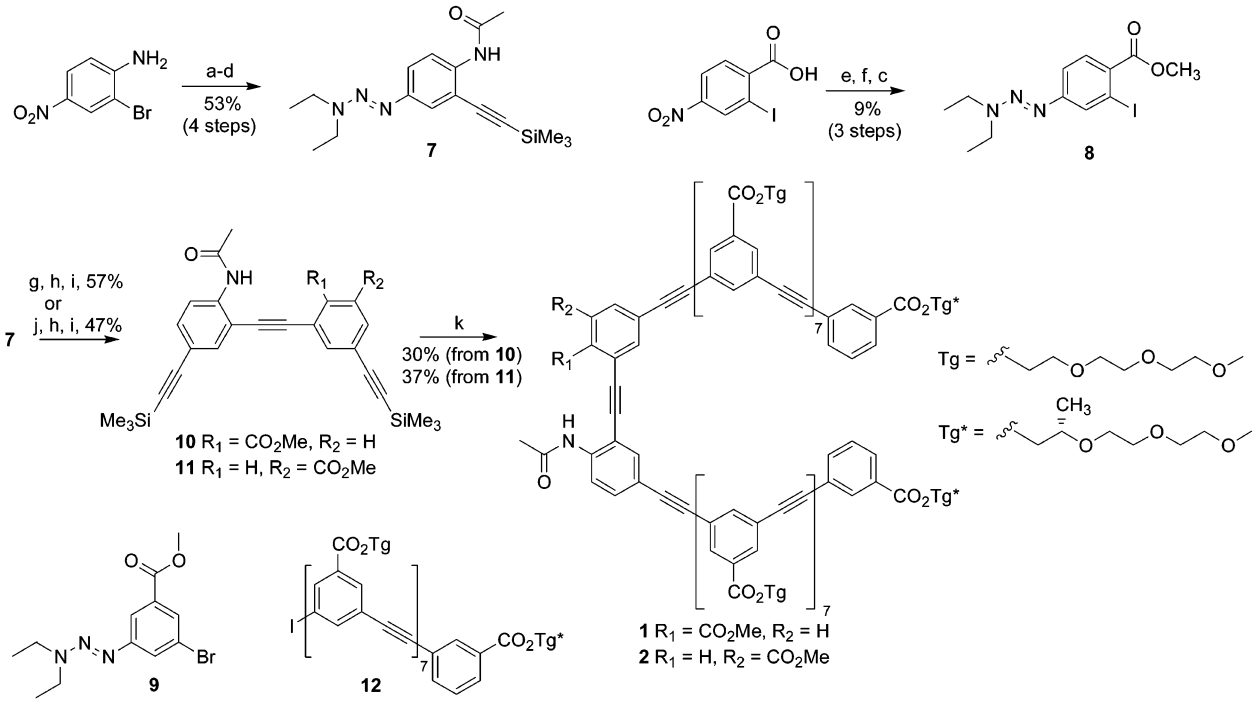
(12) Kemp, D. S.; Li, Z. Q. *Tetrahedron Lett*. **1995**, *36*, 4179−4180.

(13) Roche, D.; Prasad, K.; Repic, O.; Blacklock, T. J. *Tetrahedron Lett.* **2000**, *41*, 2083−2085.

(14) Saravanan, P.; Singh, V. K. *Tetrahedron Lett*. **1999**, *40*, 2611−2614.

(15) Katritzky, A. R.; Savage, G. P.; Gallos, J. K.; Durst, H. D. *Org. Prep. Proced. Int.* **1989**, *21*, 157−162.

Scheme 1[a]



[a] Reagents and conditions: (a) Ac_2O, $Cu(OTf)_2$, CH_2Cl_2. (b) Pt/C, HCO_2H, Et_3N. (c) (i) $NaNO_2$, HCl, H_2O, CH_3CN, 5 °C; (ii) Et_2NH, K_2CO_3, H_2O. (d) TMS acetylene, $Pd_2(dba)_3$, CuI, PPh_3, Et_3N, 70 °C. (e) MeOH, H_2SO_4, Δ. (f) $Na_2S_2O_4$, MeOH, THF, H_2O. (g) (i) TBAF, THF; (ii) **8**, $Pd_2(dba)_3$, CuI, PPh_3, Et_3N, 50 °C. (h) CH_3I, 110 °C. (i) TMS acetylene, $Pd_2(dba)_3$, CuI, PPh_3, Et_3N, 50 °C. (j) (i) TBAF, THF; (ii) **9**, $Pd_2(dba)_3$, CuI, PPh_3, Et_3N, 70 °C. (k) (i) TBAF, THF; (ii) **12**, $Pd_2(dba)_3$, CuI, PPh_3, Et_3N, 50 °C.

solvated causing the oligomer to collapse into a helical conformation, whereas in chloroform, the PE backbone is better solvated and the oligomer unfolds into a random conformation. This transition from the helix to a random conformational state was monitored by the ratio of the oligomer absorbance bands at 303 and 289 nm, with low values of $A_{303/289}$ indicating a high degree of folding.[5,6] Assuming that both **1** and **2** undergo a complete transition between their folded and unfolded states over the observed range of solvent mixtures, the UV absorbance data can be fitted to give the fraction of oligomer in the unfolded state for each solvent mixture.

Figure 2 displays the absorption data obtained for **1** and **2** as a function of solvent composition. Assuming that the free energy difference between the folded and unfolded states depends linearly on solvent composition,[16] the free energy of folding in pure acetonitrile, $\Delta G(CH_3CN)$, can be determined from eq 3 where $[CHCl_3]$ represents the chloroform composition expressed as vol % and m indicates how rapidly the stabilization energy of the helix (ΔG) changes in response to changes in solvent composition.[5]

$$\Delta G = \Delta G(CH_3CN) - m[CHCl_3] \qquad (3)$$

The composition of chloroform required to reach the midpoint of denaturation ($[CHCl_3]_{1/2}$) is given by $\Delta G(CH_3CN)/m$. A comparison of the free energy of folding and the midpoint of denaturation for **1** and **2** is presented in Table 1. These data reveal that the hydrogen bonded β-turn mimic of oligomer **1** lowers the energy of the helical conformation by ca. 1.2 kcal/mol. This stabilization results in a higher tolerance to denaturation, as the percentage of chloroform required to reach the midpoint of denaturation increases by 5.3 vol % for the hydrogen bonded oligomer.[17]

Table 1. Comparison of the Free Energy of Folding and Midpoint of Denaturation for Octadecamers **1** and **2**

oligomer	$\Delta G(CH_3CN)$ (kcal/mol)	m (cal/mol)	$[CHCl_3]_{1/2}$ (vol %)
1	−7.0 ± 0.7	90 ± 8	78.0 ± 0.8
2	−5.8 ± 0.6	80 ± 8	72.7 ± 0.8

The experimentally observed stabilization of 1.2 kcal/mol provided by the hydrogen bonded β-turn mimic is on the

(16) (a) Jasanoff, A.; Fersht, A. R. *Biochemistry* **1994**, *33*, 2129−2135. (b) Pace, C. N. *Methods in Enzymology*; Hirs, C. H. W., Timasheff, S. N., Eds.; Academic Press: New York, 1986; Vol. 131, pp 266−280. (c) Pace, C. N.; Shirley, B. A.; Thomson J. A. In *Protein Structure: A Practical Approach*; Creighton, T. E., Ed.; IRL Press: New York, 1989; pp 311−330.

(17) The predicted decrease in free energy of folding corresponding to the observed shift in denaturation midpoint is approximately 1 kcal/mol. While the calculated change in $\Delta G(CH_3CN)$ is on the edge of statistical significance, the shift in $[CHCl_3]_{1/2}$ is well beyond the error limits.

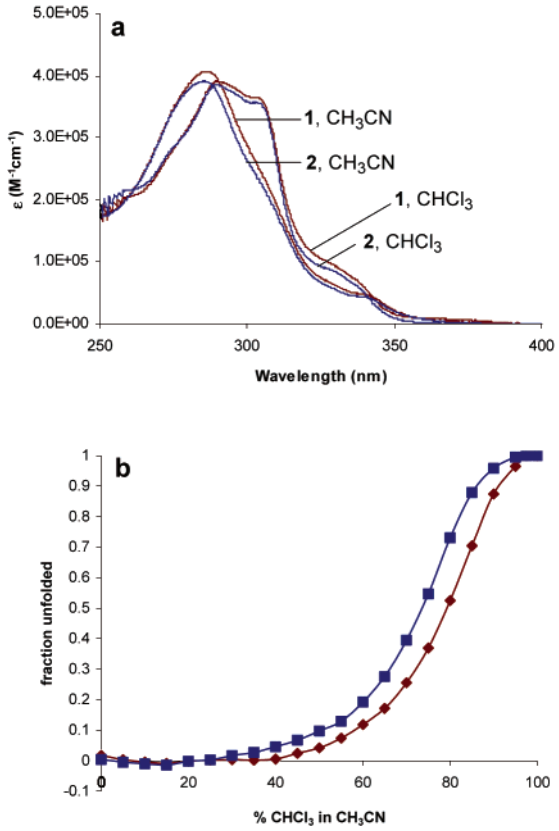


Figure 2. (a) UV spectra of **1** (red) and **2** (blue) in chloroform and acetonitrile. (b) UV titration curve of **1** (red diamonds) and **2** (blue squares).

order of the predicted value of 0.9 kcal/mol for locking the rotation about one acetylene bond in the PE backbone. In other words, the hydrogen bond of the β-turn mimic provides enough energy to lock one diphenylacetylene unit in the cisoid conformation required for helix formation. We do not yet know if the placement of the hydrogen bond in the middle of the helix rather than at the terminus leads to a more pronounced effect on the thermodynamics of helix nucleation. We are also unable to comment on the role of the slight electronic differences between the β-turn mimic and the control unit with regards to imparting greater stabilization to the helical conformation of the hydrogen bonded oligomer. Nonetheless, we believe that the results are in good agreement with the predicted behavior, and we feel that this β-turn mimic provides added versatility to our PE monomer pool.

In conclusion, the incorporation of a single hydrogen bonded β-turn mimic into a *m*-phenylene ethynylene oligomer provides an additional noncovalent interaction that can be used to control the folding properties of the oligomer. The effect on the thermodynamics of the folding reaction is apparent from a comparison of the free energy of folding and the midpoint of solvent denaturation for hydrogen bonded oligomer **1** and isomeric, non-hydrogen bonded oligomer **2**. The hydrogen bond was found to stabilize the folded conformation of the oligomer, making it more tolerant to denaturation by chloroform. As more types of noncovalent interactions are adapted for use in oligomers, these systems will better mimic proteins in their ability to use a monomer sequence to specify precise folding properties. Future studies may include use of the PE backbone as a scaffold for incorporation of multiple-peptide β-sheets to create a β-sandwich structure.

Acknowledgment. This research was funded by the National Science Foundation (CHE 00-91931). J.M.C. thanks Abbott Laboratories for a doctoral fellowship.

Supporting Information Available: Detailed descriptions of all experimental procedures and accompanying analytical data. This material is available free of charge via the Internet at http://pubs.acs.org.

OL0270982