Supplementary Material (ESI) for Chemical Communications

Optimized DNA targeting using *N*,*N*-bis(2-pyridylmethyl)-β-alanyl 2'-amino-LNA

B. Ravindra Babu, Patrick J. Hrdlicka, Christine J. McKenzie and Jesper Wengel*

Nucleic Acid Center and Department of Chemistry, Univerisity of Southern Denmark, DK-5230 Odense M, Denmark

Electronic supplementary information (ESI)

Table S1 13-mer ONs synthesized and thermal denaturation studies at different concentrations of divalent metal ions[a]

		T_m (ΔT_m)/°C						
		+ EDTA	Ni^{2+}		Cu^{2+}		Zn^{2+}	
			1 equiv.[b]	excess[c]	1 equiv.[b]	excess[c]	1 equiv.[b]	excess[c]
DNA DNA	5'-CGT GAT ATA TAA A 3'-GCA CTA TAT ATT T	31 (ref)	31	31	31	31	31	31
ON6 DNA	5'-CGT GA**X** ATA **X**AA A 3'-GCA CTA TAT ATT T	37 (+6)	47 (+16)	48 (+17)	40 (+9)	40 (+9)	45 (+14)	45 (+14)
DNA **ON7**	5'-CGT GAT ATA TAA A 3'-GCA C**X**A TA**X** ATT T	38 (+7)	48 (+17)	49 (+18)	40 (+9)	40 (+9)	46 (+15)	46 (+15)
DNA **ON8**	5'-CGT GAT ATA TAA A 3'-GCA CTA **X**AT A**X**T T	39 (+8)	47 (+16)	48 (+17)	40 (+9)	40 (+9)	44 (+13)	44 (+13)
ON6 **ON7**	5'-CGT GA**X** ATA **X**AA A 3'-GCA C**X**A TA**X** ATT T	42 (+11)	54 (+23)	29 (-2)	52 (+21)	19 (-2)	44 (+13)	35 (+4)
ON6 **ON8**	5'-CGT GA**X** ATA **X**AA A 3'-GCA CTA **X**AT A**X**T T	49 (+18)	59 (+28)	63 (+32)	51 (+20)	48 (+17)	55 (+24)	58 (+27)

[a]Thermal denaturation temperatures [T_m values/°C (ΔT_m = change in T_m value calculated relative to the reference DNA:DNA duplex) measured as the maximum of the first derivative of the melting curve (A_{260} *vs*. Temperature) recorded in medium salt buffer using 1.0 μM concentrations of the two complementary strands; see below Table 1 for other details;[b] 1 equiv. refers to 1 equivalent of metal ion per **X** monomer for experiments with DNA complements, and to 0.5 equivalent of metal ion per **X** monomer for experiments with **ON6**:**ON7** or **ON6**:**ON8**; [c] [M^{2+}] = 10 μM; MALDI-MS *m/z* ([M-H]$^-$; found/calc): **ON6**, 4542/4541; **ON7**, 4477/4483; **ON8**, 4477/4483.

Molecular Modelling Procedure. A standard B-type DNA-DNA duplex was build using the SPARTAN '02 program and subsequently modified within the MacroModel V7.2 suite of programs (R. D. Mohamadi, N. G. J. Richards, W. C. Guida, R. Liskamp, M. Lipton, C. Caufield, C. Chang, T. Hendrickson and W. C. Still, *J. Comput. Chem.*, 1990, **11**, 1301). The charge from the phosphordiester backbone was neutralized with Na^+-ions, which were placed approximately 3 Å from the negatively charged oxygens. Zn^{2+}-ions were initially placed approximately 3 Å from the nitrogen atoms of the metal chelator. All atoms were frozen except those of the sugar moiety, linker

and metal chelator part of monomer **X** and Zn^{2+}-ions. The duplexes were minimized using the Polak-Ribiere conjugate gradient method and static Merck Mechanical Force Field (T. A. Halgreen, *J. Comput. Chem.*, 1990, **11**, 1301) as implemented in MacroModel V7.2. A dielectric constant of 80 relative to vacuum was applied. Non-bonded interactions were treated with extended cut-offs (van der Waals 8.0 Å, electrostatic 20.0 Å). The minimized structure was then, using the same constraints as described above, submitted to 2 ns of stochastic dynamics (300 K, timestep of 2 fs, SHAKE all H), during which 100 structures were sampled and minimized.

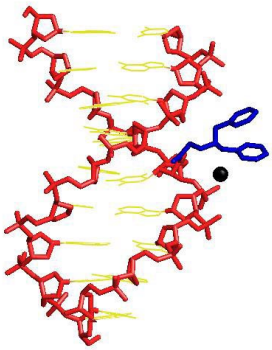

Fig. S1 Low energy structure of **ON1**:DNA including one Zn^{2+}-ion. For clarity hydrogens, sodium ions and bond orders have been omitted. Colouring scheme: nucleobases, yellow; sugar-phosphate backbone, red; *N*,*N*-bis(2-pyridylmethyl)-β-alanyl ligand, blue ; Zn^{2+}-ion, black.

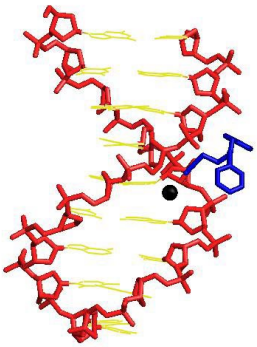

Fig. S2 Lowest energy structure of **ON1**:DNA including one Zn^{2+}-ion. Colouring scheme as in Fig S1.

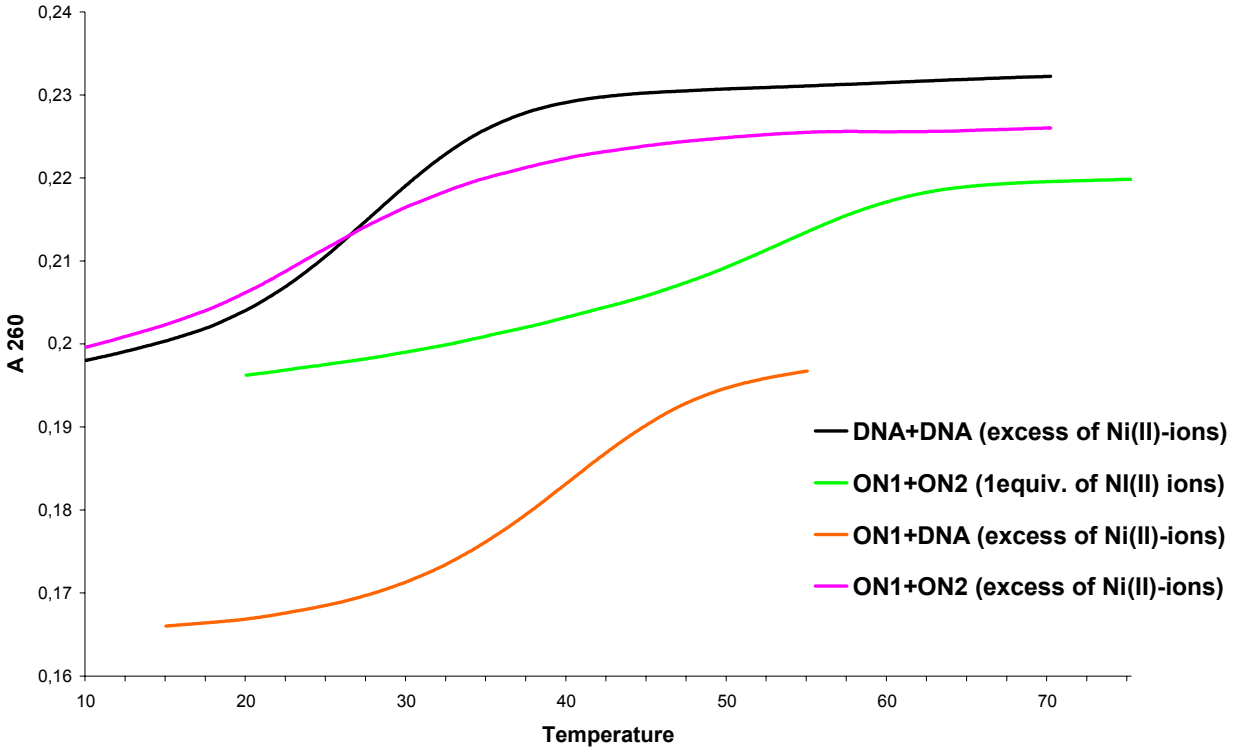

Fig. S3 Thermal denaturation curves measured at 260 nm. See Table 1 for sequence key and other details.