

Supplementary Material

Disturbance of the conformation of DNA hairpin containing the 5-GT-3' binding site caused by Zn(II)bleomycin-A5 studied through NMR spectroscopy.

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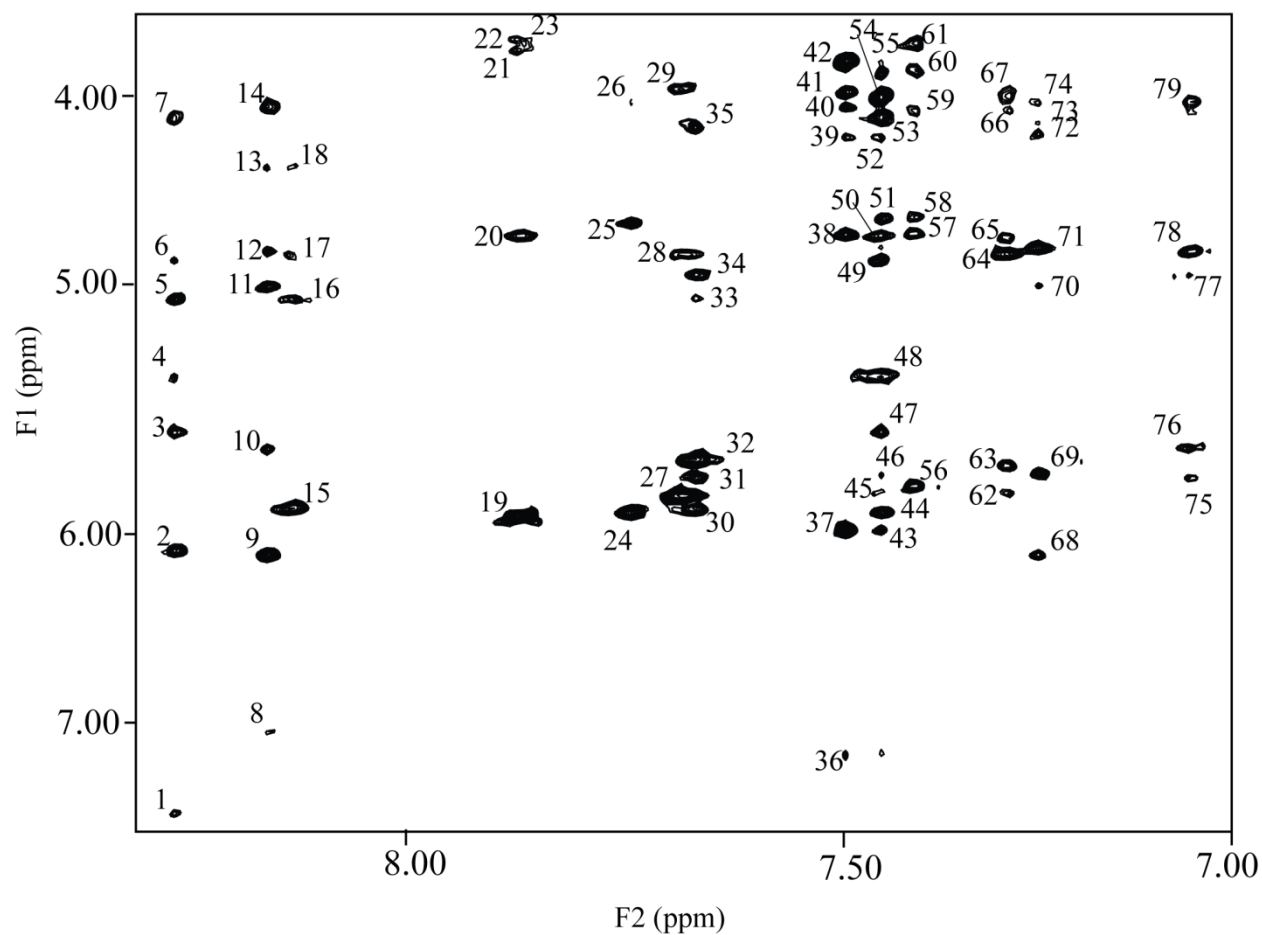


Figure S1. NOESY spectrum of free OL₂ in D₂O collected at 25 °C. This region displays the NOE connectivities between the base protons and the sugar region of the DNA backbone of free OL₂. Assignments are provided in Table S2.

Table S1

NOESY connectivities displayed by OL₂ in H₂O at 5 °C. Missing NOE (*) and new NOE (+) connections upon Zn(II)BLM binding.

Peak Label	F2 (ppm)	F1 (ppm)	Assignment	OL ₂ -Zn(II)PEP	OL ₂ -Zn(II)BLM-A ₂	OL ₂ -Zn(II)BLM-A ₅
Labels Figure 3A						
1	8.36	5.39	C2NH _{2b} -C2C5H			
2	8.36	5.63	C2NH _{2b} -C1C5H			* ^a
3	8.36	6.15	C2NH _{2b} -A3NH _{2a}	*		*
4	8.35	6.72	C2NH _{2b,a}			
5	8.35	7.27	C2NH _{2b} -C2C6H	*	*	*
6	8.35	7.61	C2NH _{2b} -A3NH _{2b}			*
7	8.06	6.44	A12C8H-C13NH _{2a}	*	*	*
8	8.06	7.78	A12C8H-C13NH _{2b}	*	*	*
9	7.79	4.95	C13NH _{2b} -C13C5H	*		
10	7.79	6.14	C13NH _{2b} - A12/A3NH _{2a}	*	*	*
11	7.79	6.44	C13NH _{2b,a}			
12	7.79	7.09	C13NH _{2b} -C13C6H	*	*	*
13	7.79	7.38	C13NH _{2b} -A12/A3C2H	*	*	*
14	7.79	7.60	C13NH _{2b} - A12/A3NH _{2b}	*	*	*
15	7.63	5.64	C1NH _{2b} -C1C5H			*
16	7.63	6.75	C1NH _{2b,a}			
17	7.60	6.14	A3/A12NH _{2b,a}			
18	7.60	6.45	A3/A12NH _{2b} - C13NH _{2a}			*
19	7.46	6.74	C1C6H-C1NH _{2a}			
20	7.38	4.98	A12/A3C2H-C13C5H	*	*	*
21	7.38	6.13	A12/A3C2H- A12/A3NH _{2a}	*	*	*
22	7.38	6.46	A12/A3C2H-C13NH _{2a}	*	*	*
23	7.28	6.31	A6NH _{2b,a}	*	*	*
24	7.28	6.73	C2C6H-C2NH _{2a}	*	*	*
25	7.11	6.44	C13C6H-C13NH _{2a}	*	*	*
26	6.72	5.39	C2NH _{2a} -C2C5H	*		
27	6.75	5.64	C1NH _{2a} -C1C5H			
28	6.44	4.94	C13NH _{2a} -C13C5H			
29	6.44	6.14	C13NH _{2a} -A12/A3NH _{2a}	*	*	*
Labels Figure 3B						
30	13.83	6.13	T14NH-A3NH _{2a}	*	*	
31	13.84	6.45	T14NH-C13NH _{2a}	*	*	*
32	13.83	6.72	T14NH-C2NH _{2a}	*	*	*
33	13.83	7.38	T14NH-A3C2H			
34	13.83	7.60	T14NH-A3NH _{2b}			*
35	13.83	7.78	T14NH-C13NH _{2b}	*	*	*
36	13.83	8.35	T14NH-C2NH _{2b}	*	*	*
37	13.75	6.32	T11NH-A6NH _{2a}	*	*	*
38	13.75	7.33	T11NH-A6NH _{2b}	*	*	*
39	13.25	6.08	T5NH-A12NH _{2a}	*	*	*

Table S1 (Cont.)

Peak Label	F2 (ppm)	F1 (ppm)	Assignment	OL ₂ - Zn(II)PEP	OL ₂ - Zn(II)BLM-A ₂	OL ₂ - Zn(II)BLM-A ₅
40	13.25	6.30	T5NH-A6NH _{2a}	*	*	*
41	13.25	7.37	T5NH-A12C2H			
42	13.25	7.57	T5NH-A12NH _{2b}	*	*	*
43	13.25	7.78	T5NH-C13NH _{2b}	*	*	*
44	12.75	5.39	G15NH-C2C5H	*	*	*
45	12.76	5.64	G15NH-C1C5H	*	*	*
46	12.75	6.72	G15NH-C2NH _{2a}	*	*	
47	12.75	7.38	G15NH-A3C2H			
48	12.75	8.35	G15NH-C2NH _{2b}			
49	12.48	4.95	G4NH-C13C5H	*	*	*
50	12.48	5.61	G16NH-C1C5H	*	*	*
51	12.48	6.14	G4NH-A12/A3NH _{2a}	*	*	*
52	12.48	6.44	G4NH-C13NH _{2a}			
53	12.48	6.77	G16NH-C1NH _{2a}	*	*	*
54	12.48	7.37	G4NH-A12/A3C2H			
55	12.48	7.59	G4NH-A12/A3NH _{2b}	*	*	*
56	12.48	7.79	G4NH-C13NH _{2b}	*	*	
Labels Figure 3C						
57	13.83	12.48	T14NH-G4NH	*	*	*
58	13.83	12.75	T14NH-G15NH	*	*	*
59	13.25	12.48	T5NH-G4NH	*	*	*
Labels Fig. 3D						
60	13.83	1.32	T14NH-T14CH ₃	*	*	*
61	13.76	1.50	T11NH-T11CH ₃	*	*	*
62	13.25	1.11	T5NH-T5CH ₃	*	*	*
Labels Figure 4						
63 ^c	8.29	5.06	C2NH _{2b} -C13C5H	+ ^b		
64'	8.29	6.39	C2NH _{2b} -C13NH _{2a}	+	+	
65'	8.06	7.39	A12C8H-A12/A3C2H		+	
66'	8.06	7.57	A12C8H-A12/A3		+	
			NH _{2b}			
67'	7.37	6.31	A12/A3C2H-Mann		+	
			NH _{2b}			
68'	7.37	6.63	A12/A3C2H-C2C5H		+	
69'	7.38	6.77	A12/A3C2H-C1 NH _{2a}		+	
70'	7.24	6.40	C2C6H-C13 NH _{2a}		+	
71'	7.19	6.66	C13C6H-C2 NH _{2a}		+	
72'	6.71	5.61	C2 NH _{2a} -C1C5H		+	
(Data not shown)	6.45	2.53	C13NH _{2a} -A12 2'		+	
74'	6.73	6.45	A12NH _{2a} -C13NH _{2a}			+
Labels Figure 5						
75'	13.83	7.33	T11NH-A12/A3C2H	+		
76'	13.81	7.61	T11NH-A12/A3NH _{2b}	+		
77'	8.28	12.57	C2NH _{2b} -G16NH	+		
78'	7.44	12.56	G16C8H-G16 NH	+		

Table S1 (Cont.)

Peak Label	F2 (ppm)	F1 (ppm)	Assignment	OL ₂ -Zn(II)PEP	OL ₂ -Zn(II)BLM-A ₂	OL ₂ -Zn(II)BLM-A ₅
79'	6.28	12.75	Mann NH _{2b} -G15NH		+	
(Data not shown)	12.50	12.72	G16NH-G15NH		+	

^aAsterisks (*) indicate that the specific NOE is missing upon complexation with the specified Zn(II)BLM,

^bPlus (+) indicates that the specific NOE connection is found upon complexation with the specified Zn(II)BLM, ^cPrime (') indicates that the NOE is not a native NOE connection for free OL₂.

Table S2Inter-base NOE connections detected in D₂O at 25 °C for free and MBLM-bound OL₂

Assignment	OL ₂ -Zn(II)PEP	OL ₂ -Zn(II)BLM-A ₂	OL ₂ -Zn(II)BLM-A ₅
C2C6H-C1 2'' ^a			+
C2C6H-C1 3'	* ^d	*	
A3C8H-C2 2''	*	*	*
A3C8H-C2 2'		*	*
A3C8H-C2 3'	*	*	*
A3C8H-C2 1'	*	*	
A3C8H-C2C6H ^b	*	*	
A3C8H-G4C8H	*	*	
G4C8H-A3 2''			*
G4C8H-A3 2'	*		*
G4C8H-A3 1'			*
G4C8H-A3 3' ^c	+ ^e	+	
G4C8H-T5 CH ₃			*
G4C8H-T5C6H	*	*	
G4 1'-T5 CH ₃	*		*
G4 2'-T5 CH ₃			*
G4 2''-T5 CH ₃			*
T5C6H-G4 2''			
T5C6H-G4 2'			
T5C6H-G4 3'	*	*	*
T5CH3-G4 3'			*
T5C6H-G4 1'	*	*	*
A6C8H-T5 2''	*		*
A6C8H-T5 2'			
A6C8H-T5 3'			
A6C8H-T5 1'		*	*
A6C8H-T5C6H	*	*	*
A6C8H-T7C6H		*	
A6C8H-T7 CH ₃			*
A6 1'-T7 CH ₃	+	+	+
A6 2''-T7 CH ₃			*
A6 2'-T7 CH ₃	*		*

Table S2 (Cont.)

Assignment	OL ₂ -Zn(II)PEP	OL ₂ -Zn(II)BLM-A ₂	OL ₂ -Zn(II)BLM-A ₅
T7C6H-A6 2''		*	*
T7C6H-A6 2'	*		
T7C6H-A6 3'		*	*
T7C6H-A6 1'			
T7 CH ₃ -A6 3'			*
T7C6H-T8 CH ₃	*	*	*
T7C6H-T9 CH ₃		*	*
T7 2'-T8 CH ₃			*
T7 2''-T8 CH ₃			*
T8 CH ₃ -T7 3'		*	*
T8C6H-T8 2'' or T7 2''			*
T8C6H-T8 2' or T7 2'			
T8 1'-T10 CH ₃			*
T8C6H-T9 3'			
T8 2''-T9 CH ₃			*
T8 CH ₃ -T9 3'			*
T8 2' - T9 CH ₃	+		
T9C6H-T7 1'	*		
T9 2'-T10 CH ₃		*	*
T9 2''-T10 CH ₃	+	+	
T10C6H-T9 2' or T10 2''			*
T10C6H-T9 2''	+		*
T10C6H-T9 2'	+		*
T10 2'-T11 CH ₃			*
T10 2''-T11 CH ₃	*	*	*
T10C6H-T11 CH ₃		+	
T11C6H-T10 2'			*
T11C6H-T10 3'		*	
T11C6H-T10 1'		*	*
T10 1'-T11 CH ₃		*	*
T11C6H-T10 2''	+	+	
A12C8H-T11C6H	*	*	
A12C8H-T11 2''	*		
A12C8H-T11 2'	*		
A12C8H-T11 3'	*	*	*
A12C8H-T11 1'		*	*
A12C8H-C13C6H	*	*	
A12C8H-C13C5H	*		
C13C5H-A12 2''			
C13C5H-A12 2'			
C13C6H-A12 2''	*		
C13C6H-A12 2'			
C13C6H-A12 1'			
C13C6H-T14 CH ₃			+

Table S2 (Cont.)

Assignment	OL ₂ -Zn(II)PEP	OL ₂ -Zn(II)BLM-A ₂	OL ₂ -Zn(II)BLM-A ₅
C13C5H-T14 CH ₃			*
C13 2''-T14 CH ₃			*
C13 2'-T14 CH ₃			*
C13C6H-T14 3'	+		
T14C6H-C13 3'	*	*	*
T14C6H-C13 1'	*	*	*
T14C6H-C13 2'			
T14 CH ₃ -C13 3'			*
T14C6H-C13 2''	+	+	
G15C8H-T14C6H		*	
G15C8H-T14 2''			*
G15C8H-T14 2'			*
G15C8H-T14 3'		*	
G16CH8-G15 2''			*
G16C8H-G15 3'	*		
G16C8H-G15 1'			
G16C8H-G15 2'	+	+	

^{a,b}Blue and green colored labels indicate base-sugar and base-base NOEs, respectively. ^cRed colored labels indicated that the NOE is only detected when the specified Zn(II)BLM is bound. ^dAsterisks (*) indicate that the specific NOE is missing upon complexation with the specified Zn(II)BLM, ^ePlus (+) indicates that the specific NOE connection is found upon complexation with the specified Zn(II)BLM. The color coding of the NOE assignments corresponds to the color coding for Figures 9 and 11.