

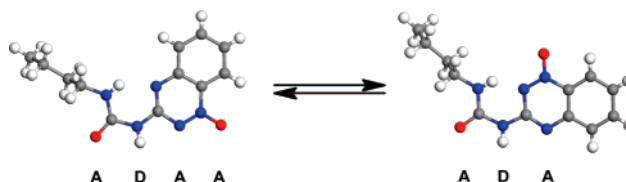
Ureidobenzotriazine Multiple H-Bonding Arrays: The Importance of Geometrical Details on the Stability of H-Bonds

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Received September 3, 2007



A 3-ureidobenzo-1,2,4-triazine 1-*N*-oxide (**1**) was synthesized successfully. The derivative displays an acceptor–donor–acceptor–acceptor (ADAA) hydrogen-bonding motif in CDCl₃ and DMSO-*d*₆ solution as well as in the solid state. Although moderately strong association of **1** was observed with DAD motifs, nonspecific binding is observed with ureidopyridines featuring a complementary DADD array. Density functional calculations of conformations **1a** and **1b** together with two complexes revealed the clearly nonplanar geometry of the multiply hydrogen-bonded complex, in which some bonds are significantly longer (3.2 Å) than is optimal for H-bonds. As a result, only very small free energies of association were calculated, in line with the experimentally observed absence of specific assembly of the components.

Introduction

Inspired by the generic storage mechanism of DNA, arrays of multiple parallel or near-parallel hydrogen bonds have been commonly used motifs in molecular recognition for the last 20 years,^{1–5} since strength, specificity, and directionality are increased compared to single hydrogen bonds. The strength of multiple hydrogen-bonded complexes has been found to depend not only on the number of hydrogen bonds—and the donor acidity and acceptor basicity of the individual hydrogen bonds^{6–9}—but also strongly on the particular arrangement of the donor and acceptor functional groups.^{10–12} During the past

decade, a large number of strongly bonded complexes held together by more than three hydrogen bonds have been described.^{13–17} In particular, strongly associating self-complementary molecules developed by our group have been of

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(1) Hamilton, A. D. *Adv. Supramol. Chem.* **1990**, 1, 1–64.

(2) Vögtle, F. *Supramolecular Chemistry*; J. Wiley & Sons: Chichester, UK, 1991.

(3) Lehn, J.-M. *Supramolecular Chemistry*; Wiley-VCH: New York, 1995.

(4) Sijbesma, R. P.; Meijer, E. W. *Chem. Commun.* **2003**, 5–16.

(5) Zimmerman, S. C.; Corbin, P. S. *Struct. Bonding (Berlin)* **2000**, 96, 63–94.

(6) Taft, R. W.; Gurka, D.; Joris, L.; Schleyer, P. v. R.; Rakshys, J. W. *J. Am. Chem. Soc.* **1969**, 91, 4801–8.

(7) Taft, R. W.; Berthelot, M.; Laurence, C.; Leo, A. J. *Chemtech* **1996**, 26, 20–29.

(8) Abraham, M. H. *J. Phys. Org. Chem.* **1993**, 6, 660–84.

(9) Platts, J. A. *Phys. Chem. Chem. Phys.* **2000**, 2, 3115–3120.

(10) Jorgensen, W. L.; Pranata, J. *J. Am. Chem. Soc.* **1990**, 112, 2008–10.

(11) Pranata, J.; Wierschke, S. G.; Jorgensen, W. L. *J. Am. Chem. Soc.* **1991**, 113, 2810–19.

(12) Murray, T. J.; Zimmerman, S. C. *J. Am. Chem. Soc.* **1992**, 114, 4010–11.

(13) Beijer, F. H.; Kooijman, H.; Spek, A. L.; Sijbesma, R. P.; Meijer, E. W. *Angew. Chem., Int. Ed.* **1998**, 37, 75–78.

(14) Beijer, F. H.; Sijbesma, R. P.; Kooijman, H.; Spek, A. L.; Meijer, E. W. *J. Am. Chem. Soc.* **1998**, 120, 6761–6769.

(15) Söntjens, S. H. M.; Sijbesma, R. P.; van Genderen, M. H. P.; Meijer, E. W. *J. Am. Chem. Soc.* **2000**, 122, 7487–7493.

(16) Chang, S. K.; Hamilton, A. D. *J. Am. Chem. Soc.* **1988**, 110, 1318–19.

(17) Zeng, H.; Miller, R. S.; Flowers, R. A., II; Gong, B. *J. Am. Chem. Soc.* **2000**, 122, 2635–2644.

eminent importance for the development of main-chain supramolecular polymers.^{18–22} However, the self-complementarity of these units imposes restrictions to the self-assembly of copolymers and the construction of supramolecular architectures consisting of more than one compound. Given our research interest in multiple hydrogen-bonding units as well as the recent use of complementary recognition motifs in supramolecular polymers, there is a clear need for strong complementary binding motifs. A number of complementary multiple hydrogen-bonding arrays have been developed, including ones based on oligoamides,¹⁷ hydrazides,²³ and pairs of heterocyclic building blocks, such as naphthyridine–ureidopyridine,²⁴ naphthyridine–ureidoguanosine,^{25,26} naphthyridine–ureidopyrimidinone,²⁷ anthyridine with dihydropyridine¹² or an azapentacene analog with dihydropyridine,²⁸ and heterocyclic homodimers combined with donor–acceptor interactions.^{29,30} In all these pairs, the individual components either lack the high synthetic accessibility required for use in supramolecular polymers or association to the heterocomplexes is not fully selective.^{31–36} In this report, a study into a new complementary hydrogen-bonding module is presented. The synthetic accessibility and binding capability of 3-ureido-1,2,4-triazine 1-*N*-oxide **1** and suitability as an acceptor–donor–acceptor–acceptor (ADAA) hydrogen-bonding scaffold is reported. *N*-Oxides of nitrogen heterocycles have a formally full negative charge on the oxygen atom, and pyridine *N*-oxide has been reported to be a stronger hydrogen-bond acceptor than pyridine.^{6,37–41} Therefore, strong binding by **1**

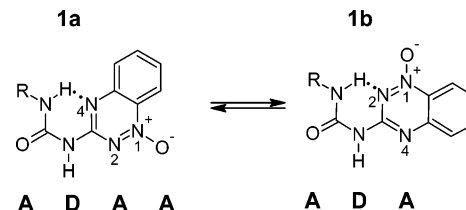


FIGURE 1. Two conformations of 3-ureido-1,2,4-triazine 1-*N*-oxide **1** and their corresponding hydrogen-bonding array.

would be expected. Although **1** can be present in two possible conformations, **1a** and **1b**, as displayed in Figure 1, the equilibrium might be shifted toward the ADAA form upon presenting to a complementary donor–acceptor–donor–donor (DADD) partner.

We complement these experimental studies by density functional quantum mechanical calculations to evaluate the experimentally found binding properties of the compound. Specifically, detailed geometric effects are studied, to reveal overall large effects on the hydrogen-bonding strength. This combination of experimental and theoretical data is then finally discussed in detail to highlight frequently overlooked aspects of binding in multiple hydrogen-bonding arrays: linearity of the array and planarity of the overall donor–acceptor complex.

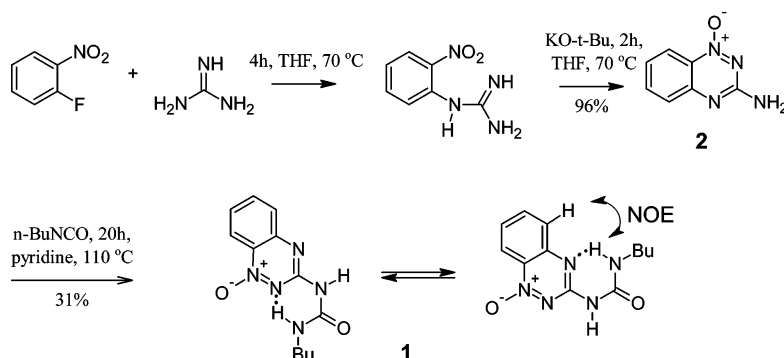
Results and Discussion

Synthesis and Characterization. Two general methods have been reported for the synthesis of 1,2,4-triazine *N*-oxides: by direct oxidation of the parent triazine with organic peroxides or by the formation of the triazine *N*-oxide ring via cyclization.⁴² 3-Aminobenzo-1,2,4-triazine 1-*N*-oxide (**2**) is a precursor to tirapazamine (3-aminobenzo-1,2,4-triazine 1,4-dioxide), which is a biochemically activated DNA-damaging agent that selectively kills the hypoxic cells found in solid tumors.^{43,44} A common method to obtain **2** involves a (violently exothermic) reaction of 2-nitroaniline with cyanamide.^{45–47} However, a more elegant approach was introduced by Suzuki et al., who reported the nucleophilic aromatic substitution between nitroarenes and free guanidine base followed by base-induced cyclization.^{48,49} The free guanidine base was prepared by passing an aqueous solution of guanidine hydrochloride through an ion exchange column (DOWEX 550A OH) and evaporation of the eluate in vacuo. 3-Aminobenzo-1,2,4-triazine 1-*N*-oxide (**2**) was subsequently obtained in 96% yield through reaction of *o*-fluoronitrobenzene with free guanidine followed by in situ cyclization with *t*-BuOK. Condensation of **2** with *n*-butyl isocyanate gave the desired product **1** as bright yellow crystals in 31% yield (Scheme 1).

- (18) 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.
- (19) Hirschberg, J. H. K. K.; Koevoets, R. A.; Sijbesma, R. P.; Meijer, E. W. *Chem. Eur. J.* **2003**, *9*, 4222–4231.
- (20) Sijbesma, R. P.; Beijer, F. H.; Brunsveld, L.; Folmer, B. J. B.; Hirschberg, J. H. K. K.; Lange, R. F. M.; Lowe, J. K. L.; Meijer, E. W. *Science* **1997**, *278*, 1601–1604.
- (21) Ky Hirschberg, J. H. K.; Beijer, F. H.; van Aert, H. A.; Magusin, P. C. M. M.; Sijbesma, R. P.; Meijer, E. W. *Macromolecules* **1999**, *32*, 2696–2705.
- (22) Folmer, B. J. B.; Sijbesma, R. P.; Versteegen, R. M.; van der Rijt, J. A. J.; Meijer, E. W. *Adv. Mater.* **2000**, *12*, 874–878.
- (23) Zhao, X.; Wang, X.-Z.; Jiang, X.-K.; Chen, Y.-Q.; Li, Z.-T.; Chen, G.-J. *J. Am. Chem. Soc.* **2003**, *125*, 15128–15139.
- (24) Luning, U.; Kuhl, C. *Tetrahedron Lett.* **1998**, *39*, 5735–5738.
- (25) Park, T.; Todd, E. M.; Nakashima, S.; Zimmerman, S. C. *J. Am. Chem. Soc.* **2005**, *127*, 18133–18142.
- (26) Ong, H. C.; Zimmerman, S. C. *Org. Lett.* **2006**, *8*, 1589–1592.
- (27) Li, X.-Q.; Jiang, X.-K.; Wang, X.-Z.; Li, Z.-T. *Tetrahedron* **2004**, *60*, 2063–2069.
- (28) Djurdjevic, S.; Leigh, D. A.; McNab, H.; Parsons, S.; Teobaldi, G.; Zerbetto, F. *J. Am. Chem. Soc.* **2007**, *129*, 476–477.
- (29) Wang, X.-Z.; Li, X.-Q.; Shao, X.-B.; Zhao, X.; Deng, P.; Jiang, X.-K.; Li, Z.-T.; Chen, Y.-Q. *Chem. Eur. J.* **2003**, *9*, 2904–2913.
- (30) Li, X.-Q.; Feng, D.-J.; Jiang, X.-K.; Li, Z.-T. *Tetrahedron* **2004**, *60*, 8275–8284.
- (31) Söntjens, S. H. M.; Meijer, J. T.; Kooijman, H.; Spek, A. L.; van Genderen, M. H. P.; Sijbesma, R. P.; Meijer, E. W. *Org. Lett.* **2001**, *3*, 3887–3889.
- (32) Brammer, S.; Luning, U.; Kuhl, C. *Eur. J. Org. Chem.* **2002**, 4054–4062.
- (33) Corbin, P. S.; Zimmerman, S. C.; Thiessen, P. A.; Hawryluk, N. A.; Murray, T. J. *J. Am. Chem. Soc.* **2001**, *123*, 10475–10488.
- (34) Mayer, M. F.; Nakashima, S.; Zimmerman, S. C. *Org. Lett.* **2005**, *7*, 3005–3008.
- (35) Yang, X.; Martinovic, S.; Smith, R. D.; Gong, B. *J. Am. Chem. Soc.* **2003**, *125*, 9932–9933.
- (36) Gong, B. *Synlett* **2001**, 582–589.
- (37) Kulevsky, N.; Lewis, L. *J. Phys. Chem.* **1972**, *76*, 3502–3.
- (38) Dega-Szafran, Z.; Kania, A.; Grundwald-Wyspianska, M.; Szafran, M.; Tykarska, E. *J. Mol. Struct.* **1996**, *381*, 107–125.
- (39) Prezhdo, V. V.; Vashchenko, E. V.; Prezhdo, O. V. *Russ. J. Gen. Chem. (Translation of Zh. Obshch. Khim.)* **2000**, *70*, 121–129.
- (40) Moreno-Fuquen, R.; De Castro, E. V. R.; Moreno, M.; De Almeida Santos, R. H.; Montano, A. M. *Acta Crystallogr.* **2000**, *C56*, 206–207.

- (41) Goswami, S.; Ghosh, K.; Mukherjee, R. *Supramol. Chem.* **2000**, *11*, 191–199.
- (42) Kozhevnikov, D. N.; Rusinov, V. L.; Chupakhin, O. N. *Adv. Heterocycl. Chem.* **2002**, *82*, 261–305.
- (43) Birincioglu, M.; Jaruga, P.; Chowdhury, G.; Rodriguez, H.; Dizdareoglu, M.; Gates, K. S. *J. Am. Chem. Soc.* **2003**, *125*, 11607–11615.
- (44) Junnotula, V.; Sarkar, U.; Sinha, S.; Inman, C.; Barnes, C. L.; Gates, K. S. *Abstracts of Papers*, 230th ACS National Meeting, Washington, DC, Aug 28–Sept 1, 2005, TOXI-075.
- (45) Arndt, F. *Ber.* **1913**, *46*, 3522–3532.
- (46) Wolf, F. J.; Pfister, K., III; Wilson, R. M., Jr.; Robinson, C. A. *J. Am. Chem. Soc.* **1954**, *76*, 3551–3.
- (47) Wolf, F. J.; Wilson, R. M., Jr.; Pfister, K., 3rd; Tishler, M. *J. Am. Chem. Soc.* **1954**, *76*, 4611–13.
- (48) Suzuki, H.; Kawakami, T. *Synthesis* **1997**, 855–857.
- (49) Suzuki, H.; Kawakami, T. *J. Org. Chem.* **1999**, *64*, 3361–3363.

SCHEME 1



Compound **1** can potentially exist in two conformers, each of which is characterized by a different intramolecular H-bond (structures **1a** and **1b**). It was anticipated that compound **1** would feature a ADAA array of hydrogen-bonding sites (conformation **1a**), presumably because in both conformers N4 bears a larger electron density than N2 (NPA charges roughly -0.6 and -0.4 , respectively). To elucidate the conformation in solution, nuclear Overhauser experiments using selective excitation were performed.⁵⁰ The presence of conformer **1a** in CDCl_3 and $\text{DMSO}-d_6$ was confirmed by observation of the intramolecular NOE contact indicated in Scheme 1. X-ray analysis of the product showed that the molecule features an intramolecular hydrogen bond from the urea NH to N4 in the triazine ring to display an AADA array of hydrogen-bonding sites in the solid state. The unit cell contains two independent molecules, one of which exhibits disorder in the butylureido group. The N–N distance between the nitrogen atoms involved in the intramolecular hydrogen bond (average distance $2.68 \text{ \AA}$) is larger than the C–C distance between the atoms connected to the central N–H (average distance $2.58 \text{ \AA}$). In addition, the corresponding C–N–C angles for the two independent molecules are 129.38°

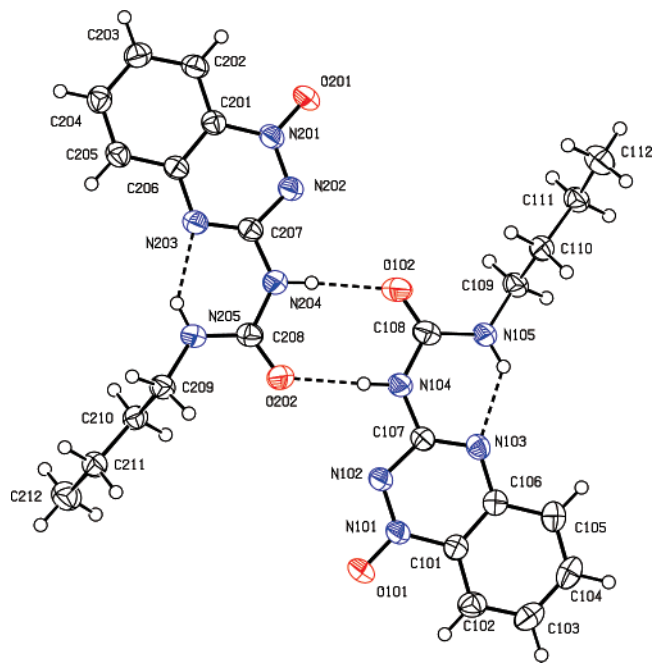


FIGURE 2. Displacement ellipsoid representation (50% probability level) of the single-crystal X-ray structure of the hydrogen-bonded dimer of compound **1**. Only the 80% disorder form of one of the two molecules is shown.

and 129.18° . These data are accurately mimicked by our calculations, with distances and angles of 2.78 and 132.19° , respectively [B3LYP/6-311G(d,p) data]. The molecule is also not fully planar, since the triazine ring is at an angle of 8.0° and 2.0° with the ureido group for the two nonequivalent molecules in the unit cell. Specifically, the observed deviations from linearity in the H-bond array, i.e., the fact that O₁₀₁, N₁₀₂, N₁₀₄, and O₁₀₂ are not on one line (see structure in Figure 2), may affect the capability of **1** to form multiple hydrogen-bonded arrays with complementary molecules.

Binding Studies. Since dimerization of **1** by double hydrogen bonds was observed in the solid state, dimerization in CDCl_3 solution was determined by ^1H NMR dilution experiments. Compound **1** was found to self-associate weakly ($K_{\text{dim}} < 5 \text{ M}^{-1}$ in CDCl_3 at 25°C). Association of **1** with compounds **3a,b** and **4a,b**, featuring DAD and DADD arrays, respectively, was investigated by ^1H NMR titration experiments (Chart 1). For comparison, association constants of compounds **3** and **4** with 1-*N*-propylthymine **5** are included in Table 1.

While binding of 1-*N*-propylthymine **5** to DAD molecules **3a** and **3b** is moderately strong (845 and 460 M^{-1} , respectively), compound **1** displays a lower binding constant (106 and 83 M^{-1} , respectively). Binding of these molecules may occur to the ADA part of the ADAA array of conformer **1a** or to the ADA array of conformer **1b** (see Figure 1). However, association of **1** with **3a** and **3b** is not accompanied with a significant shift in the intramolecular hydrogen bond, as the position of the intramolecular NH resonance does not shift by more than 0.1 ppm upon presenting the DAD partner. This indicates that no structural adjustment occurs upon binding and thus that the intramolecular hydrogen bond to N4 in the triazine ring is maintained. Moreover, no change in NOE contacts as indicated in Scheme 1 was observed. Binding of pyridylureas **4** to **1a** and **1b** forming DADD–ADAA arrays was investigated to determine the existence of favorable hydrogen bonding with the *N*-oxide. In solution, as well as in the solid state, pyridylureas are known to form a six-membered ring with an intramolecular hydrogen bond from one of the urea NHs to the nitrogen in the pyridine ring and double hydrogen bonds between two individual molecules.^{52,53} Upon complexation with a complementary binding motif, these bonds can be broken.^{53–55} The ability of **5** to bind to **4a** and **4b** confirms the low energetic penalty for

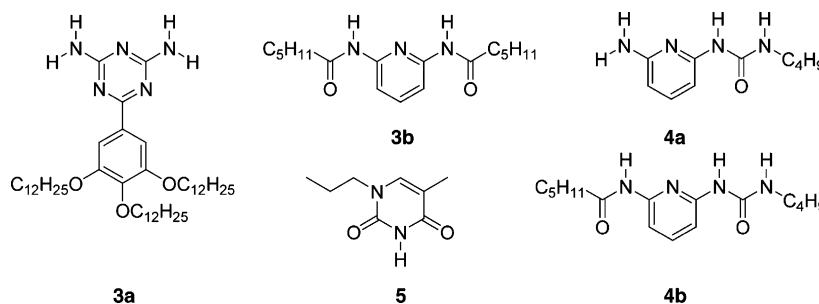
(50) Stott, K.; Keeler, J.; Van, Q. N.; Shaka, A. J. *J. Magn. Reson.* **1997**, *125*, 302–324.

(51) Beijer, F. H. Eindhoven University of Technology, Thesis, 1998.

(52) Sudha, L. V.; Sathyanarayana, D. N. *J. Mol. Struct.* **1984**, *125*, 89–96.

(53) Corbin, P. S.; Zimmerman, S. C. *J. Am. Chem. Soc.* **2000**, *122*, 3779–3780.

CHART 1

**TABLE 1.** Association Constants and Free Energies of Association of **1** and **5** with **3** and **4** Determined in CDCl₃ at 25 °C

1			5		
compd	K_a (M ⁻¹)	$-\Delta G^\circ$ (kJ/mol)	probe (signal)	K_a (M ⁻¹)	$-\Delta G^\circ$ (kJ/mol)
3a	106 ± 13	11.5 ± 0.3	3a (NH ₂) (1.7) ^a	845 ^d	16.7
3b	83 ± 17	10.9 ± 0.5	3b (NH-amide) (1.8) ^a	460 ^e	15.2
4a	<10	<5 ^b	4a (NH ₂)	57 ^d	10.0
4b	<10	<5 ^c	4b (NH-amide)	110 ^d	11.6

^a CIS values (ppm) given in parentheses. ^b The fit was obtained for five data points due to broadening of the NH₂ protons. ^c The fit was obtained for five data points due to overlap in signals. ^d Literature data from Söntjens et al.³¹ ^e Literature data from Beijer et al.⁵¹

this process. However, ¹H NMR titrations of **1** with **4** show very low association constants (<10 M⁻¹). These surprising results suggest a weak to no ability of the *N*-oxide to form a hydrogen bond.

Quantum Mechanical Calculations. Even though the presence of an AADA array in **1** is indicated in solution and the crystal structure, formation of quadruply hydrogen-bonded complexes in solution was not found with seemingly complementary agents. Two questions might be answered with computational calculations: (i) is there a large difference in energy between conformations **1a** and **1b**, and (ii) what are the stabilities of hydrogen-bonded complexes of these conformations and the corresponding partners? Finally, the combination of experimental and theoretical data can be used to delineate an explanation for the absence of intermolecular hydrogen-bonding here and hence provide deeper insight into the phenomenon of multiple hydrogen bonding.

Previously, extensive theoretical investigations have been reported on hydrogen-bonded dimers.⁵⁶ More recently, multiple hydrogen-bonded systems, including DNA base pairs,^{57–59} have also been studied theoretically, in order to rationalize relative stabilities.^{10,60–63} Validation of studies of electronic energies

TABLE 2. Energy Differences Between **1a** and **1b** from Different Methods and Basis Sets^a

entry	method and basis set	ΔE^0 (kJ/mol)
1	B3LYP/6-311G(d,p)	2.81
2	B3LYP/6-311+G(2df,2p)//B3LYP/6-311G(d,p)	4.23
3	LMP2/6-311G(d,p)//B3LYP/6-311G(d,p)	3.77
4	LMP2/6-311+G(d,p)//B3LYP/6-311G(d,p)	1.84

^a At B3LYP/6-311G(d,p) level, $\Delta H^{298} = 2.77$ kJ/mol, $\Delta G^{298} = 2.10$ kJ/mol.

TABLE 3. Calculated Binding Abilities of the Hydrogen-Bonded Complexes under Study [B3LYP/6-311G(d,p); in kJ/mol] and Dipole Moments of the Complexes

entry	complex	ΔE^0	ΔH^{298}	ΔG^{298}	dipole moment (D)
1	1a · 4a	−69.18	−63.058	−9.79	10.53
2	1a · dap	−70.769	−64.16	−10.45	
3	1b · 4a	−62.40	−55.03	−0.63	8.33
4	1b · dap	−56.68	−53.94	4.31	

TABLE 4. Calculated Hydrogen-Bond Lengths in **1**·**4b** Complexes

atom pair	1a · 4a	O1–N3	N1–N4	N2–N5	O2–N6
bond length (Å)	2.907	3.150	3.195	3.041	
atom pair	1b · 4a	N1–N3	N2–N4	O1–N5	O1–N6
bond length (Å)	3.131	3.159	2.866	3.131	

showed that quantum mechanical techniques could be used to perform calculations on molecular systems of practical significance.^{60,61} More specifically, density functional B3LYP calculations recently confirmed the strong homodimerization of ureidopyrimidinone (UPy) molecules in the keto tautomer and enol tautomer. For the keto tautomer, experimental hydrogen-bond distances $r(N-N)$ and $r(N-O)$ are 2.97 and 2.76 Å,¹⁴ respectively, while the B3LYP/6-311G(d,p)-computed values are 3.01 and 2.78 Å, respectively.⁶¹ Since in those cases agreement within experimental error was found for these parameters, the same methods were used to evaluate differences in the possible conformations of **1** and stabilities of the hydrogen-bond complexes of **1** with complementary compounds **4a** and 2,6-diaminopyridine.

Geometry Optimization. The calculated energy difference between **1a** and **1b** at specific methods and basis sets is displayed in Table 2. The calculated energy differences between **1a** and **1b** is only 4.2 kJ/mol at the B3LYP/6-311G(d,p) level (entry 1). Further stepwise increase of the basis set size up to B3LYP/6-311+G(2df,2p) (entry 2) shows no changes, suggesting that the basis set at the B3LYP/6-311G(d,p) level is appropriate. At the B3LYP/6-311G(d,p) level, the free energy of **1a** is slightly lower than that of **1b** (~2.9 kJ/mol), which means that the equilibrium between the two conformations lies

(54) Luning, U.; Kuhl, C.; Uphoff, A. *Eur. J. Org. Chem.* **2002**, 4063–4070.

(55) Quinn, J. R.; Zimmerman, S. C. *Org. Lett.* **2004**, 6, 1649–1652.

(56) Hadzi, D. *Theoretical Treatments of Hydrogen Bonding*; Wiley: New York, 1997.

(57) Guerra, C. F.; Bickelhaupt, F. M. *Angew. Chem., Int. Ed.* **2002**, 41, 2092–2095.

(58) Asensio, A.; Kobko, N.; Dannenberg, J. J. *J. Phys. Chem. A* **2003**, 107, 6441–6443.

(59) Mignon, P.; Loverix, S.; Steyaert, J. *Nucleic Acids Res.* **2005**, 33, 1779–1789.

(60) Lukin, O.; Leszczynski, J. *J. Phys. Chem. A* **2002**, 106, 6775–6782.

(61) Guo, D.; Sijbesma, R. P.; Zuilhof, H. *Org. Lett.* **2004**, 6, 3667–3670.

(62) Kobko, N.; Dannenberg, J. J. *J. Phys. Chem. A* **2003**, 107, 10389–10395.

(63) Zuilhof, H.; Morokuma, K. *Org. Lett.* **2003**, 5, 3081–3084.

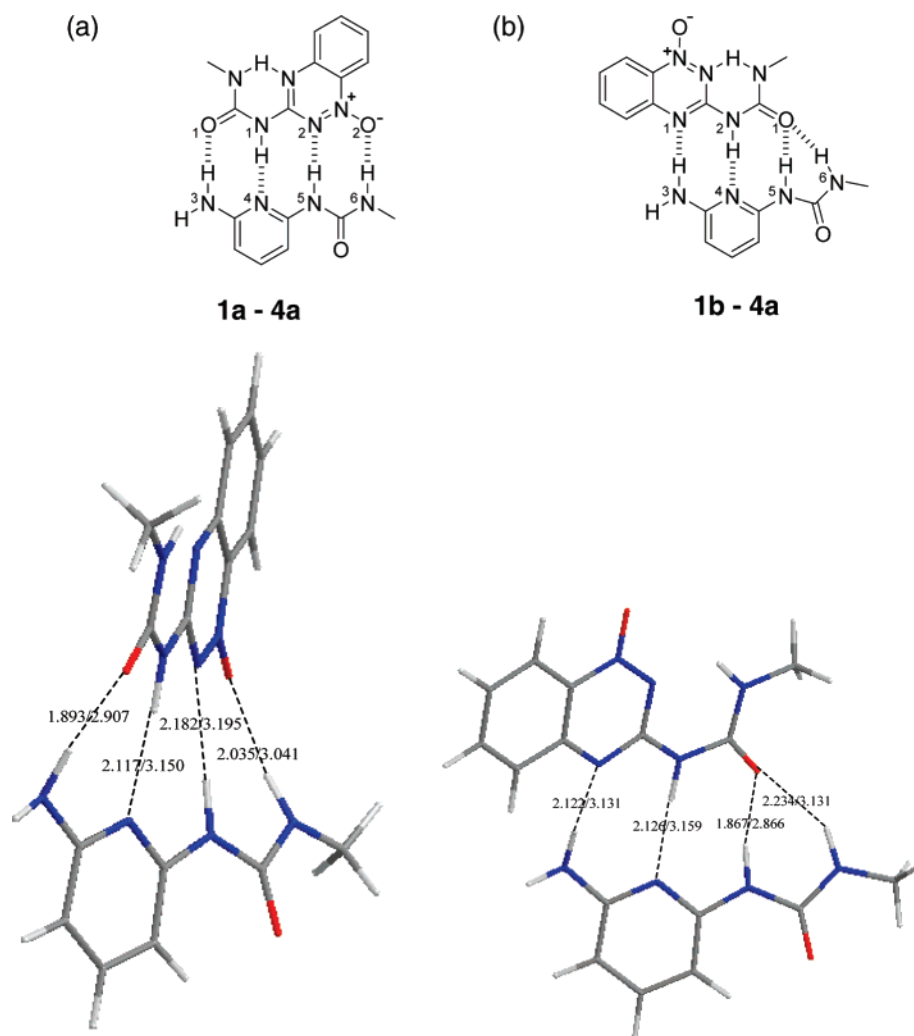


FIGURE 3. Optimized structures of complexes **1a·4a** (a) and **1b·4a** (b).

toward **1a**. Furthermore, the energy difference between the two conformations at the LMP2/6-311G(d,p)//B3LYP/6-311G(d,p) level is 3.77 kJ/mol, and at LMP2/6-311+G(d,p)//B3LYP/6-311G(d,p) level, **1a** is favored by 1.85 kJ/mol (entries 3 and 4). These data, obtained with several totally different high-level methods, all suggest that the two conformations of **1** display a similar stability, with a slightly lower energy of the ADAA array **1a**. While the NOE-NMR experiments reveal the presence of conformer **1a**, the small calculated energy differences suggest that conformations **1a** and **1b** may both exist in significant fractions in solution.

Hydrogen-Bonding Abilities of 1 with Complementary Motifs. The hydrogen-bonding combinations of **1a** and **1b** with both **4a** and 2,6-diaminopyridine (dap) have been fully optimized to obtain information about the structure and stability of the corresponding heterodimers. The calculated results are collected in Table 3. Remarkably, the free energies in all complexes of **1a** and **1b** are all close to zero, indicating that the hydrogen bonding in complexes of **1** is weak. This is unlike the situation reported earlier for the self-complexation of quadruply H-bonded ureidopyrimidone (UPy), which displays a strong H-bonding that is also computed. Therefore, the current data strongly suggest that little complex formation can be expected, as was indeed experimentally observed. Furthermore, a trend can be seen in the energy of complex formation in decreasing order:

1a·dap > **1a·4a** > **1b·4a** > **1b·dap**. This trend was mimicked when more sophistication was added in the computations, e.g., by going from total energies to free energies, i.e., inclusion of vibrational energy and intrinsic entropy effects.

Two reasons suggest that the low dimer stability should be attributed to intrinsic effects in the complex: First, the calculated dipole moments of the complexes (5–13 D) are much bigger than in UPy dimers (~0.2 D), which implies significant solvent stabilization of the dipole in complexes of **1**. However, the overall complexation energies are small, which points to an intrinsic rather than a solvent effect. Second, although differences are small, the computed data indicate that complexes of **1a** are slightly more stable than that of **1b**. Therefore, the lack of quadruple hydrogen-bond formation is not due to the absence of an ADAA array in **1**. The experimental observation that **1** prefers binding to a DAD array rather than to a DADD array is therefore due to the nature of the hydrogen-bonded complexes involved.

To compare the differences in structure, the hydrogen-bond lengths of heterodimers **1a·4a** and **1b·4a** have been collected in Table 4. A two- and three-dimensional picture of the optimized structures of the heterodimers are given in Figure 3. The computed hydrogen-bonding lengths of complex **1a·4a** are between 2.91 and 3.20 Å. These lengths are significantly longer than distances found by both X-ray determination as well as

analogous density functional and ab initio methods for strong multiple hydrogen-bonded motifs, which are between 2.70 and 3.00 Å.

An additional feature indicating weak association of the individual molecules is the significantly longer distances of the two middle hydrogen bonds. In this way, a concave molecule is formed, which is not capable of strong binding to the more linear pyridylurea **4a**. A similar explanation can be given for weak binding of **1b** to **4a**. Even though the carbonyl group of **1b** is involved in a bifurcated hydrogen bond to the urea of **4a**, hydrogen-bond lengths do not indicate strong binding. Furthermore, geometry optimization clearly shows that the individual molecules in the heterocomplexes are not planar. In fact, the angles between the two π -planes are 28.5° and 26.9° for complex **1a**·**4a** and **1b**·**4a**, respectively. These findings suggest that the electronic fine structure of **1** prevents the formation of ADAA·DADD motifs.

In addition, upon formation of the hydrogen-bond dimer, the structures of **1a** and **4a** need to be deformed with respect to their (planar) structure prior to hydrogen bonding, and this deformation energy amounts to almost 9 kJ/mol. Although this is not a major component, this deformation energy is almost zero in the case of, e.g., UPy–Upy dimers, which also contributes to the weaker hydrogen-bond formation.

Conclusions

We have shown that 3-butylureidobenzo-1,2,4-triazine 1-*N*-oxide (**1**), which is readily available by condensation of an isocyanate with 3-aminobenzo-1,2,4-triazine 1-*N*-oxide (**2**), displays an ADAA hydrogen-bonding motif. Although significant binding to a complementary DAD array is observed in chloroform solution, very weak association is found for seemingly complementary DADD arrays. Theoretical analysis of conformations **1a** (with an ADAA array) and **1b** (with an ADA array) by DFT and local MP2 calculations indicates that this is not due to a higher stability of the ADA array, as no significant differences in free energy are found. The theoretically calculated small free energies of association of **1** with a ureidopyridine and diaminopyridine (0–9 kJ/mol) are in line with the experimental data that little or no complex formation can be expected. Geometry optimization of the multiply hydrogen-bonded complexes by DFT calculations show that these are clearly nonplanar, with several long (3.2 Å), and correspondingly weak, hydrogen bonds. In other words, the hydrogen-bonding monomers have to deform themselves a bit to form the multiply hydrogen-bonded complex (cost: 9 kJ/mol), while the resulting hydrogen bonds are still long and therefore not very strong. Apparently, the formal full negative charge on the *N*-oxide oxygen atom—which could have been expected to yield strong hydrogen bonds—does not compensate for these effects. This example of the benzotriazine moiety points to the value of a combined experimental and theoretical approach in the development of novel multiply hydrogen-bonded systems.

Experimental Procedures

Single-crystal X-ray structure determination for compound 1: C₁₂H₁₅N₅O₂, *M_r* = 261.29, yellow block-shaped crystal (0.2 × 0.3 × 0.3 mm), monoclinic, space group *P*2₁/*c* (no. 14) with *a* = 10.9174(10) Å, *b* = 19.4699(10) Å, *c* = 12.785(3) Å, β = 110.518(10)°, *V* = 2545.0(7) Å³, *Z* = 8, *D_x* = 1.364 g cm^{−3}, *F*(000) = 1104, Mo K α = 0.098 mm^{−1}, 69 404 reflections measured, 5814 independent reflections, *R_{int}* = 0.0529, 1.00° < θ < 27.48°, *T* =

150 K, Mo K α radiation, graphite monochromator, λ = 0.71073 Å. Data were collected on a CCD area detector on a rotating anode. The structure was solved by direct methods (SHELXS86) and refined on *F*² using SHELXL-97-2. The NHBu moiety of one of the two independent molecules is disordered over two positions. An 80:20% disorder model was introduced. Positional parameters of the ordered N–H hydrogen atoms were refined, and all other hydrogen atoms were included in the refinement on calculated positions riding on their carrier atoms. The non-H atoms of the minor disorder component were included in the refinement with a fixed isotropic displacement parameter, set equal to the equivalent isotropic displacement parameter of the corresponding atom in the major disorder component. All other non-hydrogen atoms were refined with anisotropic displacement parameters. Hydrogen atoms were refined with a fixed isotropic displacement parameter linked to the value of the equivalent isotropic displacement parameter of their carrier atoms. Final *wR*2 = 0.1128, *w* = 1/[$\sigma^2(F^2)$ + (0.0613*P*)² + 0.2843*P*], where *P* = (max(*F_o*², 0) + 2*F_c*²)/3, *R*1 = 0.0409 (for 4205 *I* > 2 σ (*I*)), *S* = 1.032, 373 refined parameters, −0.23 < $\Delta\rho$ < 0.22 e Å^{−3}. Full details have been deposited with the CCDC (deposition number 651332).

Binding Experiments. For the ¹H NMR titrations, CDCl₃ (Cambridge Isotope Laboratories) was dried and deacidified over activated alumina (type I) and stored on 4 Å molecular sieves. DMSO-*d*₆ was dried over 4 Å molecular sieves. At least eight data points in the 20–80% saturation range were measured using temperature control at 25 °C unless stated otherwise. The association constants were evaluated by nonlinear least-squares computer fitting of the concentration dependence of the chemical shifts of NH protons to a 1:1 binding isotherm by ignoring dimerization of the individual components using standard methods.⁶⁴

Computational Methods. The molecular geometries of **1a** and **1b** and hydrogen-bonded complexes have been fully optimized using density functional theory (DFT) with the B3LYP functional as implemented in Gaussian 03⁶⁵ and the basis sets indicated in the text. To obtain the single point energies, **1a** and **1b** have also been calculated with 6-311+G(d,p), 6-311+G(2d,2p), and 6-311+G-(2df,2p) basis sets.

Synthesis. 2,4-Diamino-6-(3,4,5-tris(dodecyloxy)phenyl)-s-triazine (**3a**) was synthesized according to Beijer et al.¹³ 2,6-Bis-(hexanoylamino)pyridine (**3b**) was synthesized according to Bernstein et al.⁶⁶ Hexanoic acid (6-aminopyridin-2-yl)amide (**4a**) and hexanoic acid [6-(3-butylureido)pyridin-2-yl]amide (**4b**) were synthesized according to Söntjens et al.³¹

3-Butylureido-1,2,4-benzotriazine 1-*N*-Oxide (1). To a mixture of 3-amino-1,2,4-benzotriazine 1-*N*-oxide (**2**) (0.26 g, 1.6 mmol) in dry pyridine (4 mL) was added *n*-butyl isocyanate (1.58 g, 16.0 mmol) dropwise. The suspension was stirred at 110 °C for 16 h. The pyridine was evaporated in vacuo before chloroform was added, and the solution was washed with 1 M HCl, water, and brine and

(64) Schneider, H.-J.; Yatsimirsky, A. *Principles and Methods in Supramolecular Chemistry*; J. Wiley & Sons Ltd.: Chichester, 2000.

(65) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Montgomery, J. J. A.; Vreven, T.; Kudin, K. N.; Burant, J. C.; Millam, J. M.; Iyengar, S. S.; Tomasi, J.; Barone, V.; Mennucci, B.; Cossi, M.; Scalmani, G.; Rega, N.; Petersson, G. A.; Nakatsuji, H.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Klene, M.; Li, X.; Knox, J. E.; Hratchian, H. P.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Ayala, P. Y.; Morokuma, K.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Zakrzewski, V. G.; Dapprich, S.; Daniels, A. D.; Strain, M. C.; Farkas, O.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Ortiz, J. V.; Cui, Q.; Baboul, A. G.; Clifford, S.; Cioslowski, J.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Challacombe, M.; Gill, P. M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Gonzalez, C.; Pople, J. A. *Gaussian 03*, Revision C02; Gaussian, Inc.: Wallingford CT, 2004.

(66) Bernstein, J.; Stearns, B.; Dexter, M.; Lott, W. A. *J. Am. Chem. Soc.* **1947**, 69, 1147–50.

dried. The product could be obtained as yellow block crystals by crystallization from chloroform (0.13 g, 31%). ^1H NMR (CDCl_3): δ = 8.91 (br, 1H, NH), 8.40 (dd, 1H, J = 8.0, 0.7 Hz), 7.92 (dt, 1H, J = 7.1, 0.8 Hz), 7.80 (dd, 1H, J = 8.3, 0.7 Hz), 7.61 (dt, 1H, J = 7.0, 0.8 Hz), 7.30 (br, 1H, NH), 3.48 (q, 2H, J = 6 Hz), 1.70 (m, 2H), 1.53 (m, 2H), 1.04 (t, 3H, J = 7 Hz) ppm. ^{13}C NMR (CDCl_3): δ = 154.9, 152.9, 145.9, 136.3, 132.2, 127.8, 126.7, 120.4, 39.9, 31.6, 30.1, 13.6. ppm. ESI-MS: (m/z) calcd 261.29, observed 262.19 ($\text{M} + \text{H}^+$), 284.17 ($\text{M} + \text{Na}^+$), 545.35 ($2\text{M} + \text{Na}^+$), 806.51 ($3\text{M} + \text{Na}^+$), 1067.70 ($4\text{M} + \text{Na}^+$). FTR-IR (ATR): ν = 3251, 3096, 2931, 2869, 1678, 1579, 1518, 1490, 1419, 1357, 1303, 1280, 1198, 1135, 1050, 962, 840 cm^{-1} . Anal. Calcd for $\text{C}_{12}\text{H}_{15}\text{N}_5\text{O}_2$: C, 55.16; H, 5.79; N, 26.80. Found: C, 54.97; H, 5.53; N, 27.06.

3-Amino-1,2,4-benzotriazine 1-*N*-Oxide (2). Synthesis was according to a modified literature procedure:⁴⁸ Guanidine hydrochloride (5.74 g, 60.0 mmol) was passed through an ion exchange column (120 g, DOWEX 550A OH) and the eluate was evaporated in vacuo to yield the free guanidine base as a colorless oil (3.50 g, 59.0 mmol). To the guanidine were added THF (60 mL) and *o*-fluoronitrobenzene (0.70 g, 5.0 mmol), and the mixture was stirred

at 70 °C for 4 h. KO-*t*-Bu (0.60 g, 5.3 mmol) was subsequently added to the orange mixture followed by an additional 2 h of stirring at 70 °C. Upon addition of ethyl acetate (40 mL) to the cooled mixture, more yellow precipitate was formed which was isolated by filtration. The crude product was dissolved in hot ethyl acetate and washed with water. The resulting solid was washed with acetone and dried in vacuo to obtain the pure title compound (0.78 g, 96%). Spectroscopic data of the compound corresponded to those reported previously.⁶⁷

Supporting Information Available: General procedures, table of atomic coordinates of the calculated geometries of complexes **1a•4** and **1b•4**, copies of ^1H NMR and ^{13}C NMR spectra of compounds **1–5**, copies of ^{15}N NMR and FTIR spectra of compounds **1** and **2**, and crystallographic data of **1** (CIF). This material is available free of charge via the Internet at <http://pubs.acs.org>.

JO7019338

(67) Fuchs, T.; Chowdhury, G.; Barnes, C. L.; Gates, K. S. *J. Org. Chem.* **2001**, 66, 107–114.