Table I. Inertial Parameters of the 1-Pyridone Dimer in Its Ground Electronic State[a,b]

parameter	S_0(exptl)	S_0(3-21G)[c]
A'', MHz	2010.3 ± 0.1	2077.9
B'', MHz	319.4 ± 0.1	319.2
C'', MHz	275.9 ± 0.1	276.7
κ''	−0.950	−0.953
$\Delta I''$,[d] amu Å^2	−1.7 ± 0.8	0.0

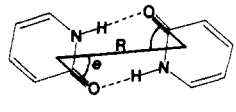
[a] Fits of the spectrum were made with use of the derivative approximation and a standard least-squares analysis. The standard deviation of the fit to a rigid-rotor Hamiltonian for both states is 9.0 MHz, significantly less than the experimental line width. Adding centrifugal distortion terms improved the standard derivation, but by less than 1 MHz, so they are neglected here. [b] The rotational constants obtained from the least-squares analysis are A'' = 2010.261 ± 0.005, B'' = 319.429 ± 0.003, and C'' = 275.888 ± 0.003. However, the precision of the measurement is reduced to ±0.1 MHz by systematic errors in the experiment (ref 6). [c] Theoretical inertial parameters calculated from the ab initio geometry of Field and Hillier (ref 9). [d] Inertial defects, $\Delta I = I_c - I_b - I_a$.

less than the corresponding spacings in the high-resolution spectra of the monomer bands, suggesting that the carrier of the high-frequency band is an aggregate structure.

We employed Watson's asymmetric rotor Hamiltonian[7] and previously described fitting strategies[8] to analyze the observed rotational structure of this band. The results for the S_0 state are given in Table I. We also list in this table the rotational constants that would be exhibited by a 2-PY dimer whose geometry is identical with that of the equilibrium structure calculated by ab initio methods.[9] The agreement between these calculated values and those measured for the electronic ground state of the carrier of the high-frequency band is good. (Typical errors in such calculations are of order 2%.) We conclude, therefore, that the aggregate structure is (2-PY)$_2$. The small, negative inertial defect shows further that the ground-state dimer is planar, or nearly so. Additionally, we find that the high-frequency band is a perpendicular-type transition polarized entirely (≥95%) along the b inertial axis of (2-PY)$_2$ (cf. I). This result is consistent with the polarization properties of the two lower frequency monomer bands.[5]

The inertial parameters of the excited-state vibronic level accessed in this experiment are very similar to those of the ground-state vibronic level. We find ΔA ($=A' - A''$) = 0.1, ΔB = −9.1, ΔC = −6.6 ± 0.1 MHz, κ' = −0.953, and $\Delta I'$ = −3.2 ± 1.2 amu Å^2. Increased vibrational amplitude along out-of-plane coordinate(s) would explain the larger magnitude of $\Delta I'$. The origin of the high-frequency band is at 30 776.479 ± 0.002 cm^{-1}.

Three rotational constants are clearly insufficient to fully characterize the structure of any electronic state of such a complex molecule. Recognizing this, we have used the experimental values of these constants to optimize two geometrical properties of the dimer, the separation of the centers of mass of the two monomer units (R), and the angle (θ) between R and the C=O bond(s) (II). Bond distances and angles for the two pyridone rings were



II

fixed at the values determined for α-pyridone by Penfold with use of X-ray crystallography.[10] The calculated rotational constants converged to within 0.15% of the experimental values for both the S_0 and S_1 states, with $R(S_0)$ = 5.30 ± 0.03 Å, $\theta(S_0)$ = 43 ± 2°, $R(S_1)$ = 5.39 ± 0.03 Å, and $\theta(S_1)$ = 43 ± 2°. The ground-state values of R and θ yield the hydrogen bond parameters r(N—H···O) = 2.75 ± 0.03 Å and ϕ[C=O···(H)—N] = 122 ± 2°. (Insufficient information is available, at present, to determine whether or not the hydrogen atom lies exactly on the N···O axis.) The corresponding solid-state values are r = 2.77 ± 0.03 Å and ϕ = 136 ± 2°.[10] Also, the gas-phase value of r lies within 1σ of the distribution found for 1357 intermolecular hydrogen bonds in a recent survey of 889 organic crystal structures.[11] On S_1 excitation of (2-PY)$_2$, r increases by 0.08 Å relative to S_0 while ϕ remains approximately the same, a result that suggests a decrease in hydrogen-bonding strength on excitation and is consistent with the large blue shift of the dimer $S_1 \leftarrow S_0$ band relative to those of the monomer.

Intermolecular forces, particularly hydrogen bonds, lie at the heart of many chemical and biochemical phenomena. There has been much speculation about the role of the environment in determining the magnitudes of these forces. Thus, the finding that r(N—H···O) in (2-PY)$_2$ is the same in both the gas phase and the condensed phase is significant. Beyond this, the high-resolution FES technique offers distinct advantages over other methods for the study of hydrogen-bonded species. Structures like I have no permanent dipole moments and so have no microwave spectrum. Additionally, isotopic substitution may be used to determine the center-of-mass coordinates of any atom in the molecule (including hydrogen atoms),[12] making possible determinations of structure and studies of tunneling and energy transfer dynamics in both ground and electronically excited states. Other degrees of freedom may be accessed at other laser wavelengths. Finally, water molecules may be attached during the expansion to probe the effect of solvent on a wide variety of structures. Work along several of these lines is in progress.

Acknowledgment. This work has been supported by NSF. We are indebted to S. A. Asher and A. D. Hamilton for helpful discussions.

(7) Watson, J. K. G. *J. Chem. Phys.* **1967**, *46*, 1935.
(8) See, for example: Champagne, B. B.; et al. *J. Phys. Chem.* **1990**, *94*, 6.
(9) Field, M. J.; Hillier, I. H. *J. Chem. Soc., Perkin Trans. II* **1987**, 617.
(10) Penfold, B. R. *Acta Crystallogr.* **1953**, *6*, 591.
(11) Taylor, R.; Kennard, O.; Versichel, W. *Acta Crystallogr.* **1984**, *B40*, 280.
(12) Johnson, J. R.; Jordan, K. D.; Plusquellic, D. F.; Pratt, D. W. *J. Chem. Phys.* **1990**, *93*, 2258.

Hydrogen Bonding in a Family of Triamides: Conformation-Directing Effects in Solution vs the Solid State

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The ability of hydrogen bonds to influence molecular conformation and to orchestrate intermolecular association is a topic of considerable current interest.[1] Our own experiments in this area are designed to elucidate the interplay among noncovalent forces that leads to discrete molecular folding patterns, with greater insight on the origins of protein tertiary structural stability as an ultimate goal.[2] We recently reported that triamide **1** experiences dramatic temperature-dependent conformational changes in a nonpolar solvent, involving the rearrangement of intramolecular hydrogen bonds.[2b] We now describe crystal structures of **1** and

(1) For leading references, see: (a) Etter, M. C. *Acc. Chem. Res.* **1990**, *23*, 120. (b) Rebek, J. *Angew. Chem., Int. Ed. Engl.* **1990**, *29*, 245. (c) Alber, T. *Prediction of Protein Structure and the Principles of Protein Conformation*; Fasman, G. D., Ed.; Plenum Press: New York, 1989; pp 161-192.
(2) Gellman, S. H.; Adams, B. R. *Tetrahedron Lett.* **1989**, *30*, 3381. (b) Gellman, S. H.; Adams, B. R.; Dado, G. P. *J. Am. Chem. Soc.* **1990**, *112*, 460.

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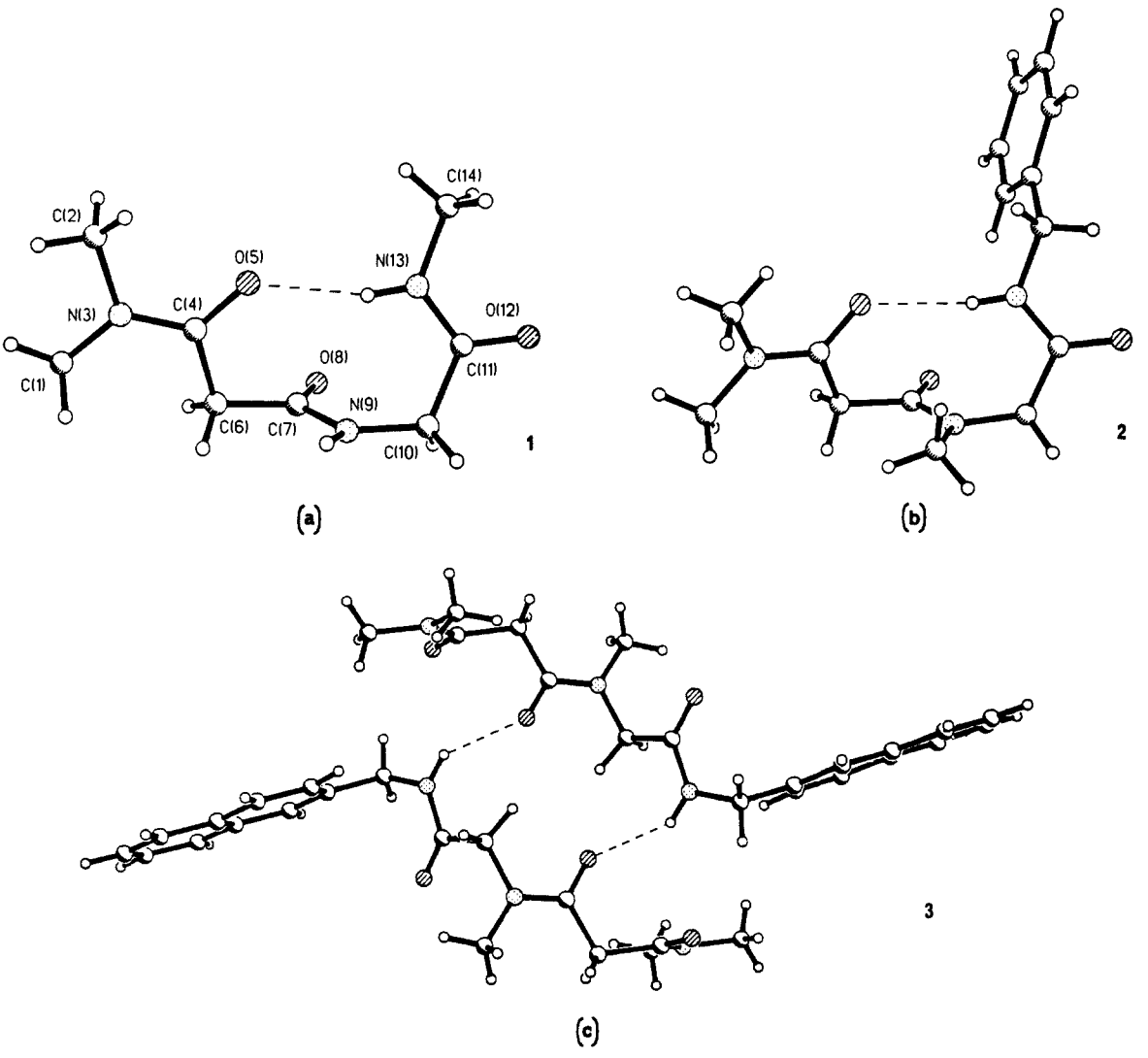
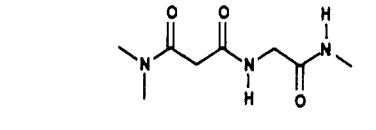


Figure 1. Ball-and-stick representation of crystallographically observed triamide conformations: (a) triamide **1**; (b) triamide **2**; (c) triamide **3**, shown as the intermolecularly amide–amide hydrogen bonded dimer. Amide–amide hydrogen bonds are indicated by dotted lines. The atomic numbering of the triamide core is the same for all three molecules. In **1**, N(9) is part of a secondary amide group, with the amide proton designated H(9); in **2** and **3**, N(9) is part of a tertiary amide group, and the attached methyl group is designated C(9).

related compounds **2**–**4**[3] and the behavior of **2**–**4** in solution. These data provide direct evidence on the favorability of the nine-membered ring hydrogen-bonded folding pattern available to this family of triamides. The present results also show that peripheral changes to a molecular array of hydrogen-bond donors and acceptors can lead to profound changes in crystalline conformation and packing.

Figure 1 shows the crystallographically observed conformations of **1**–**3**. The crystal lattice of **4** contains two independent but nearly identical molecules (not shown), each displaying an intramolecular nine-membered-ring amide–amide hydrogen bond analogous to those observed for **1** and **2**. The conformation observed for **1** in the crystal is consistent with the folding pattern we had earlier deduced to be most enthalpically stable in methylene chloride solution.[2b] Only **3** fails to adopt the nine-membered-ring folding



pattern in the solid state; instead, amide–amide hydrogen bonds occur intermolecularly, between pairs of triamides in the lattice. Intermolecular amide–amide hydrogen bonds occur in **1** as well, involving O(12) and H(9).

The O--H distances of the nine-membered-ring hydrogen bonds in **1**, **2**, and **4** (2.07–2.31 Å) are distinctly longer than the typical value of 1.95 Å for crystallographically observed intermolecular amide–amide hydrogen bonds in small molecules[4] (a tendency for intramolecular hydrogen bonds to be somewhat longer than analogous intermolecular interactions has been previously noted[5]).

(3) X-ray quality crystals of **1** were obtained by dissolving the triamide in hot methylene chloride, adding hexane to saturation, and allowing the solution to cool to room temperature. Crystals of **2** and **3** were grown by vapor diffusion of hexane into 1,2-dichloroethane solutions of the triamides at room temperature. Crystals of **4** were grown by vapor diffusion of hexane into a chloroform solution of the triamide at −20 °C. Crystallographic data were obtained at 95 K by using Cu Kα radiation for **1**, **2**, and **4** and Mo Kα radiation for **3**. $2\theta_{min/max}$ = 3.5–110° for **1**, **2**, and **4** and 3.5–50° for **3**. For structures **1**–**4**, the following data are provided below: unique reflections, observed reflections, R, weighted R (observed data), difference Fourier maximum, difference Fourier minimum. **1**: 719, 697, 3.18, 4.51, 0.12, −0.14. **2**: 1057, 1018, 2.98, 4.41, 0.16, −0.10. **3**: 2876, 1947, 6.64, 8.46, 0.27, −0.28. **4**: 4578, 3656, 6.87, 10.76, 0.25, −0.28. Synthetic and crystallographic details will be reported in a full paper that is currently in preparation.

(4) Baker, E. N.; Hubbard, R. E. *Prog. Mol. Biol. Biophys.* **1984**, *44*, 97.
(5) Taylor, R.; Kennard, O.; Versichel, W. *Acta Crystallogr.* **1984**, *B40*, 280.

The intermolecular hydrogen bonding O---H distance in crystalline **3** (2.19 Å) is also unusually long. Hydrogen-bond angles C=O---H and N—H---O in the crystalline triamides (127–131° and 165–175°, respectively) fall in the ranges commonly observed for these structural parameters,[4] except for the distinctly nonlinear intermolecular N—H---O angle in **3** (142°). The atypical geometry of the intermolecular amide–amide hydrogen bond in **3** apparently results from an attraction between O(8) and one of the hydrogen atoms on C(10) of the hydrogen-bonded partner. This O---H distance is 2.34 Å, significantly smaller than the sum of the oxygen and hydrogen van der Waals radii, and within the separation range that has been associated with C—H---O hydrogen bonds in crystal structures.[6]

The energetic factors leading to the two different types of solid-state triamide conformations are not clear because in a crystal lattice (as in the core of a folded protein) one sees only the final energetic compromise achieved as many noncovalent interactions compete to achieve optimal configurations.[7,8] The amide–amide hydrogen-bonding patterns vary between the extended and folded crystal conformations, but the intermolecular aromatic–aromatic interactions resulting from the lattice packing of these two types of conformations are similar. Small polycyclic aromatic hydrocarbons generally adopt "herringbone" crystal-packing patterns,[9] and in the crystal lattices of both **2** and **3**, the aromatic rings congregate in local herringbone arrays with a thickness of two aryl moieties (i.e., one face of each aromatic ring engages in an aromatic–aromatic interaction).

The adoption of an extended conformation by **3** in the solid state is striking because IR measurements suggest that **3** in dilute CH_2Cl_2 solution (like **2** and **4**) exists predominantly in a folded form containing the nine-membered-ring hydrogen bond.[2b,10] Triamides **2–4** remain largely intramolecularly hydrogen bonded in CH_3CN; at 10 mM, each triamide shows a major N–H stretch absorption at a position similar to that observed for the intramolecularly hydrogen bonded N–H stretch in CH_2Cl_2.[11] Triamides **2–4** also show a minor shoulder in the range 3360–3390 cm^{-1} in CH_3CN, which we assign to an amide proton hydrogen bonded to solvent nitrile.[11] In DMSO, only a single N–H stretch absorption is observed for each triamide. These absorptions are >15 cm^{-1} below the hydrogen-bonded absorptions observed in CH_2Cl_2 and CH_3CN, suggesting that the amide protons are largely engaged in N—H---O=SMe_2 rather than intramolecular N—H---O=C hydrogen bonds.

The nine-membered-ring hydrogen-bonded conformation favored by this family of triamides is related to β-turn conformations commonly observed in peptides and proteins.[12] β-Turns often contain 10-membered-ring amide–amide hydrogen bonds, but the average geometry of these intramolecular interactions is poor because the conformational preference of the dipeptide segment that links the hydrogen-bonding groups constrains the donor proton to lie outside the plane of the acceptor amide (defined by the atoms N—C=O).[4] The smaller ring size in our triamides allows a more planar (i.e., more favorable) hydrogen-bonding arrangement. The malonyl-*N*-methyl-amino acid subunit therefore represents a potential alternative to the natural dipeptide subunit at residues $i + 1$ and $i + 2$ of a β-turn.[13] This possibility is particularly intriguing in the context of recent efforts to stabilize synthetic proteins through the incorporation of unnatural "template" substructures.[14]

The amide–amide hydrogen-bonding patterns manifested by **1–4** in the solid state provide an interesting test of the rules for hydrogen bonding in organic compounds recently proposed by Etter,[1a] because these triamides have greater conformational freedom than many of the examples discussed by Etter. Amide carbonyl–amide proton pairing is maximized in all four cases, in accord with Etter's predictive guidelines. However, **1** represents an exception to Etter's second general rule ("six-membered-ring intramolecular hydrogen bonds form in preference to intermolecular hydrogen bonds"), since in this case a nine-membered ring and an intermolecular hydrogen bond are formed instead of a six-membered ring plus a second inter- or intramolecular hydrogen bond. Our earlier study of **1** in methylene chloride indicated that conformations involving the six-membered-ring hydrogen bond predominate at room temperature in solution, but give way to the more enthalpically favorable nine-membered-ring folding pattern at lower temperatures.[2b]

Our results indicate that the common triamide core of compounds **1–4** inherently favors a folding pattern containing a nine-membered-ring hydrogen bond, but that this intramolecular hydrogen bond is subject to disruption by environmental factors. The different packing patterns manifested among **2–4** suggest that it may be difficult to predict the structure-directing effects of hydrogen bonds formed by *conformationally flexible* molecules in crystals and other highly ordered states. This point is important in the context of current efforts to use hydrogen-bonding interactions to control supramolecular architecture in solids, liquid crystals, and large aggregates.[15]

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(6) Taylor, R.; Kennard, O. *J. Am. Chem. Soc.* **1982**, *104*, 5063.
(7) Considerable effort has been devoted to studying the balance of forces that determine amide crystal packing. For leading references, see: (a) Leiserowitz, L.; Hagler, A. T. *Proc. R. Soc. London A* **1983**, *388*, 133. (b) Dauber, P.; Hagler, A. T. *Acc. Chem. Res.* **1980**, *13*, 105.
(8) We cannot rule out the possibility that some or all of triamides **1–4** have alternative crystalline forms available to them.
(9) Desiraju, G. R.; Gavezzotti, A. *Acta Crystallogr.* **1989**, *B45*, 473 and references therein.
(10) The N–H stretch signatures of **2–4** in CH_2Cl_2 (1 mM; room temperature) are similar to that previously described for a related molecule (compound **4** in ref 2b): major absorption at 3300–3320 cm^{-1} (intramolecular N—H---O=C hydrogen bond), and very small absorption at 3415–3445 cm^{-1} (fully solvated NH). Comparison of these spectral data with the N–H stretch signature of a diamide that can experience only the seven-membered-ring hydrogen bond available to **2–4** (compound **3** in ref 2b) indicates that the intramolecular hydrogen bonding in solution must result from the nine-membered-ring folding pattern.
(11) In CH_3CN, at room temperature, there is no significant intermolecular hydrogen bonding among dissolved amides at 10 mM: Gellman, S. H.; Dado, G. P.; Liang, G.-B.; Adams, B. R. *J. Am. Chem. Soc.*, in press.
(12) Rose, G. D.; Gierasch, L. M.; Smith, J. A. *Adv. Protein Chem.* **1985**, *37*, 1.
(13) Malonic acid residues have been incorporated into peptide analogues that are referred to as "retro-inverso peptides"; see: Goodman, M.; Chorev, M. *Acc. Chem. Res.* **1979**, *12*, 1 and references therein.
(14) Mutter, M.; Veilleumier, S. *Angew. Chem., Int. Ed. Engl.* **1989**, *28*, 535 and references therein.
(15) For leading references, see: (a) Lehn, J.-M.; Mascal, M.; DeCian, A.; Fischer, J. *J. Chem. Soc., Chem. Commun.* **1990**, 479. (b) Brienne, M.-J.; Gabard, J.; Lehn, J.-M.; Stibor, I. *J. Chem. Soc., Chem. Commun.* **1989**, 1868. (c) Ducharme, Y.; Wuest, J. D. *J. Org. Chem.* **1988**, *53*, 5787.

^{1}H 2D Nuclear Magnetic Resonance Spectra of Oligonucleotide Phosphorodithioate, d(CGCTpS_2^-TpS_2^-AAGCG). An Unusual Hairpin Loop Structure

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In this communication we describe the synthesis, purification, NMR spectra, and structure of a dithiophosphate analogue of an oligonucleotide. Oligonucleotides and various analogues of oligonucleotides have been shown to exhibit antiviral activity. Thus addition of complementary "antisense" oligodeoxynucleotides to

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