Article

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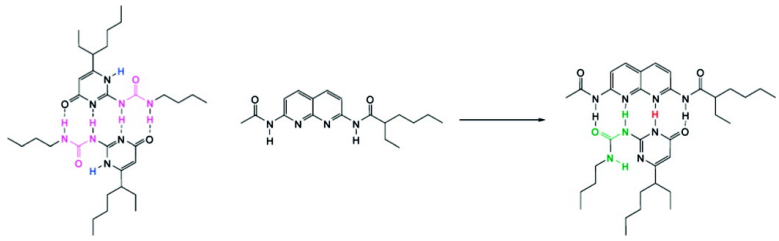

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The Mechanism of Ureido-Pyrimidinone:2,7-Diamido-Naphthyridine Complexation and the Presence of Kinetically Controlled Pathways in Multicomponent Hydrogen-Bonded Systems

Tom F. A. de Greef,[†] G. B. W. L. Ligthart,[†] Martin Lutz,[‡] Anthony L. Spek,[‡] E. W. Meijer,*[,†] and Rint P. Sijbesma*[,†]

Laboratory of Macromolecular and Organic Chemistry, Eindhoven University of Technology, P.O. Box 513, 5600 MB Eindhoven, The Netherlands, and Bijvoet Center for Biomolecular Research, Crystal and Structural Chemistry, Utrecht University, Padualaan 8, 3584 CH Utrecht, The Netherlands

Received November 12, 2007; E-mail: e.w.meijer@tue.nl; r.p.sijbesma@tue.nl

Abstract: The kinetics of association of ureido-pyrimidinone (U) dimers, present either in the 4[1*H*]-keto form or in the pyrimidin-4-ol form, with 2,7-diamido-1,8-naphthyridine (N) into a complementary heterodimer have been investigated. The formation of heterodimers with 2,7-diamido-1,8-naphthyridine from pyrimidin-4-ol dimers is much faster than from 4[1*H*]-pyrimidinone dimers. Using a combination of simple measurements and simulations, evidence for a bimolecular tautomerization step is presented. Finally, the acquired kinetic knowledge of the different pathways leading from ureido-pyrimidinone homodimers to ureido-pyrimidinone:diamido-naphthyridine (U:N) heterodimers allows the prediction and observation of kinetically determined ureido-pyrimidinone heterodimers which slowly convert back to the corresponding homodimers.

Introduction

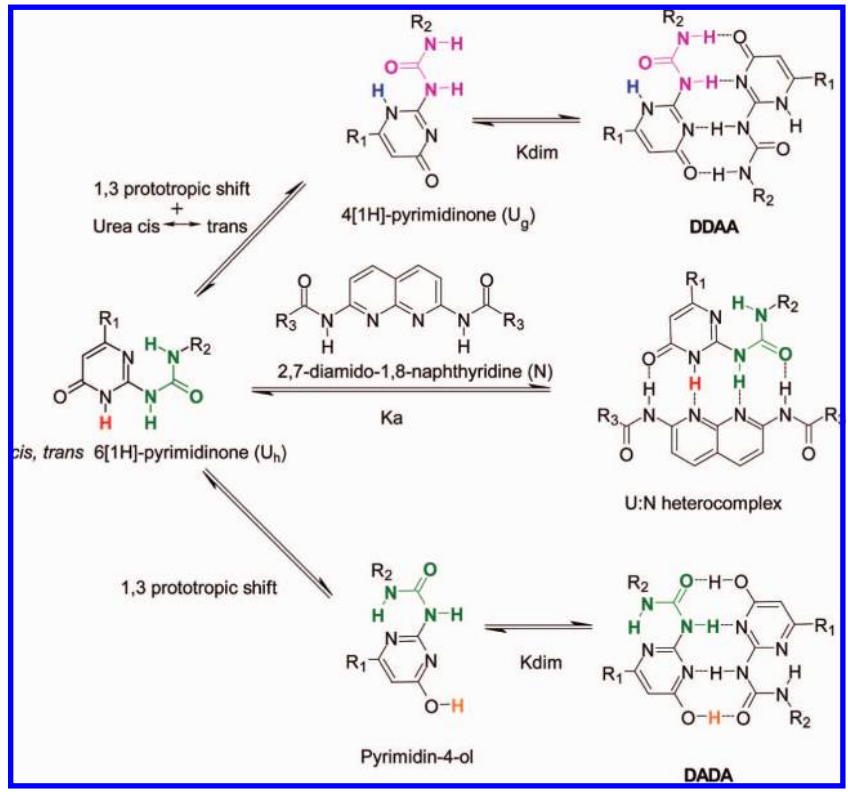
The role of kinetic control in self-assembly processes has recently attracted considerable interest. The formation of kinetically determined self-assemblies has been observed in systems of large polydisperse aggregates[1] as well as in discrete multicomponent supramolecular assemblies.[2] Although the physical basis of self-assembly under thermodynamic control is well-understood,[3] a theoretical description of the formation of supramolecular assemblies under kinetic control is less well-developed. The lack of theory makes it more difficult to analyze and understand the role of subtle effects, such as solvent shells surrounding the periphery of large supramolecular aggregates in mixed solvent (good/poor) compositions and the effect of fast cooling on the formation of supramolecular assemblies. Despite the lack of theory, the formation of kinetically controlled supramolecular products can lead to supramolecular systems displaying highly desirable features such as chiro-optical memory[4] and dynamic chiral amplification.[5] Furthermore, it is expected that an increase in the number of weak secondary interactions in a given supramolecular assembly will lead to a larger number of kinetically controlled products. Therefore, the characterization of both the ground state−ground state as well as the ground state−transition state energy differences of novel supramolecular assemblies can establish the generality of the transient formation of supramolecular complexes and provides an understanding of how the phenomenon of kinetic recognition is used in Nature to enhance biochemical specificity.[6]

The kinetics of supramolecular assemblies are closely connected to the macroscopic properties of the resulting supramo-

[†] Eindhoven University of Technology.
[‡] Utrecht University.

(1) (a) Lohr, A.; Lysetska, M.; Würthner, F. *Angew. Chem., Int. Ed.* **2005**, *44*, 5071. (b) Jyothish, K.; Hariharan, M.; Ramaiah, D. *Chem.—Eur. J.* **2007**, *13*, 5944. (c) Jonkheijm, P.; Miura, A.; Zdanowska, M.; Hoeben, F. J. M.; De Feyter, S.; Schenning, A. P. H. J.; De Schyver, F. C.; Meijer, E. W. *Angew. Chem., Int. Ed.* **2004**, *43*, 74.
(2) (a) Mathias, J. P.; Seto, C. T.; Simanek, E. E.; Whitesides, G. M. *J. Am. Chem. Soc.* **1994**, *116*, 1725. (b) Mathias, J. P.; Simanek, E. E.; Seto, C. T.; Whitesides, G. M. *Angew. Chem., Int. Ed. Engl.* **1993**, *32*, 1766. (c) Prins, L. J.; De Jong, F.; Timmerman, P.; Reinhoudt, D. N. *Nature* **2000**, *408*, 181. (d) Prins, L. J.; Neuteboom, E. E.; Paraschiv, V.; Crego-Calama, M.; Timmerman, P.; Reinhoudt, D. N. *J. Org. Chem.* **2002**, *67*, 4808. (e) Paraschiv, V.; Crego-Calama, M.; Ishi-i, T.; Padberg, C. J.; Timmerman, P.; Reinhoudt, D. N. *J. Am. Chem. Soc.* **2002**, *124*, 7638. (f) Mukhopadhyay, P.; Zavalij, P. Y.; Isaacs, L. *J. Am. Chem. Soc.* **2006**, *128*, 14093. (g) Oshikiri, T.; Takashima, Y.; Yamaguchi, H.; Harada, A. *J. Am. Chem. Soc.* **2005**, *127*, 12186. (h) Dyck, A. S. M.; Kisiel, U.; Bohne, C. *J. Phys. Chem. B* **2003**, *107*, 11652. (i) Nordel, P.; Westerlund, F.; Wilhelmsson, M.; Nord; en, B.; Lincoln, P. *Angew. Chem., Int. Ed.* **2007**, *46*, 2203. (j) Badjić, J. D.; Cantrill, S. J.; Stoddart, J. F. *J. Am. Chem. Soc.* **2004**, *126*, 2288.
(3) (a) Ercolani, G. *J. Phys. Chem. B* **1998**, *102*, 5699. (b) Ercolani, G. *J. Phys. Chem. B* **2003**, *107*, 5052.
(4) Johnson, R. S.; Yamazaki, T.; Kovalenko, A.; Fenniri, H. *J. Am. Chem. Soc.* **2007**, *129*, 5735.
(5) Prins, L. J.; Timmerman, P.; Reinhoudt, D. N. *J. Am. Chem. Soc.* **2001**, *123*, 10153.
(6) (a) Hopfield, J. J. *Proc. Natl. Acad. Sci. U.S.A.* **1974**, *71*, 4135. (b) Yamane, T.; Hopfield, J. J. *Proc. Natl. Acad. Sci. U.S.A.* **1977**, *74*, 2246.
(7) Brunsveld, L.; Folmer, B. J. B.; Meijer, E. W.; Sijbesma, R. P. *Chem. Rev.* **2001**, *101*, 4071.
(8) Lehn, J.-M. *Supramolecular Chemistry*. Wiley-VCH: Weinheim, Germany, 1995.
(9) Ciferri, A. *Supramolecular Polymers*. Marcel Dekker: New York, 2000.
(10) Zimmerman, N.; Moore, J. S.; Zimmerman, S. C. *Chem. Ind.* **1998**, 604.
(11) Ciferri, A. J. *Macromol. Sci. Polym. Rev.* **2003**, *43*, 271.

Scheme 1. Proposed Intermediates in the Exchange of the Different Tautomeric Forms of Ureido-Pyrimidinone Dimers with 2,7-Diamido-1,8-Naphthyridine (The Substituents R_1, R_2, and R_3 Can Be Any Functional Group)



lecular materials. Supramolecular polymers[7–15] based on reversible interactions between arrays of hydrogen bonds localized at the endgroups of polymeric spacers have attracted interest as a new powerful tool toward functional materials. In supramolecular polymer-based materials, stress relaxation during network deformation mainly occurs by the kinetic creation and annihilation of the reversible defining interaction.[15,16] As has been elegantly shown by Craig,[17] the dynamic relaxation rates of supramolecular polymer-based networks are often comparable to the dissociation rate constant of the defining noncovalent interaction. On the basis of the previously discussed mechanism of stress relaxation in supramolecular polymer-based networks, the creation of a kinetically stable noncovalent interaction instead of the expected thermodynamically formed interaction can lead to unexpected dynamical mechanical properties. For a rational design of supramolecular polymer-based materials, characterization of rate constants and determination of the complexation mechanism of novel developed supramolecular modules[18] is an important task.

We[19] and others[20–22] have shown the formation of supramolecular polymers based on strong complementary hydrogen bond interactions between 2,7-diamido-1,8-naphthyridine (N) and 2-ureido-pyrimidinone (U), which is also able to form homodimers (U_2). This dual complexation mode combined with the high selectivity for heterodimerization makes the U:N system eminently suitable for formation of supramolecular block copolymers[23–26] and supramolecular graft copolymers.[27]

(12) Castellano, R. K.; Clark, R.; Craig, S. L.; Nuckolls, C.; Rebek, J., Jr *Proc. Natl. Acad. Sci. U.S.A.* **2000**, *97*, 12418.
(13) Gibson, H. W.; Yamaguchi, N.; Jones, J. W. *J. Am. Chem. Soc.* **2003**, *125*, 3522.
(14) (a) Arnaud, A.; Belleney, J.; Boué, F.; Bouteiller, L.; Carrot, G.; Wintgens, V. *Angew. Chem., Int. Ed.* **2004**, *43*, 1718. (b) Arnaud, A.; Bouteiller, L. *Langmuir* **2004**, *20*, 6858.
(15) 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.
(16) Serpe, M. J.; Craig, S. L. *Langmuir* **2007**, *23*, 1626.
(17) (a) Yount, W. C.; Juwarker, H.; Craig, S. L. *J. Am. Chem. Soc.* **2003**, *125*, 15302. (b) Yount, W. C.; Loveless, D. M.; Craig, S. L. *Angew. Chem., Int. Ed.* **2005**, *44*, 2746. (c) Loveless, D. M.; Jeon, S. L.; Craig, S. L. *Macromolecules* **2005**, *38*, 10171.
(18) Selected examples discussing the importance of kinetics in supramolecular chemistry: General: (a) Davis, A. V.; Yeh, R. M.; Raymond, K. N. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 4793. (b) Nau, W. M.; Wang, X. *ChemPhysChem* **2002**, *3*, 393Organogels: (c) Huang, X.; Terech, P.; Raghavan, S. R.; Weiss, R. G. *J. Am. Chem. Soc.* **2005**, *127*, 4336. Host-guest: (d) Palmer, L. C.; Rebek, J., Jr *Org. Biomol. Chem.* **2004**, *2*, 3051. (e) Rudkevich, D. M.; Hilmersson, G.; Rebek, J., Jr *J. Am. Chem. Soc.* **1997**, *119*, 9911. (f) Castellano, R. K.; Craig, S. L.; Nuckolls, C.; Rebek, J., Jr. *J. Am. Chem. Soc.* **2000**, *122*, 7876. (g) Goshe, A. J.; Steele, I. M.; Ceccarelli, C.; Rheingold, A. L.; Bosnich, B. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 4823. (h) Gibb, C. L. D.; Li, X.; Gibb, B. C. *Proc. Natl. Acad. Sci. U.S.A.* **2002**, *99*, 4857. Nucleation−elongation: (i) Oosawa, F.; Kasai, M. *J. Mol. Biol.* **1962**, *4*, 10. (j) Pasternack, R. F.; Gibbs, E. J.; Collings, P. J.; dePaula, J. C.; Turzo, L. C.; Terracina, A. *J. Am. Chem. Soc.* **1998**, *120*, 5873. Hydrogen-bonded complexes: (k) Prins, L. J.; Neuteboom, E. E.; Paraschiv, V.; Crego-Calama, M.; Timmerman, P.; Reinhoudt, D. N. *J. Org. Chem.* **2002**, *67*, 4808. (l) Mogck, O.; Pons, M.; Böhmer, V.; Vogt, W. *J. Am. Chem. Soc.* **1997**, *119*, 5706. (m) Das, G.; Talukdar, P.; Matile, S. *Science* **2002**, *298*, 1600. Self-replicating peptides: (n) Kennan, A. J.; Haridas, V.; Severin, K.; Lee, D. H.; Ghadiri, M. R. *J. Am. Chem. Soc.* **2001**, *123*, 1797. Metallic helicates: (o) Elhabiri, L.; Hamacek, J.; Bünzli, J-.C. G.; Albrecht-Gary, A. M. *Eur. J. Inorg. Chem.* **2004**, 51Other: (p) Milic, T. N.; Chi, N.; Yablon, D. G.; Flynn, G. W.; Batteas, J. D.; Drain, C. M. *Angew. Chem., Int. Ed.* **2002**, *41*, 2117.
(19) Ligthart, G. B. W. L.; Ohkawa, H.; Sijbesma, R. P.; Meijer, E. W. *J. Am. Chem. Soc.* **2005**, *127*, 810.
(20) Park, T.; Todd, E. M.; Nakashima, S.; Zimmerman, S. C. *J. Am. Chem. Soc.* **2005**, *127*, 18133.
(21) Corbin, P. S.; Zimmerman, S. C. *J. Am. Chem. Soc.* **1998**, *120*, 9710.
(22) Wang, X.-Z.; Li, X.-Q.; Shao, X.-B.; Zhao, X.; Deng, P.; Jiang, X.-K.; Li, Z.-T.; Chen, Y.-Q. *Chemistry* **2003**, *9*, 2904.

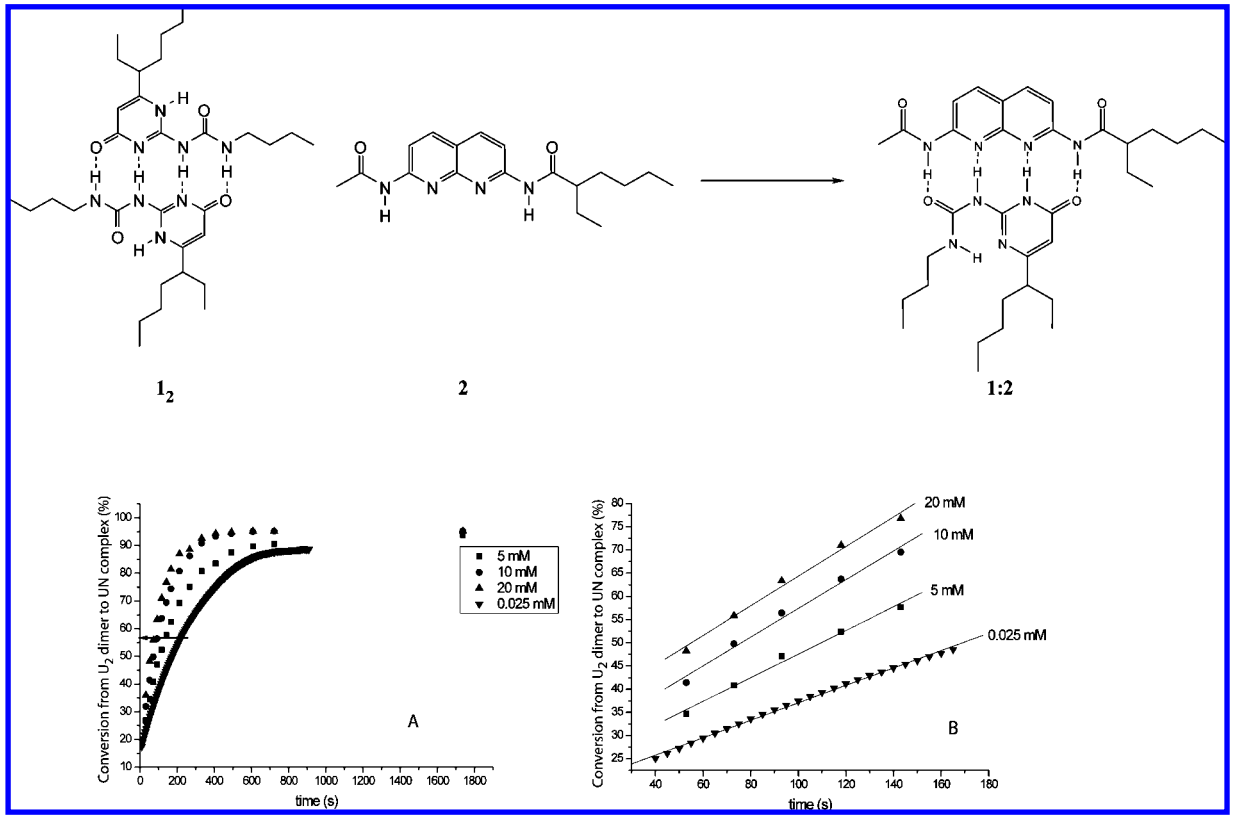


Figure 1. (A) Conversion from **DDAA** dimers to U:N heterocomplex **1**:**2** as a function of time after injection of 1 equiv of **1** for several total concentrations of **1** + **2** as determined with ^{1}H NMR (5, 10, and 20 mM) and UV–vis spectroscopy (0.025 mM). The arrow indicates an increase in the total concentration of **1** and **2**. (B) Expansion of the conversion−time plot displaying the increase of the initial conversion rate upon increasing the total concentration.

Herein we investigate and compare the association kinetics of 2,7-diamido-1,8-naphthyridine with the dimerizing tautomers of 2-ureido-4[1*H*]-pyrimidinone and 2-ureido-pyrimidin-4-ol.[28] Although several pathways for the exchange of U_2 dimers with N may be postulated, we will focus primarily on dissociative pathways in which U:N complexation starts with the dissociation of U_2 into the respective monomeric forms since kinetic measurements on the exchange of U:U homo- and heterodimers, present in their 4[1*H*]-keto tautomeric form,[29] have shown that this is a dissociative process. Scheme 1 displays the elementary steps by which either 2-ureido-4[1*H*]-pyrimidinone dimers or 2-ureido-pyrimidin-4-ol dimers can associate with 2,7-diamido-1,8-naphthyridine. The key reaction step during the formation of the U:N heterocomplex from the corresponding U_2 homodimers is in both cases a [1,3] prototropic shift resulting in the formation of the *cisoid,transoid* ureido-6[1*H*]-pyrimidinone monomer. The [1,3] prototropic shift of nitrogen-containing acyclic and heterocyclic compounds has been a subject of intense research[30] in the past few years. Considerable evidence, mainly from computational studies, has emerged, indicating that the lowest energy pathway for the [1,3] prototropic shift in such compounds proceeds via an intermolecular (dimeric, trimeric, oligomeric) proton transfer instead of a stepwise intramolecular proton transfer.[31–34]

In the present work, we investigate the mechanism of U:N complexation (*inter*molecular vs *intra*molecular tautomerization) using a combination of UV–vis and ^{1}H NMR spectroscopy.

Kinetics of Keto Dimers

^{1}H NMR spectroscopy was used to monitor the formation of U:N heterocomplex **1**:**2** from ureido-pyrimidinone dimer $\mathbf{1_2}$ in toluene-d_8 (present for 90% as keto dimers and 10% as enol dimers in this solvent) upon the addition of equimolar amounts of diamido-naphthyridine **2** (Figure 1). To rule out a solvent-assisted tautomerization pathway, we chose to perform the equilibration experiments in toluene-d_8, an apolar solvent, which lacks any acidic hydrogens or basic sites. Upon rapid injection of 50 μL of a 200 mM solution of **1** in toluene-d_8 to 1 mL of a 10 mM solution of **2**, the equilibration of both homodimeric forms of **1** to the U:N heterodimer **1**:**2** was observed with ^{1}H NMR. In order to gain more insight into the mechanism of heterocomplexation, ^{1}H NMR equilibration measurements were conducted at three different equimolar concentrations of **1** and

(23) Scherman, O. A.; Ligthart, G. B. L.; Ohkawa, H.; Sijbesma, R. P.; Meijer, E. W. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, *103*, 11850.
(24) Park, T.; Zimmerman, S. C. *J. Am. Chem. Soc.* **2006**, *128*, 13986.
(25) Park, T.; Zimmerman, S. C.; Nakashima, S. *J. Am. Chem. Soc.* **2005**, *127*, 6520.
(26) Park, T.; Zimmerman, S. C. *J. Am. Chem. Soc.* **2006**, *128*, 14236.
(27) Ohkawa, H.; Ligthart, G. B. W. L.; Sijbesma, R. P.; Meijer, R. P. *Macromolecules* **2007**, *40*, 1453.
(28) Beijer, F. H.; Sijbesma, R. P.; Kooijman, H.; Spek, A. L.; Meijer, E. W. *J. Am. Chem. Soc.* **1998**, *120*, 6761.
(29) Söntjens, S. H. M.; Sijbesma, R. P.; van Genderen, M. H. P.; Meijer, E. W. *J. Am. Chem. Soc.* **2000**, *122*, 7487.
(30) For a review on the mechanisms of a variety of prototropic tautomerizations believed to be intermolecular see Kereselidze, J. A.; Zarqua, T. Sh.; Kikalishvili, T. J.; Churgulia, E. J.; Makaridze, M. C. *Russ. Chem. Rev.* **2002**, *71*, 993.
(31) Nagy, P. I.; Tejada, F. R.; Messer, W. S., Jr *J. Phys. Chem. B* **2005**, *109*, 22588.
(32) Chou, P.-T.; Wei, C.-Y.; Hung, F.-T. *J. Phys. Chem. B* **1997**, *101*, 9119.
(33) Freeman, F.; Po, H. N. *J. Phys Chem. A* **2006**, *110*, 7904.
(34) Yang, Z.; Rodgers, M. T. *Phys. Chem. Chem. Phys.* **2004**, *6*, 2749.

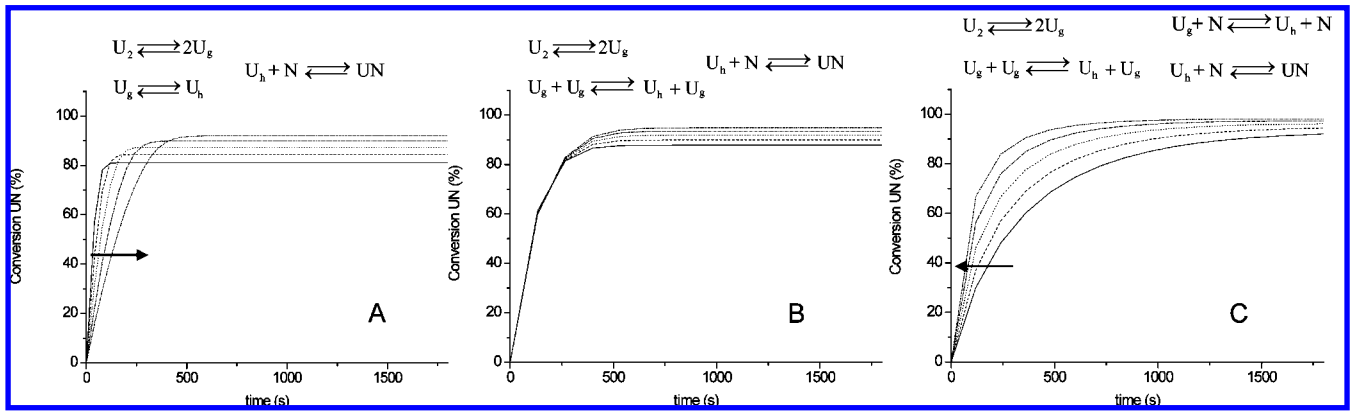


Figure 2. Simulated plots of U:N heterocomplex conversion versus time at different concentrations (from 0.05 to 50 mM) for 1:1 mixtures of **1** and **2** for (A) unimolecular tautomerization model, (B) bimolecular U assisted tautomerization, (C) N and U assisted bimolecular tautomerization. The arrow indicates an increase in the total concentration of **1** and **2**.

2 (5, 10, and 20 mM). Furthermore, we monitored the formation of the U:N heterocomplex from an equimolar mixture of **1** and **2** at a concentration of 0.025 mM for **2** using UV–vis spectroscopy.[35] Formation of the heterodimers from the enol dimer (present in 10%) was too fast to be studied with the equilibration measurements at all concentrations. In fact, all enol dimer had completely disappeared after 18 s when data collection was started.[36] The kinetics of this exchange process were therefore studied with a different technique (vide infra). Conversion of the keto dimer on the other hand could be followed conveniently using the 1:1 equilibration experiments. As is evident from the graphs in Figure 1, the initial conversion rate of the heterocomplex increases with concentration, implying that the overall order in reactants is higher than 1.

Guided by this important observation, we simulated possible dissociative kinetic mechanisms using the kinetic simulation program Gepasi.[37,38] The first minimalist kinetic representation we considered to accurately include the essential features of the dynamic system, as depicted in Scheme 1, corresponds to a mechanism in which the tautomerization from the 2-ureido-4[1*H*]-pyrimidinone monomeric form (U_g) to the 2-ureido-6[1*H*]-pyrimidinone monomeric form (U_h) is a unimolecular event (Figure 2A). As expected, simulation of the conversion of the U:N complex as a function of total concentration in equimolar mixtures of **1** and **2** using this kinetic model reveals that the initial conversion rate becomes lower at higher concentrations, in sharp contrast with the experimental results. This concentration dependence results from the fact that in a completely unimolecular tautomerization mechanism, as depicted in Figure 2A, the order in total ureido-pyrimidinone concentration (U_{tot}) is 0.5 when quasi-steady-state conditions are assumed since nearly all U is present in dimers (see Supporting Information).

A recent theoretical study[39] has shown that tautomerization via intermolecular double proton transfer has a lower energy barrier compared to a stepwise intramolecular proton transfer. The intermolecular process is facilitated by intermolecular hydrogen bonding. To study the influence of a bimolecular tautomerization event, we simulated conversion−time plots for different total concentrations of **1** and **2** in 1:1 mixtures according to the kinetic scheme depicted in Figure 2B. In this mechanism, tautomerization of the 2-ureido-4[1*H*]-pyrimidinone monomer (U_g) to the 2-ureido-6[1*H*]-pyrimidinone (U_h) monomeric form occurs via a bimolecular transition state.

Simulation of the conversion−time plots using this mechanism shows that the initial rate of conversion of U:N is independent of the total concentration in 1:1 mixtures of **1** and **2** as expected from a mechanism in which the order in U_{tot} is exactly 1 (again assuming quasi-steady-state conditions, see Supporting Information). Since the experimental results show that the initial conversion rate becomes higher at higher concentration, this kinetic model is inappropriate to describe our experimental data, as well.

To account for the lower initial conversion rate of U:N at lower concentration, there are two possibilities. First, if formation of the U:N heterocomplex from the free 6[1*H*] monomer (U_h) and N is the rate-determining step, the initial conversion rate will decrease upon decreasing the concentration. However, it has been shown by Hammes[40] that the association rate constant of double and triply hydrogen bonded complexes in organic solvents is a diffusion-controlled process. Therefore, the tautomerization from U_g to U_h is the rate-determining step in the complexation between U homodimers and N. We propose that the tautomerization of the 2-ureido-4[1*H*]-pyrimidinone

(35) Upon addition of **1** to a solution of diamido-naphthyridine **2** (2.54×10^{-5} M) in toluene, the absorption intensity at 357 nm increased. A similar red shift upon formation of the U:N heterocomplex in $CHCl_3$ has been reported by Zimmerman (see : Park, T.; Todd, E. M.; Nakashima, S.; Zimmerman, S. C. *J. Am. Chem. Soc.* **2005**, *127*, 18133) and was also observed by Leigh and co-workers (Djurdjevic, S.; Leigh, D. A.; McNab, H.; Parsons, S.; Teobaldi, G.; Zerbetto, F. *J. Am. Chem. Soc.* **2007**, *129*, 476) for a hydrogen-bonded anthyridine-based DDD−AAA dimer. In order to calculate the concentration of UN from the absorbance at 357 nm as a function of time, we conducted a separate titration experiment in toluene to determine the extinction constant of the U:N complex ($\varepsilon_{NU} = 33.1 \times 10^3$ L mol cm^{-1}) as well as an estimation of the association constant K_a (between 10^7 and 5×10^8 M^{-1}) by nonlinear curve fitting using a 1:1 binding model in which one of the components can self-associate. The full details of these experiments can be found in the Supporting Information.

(36) This can more clearly be observed from the experimental data in Figure 1 which shows that extrapolation to $y = 0$ results in an intercept with the negative *x*-axis, indicating a faster process preceding the complexation of keto dimers of **1** with **2**. Therefore, we are specifically monitoring the formation of the U:N heterocomplex from **DDAA** dimers in the 1:1 kinetic equilibration experiments.

(37) Mendes, P. *Trends Biochem. Sci.* **1997**, *22*, 361.

(38) The GEPASI simulations are available as text files as Supporting Information.

(39) Padermshoke, A.; Katsumoto, Y.; Aida, M. *J. Phys. Chem. B* **2006**, *110*, 26388.

(40) (a) Hammes, G. G.; Spivey, H. O. *J. Am. Chem. Soc.* **1966**, *88*, 1621. (b) Hammes, G. G.; Park, A. C. *J. Am. Chem. Soc.* **1969**, *91*, 956. (c) Hammes, G. G.; Park, A. C. *J. Am. Chem. Soc.* **1968**, *90*, 4151.

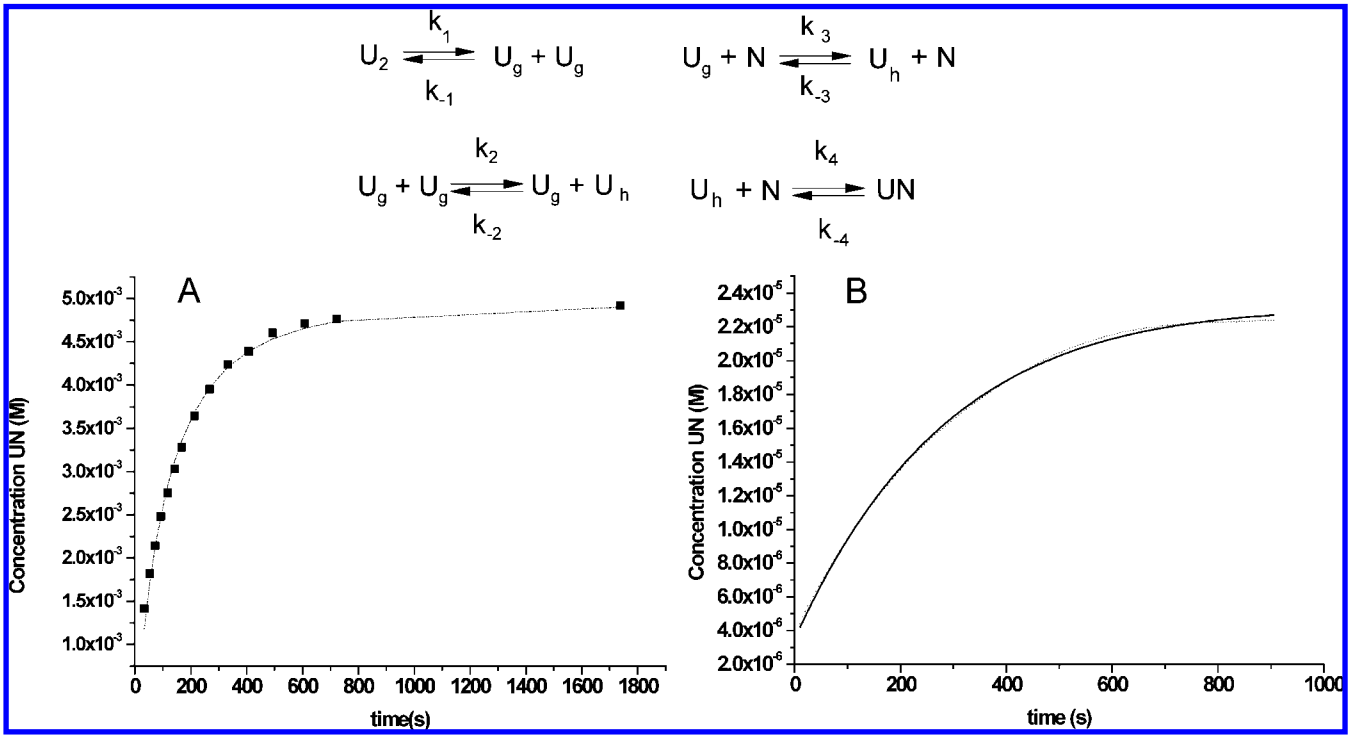


Figure 3. (A) Concentration of U:N heterocomplex upon injection of 1 equiv of **1** in toluene-d_8 to 1 mL of a 5 mM solution of **2** as calculated from the ^{1}H NMR integrals. The dotted line represents the best fitted curve ($K_{dim} = 1 \times 10^8$ M^{-1}, $K_a = 9 \times 10^7$ M^{-1}) using the kinetic model as depicted in Figure 2C. (B) Concentration of U:N heterocomplex upon titration of 1 equiv of **1** in toluene to 2 mL of a 0.025 mM solution of **2** calculated from the absorption at 357 nm as a function of time. The dotted line represents the best fitted curve ($K_{dim} = 1 \times 10^8$ M^{-1}, $K_a = 9 \times 10^7$ M^{-1}) using the kinetic model as depicted in Figure 2C.

monomeric form (U_g) to the 2-ureido-6[1*H*]-pyrimidinone monomeric form (U_h) is also catalyzed by free N. Analysis of this kinetic scheme assuming steady-state conditions (see Supporting Information) reveals that the initial rate becomes larger as the total concentration in 1:1 mixtures is increased. Further simulation of this kinetic model without any explicit steady-state assumption using Gepasi (Figure 2C) shows that the initial conversion rate of U_2 to U:N increases as the total concentration of **1** and **2** is increased only if N-catalyzed tautomerization is sufficiently fast compared to the U-catalyzed tautomerization step.

Using this kinetic model, we fitted the experimental conversion of U_2 to U:N heterocomplex as a function of time using an evolutionary programming algorithm present in Gepasi. We have argued[29] that the association rate constant (k_{-1}) of the 2-ureido-4[1*H*]-pyrimidinone monomeric form (U_g) to the 2-ureido-4[1*H*]-pyrimidinone dimeric form is a diffusion-controlled process occurring with a rate constant close to 10^{10} M^{-1} s^{-1} in toluene-d_8 and a dissociation rate constant (k_1) of 1 s^{-1}.[29] Following the results obtained by Hammes and by us, we have assumed that the association rate constant of formation of the U:N complex (k_4) from the 6[1*H*]-pyrimidinone monomeric form (U_h) with free N is also a diffusion-controlled parameter ($k_4 = 10^{10}$ M^{-1} s^{-1} for toluene[41]). The values of the rate constants k_1, k_{-1}, and k_4 were used as fixed parameters, while the estimated values of the dimerization constant (defined as $K_{dim} = [U_2]/[U_g]^2$) of **1** in toluene (between 1×10^8 and 7×10^8 M^{-1}) and the association constant (defined as $K_a = [UN]/([N] \times [U_h])$ of **1** with **2** (between 1×10^7 and 5×10^8 M^{-1}) were used as constraints in the nonlinear curve fitting of the kinetic data (Figure 3).

Because the rate-determining tautomerization step occurs via two different routes, a large number of combinations of k_2, k_{-2}, k_3, and k_{-3} are able to describe the experimental data. This results in a considerable negative covariance[42] between the different parameters and consequently results in large standard deviations. However, to test whether the mixed tautomerization model is correct, we fitted the experimental UV–vis data to a reduced model in which k_2 and k_{-2} were set to a fixed value of 0 M^{-1} s^{-1} (no U assisted tautomerization). An extra sum of squares test[43] revealed that the full model describes the data significantly better ($P < 0.0001$ for a significance level of 0.05) compared to the reduced model,[44] indicating that the both U and N contribute significantly to the tautomerization catalysis.

Complexation Kinetics of Enol Dimers

To compare the diamido-naphthyridine association kinetics of 2-ureido-4[1*H*]-pyrimidinone dimers with 2-ureido-pyrimidin-4-ol dimers, we prepared a novel ureido-pyrimidinone substituted at the C_6 position of the pyrimidinone ring with an electron-donating dibutylamino group (**3**). Compound **3** was synthesized as detailed in the Supporting Information.

(41) The diffusion-controlled rate constant was calculated using the viscosity of toluene at 293 K (η =5.58 $\times$ 10^{-4} kg m^{-1} s^{-1}) using the formula $k_{diff} = 8 \times RT/3\eta$, resulting in a value of $k_{diff} = 1.18 \times 10^{10}$ M^{-1} s^{-1}.

(42) Negative covariance indicates that higher than average values of one variable are tend to be paired with lower than average values of the other variable.

(43) Motuslky, H.; Christopoulus, A. *Fitting Models to Biological Data using Linear and Nonlinear Regression*; available at www.graphpad.com.

(44) Full model: residual sum of squares = 4.6 $\times$ 10^{-12}, 174 degrees of freedom. Reduced model: residual sum of squares = 4.52 $\times$ 10^{-11}, 176 degrees of freedom.

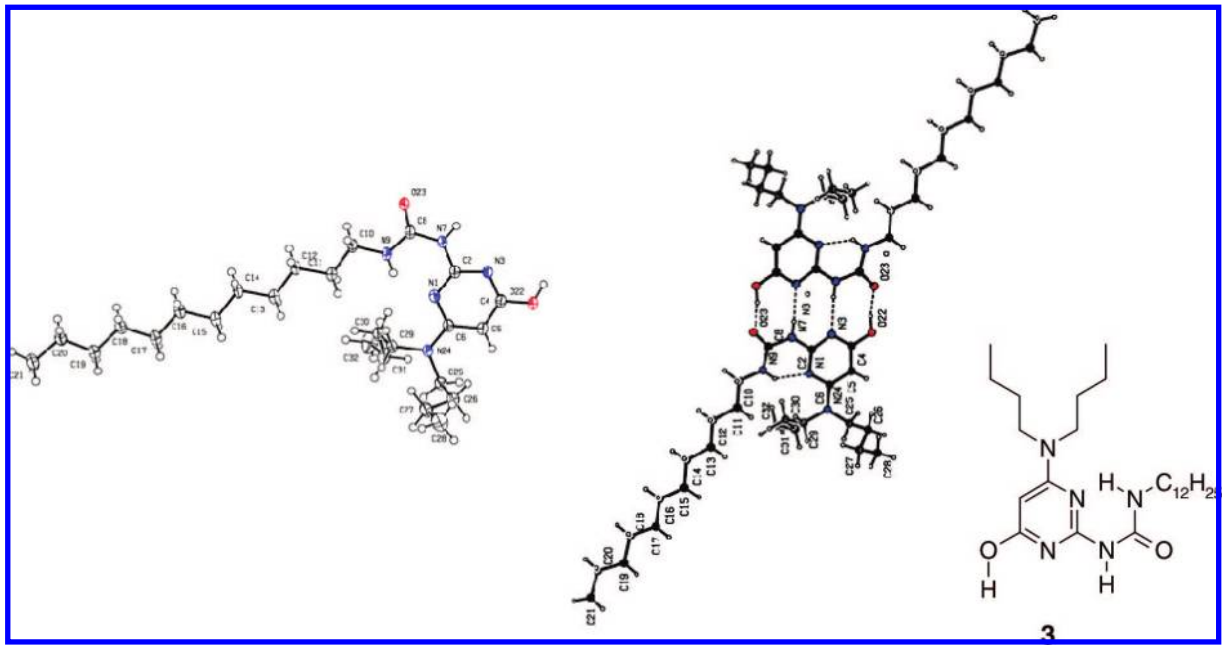


Figure 4. ORTEP diagram of the monomer geometry of **3** in the crystal. Ellipsoids represent 50% probability. PLUTON representation of the dimer geometry of **3** in the crystal.

Using a combination of X-ray crystallography (Figure 4), NOESY experiments, and FT-IR spectroscopy, the existence of an **DADA** array of the heterocycle in its pyrimidin-4-ol tautomeric form was verified both in solution (toluene and $CDCl_3$) and in the solid state (see Supporting Information).

In sharp contrast to the association kinetics of 4[1*H*]-pyrimidinone dimer $\mathbf{1}_2$, the kinetics of formation of the U:N heterocomplex **2**:**3** could not be followed by ^{1}H NMR equilibration experiments due to the extremely rapid formation of the heterocomplex after injection of a solution of **3** in toluene-d_8 to a solution of **2** in the same solvent. As the U:N complex **2**:**3** and the ureido-pyrimidinone dimer $\mathbf{3_2}$ are in slow exchange on the ^{1}H NMR time scale in this solvent, we conducted 2D-EXSY[45,46] experiments on solutions containing **3** and **2** in a 2:1 ratio (resulting in approximately equimolar amounts of $\mathbf{3_2}$ and **2**:**3**) at several concentrations of **3**.[47] The transfer functions,[48] calculated from the volume integrals of the alkylidene protons of dimer $\mathbf{3_2}$ and U:N heterodimer **2**:**3**, were fitted as a function of mixing time (τ_{mix}) according to the equation $\varphi = k_{ex}\tau_{mix}$ for all three concentrations, resulting in the pseudo-first-order rate constant k_{ex} for the exchange process (Figure 5). As can be deduced from Figure 5, the pseudo-first-order rate constant increases by 45% as the total concentration of **2** and **3** in 1:2 mixtures is increased by a factor of 4. This suggests that the kinetic pathway for association of $\mathbf{3_2}$ with **2** is similar to the association of $\mathbf{1_2}$ with **2** where tautomerization is catalyzed by monomeric components.

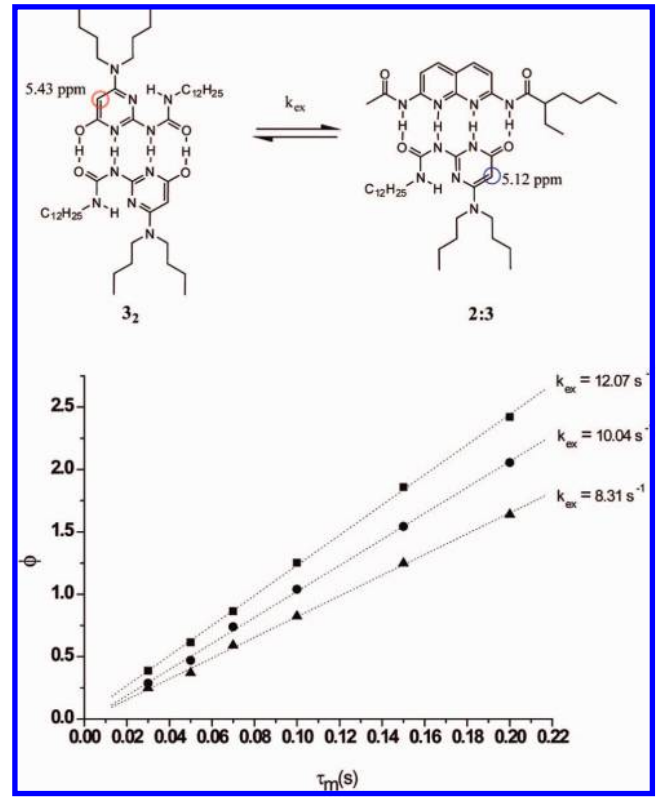


Figure 5. Plot of the transfer function φ versus mixing time for 1:2 mixtures of **2** and **3** at three different concentrations (▲ = 5 mM **2** and 10 mM **3**, ● = 10 mM **2** and 20 mM **3**, ■ = 20 mM **2** and 40 mM **3**). The dotted line represents the best fitted curve according to the equation $\varphi = k_{ex}\tau_{mix}$ (R^2 = 0.999 in all cases) in which φ represents the transfer function,[48] k_{ex} the pseudo-first-order rate constant for the exchange process, and τ_{mix} represents the mixing time during the EXSY experiment.

(45) Perrin, C. L.; Dwyer, T. J. *Chem. Rev.* **1990**, *90*, 935.
(46) Jeener, J.; Meier, B. H.; Bachmann, P.; Ernst, R. R. *J. Chem. Phys.* **1979**, *71*, 4546.
(47) 2D-EXSY measurements on a solution of 4[1*H*]-pyrimidinone dimer $\mathbf{1_2}$ and **1**:**2** in the same molar ratio did not result in cross-peaks between the diagonal peaks of the alkylidine proton of the ureido-pyrimidinone dimer and the alkylidine proton of the heterocomplex even at high mixing times (τ_{mix} > 1 s). This indicates that the site-to-site rate constant of this process is lower than 10^{-1} s^{-1} (see ref 45), in agreement with ^{1}H NMR equilibration experiments.
(48) Under the conditions that the molfraction of U:N complex is equal to the mole fraction of the U_2 dimer, the transfer function takes the following form $\varphi = \ln(r + 1/r - 1)$, where $r = (I_{AA} + I_{BB})/(I_{AB} + I_{BA})$, the ratio of diagonal and cross-peak intensities. See the Supporting Information or ref 45 for a further discussion and definition of the transfer function.

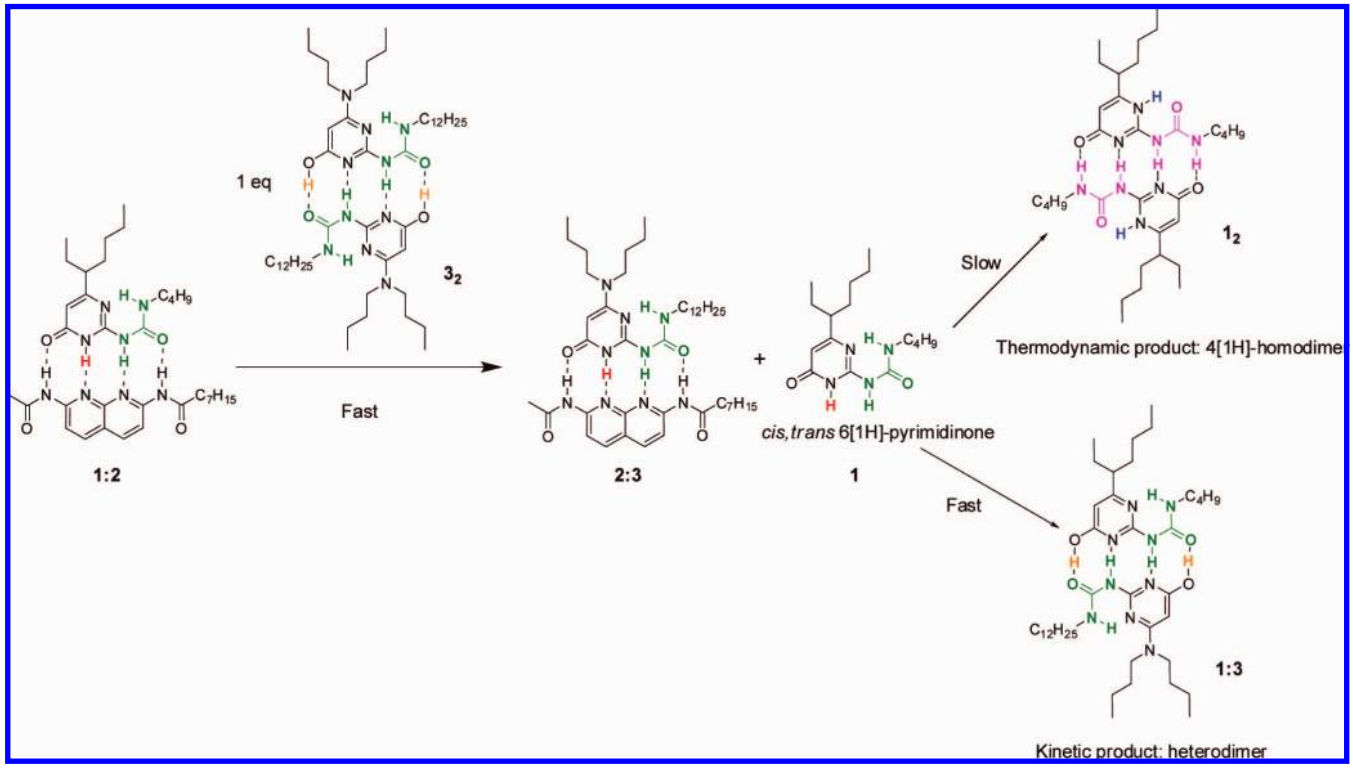


Figure 6. Kinetic and thermodynamic self-sorting in a system containing ureido-pyrimidinones **1**, **3**, and diamido-naphthyridine **2**.

Interestingly, addition[49] of 16 mol % (with respect to **3**) of benzoic acid to a solution containing 10 mM **2** and 20 mM **3** in toluene-d_8 resulted in an increase in the pseudo-first-order rate constant from 10 s^{-1} to a value of almost 40 s^{-1}. The catalytic influence of benzoic acid on the exchange between the $\mathbf{3_2}$ and the U:N complex **2**:**3** is most probably the result of a decrease in the activation barrier of the [1,3] prototropic shift by formation of an intermolecular complex between the carboxylic acid and the 2-ureido-pyrimidin-4-ol monomer. Indeed, quantum mechanical calculations on the activation barrier for tautomerization between 2-pyridone and 2-hydroxy-pyridine have indicated that the activation energy is substantially decreased by the complexation with formic acid, and double proton transfer occurs by a concerted mechanism.[50]

The fact that the kinetics of diamido-naphthyridine association are much faster for 2-ureido-pyrimidin-4-ol dimers (**DADA** array) than for 2-ureido-4[1*H*]-pyrimidinone dimers (**DDAA** array) strongly suggests that a dissociative mechanism as drawn in Scheme 1 is operative in the formation of U:N heterocomplexes. The differences in complexation kinetics are the result of one or more of the following features. First, the dissociation rate constant of $\mathbf{3_2}$ is most probably higher than the dissociation rate constant of $\mathbf{1_2}$ (1 s^{-1}).[29] Second, the activation barrier of the [1,3] prototropic shift in the 2-ureido-4[1*H*]-pyrimidinone tautomer probably is substantially higher than the activation barrier of the [1,3] prototropic shift in the 2-ureido-pyrimidin-4-ol tautomer. Finally, and most importantly, association of 2-ureido-4[1*H*] pyrimidinone dimers with N might be slow because it requires a conformational change of the ureido group that involves the breakage of an intramolecular hydrogen bond (Scheme 1).

Kinetic Product Formation in Mixtures of 1, 2, and 3

The insights gathered into the association kinetics of $\mathbf{1_2}$ and $\mathbf{3_2}$ with **2** in combination with the mechanistic scheme depicted in Scheme 1 allows us to predict that the thermodynamically disfavored heterodimer **1**:**3** can be obtained as kinetic product when **1** is present as a heterodimer with **2** (Figure 6). Because both the dissociation rate of the U:N complex **1**:**2** as well as the dissociation rate of $\mathbf{3_2}$ are fast, rapid exchange from **1**:**2** to **3**:**2** upon titration of 2 equiv of **3** will take place. This results in the release of monomeric **1** in its *cis*,*trans*-6[1*H*]-pyrimidinone tautomeric form. When **3** is added in excess, the *cis*,*trans*-6[1*H*]-pyrimidinone tautomeric form of **1** can rapidly form the heteromeric enol dimer **1**:**3** via a fast [1,3] shift, while formation of the 4[1*H*]-pyrimidinone homodimer $\mathbf{1_2}$ is slow because it requires both a [1,3] prototropic shift and a slow breakage of an intramolecular hydrogen bond. If the thermodynamic stability of homodimer $\mathbf{1_2}$ is higher than that of the heteromeric ureido-pyrimidinone complex **1**:**3**, initial kinetic self-sorting[2f] of **1** in **1**:**3** will be followed by slow equilibration to the homodimer $\mathbf{1_2}$.

Injection of 50 μL of a 200 mM solution of **3** in toluene-d_8 to a solution containing 10 mM U:N complex **1**:**2** in the same solvent indeed resulted in rapid formation of the enol heterodimer **1**:**3**. After 18 s, the kinetic product (**1**:**3**) was formed with a yield of 65%. This was followed by a slow decrease of this complex accompanied by slow formation of ureido-pyrimidinone homodimers $\mathbf{1_2}$ and $\mathbf{3_2}$ to yield an equilibrium product mixture containing 41% homodimer $\mathbf{1_2}$ and 35% heterodimer **1**:**3** (Figure 7).

(49) The ^{1}H NMR spectrum of this solution did not show any evidence of protonation of either the U_2 dimer $\mathbf{3_2}$ or the U:N complex **2**:**3**. See Supporting Information.
(50) Hazra, M. K.; Chakraborty, T. *J. Phys. Chem. A* **2006**, *110*, 9130.

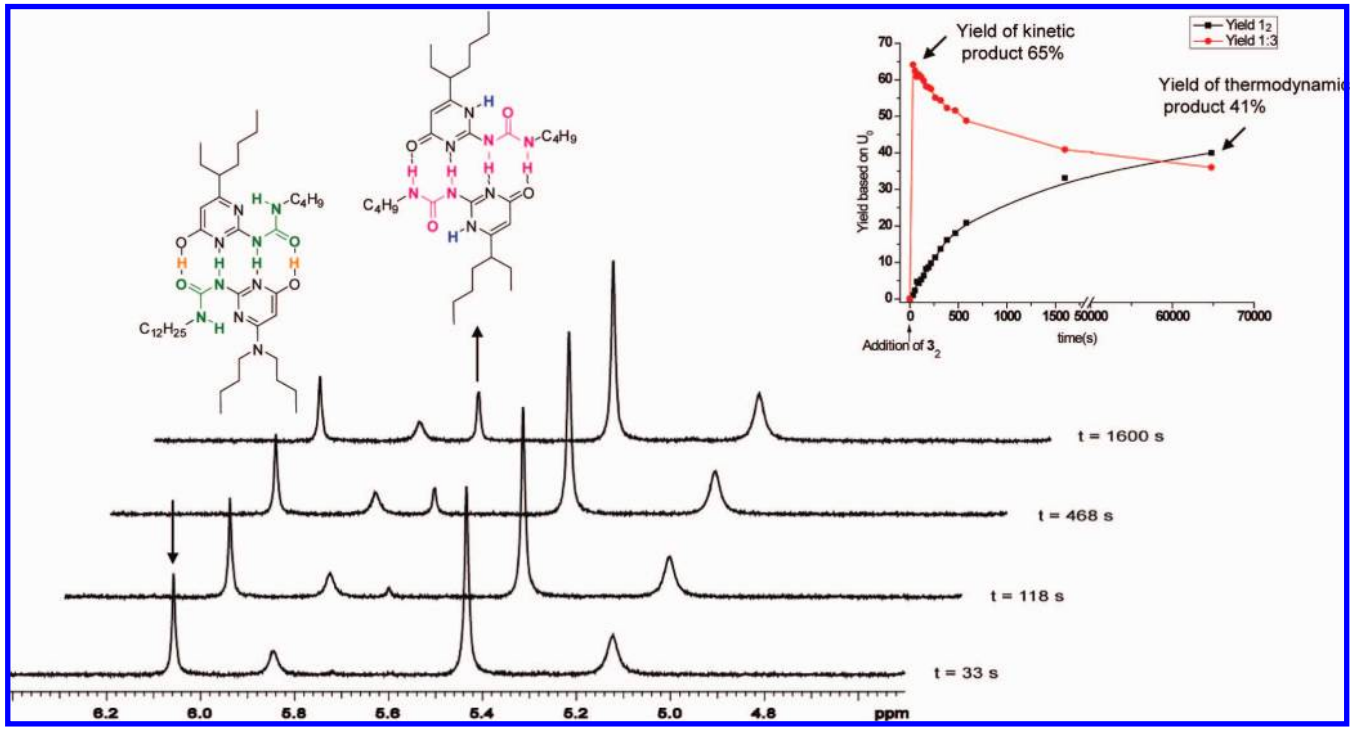


Figure 7. ^{1}H NMR spectra (500 MHz, toluene-d_8) obtained at regular time intervals after injection of 2 equiv of **3** to a solution containing 10 mM **1**:**2**. Fraction of the analytical concentration of **1** (U_0) present in the enol heterodimer **1**:**3** and homodimer $\mathbf{1_2}$ as a function of time (inset). The solid line is served to guide the eye.

Although the yield of the kinetic product is high, the selectivity for the formation of the thermodynamic product is limited because there is only a small difference in free energy between keto and enol dimers of **1** in apolar solvents.[28,51] The ^{1}H NMR spectrum of a 1:1 mixture of **1** and **3** in toluene-d_8 at equilibrium indeed reveals that approximately 33% of the total amount of **1** is incorporated in the enol heterodimer **1**:**3**. However, the experiment clearly shows the validity of the dissociative mechanism proposed in Scheme 1.

Summary and Conclusion

In summary, the kinetics of self-complementary quadruple hydrogen-bonded 2-ureido-4[1*H*]-pyrimidinone and 2-ureido-pyrimidin-4-ol dimers with 2,7-diamido-1,8-naphthyridine follow a dissociative pathway in which the tautomerization step is catalyzed in a bimolecular process. Furthermore, kinetic self-sorting in a three-component supramolecular system is reported. The analysis presented here illustrates the prominent role of kinetic product formation in supramolecular assemblies and may aid in the understanding of the relation between complexation dynamics and the mechanical properties of supramolecular polymers based upon the interaction between ureido-pyrimidinones and 2,7-diamido-1,8-naphthyridine.

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Supporting Information Available: Detailed experimental procedures. Synthesis and characterization of each compound of **3**. Crystal structure of **3** as cif file. Detailed description of kinetic experiments and GEPASI simulations. Derivation of equilibrium equations and description of the MATLAB program used to estimate K_a. This material is available free of charge via the Internet at http://pubs.acs.org.

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(51) The ^{1}H NMR spectrum of a 1:1 solution of **1** and **3** (5 mM each) in the more polar solvent $CDCl_3$ revealed that approximately 17% of the total concentration of **1** is incorporated in the ureido-pyrimidinone heterodimeric form **1**:**3**.