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Intermolecular $^{2h}J_{NN}$ Coupling in Multiply Hydrogen-Bonded Ureidopyrimidinone Dimers in Solution

Serge H. M. Söntjens, Marcel H. P. van Genderen, and Rint P. Sijbesma*

Department of Macromolecular and Organic Chemistry, Eindhoven University of Technology, P.O. Box 513, 5600 MB Eindhoven, The Netherlands

r.p.sijbesma@tue.nl

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^{15}N-Labeled ureido-4[1H]-pyrimidinones **4a** and **5a** were synthesized in order to investigate hydrogen bonding in the strongly hydrogen-bonded dimers in solution with intermolecular $^{2h}J_{NN}$ coupling. Both direct-detection ^{15}N NMR and one-dimensional ^{15}N INADEQUATE (for smaller scalar coupling constants) were employed to determine the coupling constants. For dimers of **4** in $CDCl_3$, a temperature-dependent $^{2h}J_{NN}$ was observed ranging from 2 Hz at +10 °C to 5.1 Hz at −20 °C. In dimers of more slowly exchanging bifunctional derivative **5**, the coupling constants could be determined at room temperature from an inverse-gated ^{1}H-decoupled ^{15}N NMR experiment. Coupling constants in different isomers of the dimer of **5a** (4.96, 5.13, 4.37, and 5.27 Hz) were used to test the relationship between $^{2h}J_{NN}$ values and N−N distances as proposed by Del Bene et al. The N−N distances calculated with the aid of this relationship show excellent agreement with the distances observed in the X-ray crystal structures of **5b**, particularly when the nonlinearity of the hydrogen bonds is taken into account.

Introduction

Hydrogen bonds are among the most generally employed noncovalent interactions in the self-assembly (or "noncovalent synthesis") of supramolecular structures.[1] Their reversibility, high directionality, and the possibility to create stronger interactions by combining individual hydrogen bonds in arrays[2,3] explain their widespread use in supramolecular chemistry. The presence of hydrogen bonds can be established qualitatively with spectroscopy because their formation results in significant changes in the ^{1}H NMR and IR spectrum. These spectroscopic techniques have also been used extensively to evaluate association constants of hydrogen-bonded complexes quantitatively. The study of the geometry of hydrogen bonds and of hydrogen-bonded complexes in solution is more difficult, and information is often obtained in an indirect way by determining the crystal structure of complexes in the solid state with X-ray diffraction. Intermolecular interactions in biomolecular aggregates, and also in synthetic self-assembled systems, have been studied extensively with ^{1}H NMR by making use of the nuclear Overhauser effect (NOE). This technique, however, does not provide direct evidence for the existence of hydrogen bonds. Recently, direct observation of intermolecular hydrogen bonds has become possible by making use of the scalar coupling constant between atoms across hydrogen bonds in ^{15}N-labeled nucleic acid base pairs[4,5] and in proteins.[6−11] Two excellent reviews have recently been published on the development of this technique for the analysis of biopolymers.[12,13] Observation of this interaction across hydrogen bonds provides information on hydrogen bond connectivity and complements established NMR-based correlation techniques in determining the higher order structure of biomolecules. Because the magnitude of the coupling constant is dependent on the hydrogen bond length in a predictable way,[14−17] determination of its value may eventually give very

* Author to whom correspondence should be addressed. Tel: 40-2473111. Fax: 40-2451036.

(1) Prins, L. J.; Reinhoudt, D. N.; Timmerman, P. *Angew. Chem., Int. Ed.* **2001**, *40*, 2382−2426.
(2) Zimmerman, S. C.; Corbin, P. S. *Struct. Bonding* **2000**, *96*, 63−94.
(3) Sijbesma, R. P.; Meijer, E. W. *Chem. Commun.* **2003**, 5−16.
(4) Dingley, A. J.; Grzesiek, S. *J. Am. Chem. Soc.* **1998**, *120*, 8293−8297.
(5) Pervushin, K.; Ono, A.; Fernandez, C.; Szyperski, T.; Kainosho, M.; Wuthrich, K. *Proc. Natl. Acad. Sci. U. S. A.* **1998**, *95*, 14147−14151.
(6) Cornilescu, G.; Ramirez, B. E.; Frank, M. K.; Clore, G. M.; Gronenborn, A. M.; Bax, A. *J. Am. Chem. Soc.* **1999**, *121*, 6275−6279.
(7) Cornilescu, G.; Hu, J. S.; Bax, A. *J. Am. Chem. Soc.* **1999**, *121*, 2949−2950.
(8) Cordier, F.; Grzesiek, S. *J. Am. Chem. Soc.* **1999**, *121*, 1601−1602.
(9) Hennig, M.; Geierstanger, B. H. *J. Am. Chem. Soc.* **1999**, *121*, 5123−5126.
(10) Blake, P. R.; Park, J. B.; Adams, M. W. W.; Summers, M. F. *J. Am. Chem. Soc.* **1992**, *114*, 4931−4933.
(11) Blake, P. R.; Lee, B.; Summers, M. F.; Adams, M. W. W.; Park, J. B.; Zhou, Z. H.; Bax, A. *J. Biomol. NMR* **1992**, *2*, 527−533.
(12) Dingley, A. J.; Cordier, F.; Grzesiek, S. *Concepts Magn. Reson.* **2001**, *13*, 103−127.
(13) Grzesiek, S.; Cordier, F.; Dingley, A. J. *Methods Enzymol.* **2001**, *338*, 111−133.
(14) Dingley, A. J.; Masse, J. E.; Peterson, R. D.; Barfield, M.; Feigon, J.; Grzesiek, S. *J. Am. Chem. Soc.* **1999**, *121*, 6019−6027.
(15) Benedict, H.; Shenderovich, I. G.; Malkina, O. L.; Malkin, V. G.; Denisov, G. S.; Golubev, N. S.; Limbach, H. H. *J. Am. Chem. Soc.* **2000**, *122*, 1979−1988.
(16) Del Bene, J. E.; Bartlett, R. J. *J. Am. Chem. Soc.* **2000**, *122*, 10480−10481.
(17) Del Bene, J. E.; Perera, S. A.; Bartlett, R. J. *Magn. Reson. Chem.* **2001**, *39*, S109−S114.

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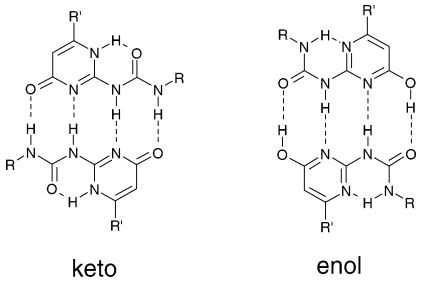
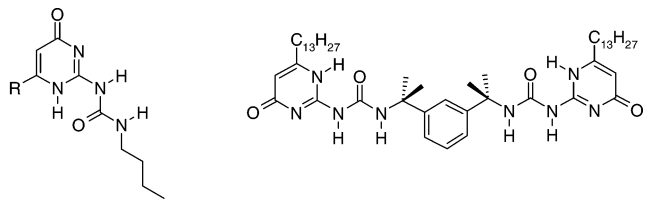


FIGURE 1. Keto and enol tautomeric forms of dimeric ureidopyrimidinones.

precise structural information on hydrogen-bonded complexes in solution, provided the theoretical correlation between distance and coupling constant can be substantiated by comparison with accurate crystals structures. The use of scalar coupling across hydrogen bonds has rapidly gained popularity in the biomolecular area, but only limited use has been made of this technique on synthetic molecules. Intermolecular coupling in synthetic systems is a little explored area, mainly because chemical exchange in most hydrogen bonded complexes is rapid, leading to loss of the spin−spin coupling interaction. Therefore, intermolecular *J* coupling in synthetic molecules has previously only been observed at very low temperatures in fluorocarbon solvents on the complex of HF and 2,4,6-trimethylpyridine,[18] in hydrogen bonded clusters of F^- and $(HF)_n$,[19] and in a dimer of a Mannich base.[20]

For our work on supramolecular polymers,[21] polymers in which the monomeric units are held together by noncovalent interactions,[22−24] we have introduced the ureidopyrimidinone (UPy) heterocyclic unit, which forms very stable dimers held together by a linear array of four hydrogen bonds.[25,26] The molecules in UPy dimers may exist in either of two tautomeric forms, the 4[1H]-pyrimidinone (keto) form or the pyrimidin-4-ol (enol) form, which feature DDAA and DADA arrays of hydrogen bonding donor (D) and acceptor (A) sites, respectively (Figure 1). Because of its high stability (the K_{dim} value of **4** in $CDCl_3$ at 298 K is 6×10^7 M^{-1}), the UPy unit has successfully been used to form supramolecular polymers with high degrees of polymerization.[27] The high stability also leads to a long lifetime of the dimer, which was shown to be 120 ms in $CDCl_3$ at 298 K with dynamic NMR studies.

Conformationally preorganized molecules such as **5** with two UPy functional groups were shown to form exceptionally stable cyclic dimers, held together by eight hydrogen bonds.[28] For bisUPy derivative **5b**, a lower limit estimate for the lifetime of the dimer was approximately 45 min. The crystal structures of both keto and enol forms of a number of UPy derivatives have been determined, and hydrogen bonding in the solid state has been investigated by double quantum magic angle spinning (DQ-MAS) NMR.[29,30] However, little direct information is available on the structure of UPy dimers in solution. The considerable lifetimes of dimers of **4** and particularly of **5** should allow the observation of *J* coupling across the hydrogen bonds. In the present paper, we report a detailed analysis of hydrogen bond length and dynamics in dimers of **4** and **5**, using hydrogen bond mediated $^{2h}J_{NN}$−coupling.



4a R = $C_{13}H_{25}$ (1x^{15}N)

4b R = Ph

4c R = CH_3

5a R = $C_{13}H_{25}$ (2x^{15}N)

5b R = CH_3

Synthesis

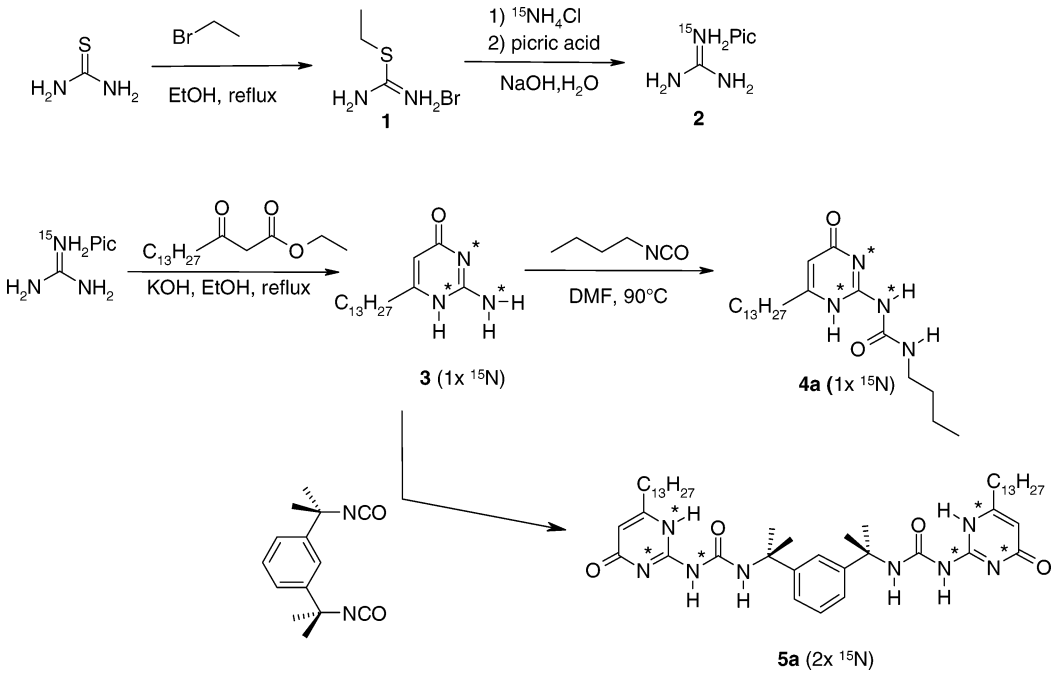
Because of the low natural abundance of ^{15}N, isotopically labeled UPy derivatives **4a** and **5a** were synthesized in order to investigate the $^{2h}J_{NN}$ coupling in their dimers. In these derivatives, each UPy group has a single ^{15}N label, randomly distributed among the excocyclic and the two endocyclic sites labeled N_a, N_b, and N_c. Although less ^{15}N spins are present than in triply labeled derivatives, leading to less signal, the interpretation of ^{15}N NMR spectra is simplified in singly labeled derivatives because intramolecular N−N coupling is effectively eliminated.

The synthesis of the ^{15}N isotope-labeled ureido-4[1H]-pyrimidinones is depicted in Scheme 1.

Guanidinium bromide with a single ^{15}N atom was prepared by reaction of isothiouronium bromide with 100% ^{15}N-enriched ammonium chloride and was isolated as its picrate salt. In contrast with the published syntheses of isocytosines employing guanidinium carbonate,[25] the addition of base to the reaction mixture was found to be necessary to effect the ring-closing reaction with the β-keto ester when guanidinium picrate was used. The singly labeled isocytosine **3** was subsequently reacted with butyl isocyanate or with 1,3-bis(1-iso-

(18) Golubev, N. S.; Shenderovich, I. G.; Smirnov, S. N.; Denisov, G. S.; Limbach, H. H. *Chem.−Eur. J.* **1999**, *5*, 492−497.
(19) Shenderovich, G. I.; Smirnov, S. N.; Denisov, G. S.; Gindin, V. A.; Gobulev, N. S.; Dunger, A.; Reibke, R.; Kirpekar, S.; Malkina, O. L.; Limbach, H.-H. *Ber. Bunsen-Ges. Phys. Chem.* **1998**, *103*, 422−428.
(20) Rospenk, M.; Sobczyk, L.; Schah-Mohammedi, P.; Limbach, H. H.; Golubev, N. S.; Melikova, S. M. *Magn. Reson. Chem.* **2001**, *39*, S81−S90.
(21) Lehn, J.-M. *Macromol. Chem., Macromol. Symp.* **1993**, *69*, 1−17.
(22) Brunsveld, L.; Folmer, B. J. B.; Meijer, E. W.; Sijbesma, R. P. *Chem. Rev.* **2001**, *101*, 4071−4098.
(23) *Supramolecular Polymers*; Ciferri, A., Ed.; Marcel Dekker: New York, 2000.
(24) Ciferri, A. *Macromol. Rapid Commun.* **2002**, *23*, 511−529.
(25) Beijer, F. H.; Sijbesma, R. P.; Kooijman, H.; Spek, A. L.; Meijer, E. W. *J. Am. Chem. Soc.* **1998**, *120*, 6761−6769.
(26) Söntjens, S. H. M.; Sijbesma, R. P.; van Genderen, M. H. P.; Meijer, E. W. *J. Am. Chem. Soc.* **2000**, *122*, 7487−7493.
(27) 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.
(28) Folmer, B. J. B.; Sijbesma, R. P.; Kooijman, H.; Spek, A. L.; Meijer, E. W. *J. Am. Chem. Soc.* **1999**, *121*, 2001−2007.
(29) Schnell, I.; Langer, B.; Sontjens, S. H. M.; van Genderen, M. H. P.; Sijbesma, R. P.; Spiess, H. W. *J. Magn. Reson.* **2001**, *150*, 57−70.
(30) Schnell, I.; Langer, B.; Soentjens, S. H. M.; Sijbesma, R. P.; van Genderen, M. H. P.; Wolfgang Spiess, H. *Phys. Chem. Chem. Phys* **2002**, *4*, 3750−3758.

SCHEME 1. Synthesis of ^{15}N-Labeled UPy Derivatives 4a and 5a[a]



[a] Each aminopyrimidine unit of **3**, **4a**, and **5a** contains a single ^{15}N atom, randomly introduced at one of the starred positions.

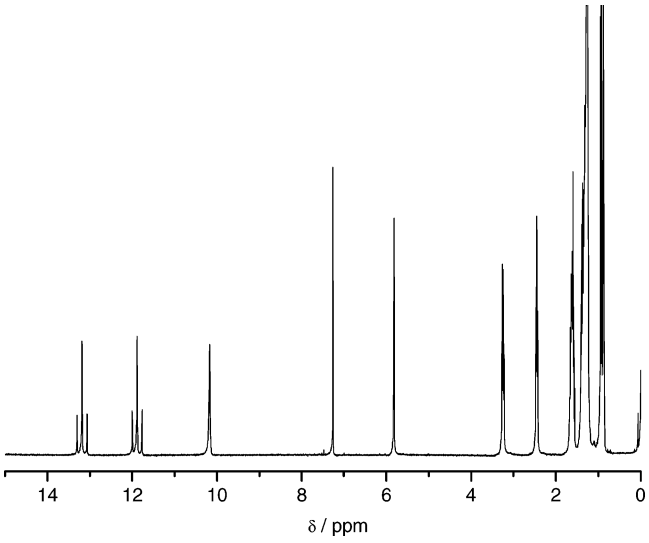


FIGURE 2. ^{1}H NMR spectrum of compound **4a**.

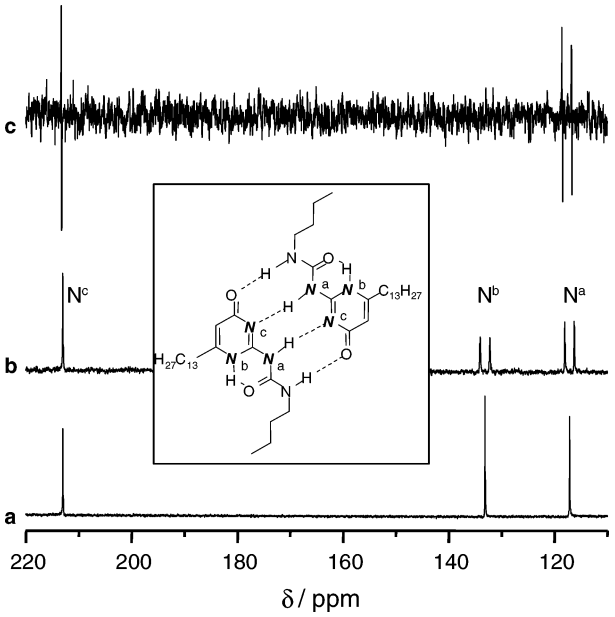


FIGURE 3. ^{15}N spectra of singly ^{15}N-labeled 2-butylureido-4[1H]-pyrimidinone **4a**. (a) Inverse-gated ^{1}H-decoupled ^{15}N NMR spectrum ($CDCl_3$, 25 °C). (b) ^{1}H-coupled ^{15}N NMR spectrum. (c) One-dimensional ^{15}N INADEQUATE spectrum.

cyanato-1-methylethyl)benzene to afford monofunctional compound **4a** and bifunctional UPy derivative **5a**, respectively.

^{1}H NMR and ^{15}N NMR Spectroscopy of Monofunctional UPy 4a. In the ^{1}H NMR spectrum of compound **4a** (Figure 2), a large intramolecular $^{1}J_{NH}$ coupling of approximately 90 Hz in the NH region is observed for one in three protons attached to $^{15}N_a$ or $^{15}N_b$; the remaining protons are bound to a ^{14}N atom and consequently resonate as a singlet.

The inverse-gated decoupled ^{15}N NMR spectrum of **4a** (Figure 3a) shows three ^{15}N signals for the three isotopic isomers present. Partial assignment of the peaks is possible based on the predicted shifts of the two sp^3-hybridized nitrogen atoms (low ppm range) and one sp^2-hybridized nitrogen (high ppm range). This assignment is confirmed by the coupling pattern in the proton-coupled ^{15}N spectrum (Figure 3b). $^{1}J_{NH}$ coupling constants of 93.3 and 90.4 Hz are observed on nitrogen atoms N_b and N_a, respectively, whereas the signal at 213 ppm does not show proton coupling. The ^{15}N spectrum was subsequently fully assigned as shown in Figure 3b with the aid of an HSQC experiment. The assignment is in agreement with a previous assignment in the solid state based on DQMAS, CPMAS, and ^{1}H−^{15}N REPT-HSQC experiments.[29,30]

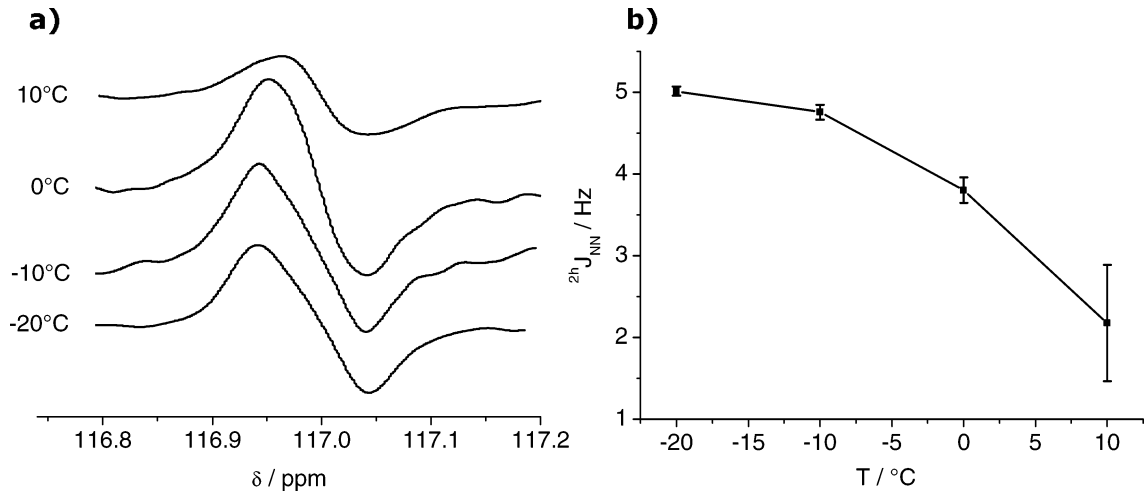


FIGURE 4. (a) Antiphase doublet of N_a in the ^{15}N INADEQUATE spectrum of **4a**. (b) The $^{2h}J_{NN}$ coupling constants determined by deconvolution as a function of temperature. The coupling constant was determined from both antiphase doublets at 117 ppm.

As can be observed in Figure 3b, no intermolecular coupling between N_a and N_c is observed at room temperature. This may be ascribed to exchange by dissociation and reassociation between dimers, as the lifetime of UPy dimers has previously been established to be in the order of 120 ms at 298 K in $CDCl_3$. In an inverse-gated ^1H-decoupled ^{15}N NMR spectrum at −20 °C, splitting of the N_a and N_c signals was observed, but strong overlap between coupled and uncoupled signals, especially at higher temperatures, did not allow quantitative analysis of the results. Therefore, a one-dimensional ^{15}N INADEQUATE experiment was performed.[31] In the INADEQUATE experiment, the interfering signal due to noncoupled nitrogens is fully removed with a double quantum filter, allowing accurate determination of $^{2h}J_{NN}$ coupling constants as low as 2 Hz. To avoid (digital) resolution problems, only the double antiphase doublet for N_a at 117 ppm was studied in subsequent measurements. In a proton-coupled one-dimensional ^{15}N INADEQUATE spectrum recorded at −20 °C (Figure 3c), the singlets of ^{15}N atoms paired with ^{14}N atoms across the hydrogen bond are eliminated and *J*-coupled signals show up as antiphase doublets for N_a and N_c with a $^{2h}J_{NN}$ coupling constant of 5.1 Hz. Because each molecule of **4a** has only a single ^{15}N atom, the observed spin−spin interaction is necessarily intermolecular and confirms the existence of the hydrogen bond array and an intermolecular *J* coupling interaction across the center hydrogen bonds.

INADEQUATE spectra at higher temperatures showed that the *J* coupling constant is temperature dependent and decreases with increasing temperature (Figure 4).

A distinct broadening of peaks is also observed with increasing temperature. Deconvolution of the antiphase doublets allowed quantitative determination of the $^{2h}J_{NN}$ coupling constants, which are plotted in Figure 3b. The broadening and decrease in coupling constant with rising temperatures can both be ascribed to the effects of chemical exchange during the evolution period of the NMR experiment. Because of the high dimerization constant of **4a**, the fraction of time spent as monomers is negligible at the concentration used for recording the spectra. However, dissociation and reassociation of dimers, which has a rate of approximately 8 s^{-1} at room temperature, leads to loss of coupling when a ^{15}N atom is exchanged for a ^{14}N atom and may lead to loss of coherence when one ^{15}N atom is exchanged for another ^{15}N atom. Because of these effects, the observed peak splittings are temperature dependent and cannot be equated with the coupling constants across hydrogen bonds. Although the change in coupling constant is leveling off when cooling below −10 °C, the values still change significantly from 4.7 to 5.1 Hz upon cooling from −10 to −20 °C. Unfortunately we were unable to determine *J* coupling constants below this temperature because the solubility of compound **4a** in $CDCl_3$ decreases rapidly below −20 °C. We can therefore not confirm that the *J* coupling constant of 5.1 Hz is indeed a limiting value for this system.

NMR Study of Bifunctional UPy Derivative 5a. Better chances of quantitative determination of coupling constants were expected for the ^{15}N-labeled derivative **5a**, with a single ^{15}N label in each of the UPy groups. Nonlabeled analogue **5b** has previously been studied in detail and was shown to form exceedingly stable dimers, held together by eight hydrogen bonds.[28] Because of the keto−enol tautomerism of the UPy groups and syn−anti isomerism in the dimer, the dimer is present as a dynamic mixture of three forms in solution: a syn keto−keto (I), an anti keto−keto (II), and a keto−enol (III) form (Figure 5), each of which has its own set of signals in the ^1H NMR spectrum. Exchange between the different forms of **5b** has previously been studied by us using ^1H NMR and was shown to be slow compared to **4a**. Although the existence of multiple isomers may complicate analysis of the ^{15}N NMR spectra in a labeled analogue, the considerably slower exchange kinetics in dimers of **5** may be very useful in the determination of the N−N coupling constants. Since the isomerization process from keto−keto to keto−enol forms was observed to be very slow ($k_{ex} = 3.8 \times 10^{-4}$ s^{-1} at 298 K), it is not anticipated to influence the measured N−N coupling constant. The exchange between syn and anti isomers of

(31) Brown, S. P.; Perez-Torralba, M.; Sanz, D.; Claramunt, R. M.; Emsley, L. *J. Am. Chem. Soc.* **2002**, *124*, 1152−1153.

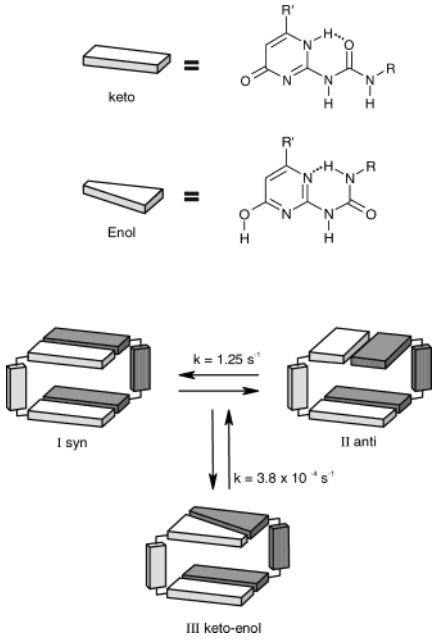
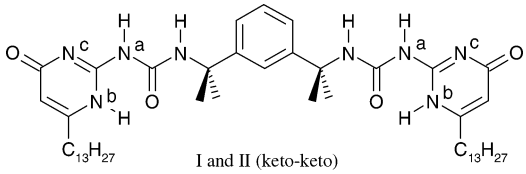
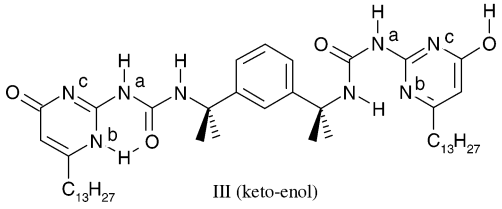


FIGURE 5. Schematical representation of the interconversion of isomers I, II, and III of **5a**.

SCHEME 2. Labeling of the Nitrogen Atoms in the Isomers of 5





the keto−keto dimers is significantly faster with a k_{ex} of 1.25 s^{-1} at 298 K.

This value is, however, approximately 4 times lower than the coupling constants across the hydrogen bonds (see below) and will therefore not lead to significant averaging of the coupling information over the two isomers. Moreover, syn−anti isomerization takes place without exchange of UPy groups and hence coupling is maintained.

The ^{1}H NMR spectrum of **5a** in $CDCl_3$ is rather complex due to coupling to ^{15}N nuclei, but the peaks were assigned using spectra of the unlabeled analogue and the unlabeled derivative **5b**. The shifts of the NH and alkylidene protons in the ^{1}H NMR spectrum of **5a** and **5b** are very similar Although the isomeric ratios in **5a** and **5b** differ slightly, the close correspondence in the peak positions of **5a** and **5b** was used to assign the signals of **5a** to the isomers I−III. With the aid of this assignment, the fraction of each isomer was determined from integration of the proton signals to be 0.12, 0.43, and 0.45, respectively.

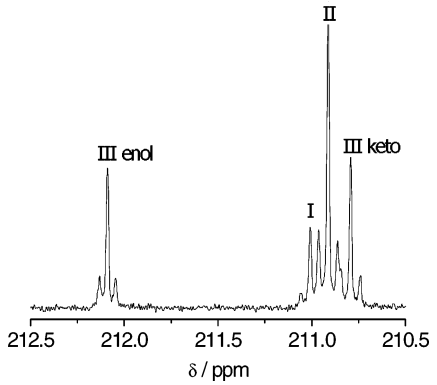


FIGURE 6. N_c region of the inverse-gated ^{1}H-decoupled ^{15}N NMR spectrum of ^{15}N-labeled **5a** in $CDCl_3$ at 25 °C.

The inverse-gated ^{1}H-decoupled ^{15}N NMR spectrum recorded at 25 °C is also complex, but in this case, coupling constants could be resolved without resorting to the less sensitive 1D-INADEQUATE experiment. In the low-field region of the spectrum, where the atoms N_c of isomers I−III resonate, four different signals are observed (Figure 6). All four signals consist of a combination of a noncoupled singlet and a $^{2h}J_{NN}$-coupled doublet in the expected 1:2 ratio. The spectrum was deconvoluted by fitting a set of Lorentzian peaks to the spectrum. The signals were subsequently assigned to isomers I−III by comparison of the ^{15}N integral ratios to the integral ratios in the ^{1}H NMR spectrum. Isomer III is asymmetric and gives rise to two signals for N_c at 212.1 and 210.8 ppm. Because its position is very close to the signals of N_c in isomers I and II, the signal at 210.8 ppm was assigned to N_c of the UPy in its keto tautomeric form. The deconvolution procedure also afforded the intermolecular $^{2h}J_{NN}$ coupling constants. The values of $^{2h}J_{NN}$ of isomer I, II, and the keto moiety of isomer III (4.96 ± 0.03, 5.13 ± 0.03, and 5.27 ± 0.03 Hz, respectively) are close to the value of 5.1 Hz found for compound **4a** at −20 °C. The coupling constant across the enol UPy dimer of III at 4.37 Hz is significantly smaller.

Correlation of Coupling Constants with Hydrogen Bond Lengths. Interpretation of the observed intermolecular coupling constants in dimers of **4a** and **5a** in terms of interatomic distances would be highly interesting and would establish the use of this parameter for structural analysis of other stable supramolecular complexes. Experimental results and theoretical calculations in the biomolecular area indicate that the magnitude of the coupling constant is closely related to the N−N distance across the hydrogen bond.[14−17] However, for a given N−N distance, the value of the coupling constant is predicted to be also dependent on the position of the hydrogen atom, with the highest coupling constant for a symmetric hydrogen bond.[15] Hydrogen bonds between neutral molecules are generally not symmetric, and usually detailed X-ray analysis of the position of the hydrogen atoms is not available. In principle, the distances in a linear N−H···N hydrogen-bonded system can be determined from the ratio of dipolar couplings $^1D_{NH}$ and $^{1h}D_{NH}$.[32] By use of the experimental data of **4a** and

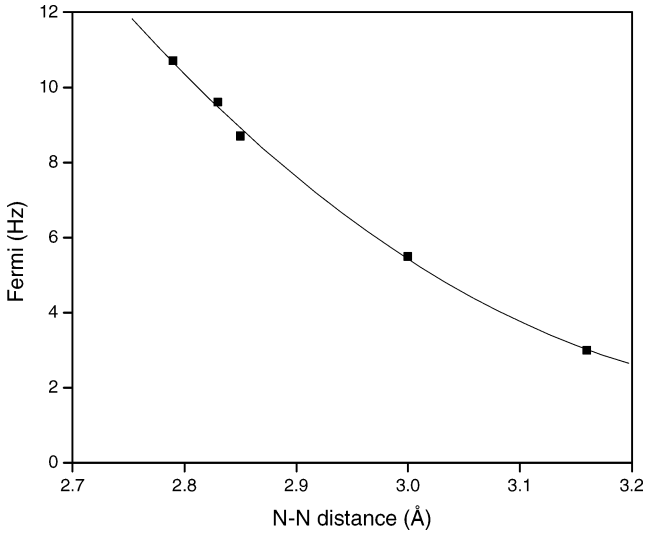


FIGURE 7. Correlation between N−N distances and the Fermi contact term of $^{2h}J_{NN}$ in equilibrium structures of neutral hydrogen bonded complexes. Data points correspond to values calculated for five different linear hydrogen-bonded complexes of CNH (adapted from ref 16).

5a, we like to test a simpler approach, using the correlation between $^{2h}J_{NN}$ coupling constants and N−N distances proposed by Del Bene and Bartlett.[16,17] These authors have calculated the value of the Fermi contact term (which was shown to dominate $^{2h}J_{NN}$) for a number of neutral hydrogen bonded complexes in their equilibrium geometry to show that the coupling constant smoothly varies with the N−N distance. This relationship, which is reproduced in Figure 7, yields $^{2h}J_{NN}$ coupling constants that are in excellent agreement with experimental values in adenine−uracil and guanine−cytosine base pairs. Nevertheless, experimental verification of this correlation in other hydrogen bonded complexes is essential to establish its general use. This prompted us to test the relationship in Figure 7 by comparing predicted N−N distances with those observed in crystal structures of **4** and **5**.

For the monofunctional and bifunctional UPy derivatives **4b** and **4c**, and for bifunctional derivative **5b**, X-ray crystal structures have been published. The average of the four hydrogen bonded N−N distances in the crystal structures of **5b**-I (containing two independent hydrogen bonded N−N distances in each of the two crystal modifications) is 2.995 Å, while the average of two independent distances in isomer II is 2.981 Å, respectively. The hydrogen bonds are, however, not linear. The N−H···N angles in the crystal structures vary between 162° and 168°. The effect of nonlinearity of the hydrogen bond on $^{2h}J_{NN}$ has been discussed in the framework of the relationship of Figure 7.[17] It can be deduced from the literature data that a deviation of 15° from linearity reduces the value of the coupling constant from 5.5 to 5.0 Hz for a complex with an equilibrium N−N distance of 3.0 Å. The value of $^{2h}J_{NN}$ measured in isomers I and II, and in the keto part of dimer III, correspond to an N−N distance of 2.98−3.0 Å if it is assumed that in solution the N−H···N angles are similar to the values in the crystal. The predictions from the relationship proposed by Del Bene et al. are therefore in perfect agreement with the experimental data on compound **5**. The smaller coupling constant across the hydrogen bond in the enol part of isomer III results in a calculated N−N distance of 3.05 Å. Unfortunately, a crystal structure of **5**-III is not available. Therefore, an estimate of the nonlinearity of this bond cannot be made nor can the calculated hydrogen bond length be compared with the crystal data. However, from the NMR data, it is predicted that the hydrogen bond length in the enol tautomer is slightly longer than that in the keto tautomer. This is in agreement with the hydrogen bond lengths in crystal structures of both keto and enol modifications of monofunctional 6-phenyl derivative **4b**. The N−N distance in the enol modification is 2.98 Å, 0.03 Å longer than that in the keto tautomer. Thus, a trend toward slightly longer hydrogen bonds in the enol tautomer found in the solid state is confirmed in the enol part of **5**-III in solution. Finally, having established a satisfactory correspondence between hydrogen-bonded distances in the solid state and those calculated with the use of coupling constants, we may conclude that the observed peak splitting in the spectrum of **4b** is similar to the coupling constants measured in **5a** and that this value must therefore be close to the actual coupling constant in the absence of chemical exchange. This conclusion is further supported by the correspondence with the hydrogen bond length in the crystal structure of 6-methyl derivative **4c**, which is 2.97 Å.

Conclusions

By use of intermolecular coupling across hydrogen bonds in ^{15}N-labeled derivatives, the present work unequivocally establishes the hydrogen bonding pattern in UPy dimers in solution. The structure of these dimers is in agreement with the solid-state structures determined previously in the solid state by X-ray diffraction and DQ-MAS NMR. The high stability of UPy dimers allows the observation of $^{2h}J_{NN}$ coupling constants at significantly higher and experimentally more easily accessible temperatures than for synthetic complexes that have been reported previously. Very slow chemical exchange of the dimers of **5** makes this the first synthetic complex for which coupling across intermolecular hydrogen bonds could be observed at room temperature. Comparison of the hydrogen bond lengths calculated from $^{2h}J_{NN}$ coupling constants with the distances in X-ray structures of **5** lends support to the use of the general relationship between N−N distance and $^{2h}J_{NN}$ proposed by Del Bene et al.

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Supporting Information Available: General experimental procedures; synthesis of compounds **2**, **3**, **4a**, and **5a**; and ^{1}H NMR and ^{13}C NMR spectra of compound **5a** in PDF. This material is available free of charge via the Internet at http://pubs.acs.org.

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(32) Wu, Z. R.; Ono, A.; Kainosho, M.; Bax, A. *J. Biomol. NMR* **2001**, *19*, 361−365.