

Electronic Supplementary Information

Double-headed nucleotides as xeno nucleic acids: information storage and polymerase recognition

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Circular dichroism spectroscopy

CD spectra were recorded at 20 °C on a Jasco J-715 spectropolarimeter as an average of 5 scans from 200–350 nm. Measurements were performed using a split width of 2.0 nm, a scan speed of 50 nm pr. min and in quartz optical cells with an optical path length of 5.0 mm. The samples contained 1.5 μ M concentrations of each strand in a medium salt buffer (2.5 mM Na₂HPO₄, 5 mM NaH₂PO₄, 100 mM NaCl, and 0.1 mM EDTA, pH 7.0) and before the measuring, the oligonucleotides were annealed by heating the samples to 75 °C followed by controlled cooling.

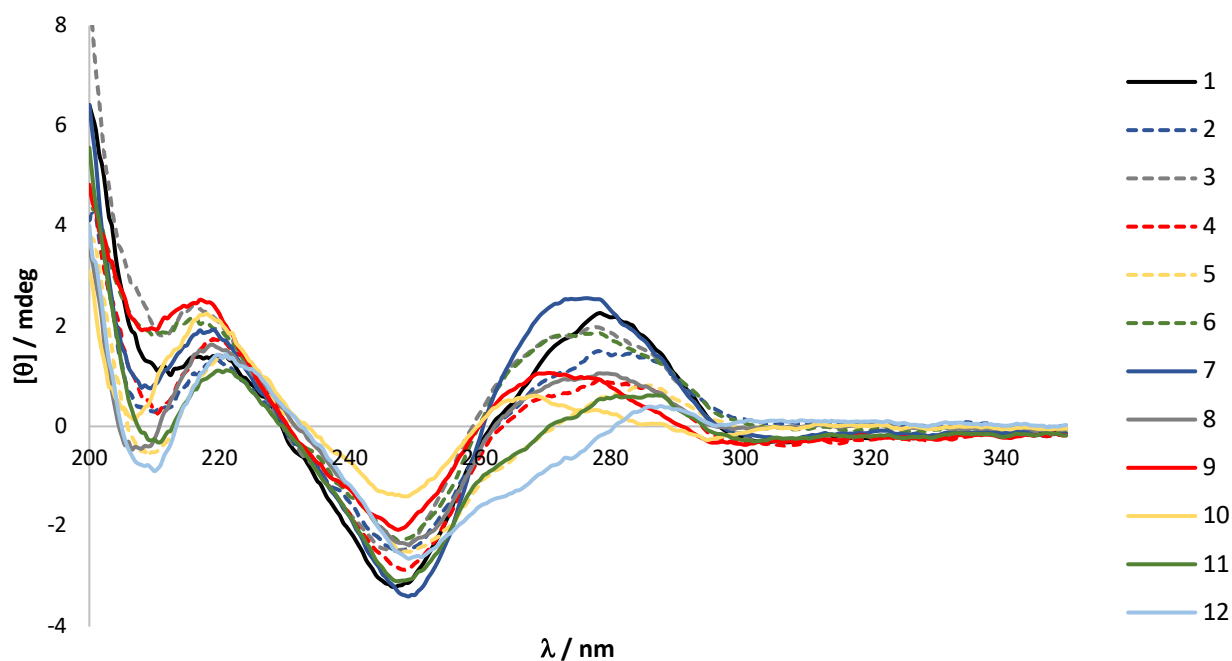


Fig. S1 CD spectra for duplexes 1-12 (entry numbers refer to Fig. 2).

MALDI-TOF MS data for synthesized oligonucleotides

Table S1: MALDI-TOF MS data for synthesized oligonucleotides.

#	Sequence	MW (calcd)	MW (found)
1	5'-d(GGTCA A _T ATGCGA)-3'	3848.6	3848.4
2	5'-d(GGTCA A _T A _T GCGA)-3'	3698.4	3698.6
3	5'-d(TCGCA U _A TTGACC)-3'	3745.5	3746.8
4	5'-d(TCGCAT A _T TGACC)-3'	3759.5	3760.4
5	5'-d(TCGCA U _A U _T GACC)-3'	3581.4	3581.5
6	5'-d(TCG CTG U _A GT AAC CGG TCT TCG CGC G)-3'	7790.1	7794.7

PAGE for primer extension experiments with Therminator DNA polymerase

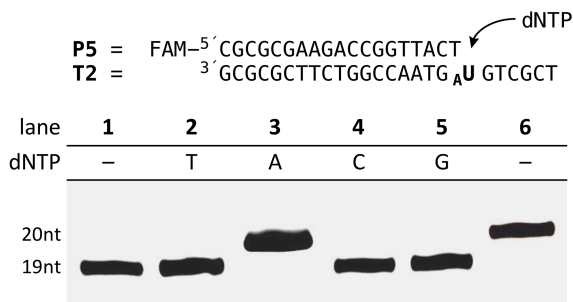


Fig. S2 PAGE analysis of a single nucleotide incorporation study with natural dNTPs and primer **P5** in the presence of template **T2** and Therminator. Lane 1 corresponds to the negative control and lane 6 corresponds to the 20 nt synthetic control **P6**. Conditions: [**P5**] = 0.1 μ M; [**T2**] = 0.1 μ M; [dNTP] = 40 μ M; [Therminator] = 0.02 U/ μ L; temperature = 75 $^{\circ}$ C; reaction time = 10 min.

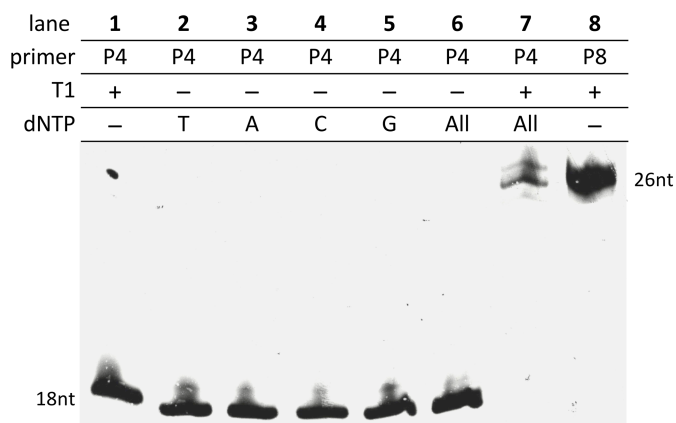


Fig. S3 PAGE analysis of template-free negative controls with natural dNTPs, primer **P4** and Therminator. Lane 1 and 8 correspond to the dNTP-free negative control with primer **P4** and **P8**, respectively. Lane 7 corresponds to positive control. Conditions: [primer] = 0.1 μ M; [**T1**] = 0.1 μ M; [dNTP] = 40 μ M; [Therminator] = 0.02 U/ μ L; temperature = 75 $^{\circ}$ C; reaction time = 15 min.

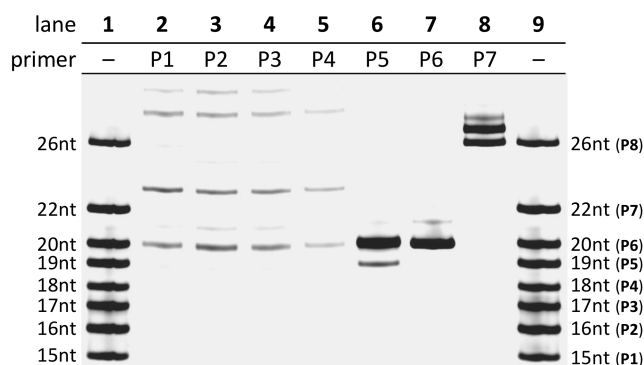


Fig. S4 PAGE analysis of full-length extension reactions with the primers **P1-P7** in the presence of **T2** and Therminator. Both lane 1 and 9 corresponds to the synthetic primers **P1-P7** and the 26 nt synthetic control **P8**. Conditions: [primer] = 0.1 μ M; [**T2**] = 0.1 μ M; [dNTP] = 200 μ M; [Therminator] = 0.02 U/ μ L; temperature = 75 $^{\circ}$ C; reaction time = 60 min.

