Supporting Information for World Wide Web Edition

Solution Structure of an O^6-[4-oxo-4-(3-pyridyl)butyl]guanine adduct in an 11mer DNA duplex: evidence for formation of a base 'triplex'

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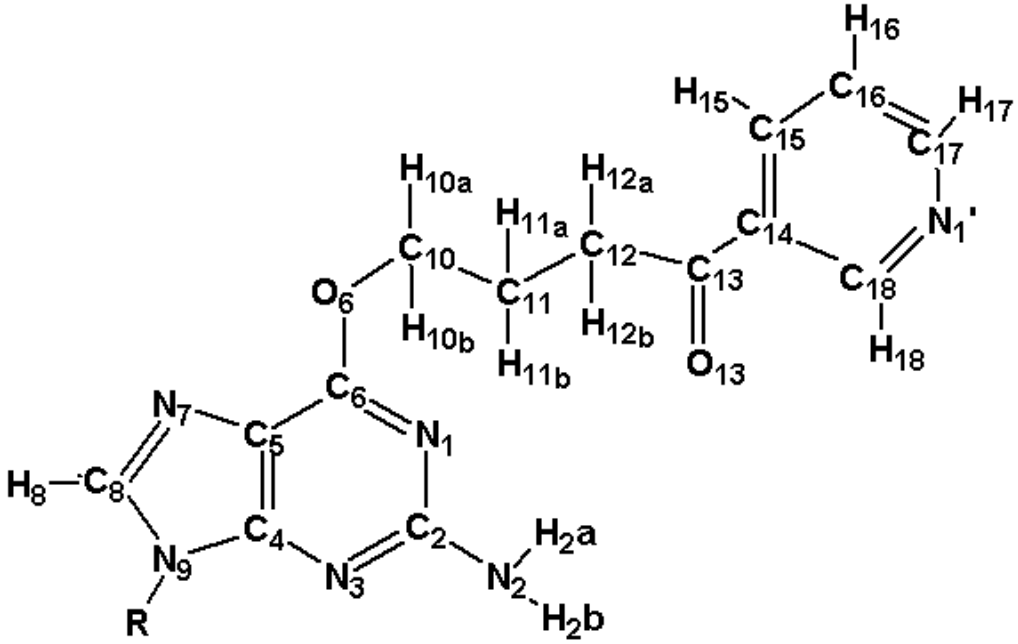
Table S1
Partial Charges for the O^6-dG-POB Residue



Name in Structure	Standard Name	Charge (Gaussian)	Name in Structure	Standard Name	Charge (Gaussian)
N1	G7(N1)	-0.340	**C15**	POB(C4')	+0.020
C2	G7(C2)	+0.419	**C16**	POB(C5')	-0.057
N2	G7(N2)	-0.400	**C17**	POB(C6')	+0.111
N3	G7(N3)	-0.334	**C18**	POB(C2')	+0.109
C4	G7(C4)	+0.279	**N1'**	POB(N1')	-0.207
C5	G7(C5)	-0.040	**H2a**	G7(H2a)	+0.164
C6	G7(C6)	+0.368	**H2b**	G7(H2b)	+0.161
O6	G7(O6)	-0.321	**H8**	G7(H8)	+0.009
N7	G7(N7)	-0.222	**H10a**	POB(H1a)	+0.011
C8	G7(C8)	+0.207	**H10b**	POB(H1b)	+0.011
N9	G7(N9)	-0.161	**H11a**	POB(H2a)	+0.041
C10	POB(C1)	+0.129	**H11b**	POB(H2b)	+0.041
C11	POB(C2)	-0.049	**H12a**	POB(H3a)	+0.038
C12	POB(C3)	-0.095	**H12b**	POB(H3b)	+0.038
C13	POB(C4)	+0.295	**H15**	POB(H4')	+0.034
O13	POB(O4)	-0.319	**H16**	POB(H5')	+0.031
C14	POB(C3')	-0.056	**H17**	POB(H6')	+0.007
			H18	POB(H2')	+0.002
The sum of the partial charges are scaled to –0.076 in order to be the same as the unmodified G					

Table S2
Torsion Angle Barriers for the POB ligand used in structure calculations

$E=V_o/2*[1 \pm \cos(m\ )]$

’	8.610/2 * (1 – cos (2* ’))	trigonal
’	-0.185/2 * (1 + cos(3* ’))	tetrahedral
’	1.07/2* (1 + cos (3* ’))	tetrahedral
’	1.07/2 * (1+ cos(3* ’))	tetrahedral
’	-0.185/2 * (1 + cos(3* ’))	tetrahedral
’	80.0/2 * (1 – cos (2 * ’))	trigonal (very stiff)

Table S3
Proton and Carbon Chemical Shifts of the POB ligand in D_2O Buffer, ppm

	^{1}H 5^{o}C	^{1}H 25 ^{o}C	ppm[a]	^{13}C 25 ^{o}C
POB(H1a/H1b)	2.90	2.71	0.19	42.43
POB(H2b)	1.73	1.70	0.03	27.78
POB(H2a)	3.69	3.81	-0.12	73.48
POB(H3a/H2b)	2.76	2.66	0.10	41.55
POB(H2’)	8.69	8.52	0.17	152.00
POB(H4')	7.92	7.73	0.19	139.85
POB(H5')	7.22	7.07	0.15	127.68
POB(H6’)	8.42	8.32	0.10	143.21
[a]Temperature dependent proton chemical shift difference (value at 5 ^{o}C – value at 25 ^{o}C)				

Table S4

Proton and Phosphorus Chemical Shifts of the O^6-dG-POB-11mer Duplex in 100% D_2O Buffer

Nonexchangeable Proton (5˚C) and Phosphorus Chemical Shifts (25˚C), ppm

	H8/H6	H5/H2/CH_3	H1'	H2', H2''	H3'	H4'	(n)-^{31}P-(n + 1)[a]
dC1	7.69	5.84	5.90	2.05,2.46	4.60	4.05	-4.15
dC2	7.56	5.64	5.35	2.16,2.39	4.81	4.08	-3.79
dA3	8.37	7.66	6.27	2.73,2.96	5.00	4.42	-4.48
dT4	7.14	1.46	5.63	2.04,2.45	4.83	4.26	-4.21
dA5	8.18	7.07	6.12	2.50,2.81	4.92	4.35	-4.35
dT6	7.01	1.23	5.65	1.86,2.25	4.77	4.06	-4.01
[POB]dG7	7.78	---	5.63	2.58,2.58	4.89	4.24	-4.19
dG8	7.55	---	5.67	2.53,2.63	4.83	4.35	-3.98
dC9	7.45	5.22	5.96	2.21,2.47	4.75	4.20	-4.00
dC10	7.56	5.57	5.90	2.15,2.40	4.76	4.08	-3.99
dC11	7.60	5.66	6.18	2.21,2.21	4.51	3.97	
dG12	7.81	---	5.62	2.51,2.66	4.77	4.12	-4.00
dG13	7.68	---	5.65	2.61,2.65	4.95	4.32	-4.13
dG14	7.64	---	5.94	2.47,2.68	4.92	4.37	-4.26
dC15	7.21	5.19	5.80	2.02,2.20	4.78	4.11	-3.67
dC16	7.60	5.67	6.23	2.30,2.61	4.88	4.22	-4.52
dA17	8.14	7.44	6.13	2.52,2.86	4.88	4.35	-3.99
dT18	7.25	1.31	5.68	2.15,2.50	4.85	4.20	-4.00
dA19	8.23	7.14	6.16	2.54,2.86	4.85	4.36	-4.46
dT20	7.07	1.30	5.63	1.78,2.17	4.78	4.06	-4.13
dG21	7.76	---	5.54	2.58,2.63	4.90	4.29	-3.91
dG22	7.74	---	6.09	2.46,2.27	4.60	4.18	

[a] ^{31}P chemical shift corresponds to residue (n) in (n)-^{31}P-(n + 1) step.

Table S5

Proton Chemical Shifts of the [POB]dG·dC-11mer Duplex in 9:1 H_2O/D_2O Buffer

Exchangeable Proton Chemical Shifts, ppm 5˚C

	G(NH1)/T(NH3)	C(NH_2-4)-exposed	C(NH_2-4)-bound
dC1·dG22	13.14	6.93	7.89
dC2·dG21	12.84	6.94	8.53
dA3·dT20	13.69	---	---
dT4·dA19	13.23	---	---
dA5·dT18	13.28	---	---
dT6·dA17	13.20	---	---
[POB]dG7·dC16	---	7.11	7.26
dG8·dC15	12.57	6.61	8.19
dC9·dG14	13.07	6.36	8.14
dC10·dG13	13.14	6.93	8.50
dC11·dG12	12.92	7.16	8.25

Table S6

Chemical Shift Differences (ppm) between the [POB]dG·dC 11mer Duplex and Control 11-mer Duplex

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Nonexchangeable Proton (5 °C) and phosphorus (25 °C)

	H8/H6	H5/H2 /CH_3	H1'	H2', H2''	H3'	H4'	(n)-^{31}P-(n + 1)
dC1	-0.07	-0.12	-0.10	-0.03,-0.04	+0.04	+0.0	+0.01
dC2	0.00	-0.04	0.00	-0.01,0.00	+0.04	+0.0	-0.02
dA3	-0.02	-0.04	-0.02	-0.02,-0.01	+0.03	+0.0	-0.01
dT4	+0.02	-0.01	0.00	+0.04,+0.01	+0.04	-0.08	-0.04
dA5	+0.03	+0.01	+0.02	+0.04,+0.04	+0.06	+0.0	-0.11
dT6	-0.01	+0.01	-0.02	-0.03,+0.01	+0.05	+0.0	-0.22
[POB]dG7	-0.05	---	-0.09	-0.01,+0.05	+0.06	+0.0	+0.18
dT8	+0.13	---	+0.12	-0.01,-0.01	+0.09	+0.0	-0.16
dC9	-0.14	-0.01	-0.10	-0.12,-0.06	+0.01	+0.0	+0.01
dC10	-0.11	-0.07	+0.09	+0.02,+0.03	+0.03	+0.0	-0.15
dC11	-0.16	-0.29	-0.07	-0.02,-0.02	+0.03	+0.0	
dG12	-0.03	---	-0.02	-0.02,-0.02	-0.02	+0.0	-0.02
dG13	+0.06	---	-0.02	+0.05,+0.10	+0.05	+0.0	+0.01
dG14	+0.04	---	-0.07	+0.04,-0.04	+0.04	+0.0	+0.03
dC15	+0.07	-0.01	+0.06	-0.01,+0.20	-0.01	+0.0	-0.56
dC16	-0.16	-0.16	-0.89	-0.22,-0.26	-0.22	-0.11	+0.62
dA17	+0.16	+0.08	+0.08	+0.13,+0.07	+0.13	+0.0	-0.50
dT18	-0.10	+0.18	-0.06	-0.07,-0.06	-0.07	0.00	-0.25
dA19	-0.02	-0.06	-0.01	-0.01,0.00	-0.01	+0.0	+0.01
dT20	-0.01	-0.01	0.00	+0.03,0.00	+0.03	+0.0	-0.01
dG21	-0.03	---	0.00	-0.01,0.00	-0.01	+0.0	-0.01
dG22	-0.07	---	-0.05	-0.18,-0.05	-0.18	+0.0	

Exchangeable Proton Chemical Shift Difference, (Control – Modified), ppm 5˚C

	G(NH1)	T(NH3)	C(NH_2-4)-e	C(NH_2-4)-b
dC1·dG22	-0.04	---	-0.06	+0.02
dC2·dG21	-0.05	---	-0.03	-0.03
dA3·dT20	---	-0.02	---	---
dT4·dA19	---	-0.03	---	---
dA5·dT18	---	-0.06	---	---
dT6·dA17	---	+0.31	---	---
[POB]dG7·dC16	---		-0.31	+1.12
dG8·dC15	+0.30		-0.30	-0.15
dC9·dG14	-0.12		+0.03	-0.06
dC10·dG13	-0.04		-0.04	-0.04
dC11·dG12	+0.05		-0.15	+0.05

Table S7

Sugar Pseudorotation (P) and Glycosidic ( ) Torsion Angles (degrees)

	Strand 1			Strand 2	
	P			P	
C1	170.21 ± 17.51	254.57 ± 5.72	**G12**	146.43 ± 36.70	231.50 ± 21.00
C2	171.43 ± 4.70	250.43 ± 1.91	**G13**	155.57± 11.21	242.43 ± 3.82
A3	156.36 ± 5.85	246.50 ± 2.38	**G14**	137.50 ± 32.63	234.07 ± 14.46
T4	155.07 ± 4.36	246.93 ± 2.89	**C15**	155.50 ± 5.19	239.50 ± 5.21
A5	157.50 ± 6.56	244.14 ± 4.44	**C16**	160.64 ± 4.29	241.64 ± 2.92
T6	115.71 ± 16.94	230.79 ± 5.09	**A17**	145.14 ± 10.85	243.36 ± 7.57
[POB]G7	129.07 ± 30.15	239.86 ± 8.68	**T18**	103.71 ± 21.28	224.07 ± 8.39
G8	145.64 ± 22.87	239.64 ± 8.54	**A19**	161.00 ± 6.68	246.14 ± 3.42
C9	117.07 ± 30.54	230.21 ± 8.46	**T20**	143.36 ± 11.08	242.86 ± 4.74
C10	101.57 ± 41.81	224.93 ± 12.65	**G20**	156.14 ± 10.16	246.71 ± 3.17
C11	112.00 ± 49.02	229.79 ± 7.98	**G21**	162.93 ± 6.03	254.36 ± 1.60

Table S8

Dihedral angles in the POB ligand

	,	,	,	,	,	,
POB slightly 5' to plane of G7						
1	95	260	232	160	175	180
2	93	261	230	159	179	179
3	96	261	230	158	179	179
4	94	260	232	164	177	180
5	93	259	232	162	178	181
Average	**94.20**	**260.20**	**231.20**	**160.60**	**177.60**	**179.80**
Standard deviation	**1.30**	**0.84**	**1.10**	**2.41**	**1.67**	**0.84**
POB slightly 3' to plane of G7						
6	292	103	136	180	167	177
7	295	103	136	176	169	177
8	294	103	137	174	170	175
9	294	104	137	174	170	175
10	292	103	138	175	167	175
Average	**293.40**	**103.20**	**136.80**	**175.80**	**168.60**	**175.80**
Standard deviation	**1.34**	**0.45**	**0.84**	**2.49**	**1.52**	**1.10**

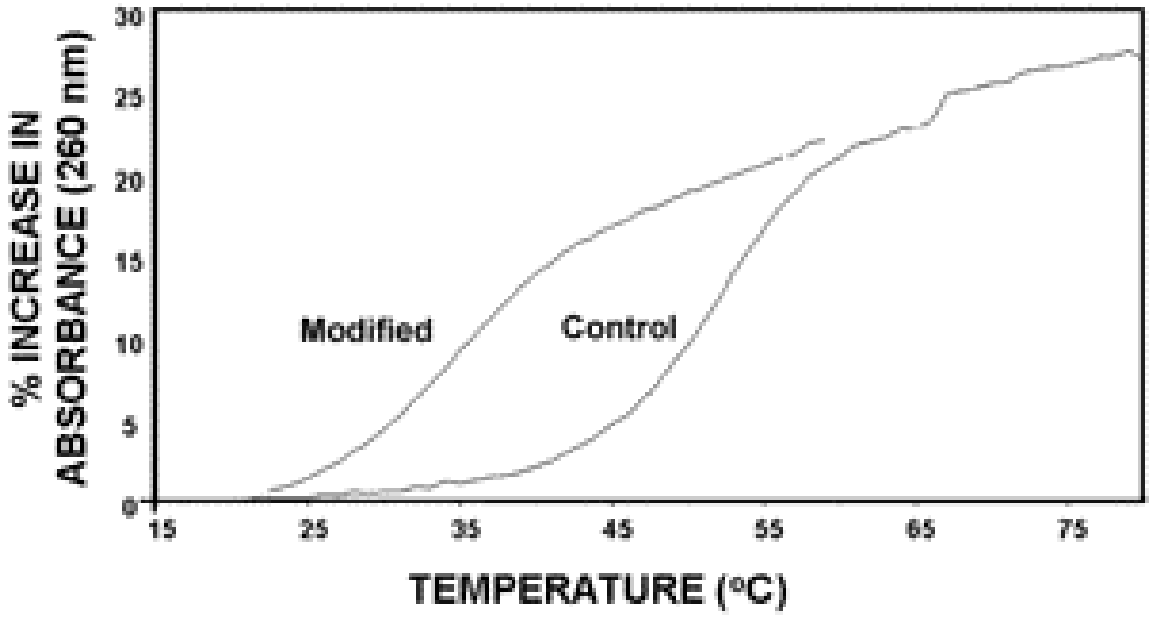

Figure S1: Melting curves for the [POB]dG.dC 11mer (left) and control 11 mer (right) duplexes dissolved in 20 mM sodium phosphate, 0.2 M NaCl, and 0.2 mM EDTA, pH 7, T_m = 35.1 1.0 C versus 52.7 C 0.9 C, respectively.

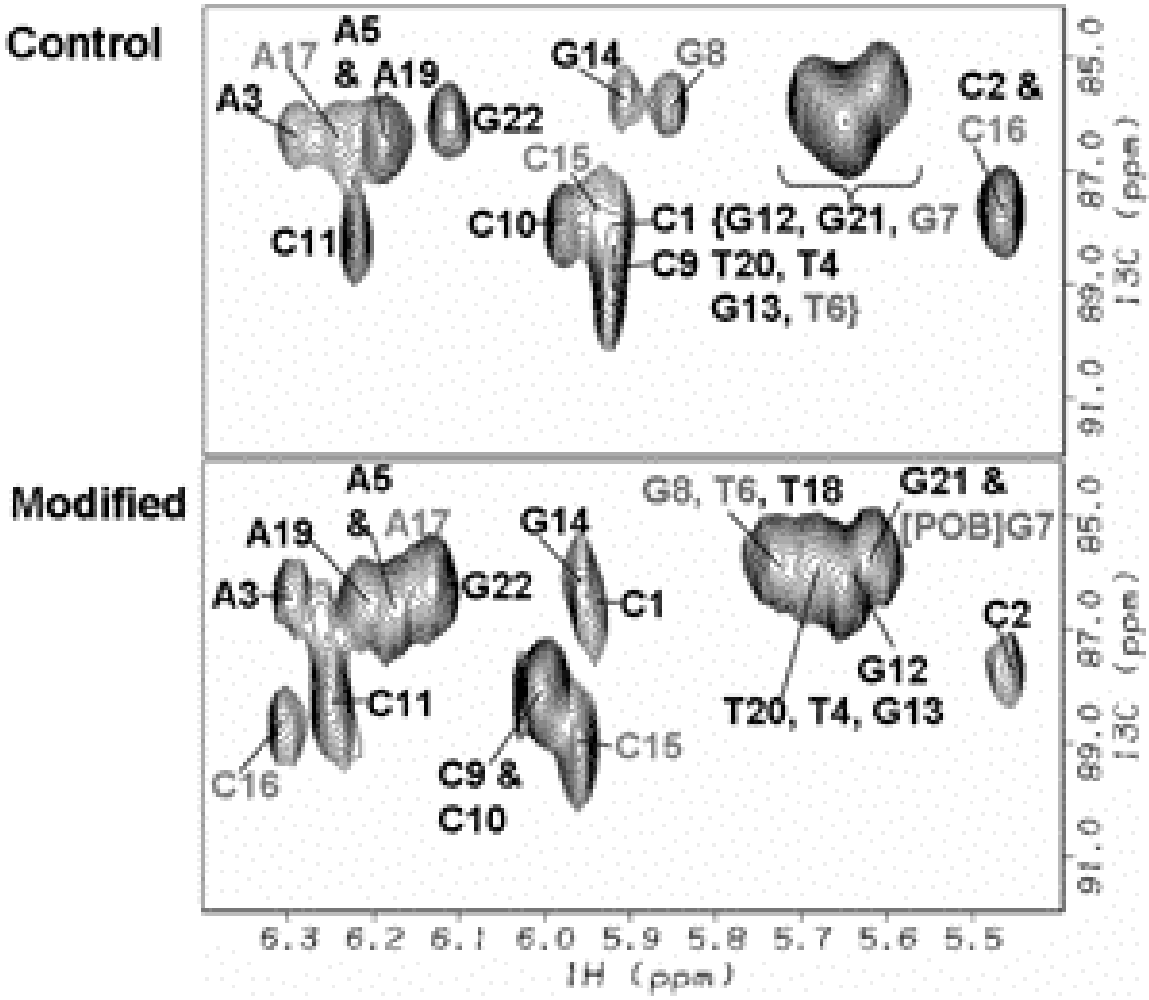

Figure S2. Expanded region of the natural abundance 13C-1H HMQC spectrum of the [POB]dG.dC duplex adduct in D2O buffer at 25 oC showing correlations between the H1' and C1' chemical shifts.

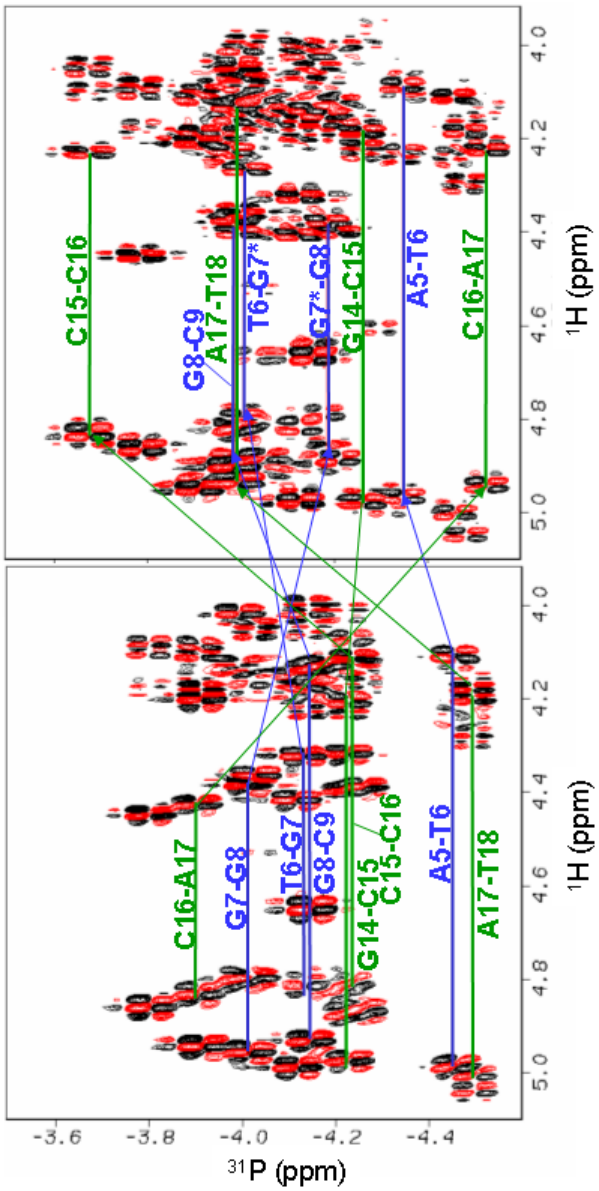

Figure S3: Proton-phosphorus HETCORR spectra for the [POB]dG.dC 11mer (top) and control 11mer (bottom) duplexes in D_2O buffer at 25 ^{o}C. The lines (blue, modified strand; green complimentary strand) designate correlations for the 5mer segment d(A5-T6-G7-G8-C9).d(G14-C15-C16-A17-T18) between the phosphorus and the nth residue's H3' proton to n+1 residue's H4' proton.

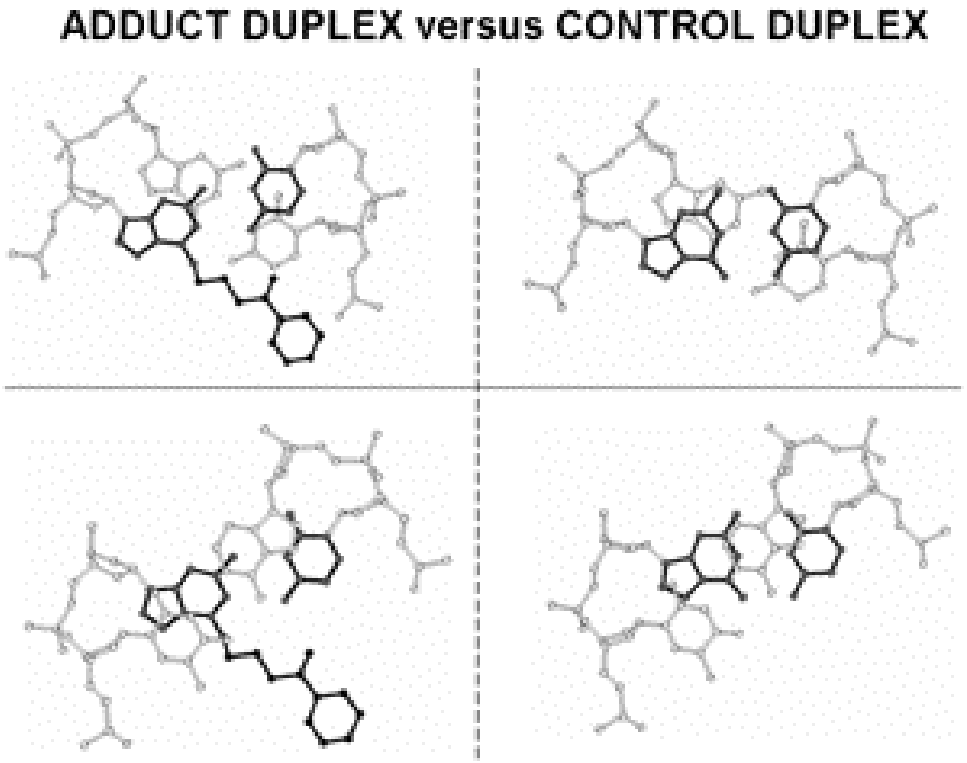

Figure S4: Comparison of the base stacking interactions between the modified (left) and control (right) duplexes at the d([POB]G7-G8).d(C15-C16) (top) and d(T6-[POB]dG7).d(C16-A17) (bottom) steps. The modified base pair and POB ligand are shown in black.

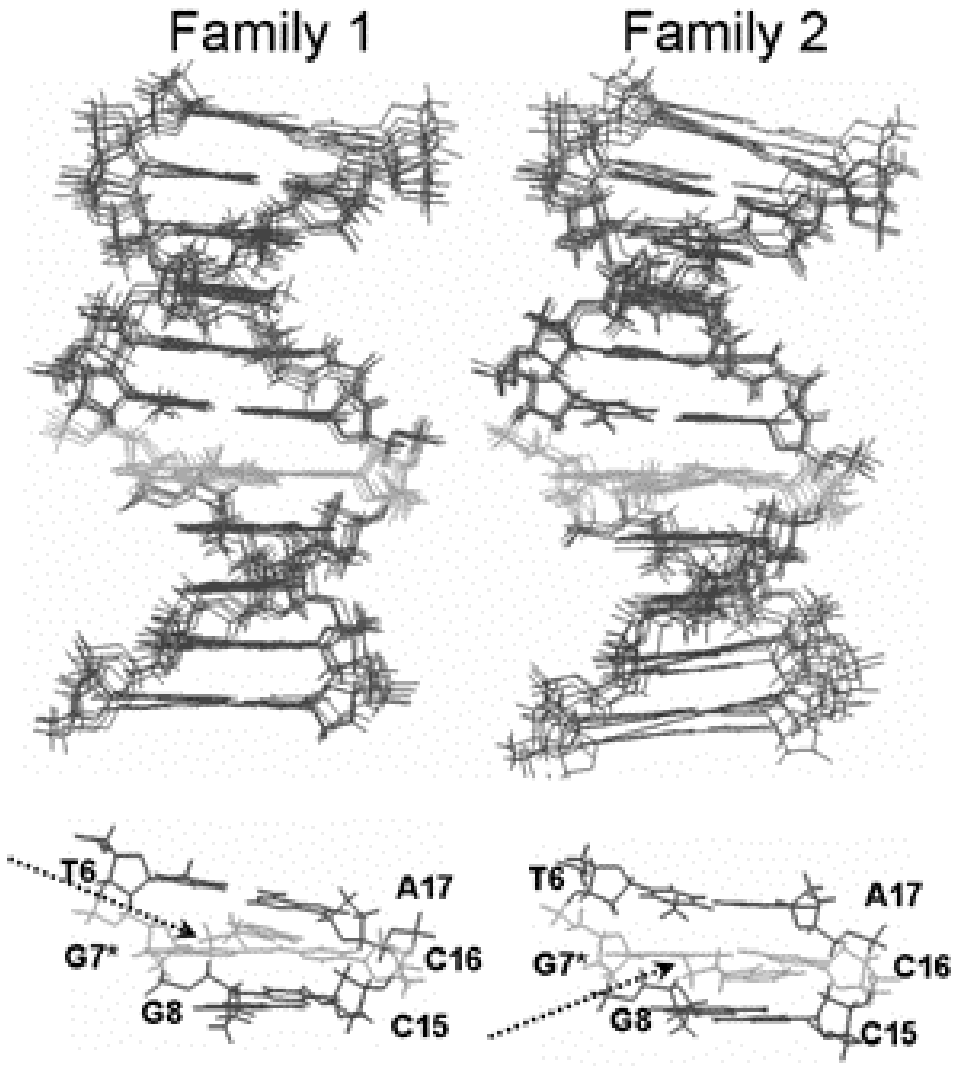

Figure S5: Family 1 is comprised of 5 structures having the lowest energy (-488.73 ± 3.84 kcal/mol), best goodness of fit to the NMR data and pairwise rmsd value of 0.68 ± 0.18. In these structures, The POB ligand is slightly 5' of the modified G, as designated by the dotted arrow. Family 2 is also comprised of 5 structures, but has higher energy (-480.14 ± 11.31 kcal/mol) and pairwise rmsd of 0.75 ± 0.20. The POB ligand is slightly 3' to the modified G, as designated by the dotted arrow.