Structure of the 1,N^2-Ethenodeoxyguanosine Adduct Opposite Cytosine in Duplex DNA: Hoogsteen Base Pairing at pH 5.2[†]

Supporting Information

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[†]This paper is dedicated to the memory of Professor Bea Singer 1922-2005.

Table S1. Non-exchangeable Proton Chemical Shifts[a] of the Unmodified Duplex[b] at 7 °C.

Nucleotide	H8	H6	H1′	H2′	H2″	H3′	H5/H2/CH_3
C^1		7.66	5.72	1.97	2.40	4.69	5.88
G^2	7.97		5.90	2.66	2.74	4.97	
C^3		7.40	5.59	2.11	2.42	4.85	5.43
A^4	8.33		6.21	2.64	2.89	5.00	7.61
T^5		7.03	5.58	1.76	2.15	4.77	1.39
G^6	7.74		5.38	2.56	2.60	4.91	
G^7	7.71		5.47	2.54	2.68	4.97	
A^8	8.07		5.95	2.66	2.89	5.02	7.18
A^9	8.09		6.12	2.50	2.89	4.95	7.65
T^{10}		7.10	5.88	2.00	2.46	4.83	1.23
C^{11}		7.55	6.02	2.17	2.46	4.81	5.59
C^{12}		7.64	6.21	2.25	2.25	4.53	5.71
G^{13}	7.81		5.60	2.48	2.66	4.79	
G^{14}	7.85		5.62	2.68	2.77	5.00	
A^{15}	8.25		6.29	2.70	2.97	5.02	7.83
T^{16}		7.20	5.96	2.01	2.58	4.83	1.31
T^{17}		7.39	6.10	2.11	2.58	4.87	1.52
C^{18}		7.51	5.92	2.09	2.44	4.83	5.55
C^{19}		7.49	5.26	2.11	2.36	4.83	5.57
A^{20}	8.35		6.23	2.70	2.93	5.00	7.64
T^{21}		7.11	5.66	1.95	2.33	4.81	1.44
G^{22}	7.85		5.78	2.60	2.62	4.79	

C^{23}		7.33	5.73	1.98	2.33	4.79	5.38
G^{24}	7.92		6.11	2.32	2.32	4.65	

[a] Values in parts per million. [b] 10 mM NaH_2PO_4, 100 mM NaCl, and 5 μM Na_2EDTA (pH 5.2).

Table S2. Non-exchangeable Proton Chemical Shifts[a] of the 1,N^2-εdG-modified Duplex[b] at 7 °C.

Nucleotide	H8/H2[c]	H6	H1′	H2′	H2′′	H3′	H5/H2/CH_3
C^1		7.65	5.73	1.97	2.40	4.69	5.88
G^2	7.97		5.89	2.66	2.74	4.97	
C^3		7.39	5.57	2.07	2.40	4.83	5.41
A^4	8.27		6.12	2.54	2.70	4.95	7.57
T^5		6.55	5.63	d	2.42	4.71	1.29
X^6	7.13		5.71	2.04	d	4.97	
G^7	7.80		d	2.64	2.64	5.12	
A^8	8.05		5.94	2.64	2.85	5.02	7.17
A^9	8.10		6.14	2.52	2.87	4.97	7.65
T^{10}		7.11	5.86	2.00	2.46	4.81	1.25
C^{11}		7.56	6.02	2.17	2.44	4.81	5.59
C^{12}		7.66	6.20	2.25	2.25	4.53	5.73
G^{13}	7.80		5.59	2.48	2.64	4.79	
G^{14}	7.84		5.59	2.68	2.77	5.00	
A^{15}	8.24		6.27	2.70	2.95	5.02	7.83
T^{16}		7.20	5.94	2.01	2.56	4.83	1.29
T^{17}		7.40	6.08	2.17	2.56	4.87	1.50
C^{18}		7.65	6.00	2.25	2.46	4.85	5.67
C^{19}		7.56	5.59	1.52	2.33	4.81	5.81
A^{20}	8.54		6.22	2.79	2.93	5.00	7.78
T^{21}		7.14	5.69	1.95	2.34	4.81	1.43
G^{22}	7.85		5.79	2.60	2.62	4.79	

C^{23}		7.33	5.71	1.89	2.33	4.79	5.38
G^{24}	7.92		6.12	2.31	2.31	4.65	

[a] Values in parts per million. [b]10 mM NaH_2PO_4, 100 mM NaCl, and 5 μM Na_2EDTA (pH 5.2). [c]Imidazole proton of X^6. Etheno protons H7 and H6 observed at 7.33 ppm. [d]not detected

Table S3. Exchangeable Proton Chemical Shifts[a] of the 1,N^2-εdG-modified duplex[b] at 7 °C (pH 5.2).

Base Pair	C(N^4H)$_{nhb}$	C(N^4H)$_{hb}$	G(N1H)	T(N3H)
C^1•G^{24}	7.16	8.14		
G^2•C^{23}	6.61	8.41	13.03	
C^3•G^{22}	6.51	8.31	12.61	
A^4•T^{21}				13.54
T^5•A^{20}				13.34
X^6•C^{19}	8.95	10.22		
G^7•C^{18}	7.19	8.14	12.34	
A^8•T^{17}				13.61
A^9•T^{16}				13.73
T^{10}•A^{15}				13.56
C^{11}•G^{14}	6.85	8.36	12.76	
C^{12}•G^{13}	7.24	8.31		

[a] Values in parts per million. [b] 10 mM NaH_2PO_4, 100 mM NaCl, and 5 μM Na_2EDTA (pH 5.2). nh and nhb refer to the hydrogen-bonded and non-hydrogen bonded cytidine amino protons, respectively.

Table S4. Exchangeable Proton Chemical Shifts[a] of the Unmodified Duplex[b] at 7 °C (pH 5.2).

Base Pair	$C(N^4H)_{nhb}$	$C(N^4H)_{hb}$	G(N1H)	T(N3H)
$C^1 \bullet G^{24}$	7.14	8.15		
$G^2 \bullet C^{23}$	6.63	8.39	13.04	
$C^3 \bullet G^{22}$	6.51	8.35	12.65	
$A^4 \bullet T^{21}$				13.53
$T^5 \bullet A^{20}$				13.61
$G^6 \bullet C^{19}$	6.82	8.47	12.69	
$G^7 \bullet C^{18}$	6.67	8.23	12.51	
$A^8 \bullet T^{17}$				13.80
$A^9 \bullet T^{16}$				13.76
$T^{10} \bullet A^{15}$				13.61
$C^{11} \bullet G^{14}$	6.86	8.39	12.79	
$C^{12} \bullet G^{13}$	7.21	8.33		

[a] Values in parts per million. [b] 10 mM NaH_2PO_4, 100 mM NaCl, and 5 μM Na_2EDTA (pH 5.2). nh and nhb refer to the hydrogen-bonded and non-hydrogen bonded cytidine amino protons, respectively.

Figure S1. ^{1}H NOESY spectrum showing cross peaks establishing connectivty between the base protons and the deoxyribose H2′ and H2′′ protons. Labeled peaks A, T^{5} H6→A^{4} H2′ (weak); B, T^{5} H6→A^{4} H2′′ (weak); C, T^{5} H6→T^{5} H2′′; D, X^{6} H2→X^{6} H2′ (weak, overlapped); E, X^{6} H2→T^{5} H2′′ (weak, overlapped); F, G^{7} H8→X^{6} H2′ (medium); G, G^{7} H8→X^{6} H2′′(medium); H, C^{18} H6→C^{18} H2′ (overlapped); I, C^{18} H6→C^{18} H2′′ (overlapped); J, C^{19} H6→C^{18} H2′ (very weak); K, C^{19} H6→C^{18} H2′′ (overlapped); L, C^{19} H6→C^{19} H2′; M, C^{19} H6→C^{19} H2′′ (medium); N, A^{20} H8→C^{19} H2′; O, A^{20} H8→C^{19} H2′′. The spectrum was recorded at 7 °C at 250 ms mixing time (pH 5.2).

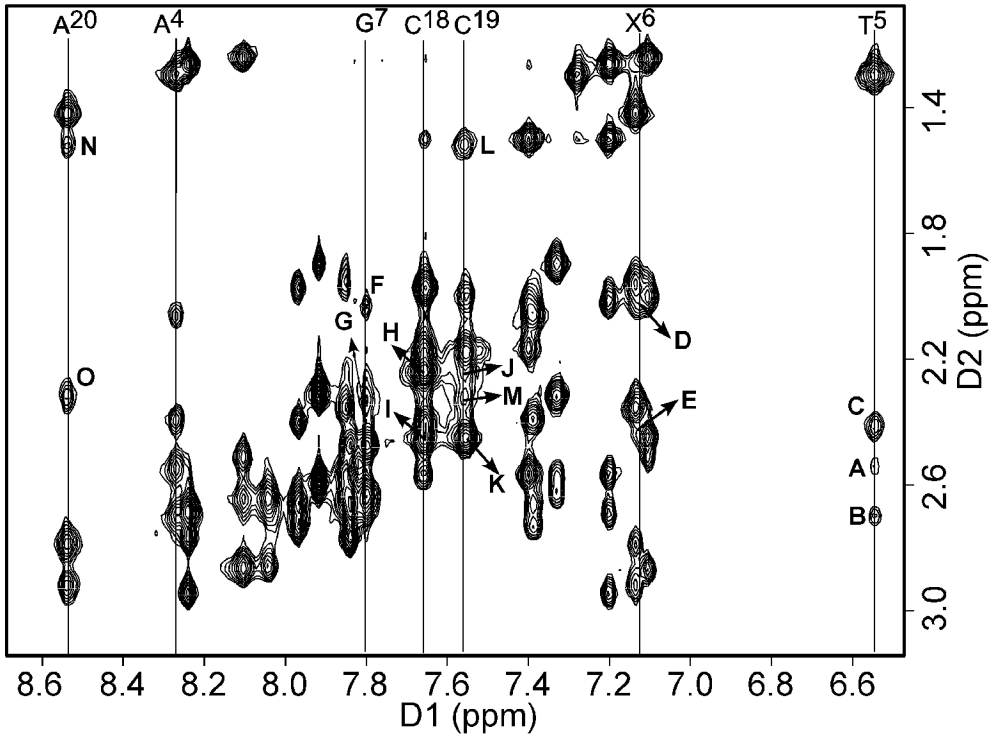
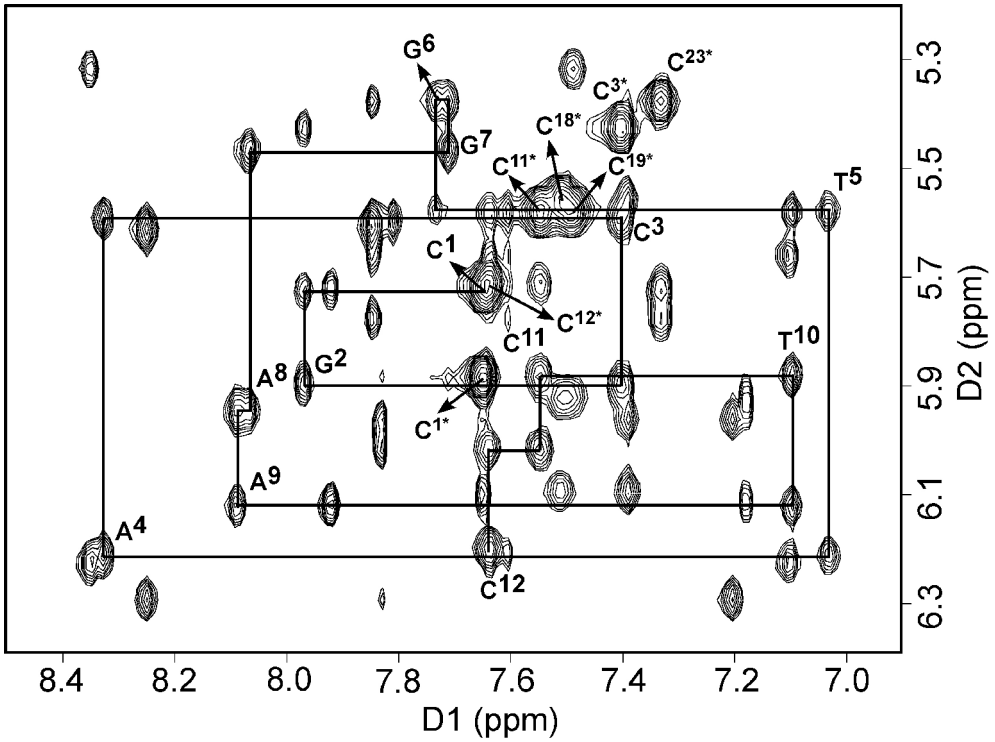


Figure S2. ^{1}H NOESY spectrum showing sequential NOE connectivty from anomeric H1′ to 3′-neighbor aromatic protons for the unmodified oligodeoxynucleotide duplex. The spectra were recorded at 7 °C, at 250 ms mixing time (pH 5.2). **A.** Nucleotides $C^{1}\rightarrow C^{12}$. **B.** Nucleotides $G^{13}\rightarrow G^{24}$. Asterisks indicate cytosine H5-H6 cross peaks.


G6
C23*
C3*
C18*
G7
C11*
C19*
T5
C3
C1
C12*
C11
T10
A8
G2
C1*
A9
A4
C12
8.4
8.2
8.0
7.8
7.6
7.4
7.2
7.0
D1 (ppm)
5.3
5.5
5.7
5.9
6.1
6.3
D2 (ppm)

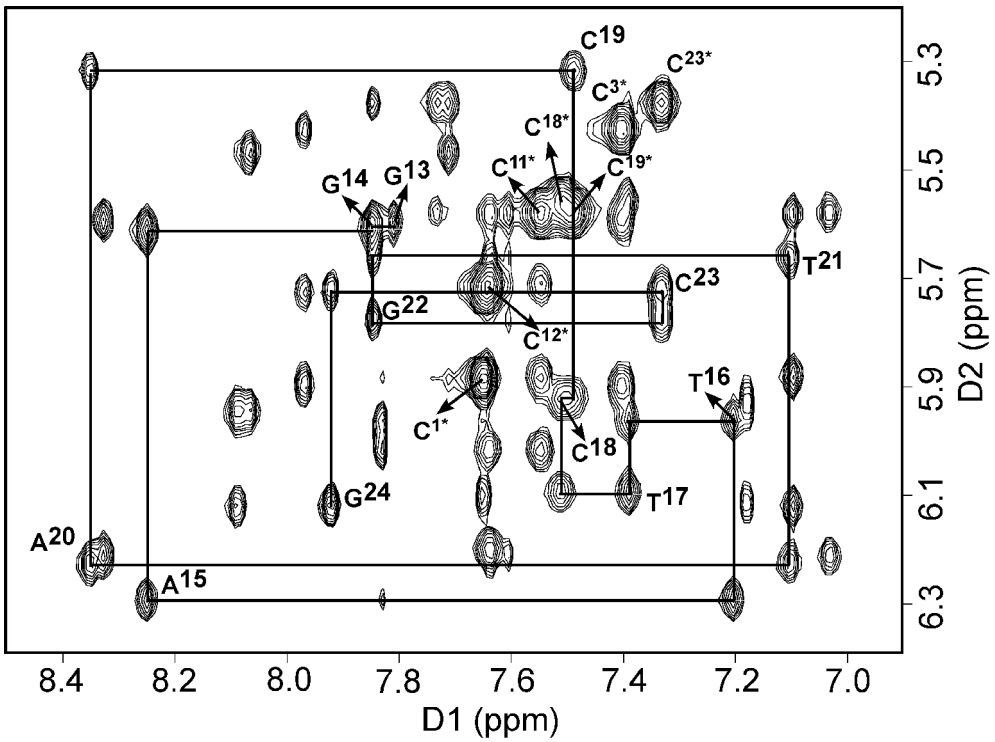

C19
C23*
C3*
C18*
G14
G13
C11*
C19*
T21
C23
G22
C12*
T16
C1*
C18
T17
G24
A20
A15
8.4
8.2
8.0
7.8
7.6
7.4
7.2
7.0
D1 (ppm)
5.3
5.5
5.7
5.9
6.1
6.3
D2 (ppm)

Figure S3. NOESY data (processed with lower threshold value than in Figure 3) showing cross peaks between the C^{19} hydrogen bonded amino proton and flanking T^5 imino proton (peak a), and between C^{19} non-hydrogen bonded amino proton and flanking T^5 imino proton (peak b). The NOESY spectrum was recorded in 10 mM phosphate buffer (9:1 H_2O:D_2O) at 7 °C (pH 5.2). Mixing time was 250 ms.

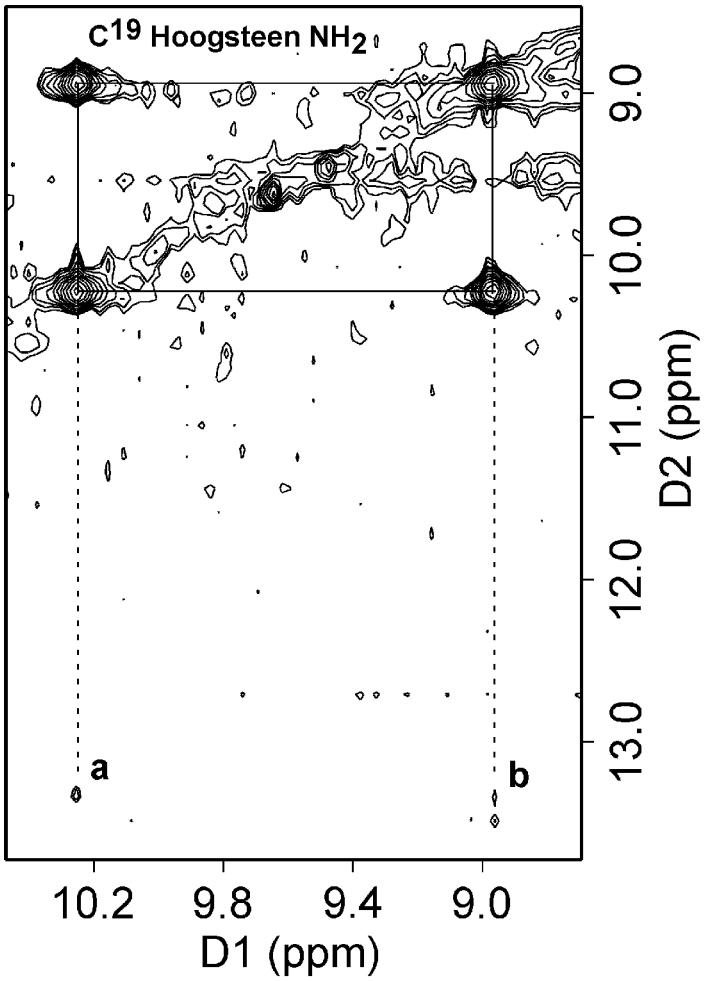


Figure S4 Chemical shift differences observed for 1,N^2-εdG-modified duplex as compared to the corresponding unmodified duplex at pH 5.2. $\Delta\delta=[\delta_{\text{unmodified oligodeoxynucleotide}} - \delta_{\text{modified oligodeoxynucleotide}}]$ (ppm).