

## Supplementary Materials

### An Unusual Monomer Recognition of Guanine Containing Mixed Sequence DNA

#### by a Dithiophene Heterocyclic Diamidine

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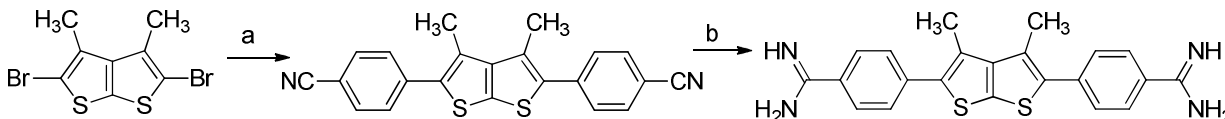
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## Scheme

### Synthesis of 2, 5-Bis (4-amidinophenyl)-3, 4-dimethyl-thieno [2, 3-*b*] thiophene dihydrochloride (DB 2297).



Reagents and conditions: a. 4-cyanophenylboronic acid, Pd(PPh<sub>3</sub>)<sub>4</sub>, Na<sub>2</sub>CO<sub>3</sub>, dioxane, reflux b. LiN[Si(CH<sub>3</sub>)<sub>3</sub>]<sub>2</sub>, THF, rt

### 2, 5-Bis (4-cyanophenyl)-3, 4-dimethyl-thieno[2,3-*b*] thiophene

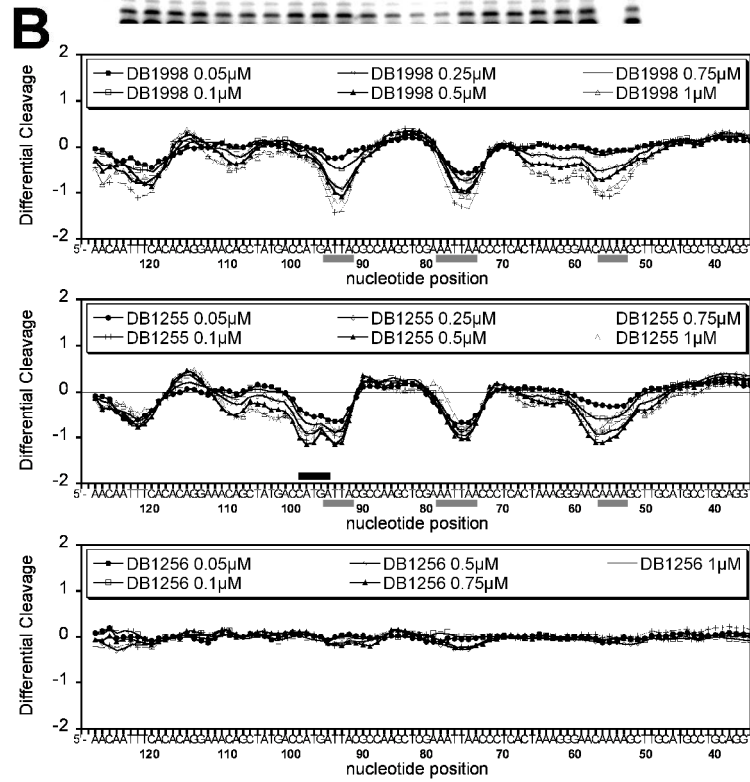
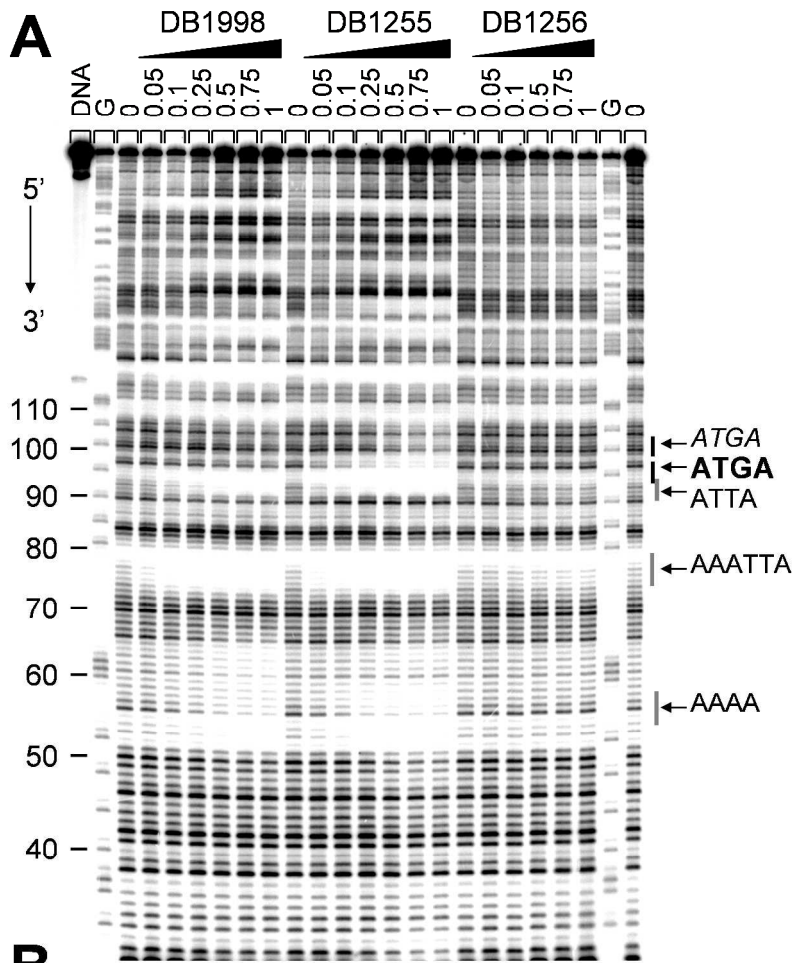
To a solution of 2, 5-dibromo-3, 4-dimethylthieno-[2, 3-*b*]thiophene<sup>1</sup> (1.63 g, 0.005 mole), 4-cyanophenyl boronic acid (g, 0.012 mole) in 80 ml dioxane, was added 2M Na<sub>2</sub>CO<sub>3</sub> (0.024 mole, 12 ml) and 5 ml ethanol (under nitrogen) was added Pd(PPh<sub>3</sub>)<sub>4</sub> (0.24 g, 2 mole %). The reaction mixture was refluxed for 16 hr (TLC monitored), concentrated under reduced pressure, diluted with water, filtered, and washed with water. The solid was dissolved in large volume of hot ethyl acetate, filtered through a bed of celite, dried over anhydrous MgSO<sub>4</sub>, concentrated under reduced pressure and collected by filtration. The solid was dried under vacuum at 50<sup>0</sup>C (12 h) yielding yellow needles 1.2 g (68%); mp 312-314°C. <sup>1</sup>H NMR (DMSO-*d*<sub>6</sub>/65°C): 7.95 (d, 2H, J=8.5Hz), 7.69 (d, 2H, J=8.5Hz), 2.56 (s, 6H); <sup>13</sup>C NMR (DMSO-*d*<sub>6</sub>/65°C): 140.3, 138.0, 137.2, 132.7, 129.6, 125.4, 118.3, 109.6, 14.8; MS (FAB): *m/z* 371 (M<sup>+</sup>); Anal. calculated for C<sub>22</sub>H<sub>14</sub>N<sub>2</sub>S<sub>2</sub>: C, 71.32; H, 3.81; N, 7.56; Found: C, 71.36; H, 3.72; N, 7.37

### 2, 5-Bis (4-amidinophenyl)-3, 4-dimethyl-thieno [2, 3-*b*] thiophene dihydrochloride (DB 2297)

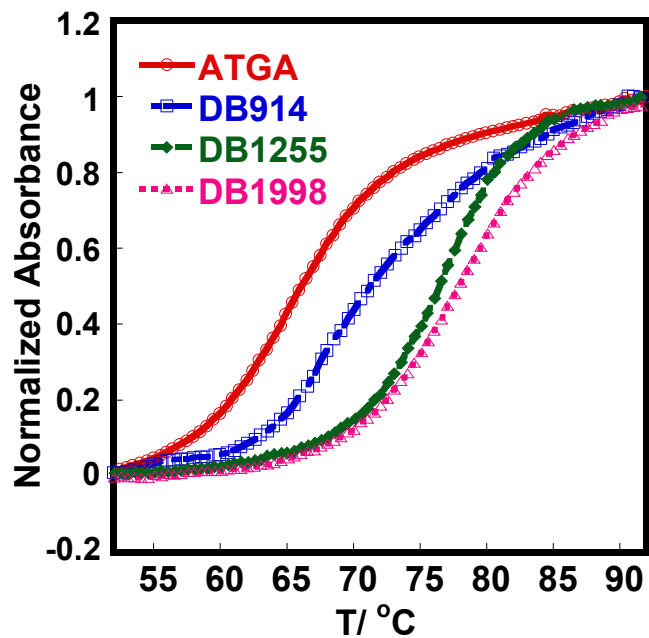
The above bis-nitrile (0.371 g, 0.001mol) was suspended in anhydrous THF (15 mL) and LiN[Si(CH<sub>3</sub>)<sub>3</sub>]<sub>2</sub> solution 1.0 M in THF (5 mL) was added to the flask and the mixture was stirred overnight at room temperature, saturated ethanolic HCl (10 mL) was carefully added while cooling and stirring, and the mixture was stirred overnight. The precipitate was filtered, washed with ether, and dried. The salt was put into water, basified with aqueous NaOH 10%, and stirred vigorously for 24 h. The precipitate was then filtered, washed with water, ether, and dried to

afford the free base as a yellow solid, which was placed in a 250 mL flask, EtOH was added to the flask, and the solution was chilled in an ice-bath. After passing dry HCl gas for 15 minutes the mixture was stirred overnight. The precipitate was filtered, washed with ether and dried under vacuum at 50<sup>0</sup>C (12 h) yielding pale yellow solid 0.35 g ( 72%); mp >320°C dec. <sup>1</sup>H NMR (DMSO-d<sub>6</sub>/75°C): 9.44 (br, 4H), 9.26 (br, 4H), 7.98 (d, 2H, J=8.5Hz), 7.73 (d, 2H, J=8.5Hz), 2.58 (s, 6H); MS (FAB): *m/z* 405 (M<sup>+</sup>); <sup>13</sup>C NMR (DMSO-d<sub>6</sub>/65°C): 165.2, 157.4, 148.23, 139.4, 138.0, 129.5, 128.7, 128.6, 126.8, 13.7; Anal. calculated for C<sub>22</sub>H<sub>20</sub>N<sub>4</sub>S<sub>2</sub>.2HCl.0.5H<sub>2</sub>O: C, 54.31; H, 4.76; N, 11.52; Found: C, 54.51; H, 4.86; N, 11.42.

<sup>1</sup>Gather, M. C., Heeney, M., Zhang, W., Whitehead, K. S., Bradle, D. D. C., McCulloch I. and Campbell, A. J., (2008) An alignable fluorene thienothiophene copolymer with deep-blue electroluminescent emission at 410 nm. *Chem. Commun.* 1079–1081



**Figure S1.** DNase I footprinting titration experiments for increasing concentration of DB1998, DB1255 and DB1256: (A) Denaturing polyacrylamide gel and (B) the corresponding densitometric analysis using lower concentration of compounds. G, grey and black boxes are as defined in the legend of Figure 2.

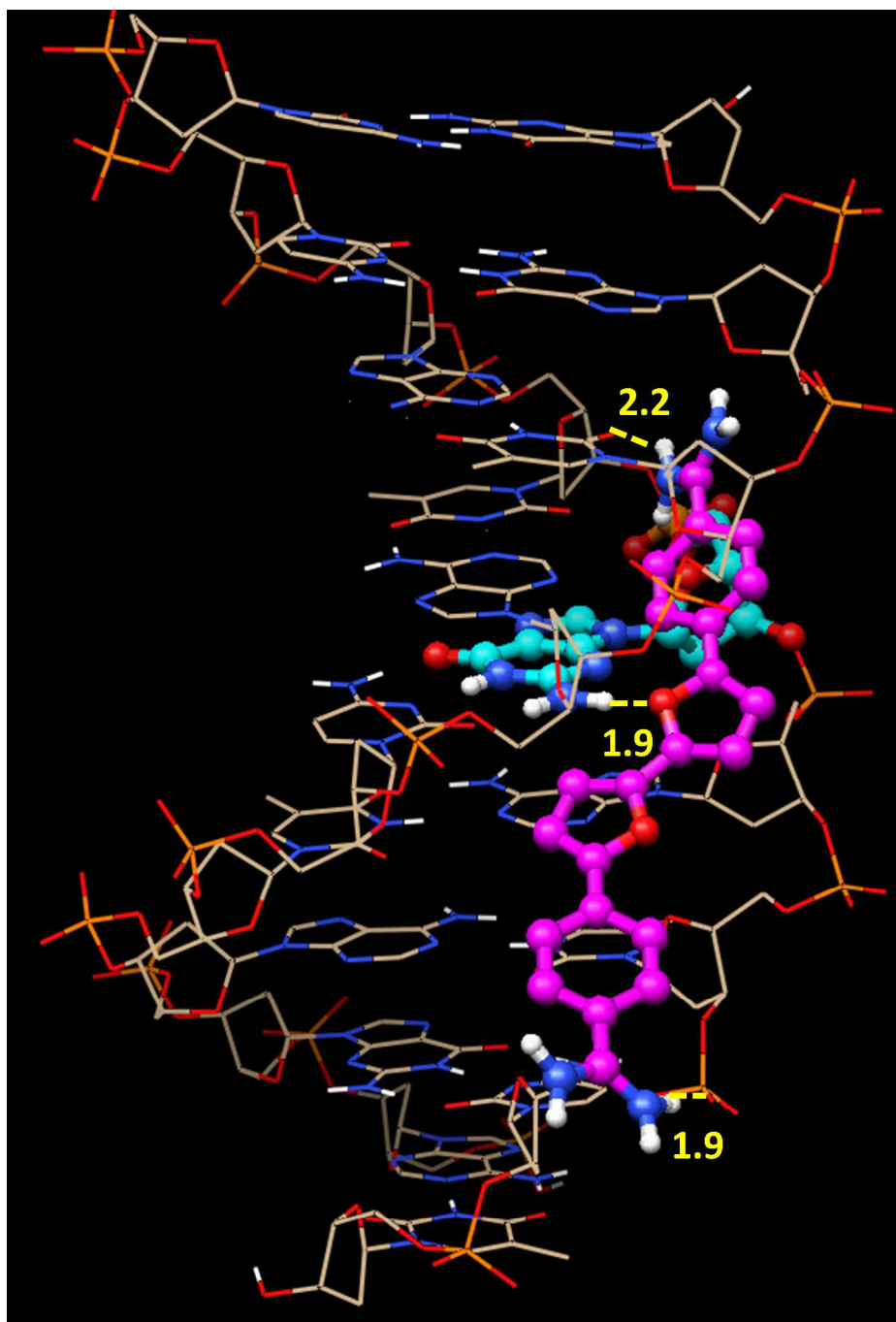


**Figure S2. Thermal melting:** Representative melting scans of free and bound ATGA sequence in the presence of DB1255, DB914, and DB1998. The experiments were conducted in cacodylic acid buffer at pH 6.25 and 25 °C

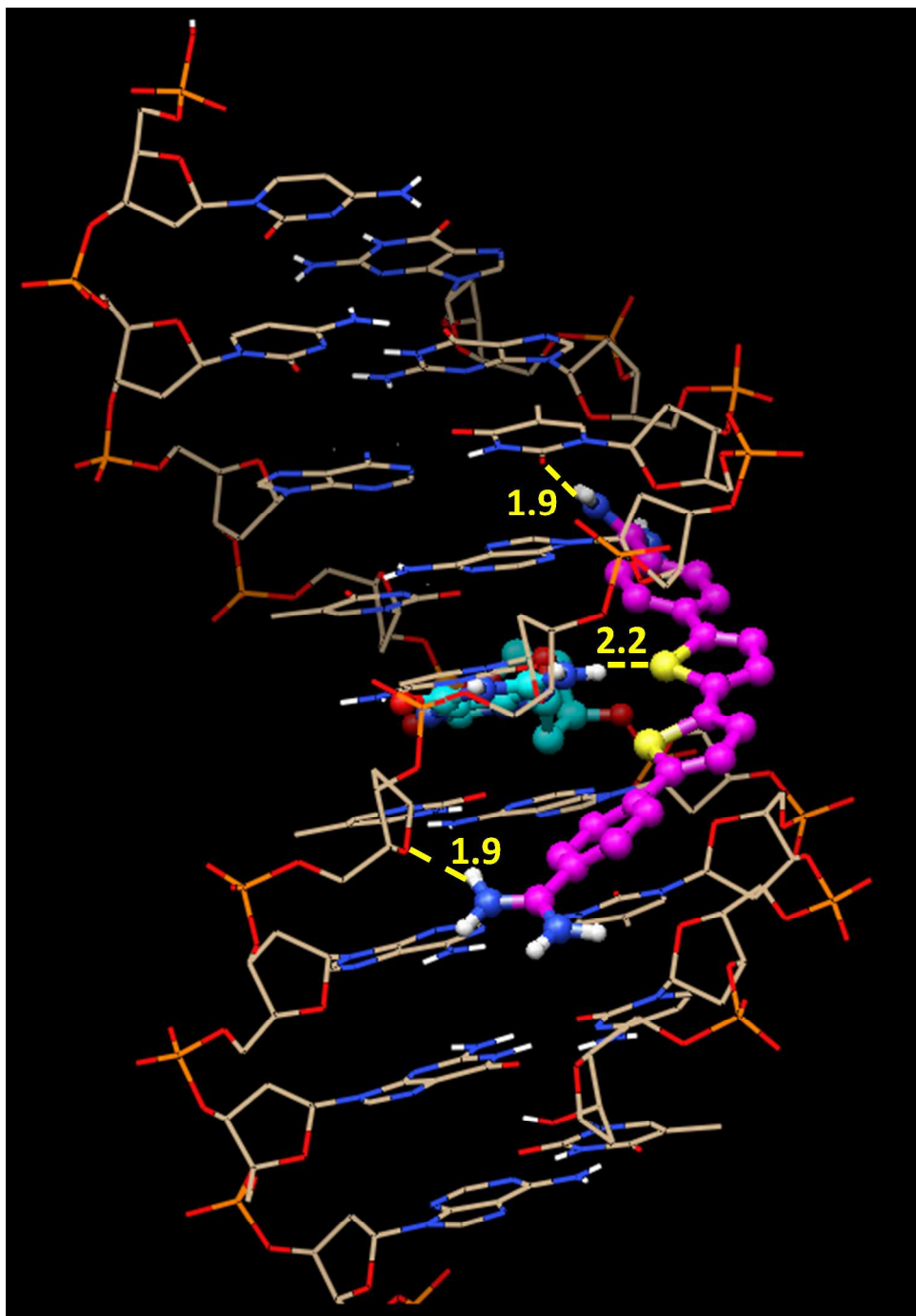
**Table S1:**  $T_m$  values for DB1255 and analogues in the AATT and ATGA sequences.

<b>Compound</b>	<b><math>\Delta T_m</math> (°C)</b>	
	<b>AATT</b>	<b>ATGA</b>
DB914	3.1	2.1
DB1255	7.5	12.2
DB1282	3.2	5.3
DB1998	15.1	14.1
DB2297	1.0	2.3
DB832	3.0	2.0
DB1450	5.4	5.2
DB1341	2.5	4.5

The experiments were conducted in cacodylic acid buffer at pH 6.25 and 25 °C. The concentration of oligomer DNA was 3  $\mu$ M and melting studies were performed at 2:1 compound to DNA ratio.



**FigureS3:** Docking results for DB914-*d*[(5'-CCATGATCT-3')(5'-AGATCATGG-3')] by using AUTODOCK v4.02. One furan O atom (red) is participating in hydrogen bonding with the G N-H (shown with a yellow dashed line). The upper amidine group forms an H-bond (weak) with the thymine carbonyl of T4 and the lower amidine forms an H-bond with the phosphate backbone. The DNA backbone, G5 base and compound are shown and colored as in Figure 7.



**FigureS4:** Docking results for DB1255-*d*[(5'-CCATGATCT-3')(5'-AGATCATGG-3')] by using AUTODOCK v4.02. One thiophene S atom (yellow) is participating in hydrogen bonding with the G N-H (shown with a yellow dashed line). The upper amidine group forms an H-bond with the thymine carbonyl of T7 (yellow dashed line). The lower amidine group forms a



hydrogen bond with the DNA backbone. The DNA backbone, DB1255 and G5 are shown and colored as in Figure 7.