Supplementary Information 1

Selective Recognition of Uracil and its Derivatives Using a DNA Repair Enzyme Structural Mimic

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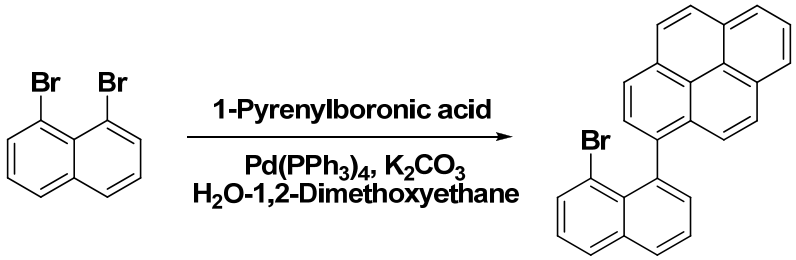
1. General Information

Chemicals were used as purchased without further purification. Flash chromatography was performed over silica gel (70–230 mesh) and monitored through thin layer chromatography (TLC) on silica gel plates. ^{1}H and ^{13}C NMR spectra were recorded using a 400 MHz instrument with $CDCl_3$, D_2O, and CD_3OD as solvents. Chemical shifts of protons are given in ppm relative to the signal of TMS as the internal standard; chemical shifts of carbon nuclei are reported in ppm relative to solvent signals used as internal standards. The structure of the complex formed between uracil and receptor **1** was calculated using Spartan'06.[1]

2. Improved synthesis of 1-[1´-(8´-bromonaphthyl)]pyrene (4)[2]



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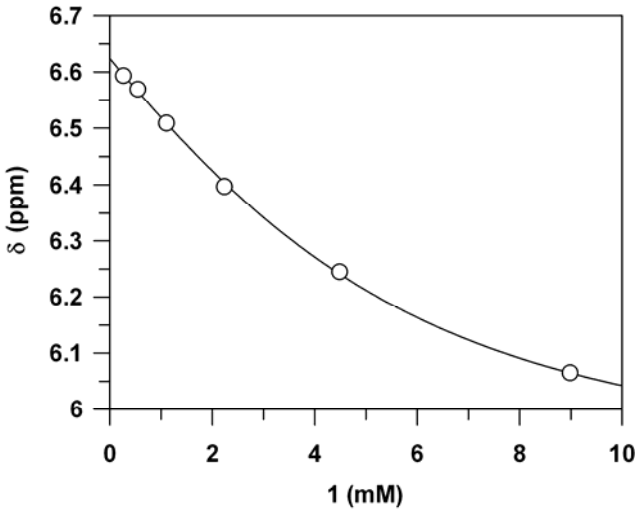
$Pd(PPh_3)_4$ (191 mg, 0.165 mmol) was added under a N_2 stream to a mixture of 1,8-dibromonaphthalene (1.40 g, 4.90 mmol), pyrenylboronic acid (1.21 g, 4.90 mmol), and 1,2-dimethoxyethane (30 mL) in a 100-mL three-neck round-bottom flask. A degassed solution of K_2CO_3 (2.92 g, 21.5 mmol) in water (15 mL) was added and the resulting mixture was heated under reflux for 13 h under N_2. After cooling to room temperature, the aqueous phase was extracted with CH_2Cl_2, the combined organic phases were dried ($MgSO_4$) and filtered, the solvents were evaporated *in vacuo*, and the residue was recrystallized from EtOAc (5–10 mL) to afford **4** as a yellow solid (1.15 g, 57%). ^{1}H NMR (400 MHz, $CDCl_3$, ppm) δ 7.33 (t, J = 7.6 Hz, 1 H), 7.58 (m, 3 H), 7.72 (d, J = 7.2 Hz), 7.89–8.22 (m, 10 H).

3. Determination of dimerization constants for 1 by dilution experiment[2]

The dimerization constant for **1** was determined by diluting a sample from its maximum solubility (9.00 mM) to the minimum concentration (0.28 mM) required for detection of a signal by ^{1}H NMR spectroscopy at 298 K. The chemical shift at 6.064 ppm at high concentration was followed. The chemical shift of **1** increased gradually to 6.592 ppm, as the concentration of **1** decreased. The data were then fitted to a nonlinear regression curve on a PC using the following equation (1):

$$\delta = \delta_m - [(\delta_m - \delta_d)/c]\,[c + 0.25/K_d - (0.5c/K_d + 0.0625/K_d)^{1/2}] \qquad (1)$$

where δ, δ_m, δ_d, and c are the observed chemical shift, the chemical shift of the monomer, the chemical shift of the dimer, and the total concentration of the receptor; K_d is the dimerization constant (Figure S1).



$\delta_m = 7.19 \pm 0.09$ ppm
$\delta_d = 5.72 \pm 0.08$ ppm
$c = 9.0$ mM
$K_d = 600 \pm 40$ M^{-1}

FIGURE S1. Measurement of the dimerization constant for **1** in D_2O using NMR spectroscopy.

4. Determination of binding stoichiometry through Job plot analysis[2]

A Job plot was used to identify the binding stoichiometry of the complex formed between uracil and **1a** at 298 K. To construct a Job plot, two stock solutions of **1** and uracil were prepared at the same concentrations (10 mM) in D_2O. For ^1H NMR spectroscopic analysis, 11 solutions were prepared by mixing the two stock solutions at volume ratios of 0:500, 50:450, 100:400, 150:350, 200:300, 250:250, 300:200, 350:150, 400:100, 450:50, and 500:0 μL. The concentrations of the complex of **1a** with uracil were estimated using equation (2) and the chemical shift of the C(5)H hydrogen atom of uracil (initially at 5.7107 ppm). The plot of the concentrations of the complex against the molar fraction of uracil created a Job plot, which revealed 1:1 binding between uracil and receptor **1a** (Figure S2).

$$[\text{complex}] = [\text{U}](\delta_{obs} - \delta_u)/(\delta_c - \delta_u) \quad (2)$$

where [U], δ_c, δ_{obs}, and δ_u are the concentration of uracil, the chemical shift of uracil in the complex, the observed chemical shift of uracil during binding, and the chemical shift of uracil in the absence of **1a**, respectively.

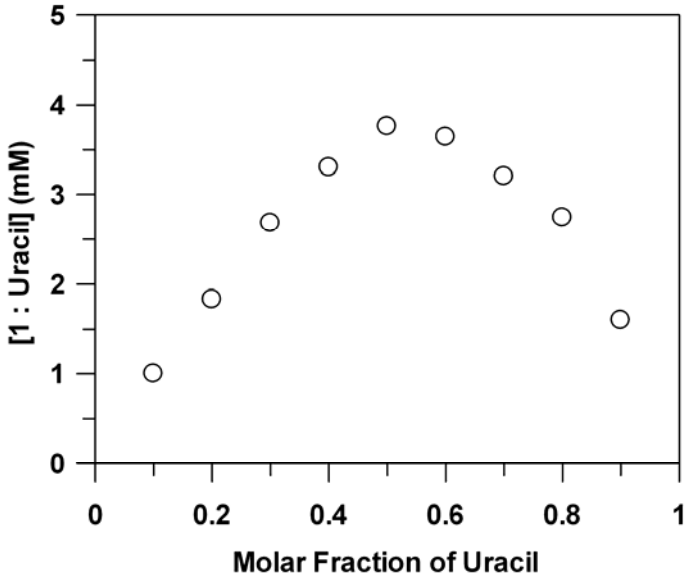


FIGURE S2. Job plot for the complex formed between uracil and **1a**.

5. Plot of logK_b with respect to values of pK_a of N(1)H units of uracil and its derivatives

Based on the data in Table S1, Figure S3 present a plot of log K_b with respect to the values of pK_a of the N(1)H units of uracil, thymine, 5-formyluracil, 5-fluorouracil, and 5-nitrouracil.

TABLE S1. Values of pK_a and logK_b for Uracil and its Derivatives[3]

compound	pK_a of N(1)H[a]	pK_a of N(3)H[a]	logK_b
Thymine (Thy)	11.23	10.04	1.78
Uracil (Ura)	10.47	9.34	2.04
5-Formyluracil (5FoU)	6.94	7.58	2.29
5-Fluorouracil (5FU)	9.05	7.26	2.50
5-Nitrouracil (5NiU)	5.66	6.91	2.62

[a] Calculated using the Poisson–Boltzmann continuum-solvation model.

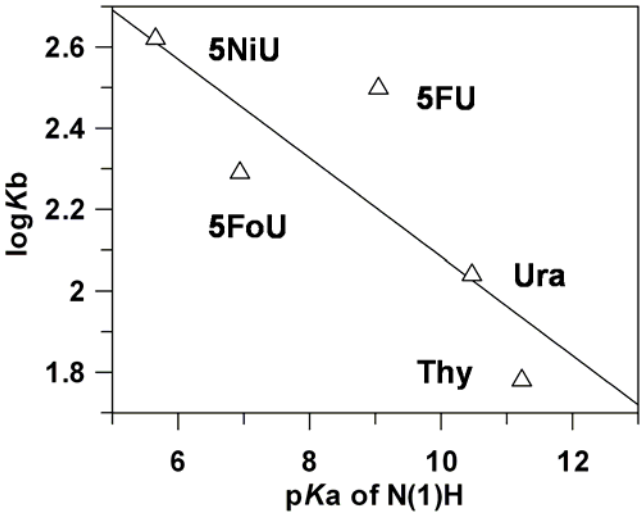


FIGURE S3. Plot of log K_b with respect to the values of pK_a of the N(1)H units of uracil derivatives. The straight line is a line of best fit.

6. Complex structure viewed from another angle

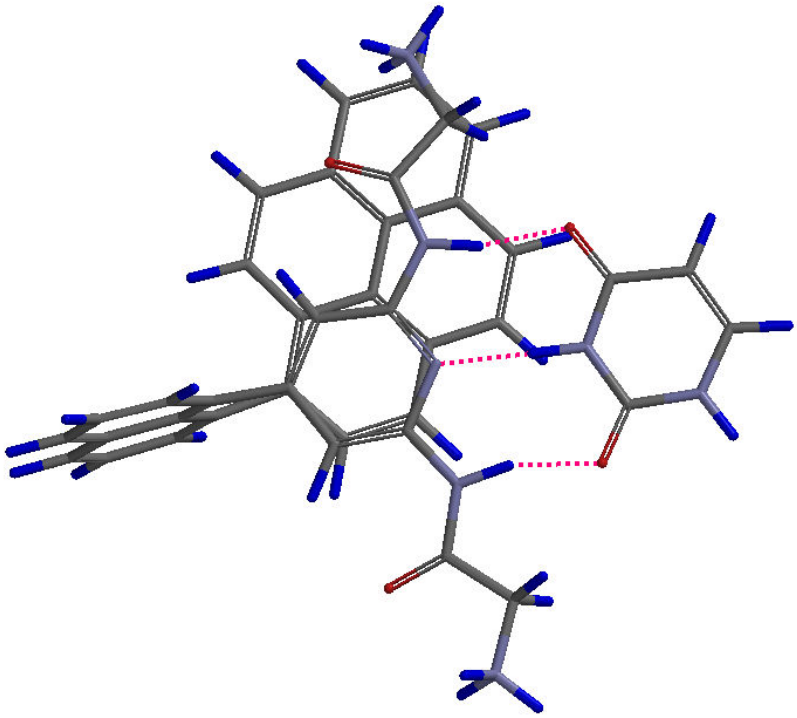


FIGURE S4. Calculated structure of the complex formed between receptor **1a** and uracil, viewed from another angle. The dotted lines represent hydrogen bonds; the two chloride ions have been omitted for clarity.

7. References

1. Shao, Y.; Fusti-Molnar, L.; Jung, Y.; Kussmann, J.; Ochsenfeld, C.; Brown, S. T.; Gilbert, A. T. B.; Slipchenko, L. V.; Levchenko, S. V.; O'Neill, D. P.; Distasio, R. A., Jr.; Lochan, R. C.; Wang, T.; Beran, G. J. O.; Besley, N. A.; Herbert, J. M.; Lin, C. Y.; Van Voorhis, T.; Chien, S. H.; Sodt, A.; Steele, R. P.; Rassolov, V. A.; Maslen, P. E.; Korambath, P. P.; Adamson, R. D.; Austin, B.; Baker, J.; Byrd, E. F. C.; Daschel, H.; Doerksen, R. J.; Dreuw, A.; Dunietz, B. D.; Dutoi, A. D.; Furlani, T. R.; Gwaltney, S. R.; Heyden, A.; Hirata, S.; Hsu, C.-P.; Kedziora, G.; Khalliulin, R. Z.; Klunzinger, P.; Lee, A. M.; Lee, M. S.; Liang, W.; Lotan, I.; Nair, N.; Peters, B.; Proynov, E. I.; Pieniazek, P. A.; Rhee, Y. M.; Ritchie, J.; Rosta, E.; Sherrill, C. D.; Simmonett, A. C.; Subotnik, J. E.; Woodcock, H. L., III; Zhang, W.; Bell, A. T.; Chakraborty, A. K.; Chipman, D. M.; Keil, F. J.; Warshel, A.; Hehre, W. J.; Schaefer, H. F., III; Kong, J.; Krylov, A. I.; Gill, P. M. W.; Head-Gordon, M. *Phys. Chem. Chem. Phys.* **2006**, *8*, 3172–3191.

2. Lamale, B.; Henry, W. P.; Daniels, L. M.; Zhang, C.; Klein, S. M.; Jiang, Y. L. *Tetrahedron* **2009**, *65*, 62–69.

3. Jang, Y. H.; Sowers, L. C.; Cagin, T.; Goddard, W.A., III. *J. Phys. Chem. A* **2001**, *105*, 274–280.