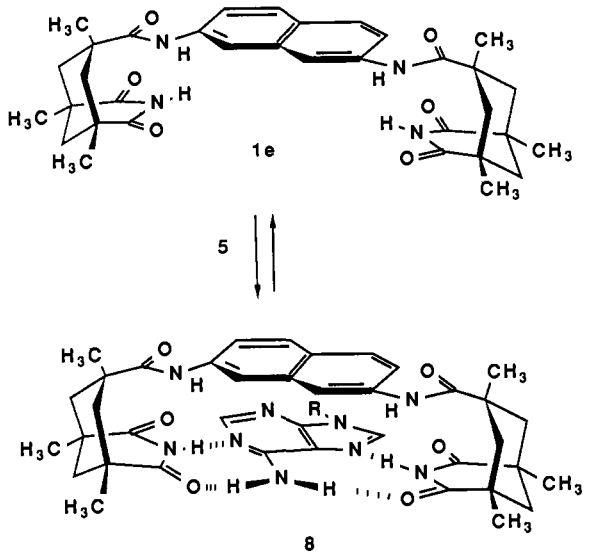

(4)

forces can result in high selectivity in model systems for molecular recognition.

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Induced Fit in Synthetic Receptors: Nucleotide Base Recognition by a "Molecular Hinge"

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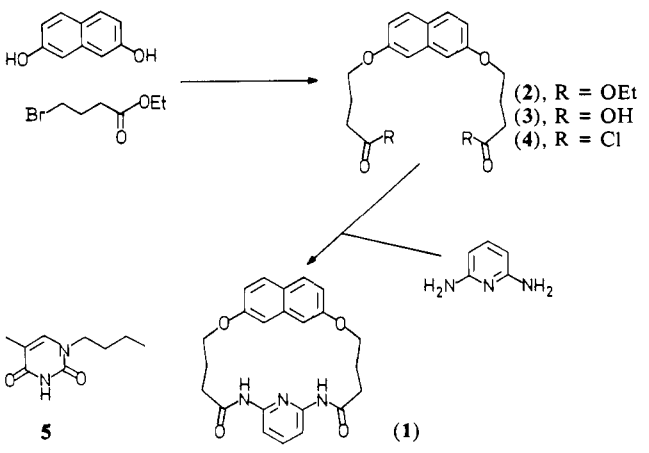
The development of molecules that recognize and bind to specific nucleotides or nucleotide base pairs provides an important goal in contempory bioorganic chemistry.[1] A key element in the design of such specific receptors concerns the incorporation of several recognition features (e.g., hydrogen bonding, hydrophobic or electrostatic) that complement the chemical characteristics of the target. This multiple point binding is dramatically seen in the enzyme ribonuclease T_1 which binds its nucleotide substrate via both hydrogen bonding and hydrophobic interactions.[2] In addition to two hydrogen bonds, formed between N-1 and O-6 of the guanine and two amide groups on the peptide backbone, hydrophobic stacking occurs between the aromatic nucleotide base and a tyrosine residue (Tyr 45).[3] Similarly many natural[5] and synthetic[6] DNA binding molecules and artificial DNA cleaving agents[7] employ both intercalation (hydrophobic) and hydrogen bonding interactions.

As part of a program aimed at the preparation of synthetic receptors for biologically active molecules[8] we have sought to incorporate multiple recognition sites into a new class of nucleic acid binding molecules.[9] Our strategy is to assemble hydrogen bonding and hydrophobic groups (and ultimately electrostatic or reactive groups) within a macrocyclic structure that can form a cavity complementary to the nucleotide base substrate. We report here the synthesis, structure, and complexation properties of a macrocycle **1**, containing 2,6-diamidopyridine and naphthalene components, that shows two-point binding to thymine derivatives. Host **1** was also designed to test the possibility of inducing a conformational change on substrate binding which would place a naphthalene ring directly above the bound substrate.

Reaction of 2,7-naphthalene diol with ethyl 4-bromobutyrate (K_2CO_3, acetone, reflux) gave diester **2**, (83% yield) which was



then hydrolyzed (acetone, HCl) to diacid **3** (98% yield). Treatment of **3** with oxalyl chloride (CH_2Cl_2) afforded diacid chloride **4** which was not isolated but directly cyclized under high dilution conditions (CH_2Cl_2, Et_3N) with 2,6-diaminopyridine[10] to form macrocyclic host **1** (26% yield).[11] The structure of **1** was confirmed by single-crystal X-ray analysis (Figure 1a) which shows an open conformation with the naphthalene poised away from the pyridine ring at an inter-plane angle of 127.5°. In addition the amide hydrogens lie in the plane of the pyridine and project under the naphthalene ring to provide a partially organized substrate binding region.

Treatment of a $CDCl_3$ solution of **1** with 1 equiv of 1-butylthymine **5**[12,13] results in several characteristic changes in the ^1H NMR spectrum.[14] The NH protons on both **1** and **5** are shifted

(1) For general reviews, see: Dervan, P. D. *Science (Washington, DC)* **1986**, *232*, 464. Wakelin, I. P. G. *Med. Res. Rev.* **1986**, *6*, 275. Neidle, S. *Prog. Med. Chem.* **1979**, *16*, 151.

(2) Heinemann, U.; Saenger, W. *Nature (London)* **1982**, *299*, 27.

(3) Distance between parallel guanine and Tyr 45 is 3.5 Å.[2] The electron density map of ribonuclease T_1[4] shows an alternative, nonstacking position for Tyr 45. This is probably due to the unbound form and suggests that on substrate binding a conformational change in the enzyme occurs to swing Tyr 45 into a stacking position. Thus, in addition to playing a role in recognition Tyr 45 also acts to lock in or "gate" the substrate.

(4) Heinemann, U.; Wernitz, M.; Pahler, A.; Saenger, W.; Menke, G.; Ruterjans, H. *Eur. J. Biochem.* **1980**, *109*, 109.

(5) For example, actinomycin see Sobell et al., (Sobell, H. M.; Jain, S. C. *J. Mol. Biol.* **1972**, *68*, 28.) or daunomycin see Quigley et al. (Quigley, G. J.; Wang, A. H.-J.; Ughetto, G.; van der Marel, G.; van Boom, J. H.; Rich, A. *Proc. Natl. Acad. Sci. U.S.A.* **1980**, *77*, 7204.).

(6) For example, mitoxantrone, see: Kotovych, G.; Lown, J. W.; Tong, J. P. K. *J. Biomolec. Struct. Dyn.* **1986**, *4*, 111. Islam, S. A.; Neidle, S.; Gandecha, B. M.; Partridge, M.; Patterson, L. H.; Brown, J. R. *J. Med. Chem.* **1985**, *28*, 857.

(7) Dervan, P. D.; Sluka, J. P. *Proceedings of the International Kyoto Conference on Organic Chemistry*; Elsevier: Amsterdam, 1986; p 307.

(8) Hamilton, A. D.; Kazanjian, P. *Tetrahedron Lett.* **1985**, 5735. Mann, M.; Hamilton, A. D.; Pant, N. *J. Chem. Soc., Chem. Commun.* **1986**, 158.

(9) For other synthetic hosts containing more than a single recognition site see, for example: Sheridan, R. E.; Whitlock, H. W., Jr. *J. Am. Chem. Soc.* **1986**, *108*, 7120. Willner, I.; Goren, Z. *J. Chem. Soc., Chem. Commun.* **1983**, 1469. Tabushi, I.; Shimizu, N.; Sugimoto, T.; Shiozuka, M.; Yamamura, K. *J. Am. Chem. Soc.* **1977**, *99*, 7100. Rebek, J.; Nemeth, D. *J. Am. Chem. Soc.* **1986**, *108*, 5637. Kimura, E.; Fujioka, H.; Kodama, M. *J. Chem. Soc., Chem. Commun.* **1986**, 1158. Schmitchen, F. P. *J. Am. Chem. Soc.* **1986**, *108*, 8249.

(10) Freshly sublimed. For another macrocycle containing 2,6-diaminopyridine units, see: Weber, E.; Vogtle, F. *Liebig Ann. Chem.* **1976**, 891.

(11) **1**: ^1H NMR ($CDCl_3$) 7.98 (2 H, d, J = 8 Hz, py 3, 5 H), 7.75 (1 H, t, J = 8 Hz, py 4 H), 7.74 (2 H, br s, NH), 7.69 (2 H, d, J = 8 Hz, naph 4, 5 H), 7.11 (2 H, d, J = 2 Hz, naph 1, 8 H), 7.05 (2 H, dd, J = 2, 8 Hz, naph 3, 6 H), 4.30 (4 H, t, J = 6.5 Hz, CH_2O), 2.52 (4 H, m. CH_2CO), 2.25 (4 H, m, CCH_2C).

(12) Prepared by alkylating thymine with butyl bromide (Me_2SO, K_2CO_3), see: Browne, D. T. In *Synthetic Procedures in Nucleic Acid Chemistry*; Zorbach, W. W., Tipson, R. S., Eds.; Interscience: New York, 1968; p 98.

(13) **5**: ^1H NMR ($CDCl_3$) 8.02 (1 H, br s, NH), 7.00 (1 H, s, th 6 H), 3.71 (2 H, t, J = 6.5 Hz, NCH_2), 1.93 (3 H, s, ring CH_3), 1.65 (2 H, m, NCH_2CH_2), 1.36 (2 H, m, CH_3CH_2), 0.96 (3 H, t, J = 8 Hz, CH_2CH_3).

(14) Complex (1:1) between **1** and **5**: ^1H NMR ($CDCl_3$) 10.58 (1 H, br s, th NH), 9.92 (2 H, br s, py NH), 8.05 (2 H, d, J = 8 Hz, py 3, 5 H), 7.79 (1 H, t, J = 8 Hz, py 4 H), 7.46 (2 H, d, J = 8 Hz, naph 4, 5 H), 7.86 (4 H, m, naph 1, 3, 6, 8 H), 6.71 (1 H, s, th H), 4.20 (4 H, t, J = 4.5 Hz, OCH_2), 3.47 (2 H, t, J = 8 Hz, th NCH_2), 2.64 (4 H, m, CH_2CO), 2.24 (4 H, m, CCH_2C), 1.74 (3 H, s, th ring CH_3), 1.55 (2 H, m, th NCH_2CH_2), 1.30 (2 H, m, CH_3CH_2), 0.96 (3 H, t, J = 8 Hz, CH_2CH_3).

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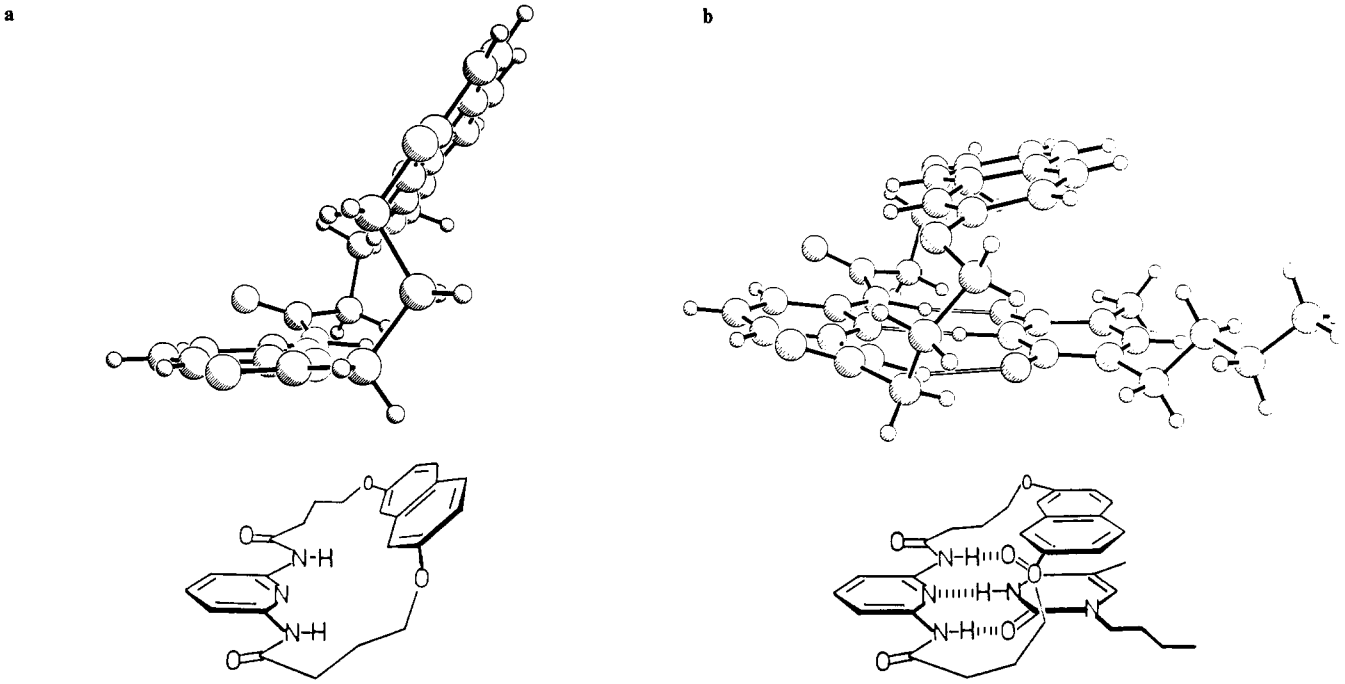

Figure 1. (a) X-ray structure of **1** and (b) X-ray structure of the complex between **1** and **5**.

downfield by 2.25 and 2.6 ppm, respectively, reflecting the formation of a triple hydrogen-bonded complex.[15] However, upfield shifts (of approximately 0.3 ppm) are seen in the thymine-6-proton, -ring methyl, and -*N*-methylene resonances while no significant shift is found for the alkyl methyl group. The selective upfield shifts of certain substrate protons are consistent with the close approach of the naphthalene to the substrate and its participation in binding.[16,17] These results are in contrast to those with 2,6-dibutyramidopyridine[15] which shows downfield shifts of its NH protons on hydrogen bonding to **5** but exhibits no upfield shifts in the substrate protons.

The structure of the complex **1**:**5** was confirmed by X-ray crystallography (Figure 1b).[18] Three hydrogen bonds are formed between the pyridine and thymine rings with distances of (N···N) 3.06, (N···O) 2.87, and 2.99 Å which are comparable to those in related complexes.[15] The naphthalene now lies approximately parallel (14°) to the plane of the thymine substrate at a closest inter-plane contact of 3.37 Å. The position of the naphthalene directly above the substrate accounts for the upfield shift of those protons on the periphery of the thymine and the absence of a shift on those protons distant from the naphthalene ring current. The angle between the pyridine and naphthalene planes is now 161.6°. Thus, on substrate complexation, **1** acts like a "molecular hinge" and swings the naphthalene unit through a 34.1° arc to within van der Waals distance of the thymine ring. This induced fit behavior in a synthetic molecule directly mimics the recognition of nucleotides by ribonuclease T_1 in which a tyrosine residue moves into place above the bound guanine.[2]

In summary, we have developed a new class of biomimetic receptors for nucleotide base substrates that employs the recognition strategy of substrate-induced organization of the binding site (induced fit). We are currently extending this approach to the other important nucleotide bases as well as to hydrogen-bonded nucleotide base pairs.

Acknowledgment. We thank the National Institutes of Health for partial support (GM-35208) of this work and Bart Kahr for his assistance with the X-ray analyses.

Supplementary Material Available: Crystallographic details for **1** and **1**:**5** including tables of atomic coordinates, thermal parameters, bond angles, and bond lengths (22 pages). Ordering information is given on any current masthead page.

(15) Feibush, B.; Figueroa, A.; Charles, R.; Onan, K. D.; Feibush, P.; Kargar, B. L. *J. Am. Chem. Soc.* **1986**, *108*, 3310.

(16) This two-point binding is confirmed by the association constants (K_s) which show an increase from 90 M^{-1} for simple 2,6-dibutyramidopyridine[15] to 290 M^{-1} for **1**. K_s measurements were made by monitoring the chemical shifts of the amide protons as a function of 1-butylthymine concentration and analyzing the data by the Foster-Fife modification of the Scatchard plot. Foster, R.; Fife, C. A. *Prog. Nucl. Magn. Reson. Spectrosc.* **1969**, *4*, 1.

(17) For other molecular complexes involving induced aromatic stacking interactions, see: Colquhoun, H. M.; Goodings, E. P.; Maud, J. M.; Stoddart, J. F.; Williams, D. J.; Wolstenholme, J. B. *J. Chem. Soc., Chem. Commun.* **1983**, 1140 and references therein.

(18) Crystals grown by slow evaporation of a 1:1 mixture of **1** and **5** in methylene chloride-heptane.

Far-Ultraviolet Resonance Raman Scattering: Highly Excited Torsional Levels of Ethylene

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Ultraviolet resonance Raman scattering[1-3] has proven to be a powerful technique for the determination of electronic state symmetry,[4-6] vibronic coupling,[7] geometry changes,[8-10] and dissociation dynamics[11,12] of excited electronic states. These Raman spectra often exhibit transitions to highly excited vibrational levels,[2,8] providing new information about the ground electronic state. In a previous study,[10] the Raman spectrum of ethylene was

(1) Hudson, B. *Spectroscopy* **1986**, *1*, 22-31.
(2) Hudson, B.; Kelly, P. B.; Ziegler, L. D.; Desiderio, R. A.; Gerrity, D. P.; Hess, W.; Bates, R. In *Advances in Laser Spectroscopy*; Garetz, B. A., Lombardi, J. R., Eds.; John Wiley & Sons: New York, 1986; Vol. 3, pp 1-32.
(3) Hudson, B. In *Time-Resolved Vibrational Spectroscopy*; Laubereau, A.; Stockburger, M., Eds.; Springer-Verlag: Berlin, 1985; pp 170-174.
(4) Gerrity, D. P.; Ziegler, L. D.; Kelly, P. B.; Desiderio, R. A.; Hudson, B. *J. Chem. Phys.* **1985**, *83*, 3209-3213.
(5) Chadwick, R. R.; Gerrity, D. P.; Hudson, B. *Chem. Phys. Lett.* **1985**, *115*, 24-28.
(6) Ziegler, L. D.; Hudson, B. *J. Chem. Phys.* **1981**, *74*, 982-992.
(7) Ziegler, L. D.; Hudson, B. *J. Chem. Phys.* **1983**, *79*, 1134-1137.
(8) Desiderio, R. A.; Gerrity, D. P.; Hudson, B. *Chem. Phys. Lett.* **1985**, *115*, 29-33.
(9) Ziegler, L. D.; Hudson, B. *J. Phys. Chem.* **1984**, *88*, 1110-1116.
(10) Ziegler, L. D.; Hudson, B. *J. Chem. Phys.* **1983**, *79*, 1197-1202.
(11) Ziegler, L. D.; Kelly, P. B.; Hudson, B. *J. Chem. Phys.* **1984**, *81*, 6399-6400.
(12) Ziegler, L. D. *J. Chem. Phys.* **1986**, *84*, 6013-6026.

0002-7863/87/1509-5036$01.50/0