Azacrown-attached *meta*-ethynylpyridine polymer: saccharide recognition regulated by supramolecular device†‡

Hajime Abe,* Shunsuke Takashima, Tsuyoshi Yamamoto and Masahiko Inouye*

Received (in Cambridge, UK) 3rd February 2009, Accepted 4th March 2009
First published as an Advance Article on the web 17th March 2009
DOI: 10.1039/b902269d

Polymeric synthetic host 2, azacrown-attached 2,6-pyridylene ethynylene polymer, was investigated for its saccharide recognition and the additive effect of triethylene tetramine–trifluoroacetic acid; heteroallosteric effects were observed on the basis of CD and UV/Vis analyses, which indicated saccharide-dependent stabilization and destabilization of helical complexes by the formation of pseudopolyrotaxanes.

Helical biomolecules carry out important functions in nature,[1] and such molecules are regulated in a sophisticated way in their structures and performance by circumstance and chemical species, sometimes showing allosterism.

Recently, we have developed artificial foldamers, 2,6-pyridylene ethynylenes or "*meta*-ethynylpyridines", which consist of 4-substituted pyridine rings linked at their 2,6-positions with acetylene bonds (Fig. 1, **1**). These artificial foldamers carry out saccharide recognition[2,3] by hydrogen bonding in organic and aqueous media, and the key feature is the formation of helices to accommodate the saccharide.[3–5] The substituent R on the pyridine rings of **1** adds optional nature to the foldamer.[5] Herein, a new type of *meta*-ethynylpyridine co-polymer **2** has been developed, which possesses a crown ether at the 4-position of every third pyridine ring (Fig. 1). Octyloxy groups were introduced at the remaining pyridine rings to improve solubility. We expected the crown ether to play a role as a scaffold[6–8] to effect structural behavior and saccharide recognition ability of the *meta*-ethynylpyridine moiety by

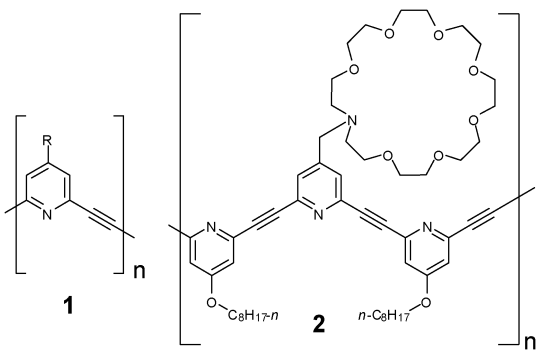


Fig. 1 Polymeric *meta*-ethynylpyridines **1** and **2**.

Graduate School of Pharmaceutical Sciences, University of Toyama, Sugitani 2630, Toyama, Toyama 930-0194, Japan.
E-mail: inouye@pha.u-toyama.ac.jp; Fax: +81 76-434-5049
† Dedicated to Professor Seiji Shinkai on the occasion of his 65th birthday.
‡ Electronic supplementary information (ESI) available: Detailed experimental procedures, additional CD and UV/Vis spectra and titration curves. See DOI: 10.1039/b902269d

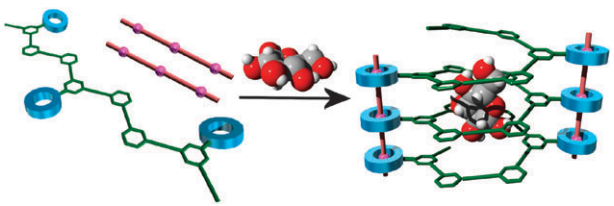


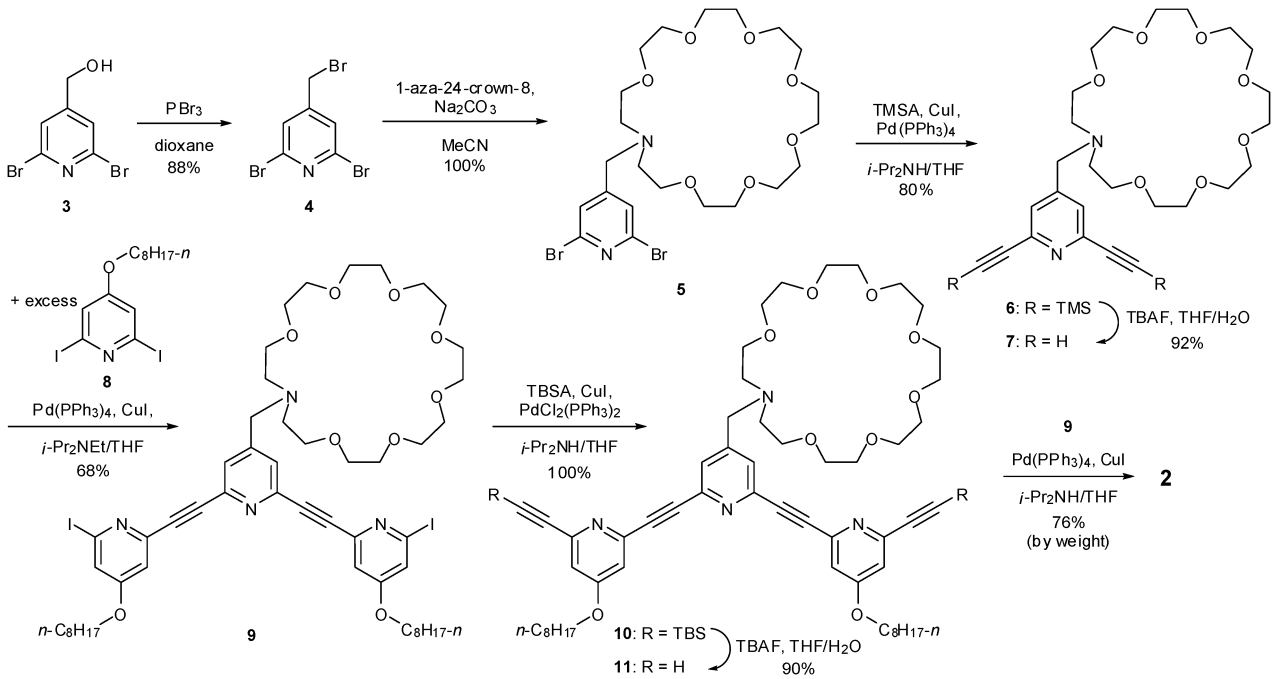
Fig. 2 Mechanical design of **2**. Stabilization of a helix by pseudopolyrotaxane formation.

the formation of pseudopolyrotaxanes with an axial guest molecule such as polyammonium cations.

The outline of the mechanical design of **2** is depicted in Fig. 2. A 24-crown-8 ring is known to make a rotaxane with a secondary ammonium axis.[7] Plural 1-aza-24-crown-8 sites (blue ring) in **2** and oligoammonium axes (purple sticks) can form pseudopolyrotaxanes to stabilize helical structures of the polymer (green) by bridging between the pitches of the helix. By distributing a crown ether at every three pyridines in **2**, it is expected that two bridges are built at opposite sides of the helix. The resulting inside hole can be suitable (or unsuitable) to accommodate a targeted guest saccharide, which biases the chiral sense of the complex to induce circular dichroism (CD). The biases might be regulated by the formation of the pseudopolyrotaxanes, *i.e.*, heterotropic allosteric behavior.

The azacrown-attached co-polymer **2** was prepared as shown in Scheme 1. Citrazinic acid was converted into 2,6-dibromo-4-(hydroxymethyl)pyridine (**3**), then it was brominated to **4**. Next, **4** was condensed with 1-aza-24-crown-8 to give **5**. After diethynylation of **5** to **7**, co-trimer **9** was made from **7** and 2,6-diiodo-4-octyloxypyridine[5e] (**8**). Further diethynylation of **9** yielded **11**, the counterpart of **9**. The final co-polymerization between the diiodide **9** and the diacetylene **11** was carried out by Sonogashira reaction to obtain **2**. The molecular weight of **2** was estimated as M_n = 8800 g mol^{-1} after brief purification using Sephadex, and the fraction of larger molecular weight (M_n = 11 500 g mol^{-1}) was taken out by preparative GPC to use in the following experiments.

The saccharide recognition manner of **2** was surveyed qualitatively by CD measurements. First, when octyl β-D-glucopyranoside (**β-D-Glc**) was added to a CH_2Cl_2 solution of **2**, an induced negative CD band appeared around 340 nm (Fig. 3A, brown solid line). The shape of this CD band strongly resembles that in the cases of other types of *meta*-ethynylpyridines associated with **β-D-Glc**.[5,9] Therefore, the CD band observed here indicates that **2** also formed a similar helical complex with **β-D-Glc**. Next, triethylene tetramine (TETA) was added to the mixture, then the induced CD band slightly increased (Fig. 3A, blue solid line). Finally,



Scheme 1 Preparation of azacrown-attached co-polymer **2**. TMSA = (trimethylsilyl)acetylene, TBSA = (*tert*-butyldimethylsilyl)acetylene, TBAF = tetrabutylammonium fluoride.

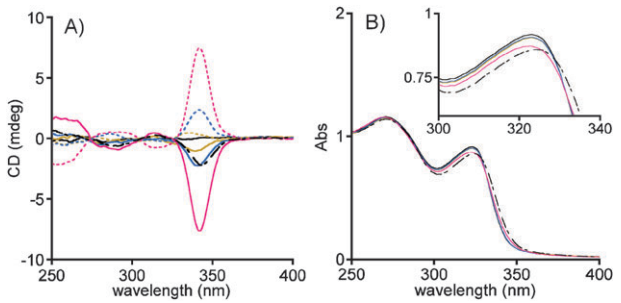


Fig. 3 The changes of (A) CD and (B) UV/Vis spectra of the mixture of **2** and **β-D-Glc** in the presence and absence of triethylene tetramine (TETA) and trifluoroacetic acid (TFA). Black solid: only **2**; brown solid: **2** + **β-D-Glc**; blue solid: **2** + **β-D-Glc** + TETA; red solid: **2** + **β-D-Glc** + TETA + TFA; brown broken: **2** + **β-L-Glc**; blue broken: **2** + **β-L-Glc** + TETA; red broken: **2** + **β-L-Glc** + TETA + TFA; black dashed: **2** + **β-D-Glc** + TFA. Conditions: **2** (1.0×10^{-3} M, unit conc.), **β-D-Glc** or **β-L-Glc** (2.0×10^{-3} M), TETA (1.7×10^{-4} M), TFA (6.7×10^{-4} M), CH_2Cl_2, 25 °C, path length = 1 mm.

trifluoroacetic acid (TFA) was added to the mixture, then significant enhancement of the CD band was observed (Fig. 3A, red solid line). The enantiomeric glycoside, octyl β-L-glucopyranoside (**β-L-Glc**), induced mirror-image CDs corresponding to those in the cases of **β-D-Glc** (Fig. 3A, broken lines), reflecting the chirality transfer from the added saccharides to the polymer. On the other hand, when diethylamine was used as the representative monoamine instead of TETA, no meaningful additive effect could be observed even after the addition of TFA (Fig. S1‡).

The changes of UV/Vis spectra were also studied. In a previous study on a strongly basic ethynylpyridine polymer, it was found that the protonation of pyridine into pyridinium causes a significant red shift in the UV/Vis spectra.[5b] On the other hand, when TETA and TFA were added to **2** or a mixture of **2** and **β-D-Glc**, such red shifts were sluggish (Fig. S2‡ and 3B). This finding means that the contribution of pyridinium species would be negligible here and most of the TFA would give its proton to TETA to form oligoammonium cation TETA·nH$^+$. Instead of a red shift, the addition of the TETA–TFA combination brought a substantial hypochromic effect on the wavelength region where the induced CD band appeared (see also Fig. S3 and S4‡). Therefore, the contribution of π-interaction seems to accompany the stabilization of the helical structure.[10] TFA alone did not enhance the glucoside recognition enough without TETA. When only TFA was added to the mixture of **2** and **β-D-Glc**, the change of CD was very little (Fig. 3A, black dashed line). In this case, a small red shift was observed in the UV/Vis spectrum, being due to the protonation of pyridine rings (Fig. 3B, black dashed line).

Quantitative evaluation of the heteroallosteric behavior of **2** with the TETA–TFA combination was carried out by CD titration experiments using **β-D-Glc** as a titrant (Fig. S5‡). In CH_2Cl_2, the formal binding constant[11] between **2** and **β-D-Glc** was obtained as $K'_0 = (1.0 \pm 0.3) \times 10^2$ M^{-1}. After the addition of TETA, the binding constant increased to $K'_1 = (8.0 \pm 0.7) \times 10^2$ M^{-1}, and further addition of TFA gave most improved binding strength as expected, $K'_2 = (1.5 \pm 0.3) \times 10^3$ M^{-1}. These results indicate that the oligoammonium TETA·nH$^+$ stabilizes the chiral helical structure of the complex **2**·**β-D-Glc** by the formation of a pseudorotaxane consisting of azacrown rings and oligoammonium axes.

Other kinds of glycosides, octyl β-D-fructopyranoside (**β-D-Fru**) and octyl β-D-mannopyranoside (**β-D-Man**), induced positive CDs in **2** around 340 nm as the previous (*meta*-ethynylpyridine)s did.[5a–c] However, the addition of

TETA–TFA to a mixture of **2** and **β-D-Fru** caused inversion of the sign of the CD band (Fig. S3‡). The formation of a pseudopolyrotaxane with the TETA–TFA combination might change the helical pitch, resulting in the geometrical rearrangement of the hydrogen-bonding pyridine nitrogens. The hydrogen-bonding array in the reversed helix is likely to interact with **β-D-Fru** more preferentially than that of the original helix. On the other hand, in the case of **β-D-Man**, the TETA–TFA combination slightly weakened the positive CD band at 344 nm and increased a negative CD band at 334 nm (Fig. S4‡). These observations illustrate that some negative heteroallosteric behavior or partial helix inversion of **2** might occur against **β-D-Man**. Thus, the additive effects of the TETA–TFA combination on saccharide recognition with **2** were demonstrated. The chirality transfer from glycosides to the helical complexes and recognition affinity of **2** were altered with the TETA–TFA combination.

Although further details of the interaction with TETA·nH$^+$ are under investigation, this collaboration of host–guest chemistry and rotaxane chemistry will broaden the capability of supramolecular chemistry of *meta*-ethynylpyridines.

Notes and references

1 (*a*) G. E. Schulz and R. H. Schirmer, *Principles of Protein Structure*, Springer-Verlag, New York, 1979; (*b*) W. Saenger, *Principles of Nucleic Acid Structure*, Springer-Verlag, New York, 1984.

2 Reviews of saccharide recognition by synthetic hosts: (*a*) *Host–Guest Chemistry—Mimetic Approaches to Study Carbohydrate Recognition*, *Top. Curr. Chem.*, ed. S. Penadés, Springer-Verlag, Berlin, 2002, vol. 218; (*b*) A. P. Davis and R. S. Wareham, *Angew. Chem., Int. Ed.*, 1999, **38**, 2978–2996. Examples of artificial saccharide receptor: (*c*) T. Ishi-i, M. A. Mateos-Timoneda, P. Timmerman, M. Crego-Calama, D. N. Reinhoudt and S. Shinkai, *Angew. Chem., Int. Ed.*, 2003, **42**, 2300–2305; (*d*) Y. Ferrand, M. P. Crump and A. P. Davis, *Science*, 2007, **318**, 619–622; (*e*) Y. Ferrand, E. Klein, N. P. Barwell, M. P. Crump, J. Jiménz-Barbero, C. Vicent, G.-J. Boons, S. Ingale and A. P. Davis, *Angew. Chem., Int. Ed.*, 2009, **48**, 1775–1779; (*f*) M. Mazik and M. Kuschel, *Chem.–Eur. J.*, 2008, **14**, 2405–2419; (*g*) W. Lu, L.-H. Zhang, X.-S. Ye, J. Su and Z. Yu, *Tetrahedron*, 2006, **62**, 1806–1816; (*h*) C. Nativi, M. Cacciarini, O. Francesconi, A. Vacca, G. Moneti, A. Ienco and S. Roelens, *J. Am. Chem. Soc.*, 2007, **129**, 4377–4385; (*i*) P. B. Palde, P. C. Gareiss and B. L. Miller, *J. Am. Chem. Soc.*, 2008, **130**, 9566–9573.

3 Saccharide recognition by synthetic helical foldamers: (*a*) C. Li, G.-T. Wang, H.-P. Yi, X.-K. Jiang, Z.-T. Li and R.-X. Wang, *Org. Lett.*, 2007, **9**, 1797–1800; (*b*) W. Cai, G.-T. Wang, P. Du, R.-X. Wang, X.-K. Jiang and Z.-T. Li, *J. Am. Chem. Soc.*, 2008, **130**, 13450–13459; (*c*) H. Goto, Y. Furusho and E. Yashima, *J. Am. Chem. Soc.*, 2007, **129**, 9168–9174.

4 Reviews of helical foldamers: (*a*) *Foldamers*, ed. S. Hecht and I. Huc, Wiley-VCH, Weinheim, 2007; (*b*) D. J. Hill, M. J. Mio, R. B. Prince, T. S. Hughes and J. S. Moore, *Chem. Rev.*, 2001, **101**, 3893–4011; (*c*) T. Nakano and Y. Okamoto, *Chem. Rev.*, 2001, **101**, 4013–4038; (*d*) E. Yashima, K. Maeda and T. Nishimura, *Chem.–Eur. J.*, 2004, **10**, 42–51; (*e*) I. Huc, *Eur. J. Org. Chem.*, 2004, 17–29; (*f*) C. R. Ray and J. S. Moore, *Adv. Polym. Sci.*, 2005, **177**, 91–149; (*g*) K. Maeda and E. Yashima, *Top. Curr. Chem.*, 2006, **265**, 47–88; (*h*) E. Yashima and K. Maeda, *Macromolecules*, 2008, **41**, 3–12; (*i*) H.-J. Kim, Y.-B. Lim and M. Lee, *J. Polym. Sci., Part A: Polym. Chem.*, 2008, **46**, 1925–1935; (*j*) B. Gong, *Acc. Chem. Res.*, 2008, **41**, 1376–1386.

5 (*a*) M. Inouye, M. Waki and H. Abe, *J. Am. Chem. Soc.*, 2004, **126**, 2022–2027; (*b*) H. Abe, N. Masuda, M. Waki and M. Inouye, *J. Am. Chem. Soc.*, 2005, **127**, 16189–16196; (*c*) M. Waki, H. Abe and M. Inouye, *Chem.–Eur. J.*, 2006, **12**, 7839–7847; (*d*) M. Waki, H. Abe and M. Inouye, *Angew. Chem., Int. Ed.*, 2007, **46**, 3059–3061; (*e*) H. Abe, H. Machiguchi, S. Matsumoto and M. Inouye, *J. Org. Chem.*, 2008, **73**, 4650–4661.

6 (*a*) S. Shinkai and O. Manabe, *Top. Curr. Chem.*, 1984, **121**, 67–104; (*b*) G. W. Gokel, W. M. Leevy and M. E. Weber, *Chem. Rev.*, 2004, **104**, 2723–2750; (*c*) V. Balzani, M. Venturi and A. Credi, *Molecular Devices and Machines*, Wiley-VCH, Weinheim, 2003; (*d*) E. R. Kay, D. A. Leigh and F. Zerbetto, *Angew. Chem., Int. Ed.*, 2007, **46**, 72–191.

7 (*a*) P. R. Ashton, E. J. T. Chrystal, P. T. Glink, S. Menzer, C. Schiavo, N. Spencer, J. F. Stoddart, P. A. Tasker, A. J. P. White and D. J. Williams, *Chem.–Eur. J.*, 1996, **2**, 709–728; (*b*) S. J. Rowan and J. F. Stoddart, *Polym. Adv. Technol.*, 2002, **13**, 777–787; (*c*) W. R. Dichtel, O. S. Miljanić, W. Zhang, J. M. Spruell, K. Patel, I. Aprahamian, J. R. Heath and J. F. Stoddart, *Acc. Res. Chem.*, 2008, **41**, 1750–1761; (*d*) F. Huang and H. W. Gibson, *Prog. Polym. Sci.*, 2005, **30**, 982–1018; (*e*) S. Kawano, N. Fujita and S. Shinkai, *Chem. Commun.*, 2003, 1352–1353.

8 Examples of helical polymers bearing crown moieties: (*a*) K. Morino, M. Oobo and E. Yashima, *Macromolecules*, 2005, **38**, 3461–3468; (*b*) T. Miyagawa, M. Yamamoto, R. Muraki, H. Onouchi and E. Yashima, *J. Am. Chem. Soc.*, 2007, **129**, 3676–3682; (*c*) R. Sakai, T. Satoh, R. Kakuchi, H. Kaga and T. Kakuchi, *Macromolecules*, 2003, **36**, 3709–3713; (*d*) R. Kakuchi, R. Sakai, I. Otsuka, T. Satoh, H. Kaga and T. Kakuchi, *Macromolecules*, 2005, **38**, 9441–9447.

9 H. Abe, D. Murayama, F. Kayamori and M. Inouye, *Macromolecules*, 2008, **41**, 6903–6909.

10 Examples of helix stabilization accompanied by hypochromism: (*a*) J. C. Nelson, J. G. Saven and J. S. Moore, *Science*, 1997, **277**, 1793–1796; (*b*) L. Brunsveld, E. W. Meijer, R. B. Prince and J. S. Moore, *J. Am. Chem. Soc.*, 2001, **123**, 7978–7984; (*c*) M. Ohkita, J.-M. Lehn, G. Baum and D. Fenske, *Chem.–Eur. J.*, 1999, **5**, 3471–3481; (*d*) S. Hecht and A. Khan, *Chem.–Eur. J.*, 2006, **12**, 4764–4774; (*e*) R. M. Meudtner and S. Hecht, *Angew. Chem., Int. Ed.*, 2008, **47**, 4926–4930.

11 Binding constants were determined on the assumption that the molecular weight of the polymer was uniform and that the polymer and the guest associate in a 1 : 1 ratio. Deviation was observed between plots and fitted curves in Fig. S5‡ possibly due to variation of the molecular length.