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Hydrogen-Bonding Complexes of 5-Azauracil and Uracil Derivatives in Organic Medium

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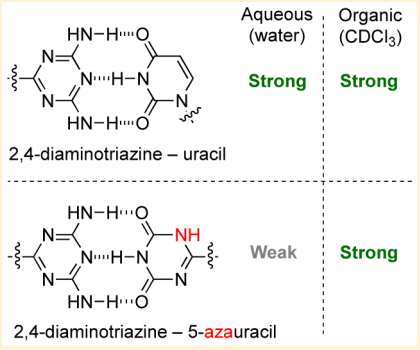
Supporting Information

ABSTRACT: Uracil derivatives form strong complexes with complementary 2,4-diaminotriazine and adenine compounds, whereas derivatives of 5-azauracil (2,4-dioxotriazine) are known to form weak complexes in aqueous medium. However, herein we report that in organic medium ($CDCl_3$), the 5-azauracil moiety forms hydrogen-bond-mediated complexes with complementary 2,4-diaminotriazine and adenine compounds, with strengths comparable to those formed by uracil compounds. Such dichotomous base-pairing behavior of the 5-azauracil moiety, in organic versus aqueous media, is found to be consistent with the ionization of the 5-azauracil moiety in aqueous medium leading to competitive interference from water molecules (via solvation), which is absent (lack of such ionization and solvent interference) in organic medium. This discriminating role of solvent (e.g., water) could have been an important factor in the selection of molecules, based on their physicochemical properties, and subsequently in the emergence of potential primordial informational oligomers that would have played a role in the origins of life.



INTRODUCTION

Hydrogen-bond-mediated associations in organic medium have long been used as surrogates to mimic and investigate the complexes formed between nucleobases within the hydrophobic stacks of RNA and DNA.[1] Initially, the base-pairing propensity of canonical nucleobase derivatives in organic solvents was investigated.[1a] Inspired by the acceptor−donor (A-D) type arrangement of the canonical nucleobases, there have been numerous studies of assemblies formed from the 2,4-diaminotriazine derivatives.[2] However, all of the studies with 2,4-diaminotriazine derivatives have been conducted with complementary uracil- and thymine-based-hydrogen-bonding partners and not with the exact complement, 5-azauracil.[2] The corresponding hydrogen-bonding behavior of 5-azauracil (2,4-dioxotriazine) with complementary 2,4-diaminotriazines derivatives have been, surprisingly, absent. Herein, we report for the first time on the association properties of 5-azauracil with 2,4-diaminotriazine and adenine derivatives in organic solvent (Figure 1).

We have been interested in the base-pairing behavior of alternative nucleobases within the context of mapping the landscape of potential primordial informational oligomers.[3] In that frame of reference, it was shown that 2,4-diaminotriazine containing dipeptide oligomers are able to base-pair, in neutral aqueous media, with complementary thymine and uracil containing DNA and RNA strands. On the contrary, 5-azauracil tagged dipeptide-containing oligomers were found to pair very weakly (or not pairing) with complementary adenine and 2,4-diaminopurine sequences of RNA/DNA. Moreover, 2,4-diaminotriazine containing oligodipeptides were found to pair extremely weakly with complementary 5-azauracil tagged

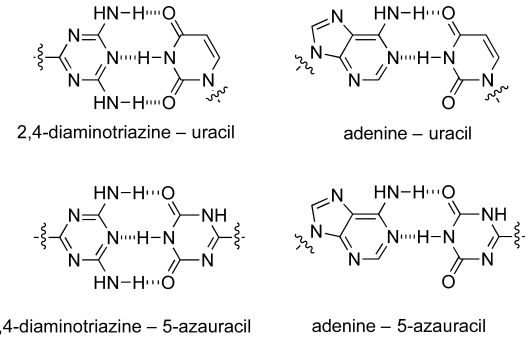


Figure 1. Hydrogen-bond-mediated associations of uracil and 5-azauracil (2,4-dioxotriazine) with 2,4-diaminotriazine and adenine. It should be noted that in organic medium the pairing mode with adenine could be in the Hoogsteen mode[1] instead of the Watson−Crick mode that has been shown for convenience of comparison with the 2,4-diaminotriazines.

oligodipeptides. Such contrasting behavior between 2,4-diaminotriazine and 5-azauracil alternative nucleobases was rationalized by the ionization of 5-azauracil ($pK_a \approx 6-7$)[3a] at neutral pH in aqueous milieu.[4] The resulting solvation (by water) of the ionized 5-azauracil moiety was thought to interfere competitively with hydrogen-bond-mediated associations, hindering the duplex formation of the complementary strands.[5]

This explanation suggested that in a nonpolar organic medium (such as chloroform), where there is no deprotonation

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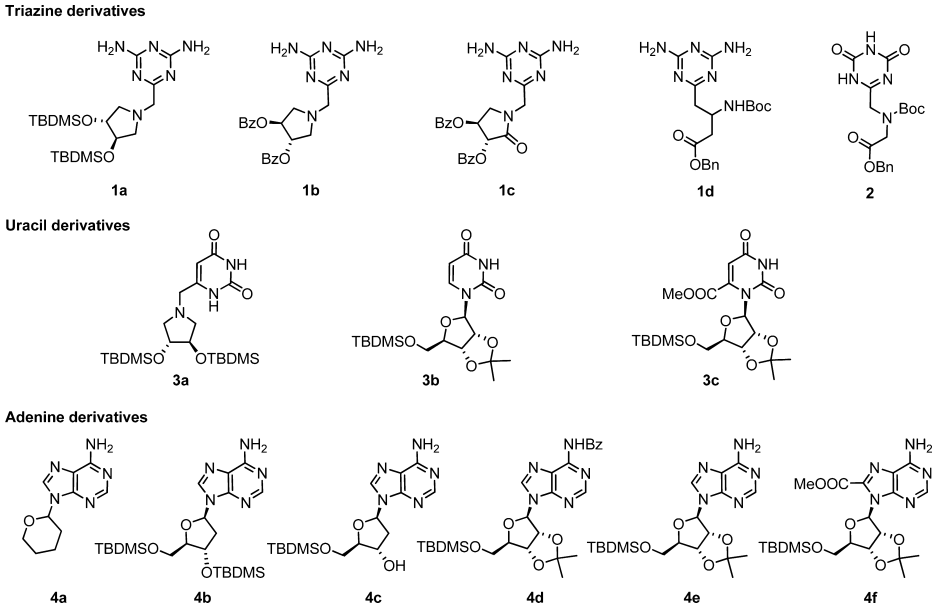
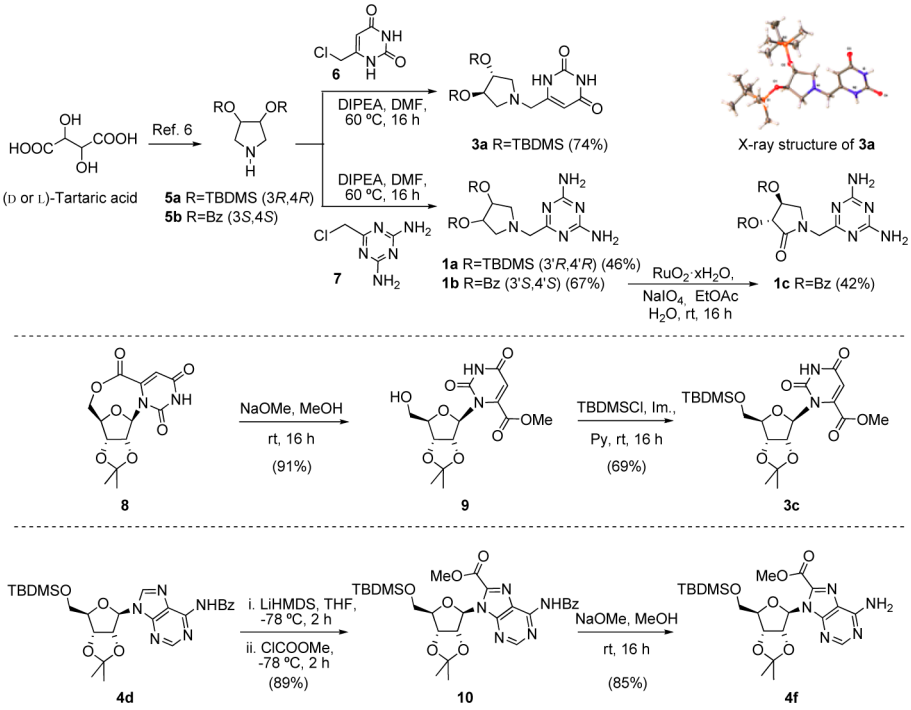


Figure 2. Monomers used in this study.

Scheme 1. Synthesis of 1a−1c, 3a, 3c, and 4f Used in This Study[a]



[a]On the top: **3a** and triazines **1a−1c** starting from tartaric acid. In the middle: synthesis of orotidine derivative **3c** starting from the lactone **8**. On the bottom: synthesis **4f** starting from **4d**.

of 5-azauracil, interference from the solvent (i.e., solvation by chloroform) should be minimal. As a consequence, 5-azauracil derivatives with an ADA arrangement should be able to associate in chloroform (unlike in water) with complementary (DAD) hydrogen bond partners, like 2,4-diaminotriazine derivatives. Herein, we tested this prediction and report on the association properties of 5-azauracil derivatives with 2,4-diaminotriazine and adenine derivatives, and compare their strength of association with those of uracil derivatives with 2,4-diaminotriazine and adenine hydrogen-bonding partners (Figure 1). We also document the unique effects of protecting groups on the ^{1}H NMR signals of the exchangeable protons involved in complexation, which could be of practical value in the design of substrates for future complexation studies.

RESULTS AND DISCUSSION

Monomers. The choice of monomers used in this study was dictated by our previous work[3a] and the hypothesis stemming from it. They consisted mainly of 2,4-diaminotriazine (**1a−1d**) and its complementary partner 5-azauracil (**2**) attached to suitably protected sugars and amino acids (Figure 2). The

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pyrrolidine skeleton was chosen for mimicking nucleoside derivatives when tagged with a 2,4-diaminotriazine moiety, and for the ease of synthesis. We did not use the comparable tetrahydrofuran skeleton due to the difficulties encountered in the synthesis of corresponding triazine C-nucleosides. For comparison purposes, we also synthesized uracil (**3a**–**3c**) and adenine (**4a**–**4f**) nucleoside derivatives shown in Figure 2. Compounds **3a**, **1a**, **1b**, and **1c** were synthesized from the corresponding pyrrolidine moieties **5a** and **5b**.[6] Compound **3c** was synthesized from the lactone **8**[7a] and **4f** from the adenosine derived **4d**. Details of the syntheses of the new compounds (Scheme 1) are provided in the Experimental Section. Compounds **1d** and **2** were obtained from our previous work.[3] Compounds **3b**,[7b] **4a**,[7c] **4b**,[7d] **4c**,[7d] **4d**,[7e] and **4e**[7f] were synthesized according to reported procedures.

Pairing Studies. The complexation between guests (**1a**–**1d** and **4a**–**4f**) and hosts (**2** and **3a**–**3c**) using $CDCl_3$ as solvent was investigated employing ^{1}H NMR spectroscopy, following literature protocols.[8] In this investigation, we focused on assessing the strengths of association of 5-azauracil **2** with various guests and comparing them with values obtained from uracil derivatives (**3a**–**3c**). We also explored the influence of protecting groups on the strength of complexation, once we became aware of the unusual effects that they exhibited.

Self-association. We began with investigating the self-association behavior of the monomers in order to consider the effects on the calculated heterocomplexation constants. The self-association behavior of the compounds was measured by monitoring the N*H* proton shift at different concentrations at 298 K. All of the compounds exhibited negligible self-association (as dimers, $K_a \approx 10$ M^{-1} or less; Table 1), except for two compounds, **1c** and **1d**, which had weak self-association constants around 22 and 76 (Table 1, entries 3 and 4). The weak to very weak self-associations in all these systems could be the result of the requirement for an acceptor–donor arrangement, which leads to two hydrogen-bond-mediated associations; however, it is difficult to rationalize the differences between, for example, **1a** and **1c** or **1d**. For the 5-azauracil derivative **2**, we were unable to obtain a self-association constant since the N*H*-protons were found to disappear (by broadening) after a certain point with increasing concentration (Figure S44, Supporting Information). Interestingly, the self-association of 3′,4′-di-O-TBDMS pyrrolidine tagged 2,4-diaminotriazine (**1a**) exhibited a curious effect as its concentration was increased; the exocyclic methylene protons, which were well separated at low concentrations, began pronouncedly to shift toward each other (Figure 3A) and even cross over as the concentration was increased (Figure S1, Supporting Information). The corresponding 3′,4′-di-OBz derivative **1b** did not exhibit this crossover effect at these concentrations (Figure 3B). It is of interest to note that 3′,4′-di-O-TBDMS pyrrolidine tagged uracil (**3a**) also did not exhibit this shift, though both **1a** and **3a** have association constants less than 10 M^{-1}. Thus, this behavior cannot be attributed to the nucleobase or the protecting group alone. It seems to be a combined effect that is manifested perceptibly in **1a**. While such a behavior and the effect of distal protecting groups (in **1a**) is not easy to rationalize, it nevertheless points to the unique role protecting groups could play in affecting ^{1}H NMR signals in titration studies, making the choice of protecting groups critical for successful experimental design.

Table 1. Calculated Self-association Constant Values (K_a) and Complexation-Induced Shift (CIS) of N*H* for Dimers of Uracil, Adenine, and Triazine Derivatives in $CDCl_3$

entry	compound	K_a (M^{-1})[a]	CIS (ppm)
1	**1a**	7	0.982
2	**1b**	8	0.814
3	**1c**	22	0.505
4	**1d**	76	0.170
5	**4a**	1	1.076
6	**4b**	1	0.463
7	**4c**	1	0.455
8	**4d**	*b*	0.165
9	**4e**	2	0.710
10	**4f**	1	0.683
11	**3a**	9	1.523
12	**3b**	8	1.541
13	**3c**	5	1.340
14	**2**	*b*	*c*

[a]Estimated error ±1. [b]It was not possible to calculate the self-association constant since saturation was not achieved. [c]Exchangeable protons disappeared during the titration.

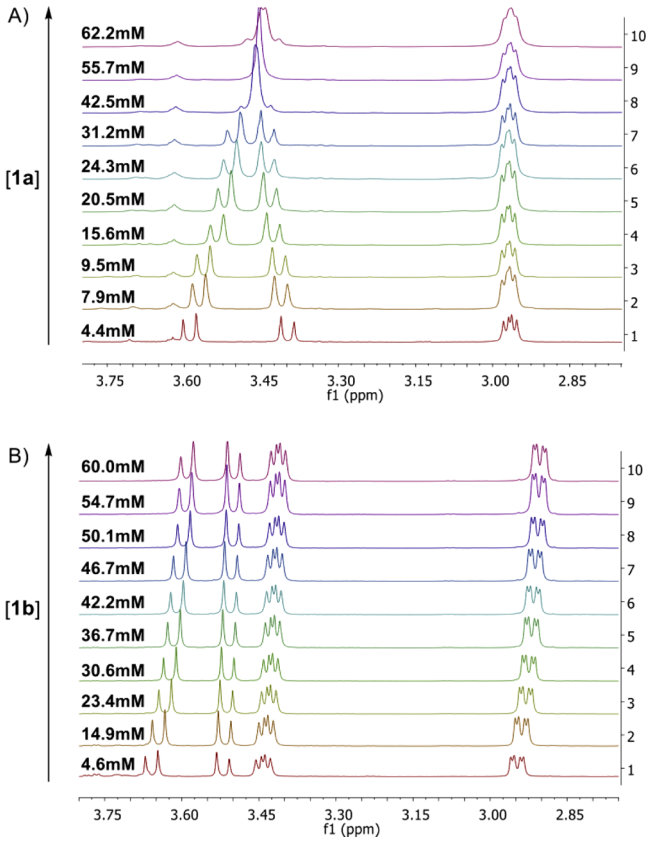

Figure 3. ^{1}H NMR self-association titration studies ($CDCl_3$) of **1a** (A) showing the unexpected shifting and crossover of the exocyclic methylene diastereomeric protons attached to the 2,4-diaminotriazine nucleobase with increasing concentrations. Corresponding NMRs for compound **1b** (B) are devoid of such effects.

Cross-Association. *Adenine and 2,4-Diaminotriazine with Uracil.* We initially investigated the base-pairing strength by ^{1}H NMR, titrating **3a** (guest, component whose concentration is varied) into a solution of **1a** (host, component whose concentration is constant). However, we observed the

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disappearance of the NH_2 protons of **1a** and, unexpectedly, the concomitant upfield shift of the N*H* protons of **3a** (Figure S47, Supporting Information). So, we reversed the roles of **1a** as the guest and **3a** as the host and observed downfield shifting of the exchangeable N*H* protons of **1a**, indicative of guest−host complexation and amenable to the determination of K_a values (Figure S45, Supporting Information). The cross-association of adenine derivatives **4a**−**4f** with uracil derivatives **3a**−**3c** was weaker (ranging from $K_a \approx 6$ M^{-1} to 87 M^{-1}) when compared to the 2,4-diaminotriazines **1a**−**1d**, $K_a \approx 230$ M^{-1} to 754 M^{-1} (Table 2 and Figure 4). The stoichiometry of complexation in all cases was shown to be 1:1 by Job plots (Supporting Information). There is also a wider spread of the complexation-induced NMR-shift (CIS) values ranging from 2.2 to 5.2 ppm for the adenine derivatives when compared to those of the 2,4-diaminotriazine derivatives, ranging from 4.3 to 5.2 ppm (Figure 4). This seems to indicate that the weaker adenine derived complexes are more perturbed by changes in the nature of the substituents attached to the adenine. This follows the expectation that weaker complexes are more sensitive to perturbations. Accordingly, the stronger 2,4-diaminotraizine complexes are relatively insensitive to changes in the type of substituents that they are attached to (within the range of substrates investigated).

Table 2. Calculated Cross-Association Constant Values (K_a) and CIS of Uracil and 5-Azauracil N*H* for the Complexation with Adenine and Diaminotriazine Derivatives in $CDCl_3$

entry	host	guest	K_a (M^{-1})	CIS (ppm)	$\Delta G°$(kcal/M)
1	**3a**	**1a**	317 ± 7	5.100	−3.41
2	**3a**	**1b**	550 ± 2	5.228	−3.74
3	**3a**	**1c**	496 ± 6	4.279	−3.68
4	**3a**	**1d**	521 ± 10	5.123	−3.70
5	**3a**	**4a**	83 ± 2	5.190	−2.61
6	**3a**	**4b**	38 ± 1	4.482	−2.14
7	**3a**	**4c**	33 ± 1	4.319	−2.07
8	**3a**	**4d**	10[a] ± 4	2.212	−1.36
9	**3a**	**4e**	38 ± 2	3.811	−2.15
10	**3a**	**4f**	26 ± 1	2.875	−1.93
11	**3b**	**1a**	350 ± 4	4.991	−3.47
12	**3b**	**1b**	467 ± 1	5.243	−3.64
13	**3b**	**1c**	230 ± 11	4.235	−3.22
14	**3b**	**1d**	336 ± 24	5.059	−3.44
15	**3b**	**4d**	6[a] ± 2	2.498	−1.06
16	**3b**	**4e**	59 ± 2	4.427	−2.41
17	**3b**	**4f**	45 ± 2	3.339	−2.25
18	**3c**	**1a**	*b*	*b*	
19	**3c**	**1b**	480 ± 3	5.295	−3.66
20	**3c**	**1c**	633 ± 12	4.253	−3.82
21	**3c**	**1d**	754 ± 11	5.082	−3.92
22	**3c**	**4e**	87 ± 2	5.033	−2.64
23	**3c**	**4f**	47 ± 1	4.647	−2.28
24	**2**	**1a**	*b*	*b*	
25	**2**	**1b**	711[c] ± 3	5.735, 5.813	−3.89
26	**2**	**1c**	484[c] ± 10	4.848, 4.880	−3.66
27	**2**	**1d**	320[c] ± 24	5.320, 5.356	−3.42
28	**2**	**4e**	88[c] ± 2	4.832, 4.976	−2.66
29	**2**	**4f**	39[c] ± 1	4.035, 4.080	−2.17

[a]The value of the self-association constant of the guest was not available. [b]Exchangeable protons disappeared during the titration. [c]The value of the self-association constant of the host was not available.

While this was understandable in terms of the two-hydrogen bond versus three-hydrogen bond association, the extreme weak association of **4d** (Table 2, entries 8 and 15) was particularly striking. Usually, an electron-withdrawing group on an amino moiety increases the strength of association due to the better hydrogen bond donating capability of the N*H* group.[9] However, there are examples in literature that have documented the opposite effect and are attributed to electrostatic repulsions.[10] In our case, it seems that there is only one hydrogen bond (instead of two) in the complexation involving **4d**. It is proposed that the steric hindrance of the (*N*6)-benzoate group leads to a twist in the *C*(6)-position-NHBz bond (Figure 5C, 4**d**-S-trans **II**) such that the N−H bond is not in same plane as the N(1)-atom for the Watson and Crick-mode (or N(7)-atom in the Hoogsteen mode in the purine ring), and therefore, unable to participate in hydrogen bonding in the complex formation. This effect lowers the strength of association from $K_a \approx 38$−59 M^{-1} to $K_a \approx 6$−10 M^{-1}. This hypothesis seems to be in agreement with the qualitative rationalizations based on calculations of ΔG values (Tables S1 and S2, and ΔG/number of expected hydrogen bonds, Supporting Information), which indicates that **4d** participates in complexation with only one hydrogen bond, presumably via the N(1)- or N(7)-position, with the C(6)-NHBz unable to participate due to the steric factors discussed above.

In another twist, the 6-carbomethoxy uridine derivative **3c** and the 8-carbomethoxy adenosine derivative **4f** were observed to have contrasting behavior compared to the unmodified parent compounds **3b** and **4e**, respectively. While association strengths of 8-carbomethoxy adenosine **4f** complexes were always slightly weaker than adenosine **4e** complexes, association strengths of 6-carbomethoxy uridine **3c** complexes were always stronger than **3b** complexes. The opposing effect of the carbomethoxy group on the adenine versus the uracil may stem, in part, from the electron withdrawing nature of the ester group, skewing the electron distribution of the contributing tautomer involved in complex formation as depicted in Figure 5. In the case of adenine, it apparently lowers the electron donation capability of N(1) by stabilizing a negative charge at the C(8)-position (Figure 5A, **IV** has a higher contribution than **III**), as opposed to the parent adenosine **4e**, in which the electron donating capability of N(1) is higher (Figure 5A, **I**). In the case of uracil, it seems to increase the hydrogen donation capability of N(3)-H by disfavoring the distribution of the positive charge to the C(6)-position (Figure 5B, **VI** versus **VIII**). While these are plausible explanations, the exact mechanisms by which such effects manifest themselves needs to be studied further.[11]

Effect of Protecting Groups on the Broadening and the Disappearance of the Exchangeable Protons Involved in the Complexation of 5-Azauracil. With these data in hand, we were in a position to investigate the ability of the 5-azauracil derivative to form hydrogen-bonded complexes with its complementary partners, 2,4-diaminotriazine (**1a**−**1d**) and adenine (**4e** and **4f**) derivatives, in organic media. We initially studied the interaction of **1a** with **2** and found that as the concentration of either **1a** or **2** was increased (Figure S70a and S71a, Supporting Information), it led to the disappearance of the exchangeable N*H* protons of **2** (similar to what was observed in the self-association measurements of **2**). Lowering

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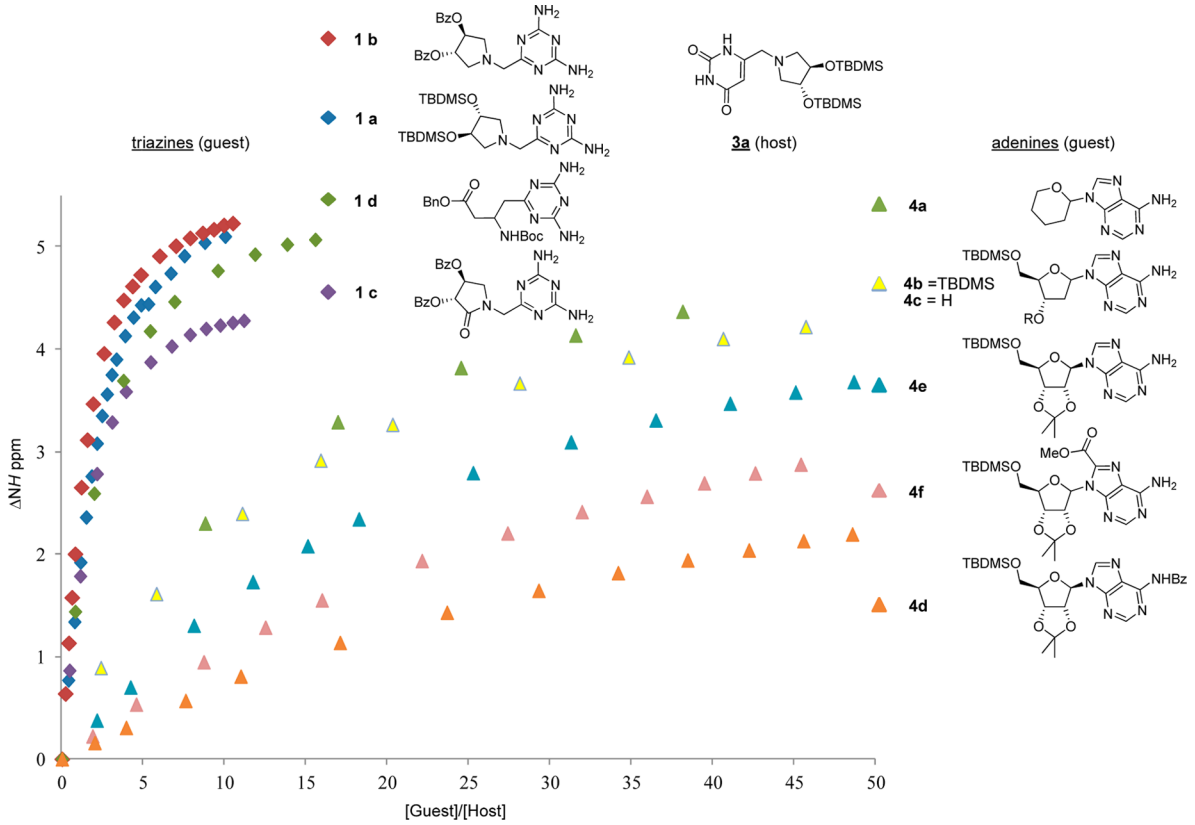

Figure 4. Comparison of the binding isothermal curves of the ^{1}H NMR of for 2,4-diaminotriazine derivatives (**1a**–**1d**) versus adenine derivatives (**4a**–**4f**) with uracil derivative **3a** (measured in $CDCl_3$). The CIS trends are consonant with the greater strength of the three-hydrogen bonded versus two-hydrogen bonded complexes.

the temperature (3 °C) did not enable the observation of the proton signals of interest (Figure S70b and S71b, Supporting Information). Since the exchangeable protons were disappearing, the association constant could not be measured; but this does not imply that there is no association between **1a** and **2**.

Since TBDMS derivative **1a** was shown to associate with **3a** and **3b**, the disappearance of the exchangeable N*H* protons was thought to be inherent to the behavior of 5-azauracil **2** itself; that is, the acidic N*H* proton of 5-azauracil **2** was interfering with the titration studies. However, when the N*H* proton of uracil derivative **3c** also disappeared when titrated with silylated derivative **1a**, we re-evaluated our original reasoning. We had originally reasoned that the nitrogen in the pyrrolidine ring of **1a** was basic enough to promote an acid–base reaction with the acidic proton of 5-azauracil **2**. However, the basicity of the analogous nitrogen in pyrrolidinone **1c** is reduced being part of the lactam, and therefore, such a proton transfer is not expected. The reasons for the disappearance of exchangeable protons of **2** in the titration of **2** with **1a** are not clear and at present not easily understood.

In parallel, we also noticed an interesting trend in the ^{1}H NMR studies of silylated **1a** versus benzoylated **1b** and **1c**: the N*H* protons of the host **3a** were always sharper and visible throughout the titration experiments when benzoate derivative **1b** (or **1c**) was used, when compared with the titration of **3a** with the TBDMS derivative **1a** (Figure 6). This was also found to be true for guest **3b**. This led us to test the combination of **1c** with **2**, and to our pleasant surprise, we found that the exchangeable protons of **2** did not disappear with increasing concentrations of **1c** (Figure S73, Supporting Information). The titration of 5-azauracil **2** with 2,4-diaminotriazine **1b** also confirmed that the exchangeable N*H* proton of **2** did not disappear, affording an association constant of 711 M^{-1}, which was much more than the uracil derivatives **3a** and **3b** we had studied. The fact that a simple change of the distal protecting groups at the 3′,4′-O-positions, from the TBDMS group (**1a**) to a benzoate group (**1b**), would have such an effect on the behavior of ^{1}H NMR signal of the exchangeable proton of **2** was, a priori, not evident, nor could be expected. Since all of the compounds investigated in this study were soluble in $CDCl_3$, these results do seem to suggest that the use of benzoate (as protecting groups) may provide better results for experiments monitored by ^{1}H NMR when compared to the use of TBDMS as protecting/solubilizing groups; the general applicability of benzoate in place of TBDMS needs to be verified by future studies.

5-Azauracil (2,4-Dioxotrazine) with 2,4-Diaminotriazine and Adenine. As stated in the introduction, the base-pairing studies of AspGlu-oligodipeptides tagged with 5-azauracil in *aqueous media* were shown from our previous work to have very weak (or no) duplex forming capability (Figure 7 and Table 3, entries 3–5).[3a] The weak pairing of 5-azauracil is attributed to its propensity to ionize ($pK_a \approx 6$–7) and get solvated in the aqueous environment (pH ≈ 7).[5] This observation is also consistent with what is known for other 5-aza-heterocycles such as 5-azacytidine, whose insertion into a oligonucleotide duplex generally leads to weakened duplex stability.[12] In contrast, the 2,4-diaminotriazine tagged oligodipeptides were shown to pair strongly with uracil and thymine-containing oligonucleotides (Figure 7, Table 3, entries 1–2),[3a]

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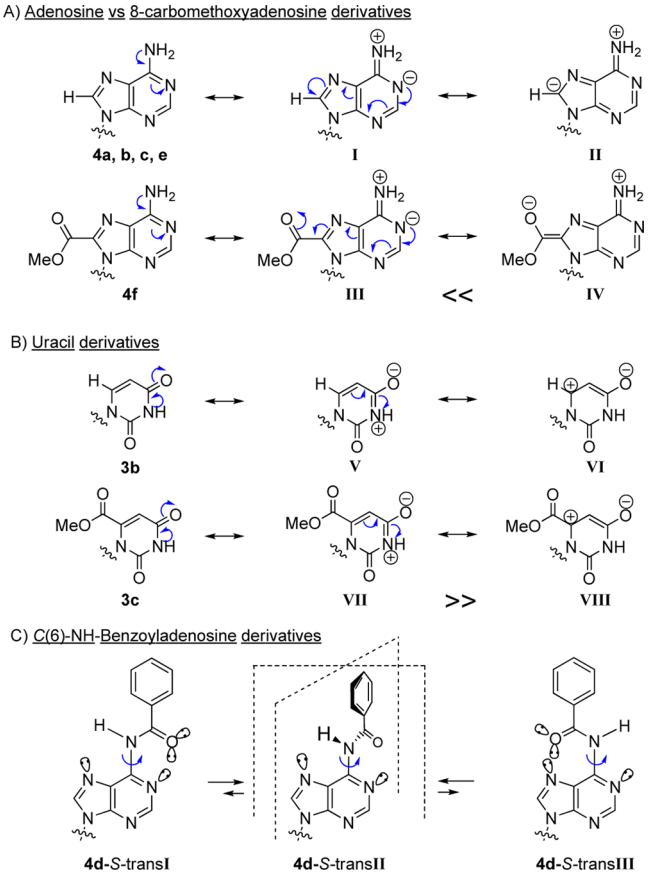

Figure 5. Plausible explanation for the effect of substituents on strengths of complexation. (A and B) Opposing effect of the carbomethoxy group on association strengths of 8-carbomethoxy adenine versus 6-carbomethoxy uracil. (C) The orthogonal disposition of the *C*(6)-*NH*-benzoyl substituent on adenine seems to be responsible for the weaker association compared to that of the parent adenine derivative.

consistent with the pK_a−pH correlations.[5] Since such ionization and solvent interaction would be absent in a nonpolar solvent like chloroform, we expected 2,4-dioxotriazines to form hydrogen-bond-mediated complexes with its complementary counterparts, and indeed, this was found to be the case.

The 5-azauracil derivative **2** formed relatively moderate to strong associations (K_a ≈ 320 to 711 M^{-1}) with 2,4-diaminotriazine derivatives **1b**, **1c**, and **1d** (Table 2, entries 25−27) and correspondingly weaker complexes with adenine derivatives **4e** and **4f** (Table 2, entries 28, 29). The N(3)-*H* proton of 5-azauracil **2** appeared separately in the ^{1}H NMR spectrum (as two peaks around 7.7 ppm), and both signals shifted downfield in tandem as the hydrogen-bonded complexes formed with **1b** (Figure 8a). As shown in Figure 8b, 1:1 stoichiometry of complexation was confirmed by a Job plot in all cases (see also Supporting Information). The strength of the complexes formed with the 5-azauracil derivatives turned out to be on par with uracil derivatives, having one of the highest (711 M^{-1} versus 754 M^{-1}) and the lowest (320 M^{-1}versus 230 M^{-1}) values. This observation points out that in a nonpolar medium, 5-azauracil behaves almost the same as uracil in terms of the relative strengths of hydrogen-bond-mediated association (Figure 9).

The above results demonstrate that 5-azauracil is capable of hydrogen bonded association not only with its counterpart, 2,4-diaminotriazine derivatives, but also with adenine derivatives. The strength of association of the 5-azauracil moiety, in nonpolar organic solvent, is similar to that of uracil, and the presence of the extra nitrogen at the 5-position seems to not affect the hydrogen-bonding (donating and accepting) capability when compared with its parent uracil moiety. This observation is in contrast to what is known in neutral aqueous media, where 5-azauracil has sharply reduced hydrogen-bonding capability (with 2,4-diaminotriazine and adenine) when compared with that of uracil. This observation gives credence to the hypothesis that the interference from the

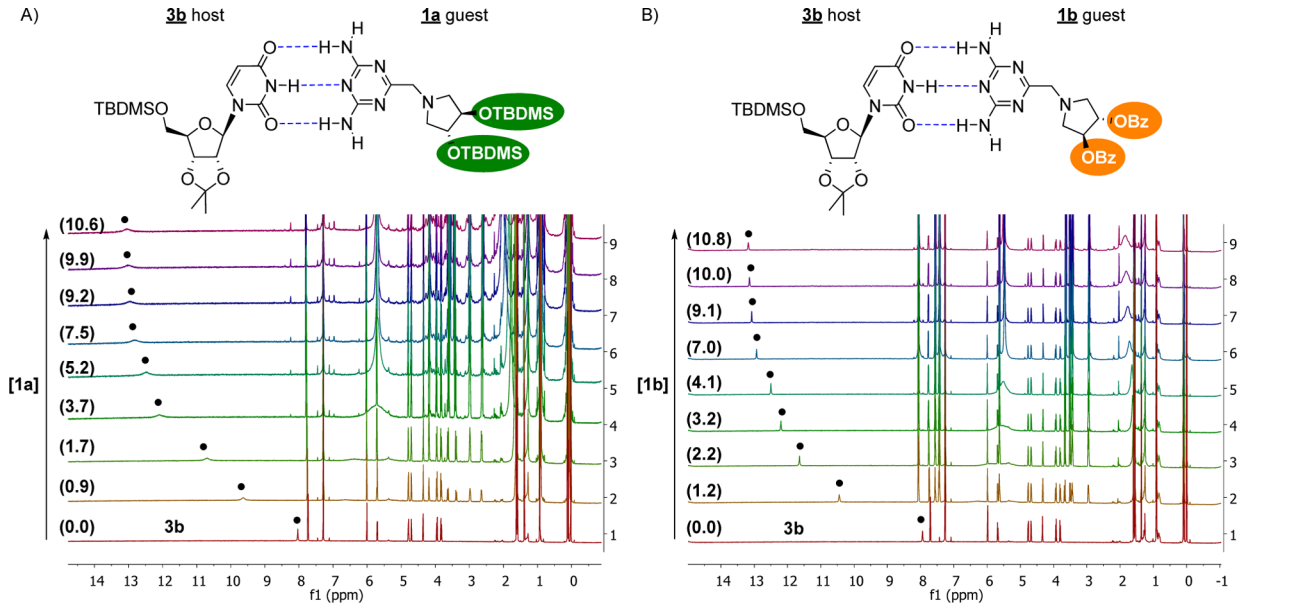

Figure 6. ^{1}H NMR spectra of titration experiments of **3b** with pyrrolidines **1a** (A) and **1b** (B) documents the unpredictable effect of the distal protecting groups on the ^{1}H NMR signal of the exchangeable proton of **3b**. Titration of **3b** with **1a** results in a successively broadening signal, while the same signal remains sharp when titrated with **1b**. The number of equivalents of the guest in each spectrum is given in parentheses.

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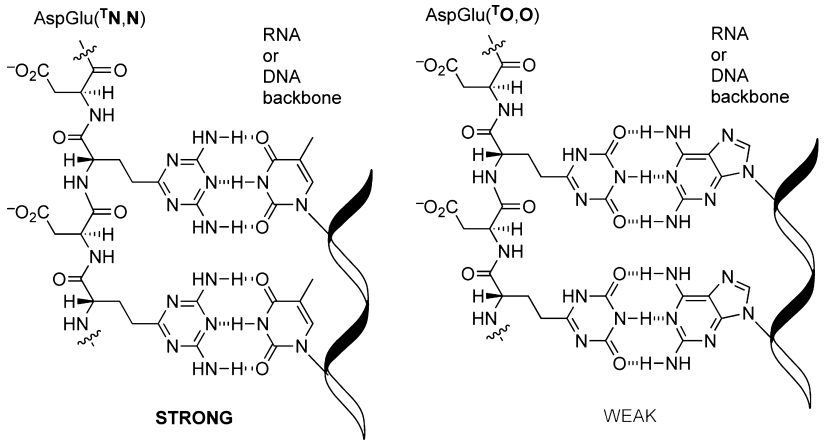

Figure 7. Aspartyl-glutamyl-oligodipeptide backbone tagged with 2,4-diaminotriazine AspGlu(TN,N) shows strong base pairing with complementary uracil (RNA) or thymine (DNA), while the aspartyl-glutamyl-oligodipeptide backbone tagged with 5-azauracil AspGlu(TO,O) shows weak base-pairing behavior with complementary purines (adenine or 2,6-diaminopurine) of RNA and DNA, in aqueous medium, pH ≈ 7.[3a]

Table 3. Previously Published UV-T_m Data of 5-Azauracil (^{T}OO) versus 2,4-Diaminotriazine (^{T}NN) Containing Oligodipeptides in Aqueous Media[a]

entry	pairing system	UV-T_m °C[a]
	2,4-Diaminotriazine	
1	^{HOOC}AspGlu(^{T}NN)$_{12}$ + poly-**r**(U)	51.4
2	^{HOOC}AspGlu(^{T}NN)$_{12}$ + **d**(T)$_{12}$	49.2
	5-Azauracil	
3	^{HOOC}AspGlu(^{T}OO)$_{12}$ + poly-**r**(A)	<10
4	^{HOOC}AspGlu(^{T}OO)$_{12}$ + **d**(D)$_{12}$	19.4
5	^{HOOC}AspGlu(^{T}OO)$_{12}$+ ^{NH2}AspGlu(^{T}NN)$_{12}$	≈ 13

[a]Data from ref 3a. UV-T_m measures the strength of duplexes; higher values reflect stronger base-pairing duplexes.

solvent is mainly responsible for the contrasting behavior of 5-azauracil versus uracil. It is known that 5-azauracil exists in the keto-form.[13] Therefore, interference from the (nonexistent) enol form is not a plausible explanation for this contrasting base-pairing behavior.

The weaker association of 5-azauracil in water can be attributed to its pK_a (≈ 6−7), which is close to the pH of the neutral aqueous medium, leading to the deprotonation of the N*H* proton of 5-azauracil. The ionization of the 5-azauracil moiety (also known from pH dependent UV-studies)[3a] leads to the solvation of the ions by water molecules. The competition from water molecules interferes with the association of the ionized 5-azauracil moiety with its complementary pairing partners and results in weak (or no) associations. However, the uracil N*H* moiety (with a pK_a 9.5) is more than 2 units away

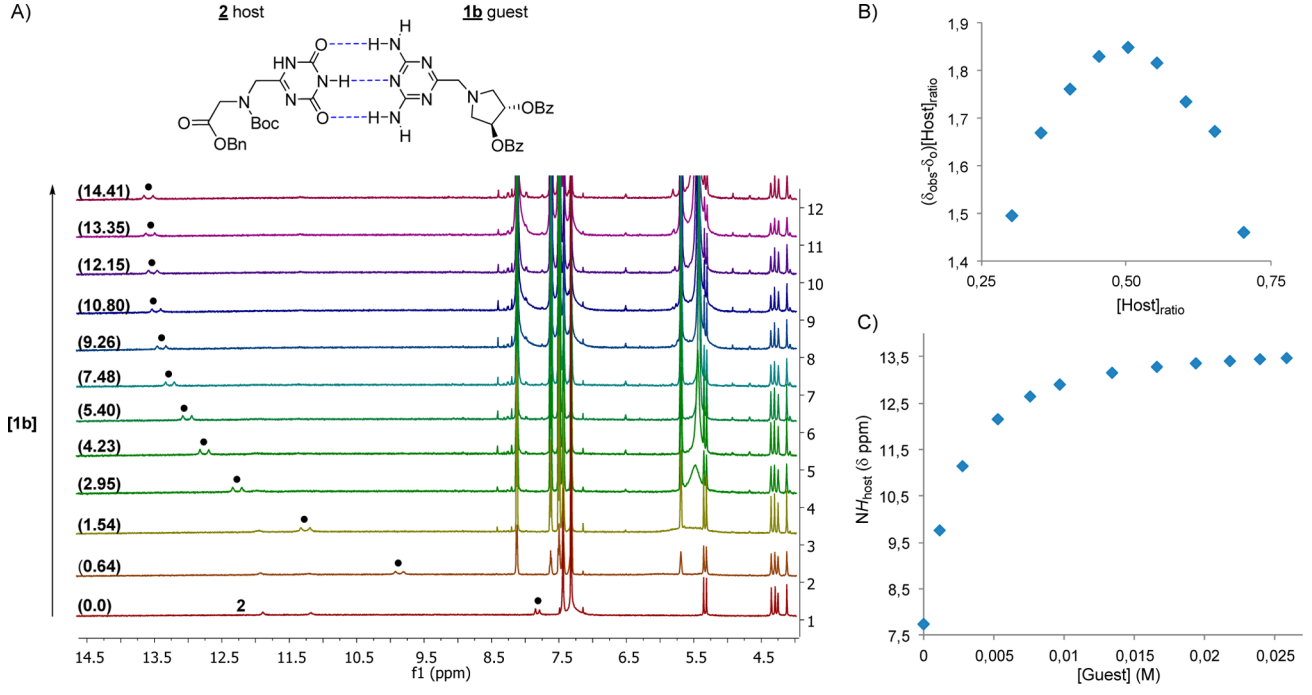

Figure 8. Representative ^{1}H NMR (600 MHz, 298 K, $CDCl_3$) titration data for 5-azauracil **2** with **1b**. (A) Stacked ^{1}H NMR titration spectra. The number of equivalents of the guest **1c** in each spectrum is given in parentheses. (B) Job plot for 5-azauracil **2** binding to **1b** confirming a 1:1 binding stoichiometry. (C) Plot of the N*H* chemical shift data of 5-azauracil derivative **2** from ^{1}H NMR titration.

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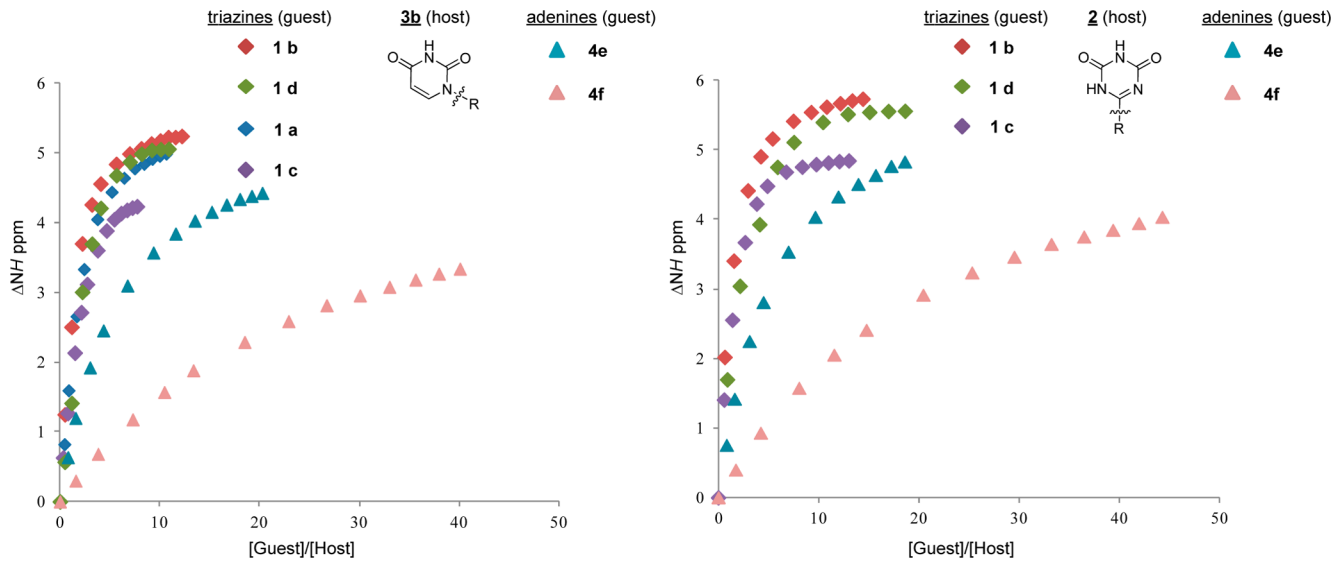

Figure 9. Binding association curves of the uracil derivative **3b** and 5-azauracil derivative **2** with triazines and adenines demonstrating that the complex induced shifts (CIS) formed by uracil and 5-azauracil are comparable. This parallels their similar association strengths (K_a) (Table 2, entries 11−17 and 24−29).

from the neutral pH of the medium, which makes uracil "un-ionized" in neutral media and, thus, hydrophobic. This allows for the uracil moiety to interact with its complementary counterparts (2,4-diaminotriazine and adenine) without any hindrance from the solvent. In a nonpolar organic solvent, like chloroform, such ionizations and interference from solvents are absent, allowing both the 5-azauracil and uracil to interact with their complementary counterparts.

These observations reinforce the critical role that the nature of the solvent has (or must have) played in selecting the types of heterocycles (nucleobases) of RNA and DNA, both of which rely on hydrophobicity-driven and hydrogen-bond-mediated recognition for structure and function. In other words, water as a solvent, unlike a nonpolar organic solvent, is able to discriminate between those heterocycle derivatives that can form hydrogen-bond-mediated associations without ionization versus those that cannot form hydrogen bonds due to ionization. Such involvement and discrimination by the solvent highlights an important selection mechanism by which mixtures of heterocyclic derivatives, in an aqueous medium, can be differentiated by the type of functional capabilities they "acquire" in a particular solvent like water. These types of mechanisms could have been crucial for the selection and chemical evolution of molecules on early earth.[14]

CONCLUSIONS

The hydrogen-bonding behavior of the 5-azauracil (2,4-dioxotraizine) moiety, in chloroform, with its complementary counterpart, 2,4-diaminotriazine and adenine derivatives has been described for the first time, and the strength of association is found to be comparable to that of uracil with 2,4-diaminotriazine and adenine in chloroform. This base-pairing behavior of 5-azauracil versus uracil seems to be the opposite of what is known in aqueous media, wherein the 5-azauracil forms much weaker complexes when compared to those formed by uracil. These results, in conjunction with earlier studies of base-pairing propensity in neutral aqueous media, emphasize the role of the solvent in controlling hydrogen-bond-mediated associations. The "enabling" and "disabling" roles of water could be viewed as an additional "selection mechanism" in the context of origins of life studies.[15] Such mechanisms could have selected certain heterocycle derivatives from a complex mixture for further processing by chemical evolution on early earth.[16]

EXPERIMENTAL SECTION

General Experimental. All experiments were performed under a nitrogen or argon atmosphere. Thin layer chromatography (TLC) was performed on silica gel 60 Å F254, and it was visualized by UV lamp and/or a stain solution of phosphomolybdic acid (PMA) in ethanol. Flash column chromatography was performed on silica gel 60 Å with a particle size of 35−70 μm. Mass spectra were measured with ESI-TOF or an LTQ ion trap mass spectrometer.

General Procedure for Determining Binding Constant. A binding study was performed in $CDCl_3$ for each host molecule (**2**, **3a**, **3b**, or **3c**) and monitored by ^{1}H NMR spectroscopy. In a typical $CDCl_3$ titration, 1 mL of x mM (see the corresponding titration experiment in Supporting Information) of the host was prepared. The host solution was then divided in half such that 0.5 mL was placed into an NMR tube, and the other 0.5 mL was used to create a second solution containing from 10 to 100 mM of the guest solution (depending on the guest, see also the corresponding titration experiment in the Supporting Information). The host was also placed in the guest solution so as to maintain constant concentration of the host throughout the titration experiment. An initial spectrum of the free host was recorded, after which aliquots (5−50 μL) of the guest solution (with the host) were added until the N*H* resonance of the host was no longer shifted (saturation was reached). All NMR spectra were processed using MestReNova NMR software. Association constants (K_a) were calculated by nonlinear curve fitting of the obtained titration isotherms and the values of the self-association by using HypNMR 2006.[17] The association constants were calculated from the downfield shifting of the N*H* proton resonances in all cases.

Job Plot Procedure. Job plots were performed in $CDCl_3$ and monitored by ^{1}H NMR for each host molecule. For a typical Job plot, 3 mL of a host solution and 3 mL of a guest solution were prepared and then divided between 10 NMR tubes in corresponding mol % increments. After equilibration, the ^{1}H NMR spectrum for each sample was recorded, and the shift in the host N*H* resonance was used to construct the Job plot.

General Procedure for the Synthesis of the Pyrrolidine-*N*-yl-methyl-derivatives. To a suspension of 6-chloromethyl-uracil (**6**) or 6-chloromethyl triazine (**7**)[3a] (1.0 mmol, 1.0 equiv) and diisopropy-

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lethylamine (1.0 mL, 6.0 mmol, 6.0 equiv) in DMF (10 mL) at rt, the appropriate 3,4-bis-protected-pyrrolidine[6] (1.0 mmol, 1.0 equiv) was added. The reaction mixture was stirred at 60 °C overnight. After evaporation of the solvent at reduced pressure, the residue was redissolved in EtOAc/MeOH (40 mL, 9:1, v/v) and filtered through a bed of silica and Celite. Then, the bed was washed several times with the same mixture of solvents. The filtrate and washings were combined and concentrated to dryness to yield a solid that was used without any further purification.

*6-(((3R,4R)-3′,4′-Bis(tert-butyldimethylsilyloxy)pyrrolidine-N-yl)-methyl)-1,3,5-triazine-2,4-diamine (**1a**).* Pale yellow solid (205 mg, 46%). ^{1}H NMR (600 MHz, $CDCl_3$, 4.4 mM) δ ppm: 0.07 (s, 6H, 2 × SiCH_3), 0.10 (s, 6H, 2 × SiCH_3), 0.91 (s, 18H, 2 × SiC(CH_3)$_3$), 2.57 (dd, J = 9.4, 3.8 Hz, 2H, 2 × CHHN), 2.99 (dd, J = 9.4, 5.8 Hz, 2H, 2 × CHHN), 3.38 (d, J = 15.3 Hz, 1H, CHH-triazine), 3.62 (d, J = 15.3 Hz, 1H, CHH-triazine), 4.16 (dd, J = 3.8, 5.8 Hz, 2H, 2 × CHOSi), 5.62 (bs, 4H, 2 × NH_2). ^{13}C NMR (150 MHz, $CDCl_3$, 4.46 mM) δ ppm: −4.6 (2 × SiCH_3), −4.5 (2 × SiCH_3), 18.2 (SiC(CH_3)$_3$), 26.0 (SiC(CH_3)$_3$), 61.2 (CH_2N), 62.2 (CH_2-triazine), 79.9 (CHOSi), 167.3 (C-triazine), 176.3 (C-triazine).^{1}H NMR (600 MHz, DMSO-d_6) δ ppm: 0.03 (s, 6H, 2 × SiCH_3), 0.04 (s, 6H, 2 × SiCH_3), 0.86 (s, 18H, 2 × SiC(CH_3)$_3$), 2.43 (dd, J = 9.5, 4.7 Hz, 2H, 2 × CHHN), 2.83−2.93 (dd, J = 9.5, 4.5 Hz, 2H, 2 × CHHN), 3.24 (s, 2H, CH_2-triazine), 4.00 (dd, J = 4.7, 4.5 Hz, 2H, 2 × CHOSi), 6.49−6.74 (m, 4H, NH_2). ^{13}C NMR (150 MHz, DMSO-d_6) δ ppm: −4.9 (2 × SiCH_3), −4.6 (2 × SiCH_3), 17.7 (SiC(CH_3)$_3$), 25.7 (SiC(CH_3)$_3$), 60.2 (CH_2N), 61.2 (CH_2-triazine), 79.3 (CHOSi), 167.1 (C-triazine), 174.4 (C-triazine). ESI(+)-HRMS: m/z calcd for $C_{20}H_{42}N_6O_2Si_2$ (M + H)$^+$ calcd, 455.2981; found, 455.2980. ESI(+)-HRMS: m/z calcd for $C_{20}H_{41}N_6NaO_2Si_2$ (M+Na)$^+$ calcd, 477.2803; found, 477.2803.

*6-(((3S,4S)-3′,4′-Dibenzoyloxypyrrolidine-N-yl)methyl)-1,3,5-triazine-2,4-diamine (**1b**).* (291 mg, 67%) White solid. ^{1}H NMR (600 MHz, $CDCl_3$, 4.6 mM) δ ppm: 2.95 (dd, J = 10.5, 4.1 Hz, 2H, 2 × NCHH), 3.44 (dd, J = 10.5, 6.3 Hz, 2H, 2 × NCHH), 3.52 (d, J = 14.7 Hz, 1H, CHH-triazine), 3.66 (d, J = 14.7 Hz, 1H, CHH-triazine), 5.12 (br s, 4H, 2 × NH_2), 5.62 (dd, J = 6.3, 4.1 Hz, 2H, CHOBz), 7.43 (t, J = 7.8 Hz, 4H, H-Bz), 7.55 (d, J = 7.4 Hz, 2H, H-Bz), 8.03−8.09 (m, 4H, H-Bz). ^{13}C NMR (150 MHz, DMSO-d_6) δ ppm: 58.8 (CH_2N), 61.4 (CH_2-triazine), 78.4 (CHOBz), 128.6 (C-Bz), 129.7 (C-Bz), 130.0 (C-Bz), 133.4 (C-Bz), 166.2 (COPh), 167.2 (C-triazine), 175.5 (C-triazine). ESI(+)-HRMS: m/z calcd for $C_{22}H_{23}N_6O_4$ (M + H)$^+$ calcd, 435.1775; found, 435.1773.

*6-(((3R,4R)-3′,4′-Bis(tert-butyldimethylsilyloxy)pyrrolidine-N-yl)-methyl)-uracil (**3a**).* Pale yellow solid (337 mg, 74%). R_f (DCM/MeOH, 95:5) = 0.30. ^{1}H NMR (600 MHz, $CDCl_3$, 4.4 mM) δ ppm: 0.05 (s, 6H, SiCH_3), 0.07 (s, 6H, 2 × SiCH_3), 0.88 (s, 18H, 2 × SiC(CH_3)$_3$), 2.53 (dd, J = 10.0, 3.0 Hz, 2H, 2 × CHHN), 2.96 (dd, J = 10.0, 4.9 Hz, 2H, 2 × CHHN), 3.46 (d, J = 16.0 Hz, 1H, CHH-uracil), 3.52 (d, J = 16.0 Hz, 1H, CHH-uracil), 4.05−4.07 (m, 2 × CHOSi), 5.51 (s, 1H, CH-uracil), 7.86 (bs, 2H, 2 × NH). ^{13}C NMR (150 MHz, $CDCl_3$, 4.46 mM) δ ppm: −4.6 (2 × SiCH_3), −4.5 (2 × SiCH_3), 18.1 (SiC(CH_3)$_3$), 25.8 (SiC(CH_3)$_3$), 56.9 (CH_2N), 60.8 (CH_2−Uracil), 79.2 (CHOSi), 98.6 (CH-uracil), 151.0 (C-uracil), 153.2(C-uracil), 164.3 (C-uracil). ESI(+)-HRMS: m/z calcd for $C_{21}H_{42}N_3O_4Si_2$ (M + H)$^+$ calcd, 456.2708; found, 456.2711. ESI(−)-HRMS: m/z calcd for $C_{21}H_{40}N_3O_4Si_2$ (M − H)$^-$ calcd, 454.2563; found, 454.2572. CCDC-1056406 contains the supplementary crystallographic data for this article. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data%5Frequest/cif.

*6-(((3S,4S)-3′,4′-Dibenzoyloxy-2′-oxopyrrolidine-N-yl)methyl)-1,3,5-triazine-2,4-diamine (**1c**).* To a solution of $NaIO_4$ (330 mg, 1.54 mmol, 3.0 equiv) in H_2O (6 mL) was added $RuO_2{\cdot}xH_2O$ (17 mg, 0.11 mmol, 25 mol %). The mixture was stirred at room temperature for 5 min, and a solution of **1b** (195 mg, 0.45 mmol, 1.0 equiv) in EtOAc (6 mL) was added dropwise. Then, the mixture was stirred at room temperature for 16 h. H_2O (15 mL) and EtOAc (15 mL) were added, and the aqueous layer was extracted with EtOAc (3 × 20 mL). The combined organic phases were washed with brine and dried over Na_2SO_4. After filtration and removal of the solvent, the resulting residue was purified by flash chromatography (EtOAc/MeOH, 95:15 to 85:15) to afford the expected lactam as a white solid (188 mg, 42%). R_f (DCM/MeOH, 95:5) = 0.40. Mp: 108 °C−110 °C. ^{1}H NMR (600 MHz, DMSO-d_6) δ ppm: 3.70 (dd, J = 9.6, 7.0 Hz, 1H, CHHN), 4.06 (d, J = 9.6, 8.5 Hz, 1H, CHHN), 4.10 (d, J = 16.6 Hz, 1H, CHH-triazine), 4.30 (d, J = 16.6 Hz, 1H, CHH-triazine), 5.79 (dd, J = 15.0, 7.0 Hz, 1H, CHBz), 6.00 (d, J = 7.0 Hz, 1H, CHBz), 6.67−6.88 (br s, 4H, 2 × NH_2), 7.53−7.59 (m, 4H, H-Bz), 7.67−7.67 (m, 2H, H-Bz), 7.97−8.06 (m, 4H, H-Bz). ^{13}C NMR (150 MHz, DMSO-d_6) δ ppm: 47.1 (CH_2-triazine), 48.7 (CH_2N), 72.2 (C$^{4'}$HOBz), 74.6 (C$^{3'}$HOBz), 128.7 (C-Bz), 128.8 (C-Bz), 128.9 (C-Bz), 128.9 (C-Bz), 129.5 (C-Bz), 129.6 (C-Bz), 133.9 (C-Bz), 134.0 (C-Bz), 165.0 (COPh), 165.3 (COPh), 166.6 (C-triazine), 167.0 (NCO), 172.4 (C-triazine). ESI(+)-HRMS: m/z calcd for $C_{22}H_{21}N_6O_5$ (M + H)$^+$: 449.1568; found, 449.1568.

*2′,3′-O-Isopropylidene-5′-O-(tert-butyldimethylsilyl)-orotidine Methyl Ester (**3c**).* To a solution of 2′,3′-O-isopropylideneorotidine 5′-lactone **8**[7a] (1.42 g, 4.57 mmol, 1.0 equiv) in anhydrous MeOH (45 mL) was added 1 M NaOMe in MeOH (0.92 mL, 0.92 mmol, 0.2 equiv), and the mixture was stirred at room temperature overnight. The residue was absorbed onto silica and purified by column chromatography (CH_2Cl_2/acetone, 3:1 to1:1) to give 2′,3′-O-isopropylideneorotidine methyl ester **9** as colorless foam (1.42 g, 91%). To a solution of **9** (270 mg, 0.79 mmol, 1.0 equiv) and imidazole (107 mg, 1.57 mmol, 2.0 equiv) in dry pyridine (20 mL) was added TBDMSCl (142 mg, 0.94 mmol, 1.2 equiv) at room temperature. The mixture was stirred at room temperature overnight. Then, the mixture was partitioned between EtOAc (50 mL) and sat. aq. $NaHCO_3$ (50 mL). The aqueous layer was extracted with EtOAc (2 × 30 mL), and the combined organic extracts were dried over Na_2SO_4 and concentrated. The residue was purified by a flash purification system using 6−70% gradient EtOAc in hexanes to give **3c** as colorless foam (247 mg, 69%). R_f (Hexanes/EtOAc, 1:2) = 0.78. ^{1}H NMR (600 MHz, $CDCl_3$) δ ppm: 0.03 (s, 6H, 2 × SiCH_3), 0.86 (s, 9H, SiC(CH_3)$_3$), 1.33 (s, 3H, CH_3), 1.53 (s, 3H, CH_3), 3.75 (dd, J = 10.9, 7.3 Hz, 1H, C$^{5'}$$H$H), 3.80 (dd, J = 10.9, 4.9 Hz, 1H, C$^{5'}$HH), 3.93 (s, 3H, OCH_3), 4.11−4.06 (m, 1H, C$^{4'}$$H$), 4.73 (dd, J = 6.6, 4.8 Hz, 1H, C$^{3'}$$H$), 5.18 (dd, J = 6.6, 1.9 Hz, 1H, C$^{2'}$$H$), 5.90 (d, J = 1.9 Hz, 1H, C$^{1'}$$H$), 6.07 (s, 1H, C^5$H$), 9.95 (s, 1H, N$H$). ^{13}C NMR (150 MHz, $CDCl_3$) δ ppm: −5.2 (SiCH_3), −5.2 (SiCH_3), 18.5 (C(CH_3)$_3$), 25.4 (CH_3), 26.0 (C(CH_3)$_3$), 27.3 (CH_3), 53.9 (OCH_3), 64.0 (C$^{5'}$H$_2$), 81.6 (C$^{3'}$H), 84.8 (C$^{2'}$H), 88.8 (C$^{4'}$H), 93.4 (C$^{1'}$H), 106.1 (C^5), 114.4 (C(CH$_3$)$_2$), 145.2 (CO-Orotate), 150.1 (CO-Orotate), 162.0 (COOMe), 162.6 (C^6). ESI(+)-HRMS: m/z calcd for $C_{20}H_{33}N_2O_8Si$ (M + H)$^+$: 457.2001, found, 457.2001.

*2′,3′-O-Isopropylidene-5′-O-(tert-butyldimethylsilyl)-8-carboxymethyl-adenine (**4f**).* LiHMDS (1 M solution in THF, 5 mL, 5.0 mmol, 5.0 equiv) was slowly added to a stirring solution of **4d** (525 mg, 1.0 mmol, 1.0 equiv) in THF (10 mL) at −78 °C in Ar atmosphere. The solution was stirred for 2 h at the same temperature. Then, methyl chloroformate (0.4 mL, 5.0 mmol, 5.0 equiv) was added dropwise, and the mixture was stirred for further 2 h. The solution was quenched with AcOH (2 mL), allowed to reach to room temperature, treated with sat. aq. NH_4Cl (40 mL), and concentrated. The residue was dissolved in CH_2Cl_2 (25 mL) and washed with H_2O (20 mL) and brine (20 mL), dried over Na_2SO_4, filtered, and concentrated. The residue was purified by flash chromatography (hexanes/EtOAc, 60:40) to afford N^6-benzoyl-2′,3′-O-isopropylidene-5′-O-(tert-butyldimethylsilyl)-8-carboxymethyl-adenine **10** (490 mg, 89%) as a sticky yellow oil. R_f (hexanes/EtOAc, 1:1) = 0.57. ^{1}H NMR (600 MHz, $CDCl_3$) δ ppm: −0.04 (s, 3H, SiCH_3), −0.03 (s, 3H, SiCH_3), 0.84 (s, 9H, SiC(CH_3)$_3$), 1.40 (s, 3H, CH_3), 1.63 (s, 3H, CH_3), 3.73 (dd, J = 10.7, 6.4 Hz, 1H, C$^{5'}$$H$H), 3.82 (dd, J = 10.7, 6.2 Hz, 1H, C$^{5'}$HH), 4.09 (s, 3H, OCH_3), 4.28−4.33 (m, 1H, C$^{4'}$$H$), 5.16 (dd, J = 6.4, 3.9 Hz, 1H, C$^{3'}$$H$), 5.75 (dd, J = 6.4, 2.2 Hz, 1H, C$^{2'}$$H$), 7.10 (d, J = 2.2 Hz, 1H, C$^{1'}$$H$), 7.54 (t, J = 7.7 Hz, 2H, H-Bz), 7.63 (t, J = 7.6 Hz, 1H, H-Bz), 8.01 (d, J = 7.5 Hz, 2H, H-Bz), 8.92 (s, 1H, C^2H), 9.20 (s, 1H, NH).^{13}C NMR (150 MHz, $CDCl_3$) δ ppm: −5.3 (SiCH_3), −5.2 (SiCH_3), 18.5 (C(CH_3)$_3$), 25.7 (CH_3), 26.0 (C(CH_3)$_3$), 27.4 (CH_3), 53.9 (OCH_3), 63.4 (C$^{5'}$H$_2$), 82.1 (C$^{3'}$H), 83.7 (C$^{2'}$H), 88.0 (C$^{4'}$H),

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90.9 ($C^{1'}H$), 114.6 ($C(CH_3)_2$), 121.7 (C^5), 128.1 (*C*-Bz), 129.0 (*C*-Bz), 133.1 (*C*-Bz), 133.7 (*C*-Bz), 141.0 (C^8), 151.6 (C^4), 152.0 (C^6), 154.9 (C^2), 159.2 (COOMe), 164.7 (NCO). ESI(+)-HRMS: *m*/*z* calcd for $C_{28}H_{38}N_5O_7Si$ $(M + H)^+$: 584.2535, found, 584.2536. ESI(+)-HRMS: *m*/*z* calcd for $C_{28}H_{37}N_5NaO_7Si$ $(M+Na)^+$: 606.2356, found, 606.2355. To a solution of **10** (583 mg, 1.0 mmol, 1.0 equiv) in anhydrous MeOH (10 mL), NaOMe (54 mg, 1.0 mmol, 1.0 equiv) was added, and the mixture was stirred at room temperature overnight. While the mixture was concentrated, the residue was absorbed onto silica and purified by column chromatography (hexanes/EtOAc 4:6) to give **4f** as a colorless foam (407 mg, 85%). R_f (hexanes/EtOAc, 4:6) = 0.35. ^{1}H NMR (600 MHz, $CDCl_3$) δ ppm: −0.06 (s, 6H, 2 × $SiCH_3$), 0.81 (s, 9H, $SiC(CH_3)_3$), 1.37 (s, 3H, CH_3), 1.59 (s, 3H, CH_3), 3.67 (dd, *J* = 10.4, 6.6 Hz, 1H, $C^{5'}HH$), 3.76 (dd, *J* = 10.4, 6.7 Hz, 1H, $C^{5'}HH$), 4.01 (s, 3H, OCH_3), 4.22−4.27 (m, 1H, $C^{4'}H$), 5.10 (dd, *J* = 5.1, 3.8 Hz, 1H, $C^{3'}H$), 5.69 (d, *J* = 5.1 Hz, 1H, $C^{2'}H$), 6.66 (br s, 2H,NH_2), 7.03 (s, 1H, $C^{1'}H$), 8.33 (s, 1H, C^2H). ^{13}C NMR (150 MHz, $CDCl_3$) δ ppm: −5.3 ($SiCH_3$), −5.2 ($SiCH_3$), 18.5 ($C(CH_3)_3$), 25.7 (CH_3), 26.0 ($C(CH_3)_3$), 27.4 (CH_3), 53.6 (OCH_3), 63.5 ($C^{5'}H_2$), 82.3 ($C^{3'}H$), 83.7 ($C^{2'}H$), 88.0 ($C^{4'}H$), 90.7 ($C^{1'}H$), 114.3 ($C(CH_3)_2$), 119.2 (C^5), 138.7 (C^8), 150.8 (C^4), 155.1 (C^2), 156.9 (C^6), 159.4 (CO). ESI(+)-HRMS: *m*/*z* calcd for $C_{21}H_{34}N_5O_6Si$ $(M + H)^+$: 480.2273, found, 480.2273. ESI(+)-HRMS: *m*/*z* calcd for $C_{21}H_{33}NaN_5O_6Si$ $(M + Na)^+$: 502.2092, found, 502.2083. ESI(−)-HRMS: *m*/*z* calcd for $C_{21}H_{32}N_5O_6Si$ $(M - H)^-$: 478.2127, found, 478.2134.

■ ASSOCIATED CONTENT

Supporting Information

^{1}H and APT NMR spectral data for all compounds, NMR data of titration experiments, Job's plot, fit of binding curves, summary of binding isothermal data, free energy (ΔG) calculations of complexation, and crystal data and X-ray structure for **3a**. The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.joc.5b00911.

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Notes

The authors declare no competing financial interest.

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■ DEDICATION

This work is dedicated to Professor Julius Rebek on the occasion of his 71st birthday.

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