SUPPORTING INFORMATION

An Improved Route for the Synthesis of Guanine Quadruplex Ligand Phen-DC3
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p. 10 - 11 Experimental details and spectra for quantitative $^{19}$F NMR analysis of Phen-DC3 (triflate salt)
A. NMR of Synthesized Compounds

Fig. S1: $^1$H NMR spectrum of compound 2 (CDCl$_3$, 25 °C, 400 MHz). CDCl$_3$ was neutralized by filtration through basic alumina prior to use.

Fig. S2: $^1$H NMR spectrum of compound 3 (DMSO-d$_6$, 25 °C, 300 MHz).
Fig. S3: $^1$H NMR spectrum of compound 4 (DMSO-d$_6$, 25 °C, 300 MHz).

Fig. S4: COSY NMR spectrum of compound 4 (DMSO-d$_6$, 25 °C, 300 MHz).
**Fig. S5:** $^1$H NMR spectrum of compound 5 (DMSO-$d_6$, 25 °C, 500 MHz).

**Fig. S6:** COSY NMR spectrum of compound 5 (DMSO-$d_6$, 25 °C, 500 MHz).
Fig. S7: $^1$H NMR spectrum of Phen-DC3 (triflate salt) produced by metathesis with silver(I) triflate (DMSO-d$_6$, 25 °C, 500 MHz).

Fig. S8: $^1$H NMR spectrum of Phen-DC3 (triflate salt) produced by metathesis with calcium(II) triflate (DMSO-d$_6$, 25 °C, 500 MHz).
B. Synthesis of Compound 2 by Radical Chlorination: Reaction Optimization and Analysis of Byproducts

Scheme S1: Radical chlorination of 2,9-dimethyl-1,10-phenanthroline, with proposed byproducts 6 and 7.

Following literature protocols, 10 equivalents of NCS in CH₂Cl₂ (14 mL) were added dropwise to a solution of 2,9-dimethyl-1,10-phenanthroline (100 mg, 0.49 mmol, 1.0 equiv.) in CHCl₃ (7 mL). The reaction was protected from light, placed under argon, and stirred under reflux (46 °C, 48 hours). Purification by flash column chromatography (SiO₂, 1:1 CHCl₃:hexanes as eluent, Rf = 0.28) afforded compound 2 in high yields (see Table S1). Increasing the scale of reaction to 750 mg of 2,9-dimethyl-1,10-phenanthroline resulted in a significant drop in yield and a notable increase in the formation of several byproducts. Increasing the number of equivalents of NCS used did not improve matters. Two of the byproducts, compounds 6 and 7, were also isolated by column chromatography (SiO₂, 1:1 to 2:1 CHCl₃:hexanes as eluent). NMR and EI-MS analysis suggests that under-chlorination at the methyl positions and over-chlorination of the phenanthroline unit may be occurring (see Figs. S9 – S14). NMR assignments were verified using COSY, HSQC, and HMBC experiments. The formation of byproducts was minimized by (i) reducing the number of equivalents of NCS to 8.0, and (ii) substituting CH₂Cl₂ with CHCl₃ to increase the temperature of reflux (61 °C), allowing the reaction to reach completion within 6 hours.

Table S1: Exploration of reagent conditions for the synthesis of 4 through radical chlorination with NCS

<table>
<thead>
<tr>
<th>Scale of Reaction (amount of 2,9-dimethyl-1,10-phenanthroline)</th>
<th>Equivalents of NCS</th>
<th>Percent Yield of Compound (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>100 mg, 0.49 mmol</td>
<td>10</td>
<td>73</td>
</tr>
<tr>
<td>750 mg, 3.6 mmol</td>
<td>10</td>
<td>51</td>
</tr>
<tr>
<td>750 mg, 3.6 mmol</td>
<td>15</td>
<td>26</td>
</tr>
<tr>
<td>430 mg, 2.1 mmol</td>
<td>8</td>
<td>68</td>
</tr>
</tbody>
</table>

6. ¹H NMR (500 MHz, CDCl₃) δ 7.83 (d, 3J = 9.0 Hz, 1 H), 7.94 (d, 3J = 9.0 Hz, 1 H), 8.29 (d, 3J = 8.5 Hz, 1 H), 8.38 – 8.40 (m, 2 H). ¹³C NMR (126 MHz, CDCl₃) δ 97.1, 98.2, 120.6, 126.3, 128.8, 129.1, 129.1, 130.5, 138.3, 140.0, 140.9, 143.1, 151.0, 158.4. R.f. (1:1 CHCl₃:hexanes) = 0.33. EI(+)HRMS calc. for [M]+ (C₁₄H₂₅Cl₁₂N₂): 445.8272, found: 445.8277; calc. for [M - Cl]+ (C₁₄H₂₅Cl₁₁N₂): 410.8583, found: 410.8544; calc. for [M - 2 Cl]+ (C₁₄H₂₀Cl₁₀N₂): 375.8895, found: 375.8873. M.p. 258-260 °C.

7. ¹H NMR (500 MHz, CDCl₃) δ 7.38 (s, 1 H), 7.83 (d, 3J = 9.0 Hz, 1 H), 7.92 (d, 3J = 9.0 Hz, 1 H), 8.32-8.33 (m, 2 H), 8.40 (d, 3J = 8.5 Hz, 1 H). ¹³C NMR (126 MHz, CDCl₃) δ 70.2, 98.2, 120.6, 126.9, 128.5, 129.1, 129.2, 130.4, 130.8, 143.0, 143.2, 152.5, 158.4. R.f. (2:1 CHCl₃:hexanes) = 0.19. EI(+)HRMS calc. for [M]+ (C₁₄H₂₅Cl₁₂N₂): 411.8662, found: 411.8669; calc. for [M - Cl]+ (C₁₄H₂₄Cl₁₁N₂): 376.8974, found: 376.8933; calc. for [M - 2 Cl]+ (C₁₄H₂₂Cl₁₀N₂): 341.9285, found: 341.9265. M.p. 243-245 °C.

\(^1\)H and \(^{13}\)NMR Spectra of Compounds 6 and 7:

**Fig. S9:** \(^1\)H NMR spectrum of byproduct 6 from the radical chlorination of 2,9-dimethyl-1,10-phenanthroline using 10 equivalents of NCS (CDCl\(_3\), \(25^\circ\)C, 500 MHz).\(^2\)

**Fig. S10:** \(^{13}\)C NMR spectrum of byproduct 6 (CDCl\(_3\), \(25^\circ\)C, 500 MHz).

Fig. S11: $^1$H NMR spectrum of byproduct 7 from the radical chlorination of 2,9-dimethyl-1,10-phenanthroline using 15 equivalents of NCS (CDCl$_3$, * 25 $^\circ$C, 500 MHz).

Fig. S12: $^{13}$C NMR spectrum of byproduct 7 (CDCl$_3$, * 25 $^\circ$C, 500 MHz).
High Resolution Electron Impact Mass Spectra of Compounds 6 and 7:

**Fig. S13**: EI-HRMS (positive mode) spectrum of compound 6 (C₁₄H₂Cl₂N₂). *Inset*: comparison of predicted and experimental [M]⁺ peak splitting patterns.

**Fig. S14**: EI-HRMS (positive mode) spectrum of compound 7 (C₁₄H₂Cl₂N₂). *Inset*: comparison of predicted and experimental [M]⁺ peak splitting patterns.
C. Quantitation of Phen-DC3 Triflate Counter-Ion by $^{19}$F NMR

To a solution of a known quantity of Phen-DC3 (4.0 or 2.8 mg respectively) in DMSO-$d_6$ was added α,α,α-trifluorotoluene as an internal standard (5 µL, or 5 µL of a 1:4 dilution in DMSO-$d_6$ respectively). The NMR sample was locked on DMSO-$d_6$ for $^1$H NMR analysis and the spectrometer was re-tuned for $^{19}$F NMR. The theoretical number of equivalents was calculated from the ratio between the number of moles of added internal standard and the number of moles of compound. This was then compared to the experimental ratio of integration between the standard and Phen-DC3 (see Figs. S15 and S16), as is outlined in Table S2. Values remain within the limits of experimental error.

<table>
<thead>
<tr>
<th>Source of Triflate Salt</th>
<th>Calculated Molar Ratio (n_{standard}/n_{Phen-DC3})</th>
<th>Experimental Ratio of Integration (standard/Phen-DC3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AgOTf</td>
<td>8.6</td>
<td>8.2</td>
</tr>
<tr>
<td>Ca(OTf)$_2$</td>
<td>3.1</td>
<td>3.2</td>
</tr>
</tbody>
</table>

$^{19}$F NMR Spectra of Phen-DC3 (triflate salt):  

![19F NMR spectra of Phen-DC3](image)

Fig. S15: $^{19}$F NMR spectrum of Phen-DC3 (4 mg, 4.7 µmol, 1.0 equiv.) produced from metathesis with silver(I) triflate with trifluorotoluene (5 µL, 41 µmol, 8.6 equiv.) as internal standard (DMSO-$d_6$, 25 °C, 500 MHz).
Fig. S16: $^{19}$F NMR spectrum of Phen-DC3 (2.8 mg, 3.3 µmol, 1.0 equiv.) produced from metathesis with calcium(II) triflate with trifluorotoluene (5 µL of trifluorotoluene in DMSO-d$_6$ (1:4), 10 µmol, 3.1 equiv.) as internal standard (DMSO-d$_6$, 25 ºC, 500 MHz).