

Supplementary Information

Thermodynamic Stability of DNA Duplexes Comprising the Simplest T → dU Substitutions

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Table S1. Thermodynamic Profiles for the Unfolding of Dodecamers with dT → dU Substitutions.

Duplex	T_M (°C)	ΔH_{cal} (kcal/mol)	$T\Delta S_{cal}$ (kcal/mol)	ΔG°_{20} (kcal/mol)	Δn_{Na^+} (mol Na ⁺ /mol)	Δn_W (mol H ₂ O/mol)	ΔC_P (cal/K-mol)
<i>AATT/TTAA</i>	54.8 (67.0)	90.8 (101.0)	81.1 (87.0)	9.6 (14.0)	2.2	69	840
<i>AATU/TTAA</i>	52.0 (66.9)	86.5 (84.3)	78.0 (72.7)	8.5 (11.6)	2.3	49	-150
<i>AAUT/TTAA</i>	49.6 (66.3)	84.4 (84.6)	76.7 (73.1)	7.7 (11.5)	3.0	52	10
<i>AAUU/TTAA</i>	49.0 (65.7)	81.9 (84.4)	74.5 (73.0)	7.4 (11.4)	3.0	51	150
<i>AATT/UTAA</i>	51.9 (66.8)	84.8 (84.3)	76.5 (72.7)	8.3 (11.6)	2.3	48	70
<i>AATT/TUAA</i>	48.8 (66.6)	79.0 (84.3)	71.9 (72.7)	7.1 (11.6)	2.8	50	300
<i>AATT/UUAA</i>	48.7 (65.1)	78.7 (85.2)	71.7 (73.8)	7.0 (11.4)	2.8	49	400
<i>AATU/UTAA</i>	50.8 (65.6)	85.8 (84.2)	77.6 (72.9)	8.2 (11.3)	2.4	50	-110
<i>AAUT/TUAA</i>	48.8 (64.6)	81.0 (84.2)	73.8 (73.1)	7.2 (11.1)	2.9	43	200
<i>AAUU/UUAA</i>	47.9 (64.0)	80.8 (83.6)	73.8 (72.7)	7.0 (10.9)	2.9	48	170

All parameters are measured in 10 mM NaPi buffer at pH 7.0. Values in parenthesis correspond to oligonucleotides in 0.2 M NaCl. Experimental uncertainties are as follow: T_M (± 0.5 °C), ΔG° ($\pm 7\%$), ΔH_{cal} ($\pm 5\%$), $T\Delta S$ ($\pm 5\%$), Δn_{Na^+} ($\pm 12\%$), Δn_W ($\pm 12\%$), ΔC_P ($\pm 20\%$).

Table S2. ΔC_p Corrected Thermodynamic Profiles for the Unfolding of Dodecamers with dT $\rightarrow$ dU Substitutions.

Duplex	T_M (°C)	ΔH_{cal} (kcal/mol)	$T\Delta S_{cal}$ (kcal/mol)	ΔG°_{20} (kcal/mol)
<i>AATT/TTAA</i>	54.8 (67.0)	120.0 (140.5)	101.8 (114.9)	18.2 (25.6)
<i>AATU/TTAA</i>	52.0 (66.9)	81.7 (77.3)	69.9 (63.1)	11.8 (14.1)
<i>AAUT/TTAA</i>	49.6 (66.3)	84.7 (85.1)	73.0 (69.7)	11.7 (15.4)
<i>AAUU/TTAA</i>	49.0 (65.7)	86.3 (91.3)	74.5 (64.9)	11.8 (16.4)
<i>AATT/UTAA</i>	51.9 (66.8)	87.0 (87.6)	74.4 (71.7)	12.6 (15.9)
<i>AATT/TUAA</i>	48.8 (66.6)	87.6 (98.3)	75.7 (80.5)	11.9 (17.8)
<i>AATT/UUAA</i>	48.7 (65.1)	90.2 (103.2)	78.0 (84.9)	12.2 (18.3)
<i>AATU/UTAA</i>	50.8 (65.6)	82.4 (79.2)	70.8 (65.0)	11.6 (14.2)
<i>AAUT/TUAA</i>	48.8 (64.6)	86.8 (93.1)	75.0 (76.7)	11.8 (16.4)
<i>AAUU/UUAA</i>	47.9 (64.0)	85.5 (91.1)	74.1 (75.2)	11.4 (15.9)

All parameters are measured in 10 mM NaPi buffer at pH 7.0. Values in parenthesis correspond to oligonucleotides in 0.2 M NaCl. Experimental uncertainties are as follow: T_M (± 0.5 °C), ΔG° ($\pm 7\%$), ΔH_{cal} ($\pm 5\%$), $T\Delta S$ ($\pm 5\%$).

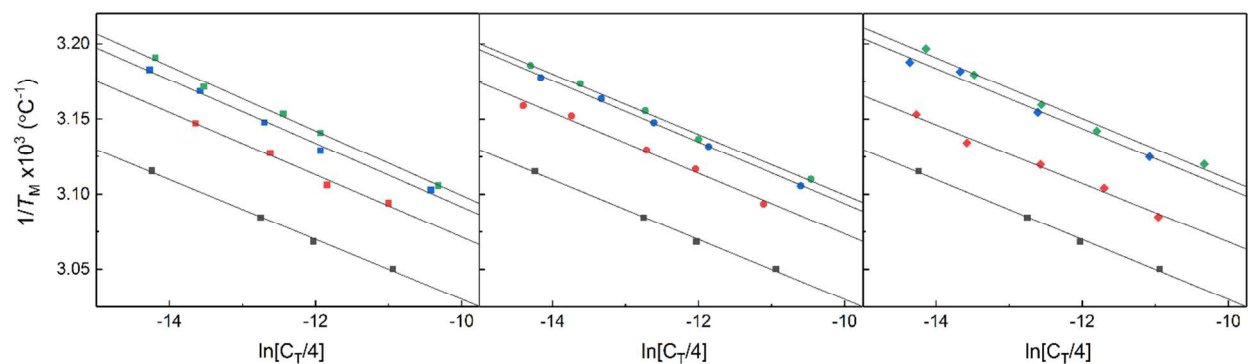


Figure S1. T_M dependence on total strand concentration at 260 nm for duplexes in 10 mM NaPi buffer at pH 7.0. (Left) *AATT/TTAA* (black squares), *AATU/TTAA* (red squares), *AAUT/TTAA* (blue squares), *AAUU/TTAA* (green squares) (Middle) *AATT/TTAA* (black squares), *AATT/UTAA* (red circles), *AATT/TUAA* (blue circles), *AATT/UUAA* (green circles) (Right) *AATT/TTAA* (black squares), *AATU/UTAA* (red diamonds), *AAUT/TUAA* (blue diamonds), *AAUU/UUAA* (green diamonds).

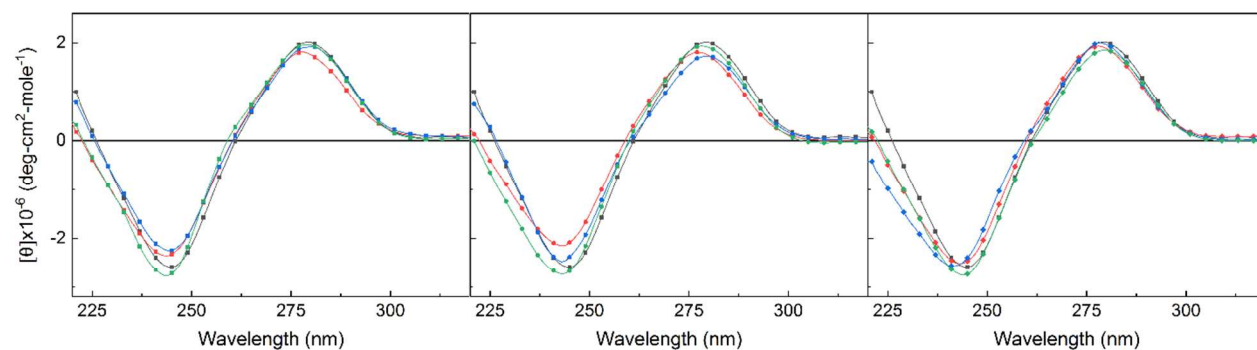


Figure S2. CD spectra of duplexes in 10 mM NaPi buffer at pH 7.0. (Left) *AATT/TTAA* (black squares), *AATU/TTAA* (red squares), *AAUT/TTAA* (blue squares), *AAUU/TTAA* (green squares) (Middle) *AATT/TTAA* (black squares), *AATT/UTAA* (red circles), *AATT/TUAA* (blue circles), *AATT/UUAA* (green circles) (Right) *AATT/TTAA* (black squares), *AATU/UTAA* (red diamonds), *AAUT/TUAA* (blue diamonds), *AAUU/UUAA* (green diamonds).

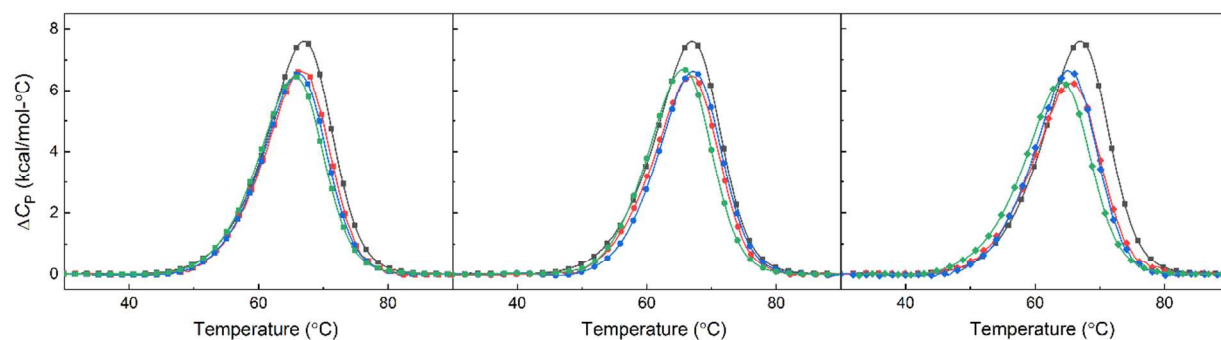


Figure S3. DSC thermograms of duplexes in 10 mM NaPi buffer, 0.2 M NaCl at pH 7.0. (Left) *AATT/TTAA* (black squares), *AATU/TTAA* (red squares), *AAUT/TTAA* (blue squares), *AAUU/TTAA* (green squares) (Middle) *AATT/TTAA* (black squares), *AATT/UTAA* (red circles), *AATT/TUAA* (blue circles), *AATT/UUAA* (green circles) (Right) *AATT/TTAA* (black squares), *AATU/UTAA* (red diamonds), *AAUT/TUAA* (blue diamonds), *AAUU/UUAA* (green diamonds).

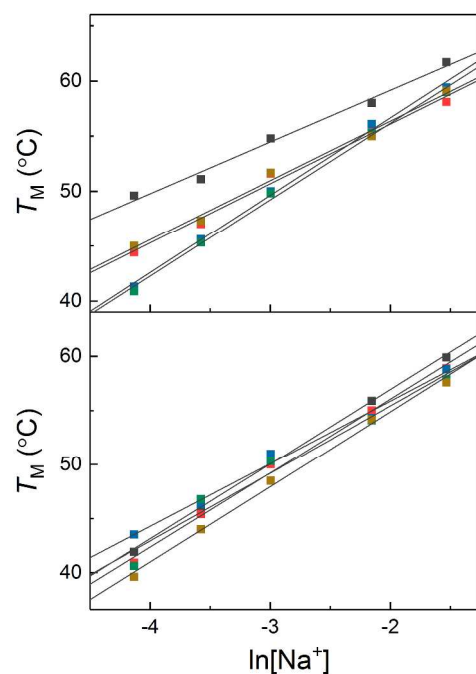


Figure S4. T_M dependence on salt concentration of duplexes in 10 mM NaPi buffer at pH 7.0. (Top) *AATT/TTAA* (black squares), *AATU/TTAA* (red squares), *AAUT/TTAA* (blue squares), *AAUU/TTAA* (green squares), *AATT/UTAA* (gold squares) (Bottom) *AATT/TUAA* (black squares), *AATT/UUAA* (red squares), *AATU/UTAA* (blue squares), *AAUT/TUAA* (green squares), *AAUU/UUAA* (gold squares).

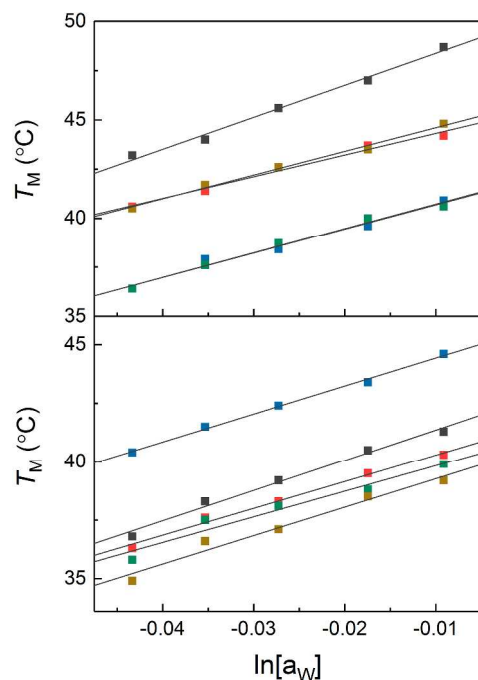


Figure S5. T_M dependence on ethylene glycol concentration of duplexes in 10 mM NaPi buffer at pH 7.0. (Top) *AATT/TTAA* (black squares), *AATU/TTAA* (red squares), *AAUT/TTAA* (blue squares), *AAUU/TTAA* (green squares), *AATT/UTAA* (gold squares) (Bottom) *AATT/TUAA* (black squares), *AATT/UUAA* (red squares), *AATU/UTAA* (blue squares), *AAUT/TUAA* (green squares), *AAUU/UUAA* (gold squares).