Supporting Information

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Experimental part:

Synthesis and analysis of oligonucleotides. Cyanoethyl phosphoramidites from *Transgenomic* (Glasgow, UK) were used for oligonucleotide synthesis. Oligonucleotides **1, 2** were obtained from *Microsynth* (Switzerland) and were used without additional purification. Oligonucleotides **3-6** were prepared via automated oligonucleotide synthesis by a standard synthetic procedure ('trityl-off' mode) on a 394-DNA/RNA synthetizer (Applied Biosystems). Cleavage from the solid support and final deprotection was done by a treatment with 33% aqueous NH_3 at 55°C overnight. All oligonucleotides were purified by reverse phase HPLC (LiChrospher 100 RP-18, 5 μm, Merck, *Bio-Tek instrument Autosampler 560*); eluent A = $(Et_3NH)OAc$ (0.1 M, pH 7.4); eluent B = 80 % MeCN and 20% eluent A; gradient 5-35% B over 20 min at 25°C. ESI-MS (negative mode, CH_3CN/H_2O/TEA) of oligonucleotides was performed with a *Sciex QSTAR pulsar* (hybrid quadrupole time-of-flight mass spectrometer, *Applied Biosystems*).

Thermal denaturation experiments were carried out on a *Varian Cary-100 Bio-UV/VIS* spectrometer equiped with a Varian Cary-block temperature controller and data were collected with Varian WinUV software at 260 nm (cooling-heating-cooling cycles in the temperature range of 10-90°C, temperature gradient of 0.5°C/min). Experiments were carried out for 1.0 μM oligonucleotide concentration (each strand), 10 mM phosphate buffer and 100 mM NaCl at pH 7.4. Data were analyzed with Kaleidagraph software from Synergy software. Melting temperature (Tm) values were determined as the maximum of the first derivative of the smoothed melting curve.

Fluorescence data were collected for 1.0 μM oligonucleotide solutions (1.0 μM of each strand in case of double strands) in phosphate buffer (10 mM) and NaCl (100 mM) at pH 7.4 on a Varian Cary Eclipse fluorescence spectrophotometer equipped with a Varian Cary-block temperature controller (a) pyrene fluorescence: excitation at 354 nm, excitation and emission slit width 5 nm and 5 nm respectively, PMT detector voltage at medium sensitivity, 600 V; b) perylene diimide: excitation at 505 nm, ex. slit 10 nm, em. slit. 10 nm, PMT detector voltage at high sensitivity, 800 V)

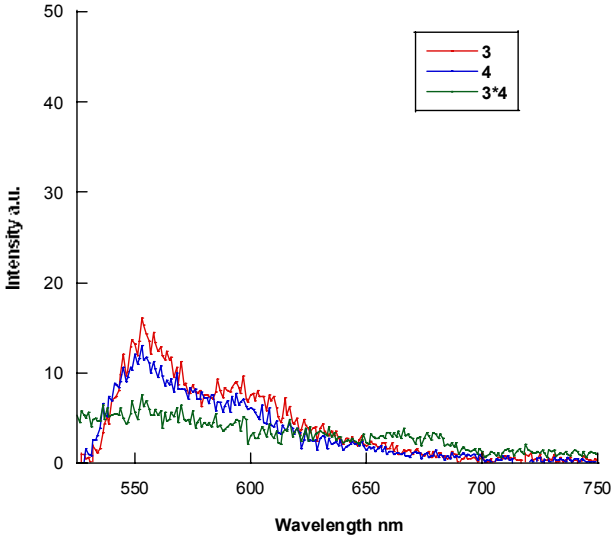
CD spectra were recorded on a JASCO J-715 spectrophotometer using quartz cuvettes with an optical path of 1 cm.

UV-Vis data were collected in the range of 220-700 nm at 30 ºC on *Varian Cary-100 Bio*-UV/VIS spectrophotometer equipped with a *Varian Cary*-block temperature controller. All experiments were carried out for 2.5 μM oligonucleotide concentration (each strand) in phosphate buffer (10 mM) and NaCl (100 mM) at pH=7.4.

Table 1. Mass spectrometry data (molecular formula, calc. average mass and found mass).

Oligo.		*Molecular formula*	*Calc. aver.mass*	*Found*
3	(5') AGC TCG GTC A**E**C GAG AGT GCA	$C_{225}H_{265}N_{83}O_{124}P_{20}$	6735.6	6735
4	(3') TCG AGC CAG T**E**G CTC TCA CGT	$C_{223}H_{267}N_{73}O_{128}P_{20}$	6637.5	6638
5	(5') AGC TCG GTC **EE**C GAG AGT GCA	$C_{245}H_{274}N_{80}O_{127}P_{20}$	6990.9	6992
6	(3') TCG AGC CAG **EE**G CTC TCA CGT	$C_{243}H_{275}N_{73}O_{129}P_{20}$	6901.8	6903

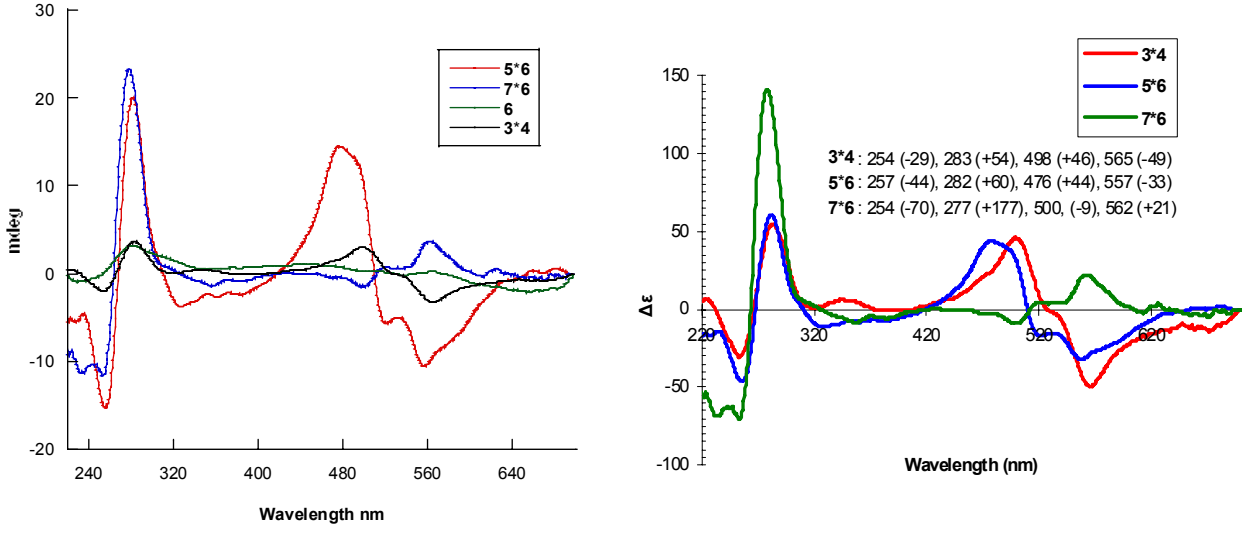
Figure 1. Fluorescence spectra of PDI containing single strands **3**, **4** and hybrid **3*4**



Conditions: 1 μM oligonucleotide concentration (each strand), 10 mM phosphate buffer (pH 7.4) and 100 mM NaCl; excitation at 505 nm.

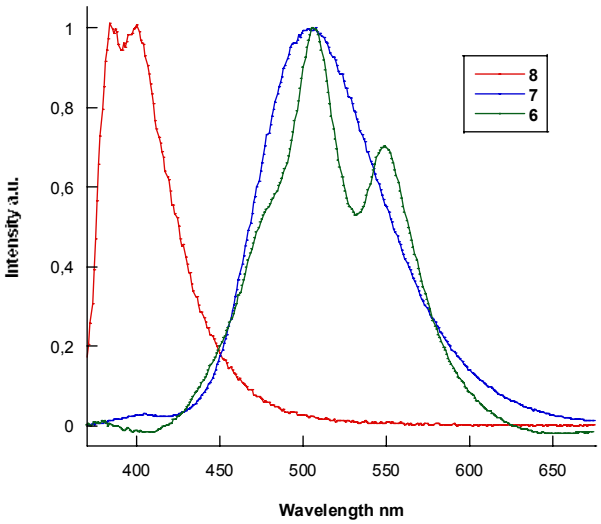
Note: please note, that such low signal was observed with enlarged slits (ex. slit 10; em. slit 10) and high detector sensitivity (800 V), where all other experiments in this work are described for slits (5 and 5) and middle sensitivity, 600 V).

Figure 2 CD spectra of hybrids **3*4**, **5*6**, **7*6** and single strand **6** at 30°C.



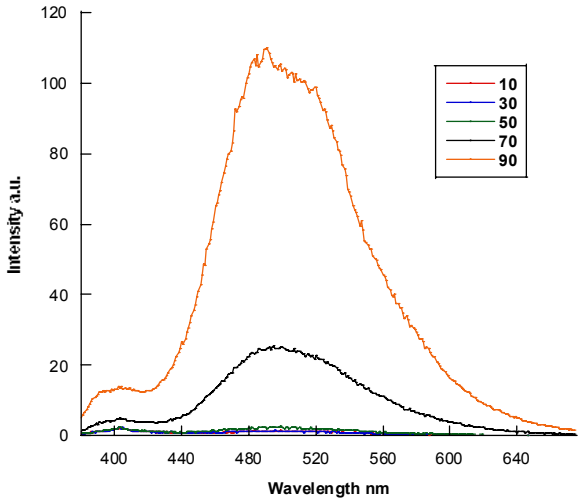
Conditions : a) CD/mdeg: 1 μM (**3*4**) and 2.5 μM (**5*6**, **7*6** and **6**) oligonucleotide concentration (each strand), 10 mM phosphate buffer (pH 7.4) and 100 mM NaCl; b) CD/ Δε , λ (Δε, M^{-1} cm^{-1}).

Figure 3. Normalized absorbance spectra of PDI-containing strand **6** and fluorescence spectra of pyrene monomer and excimer in oligonucleotides **8** and **7**, respectively.



Note: single strand **8** (5') AGC TCG GTC A**S**C GAG AGT GCA was taken as a reference sequence that contains one pyrene unit.

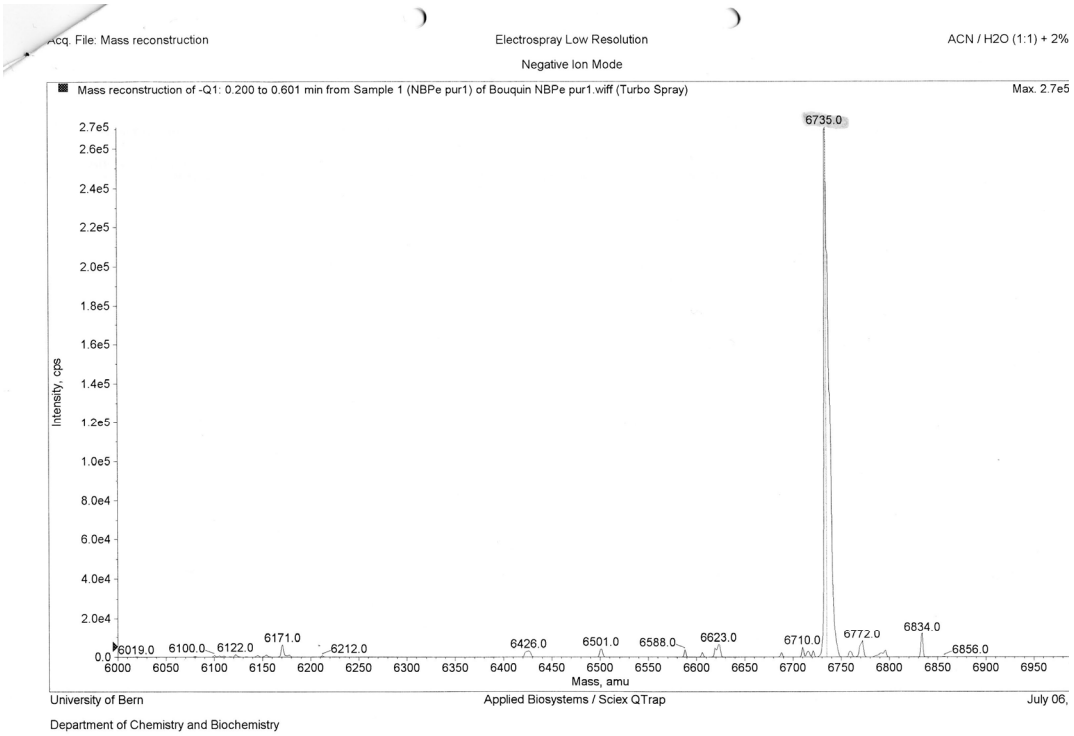
Figure 4. Temperature-dependent fluorescence of hybrid **7*6**.



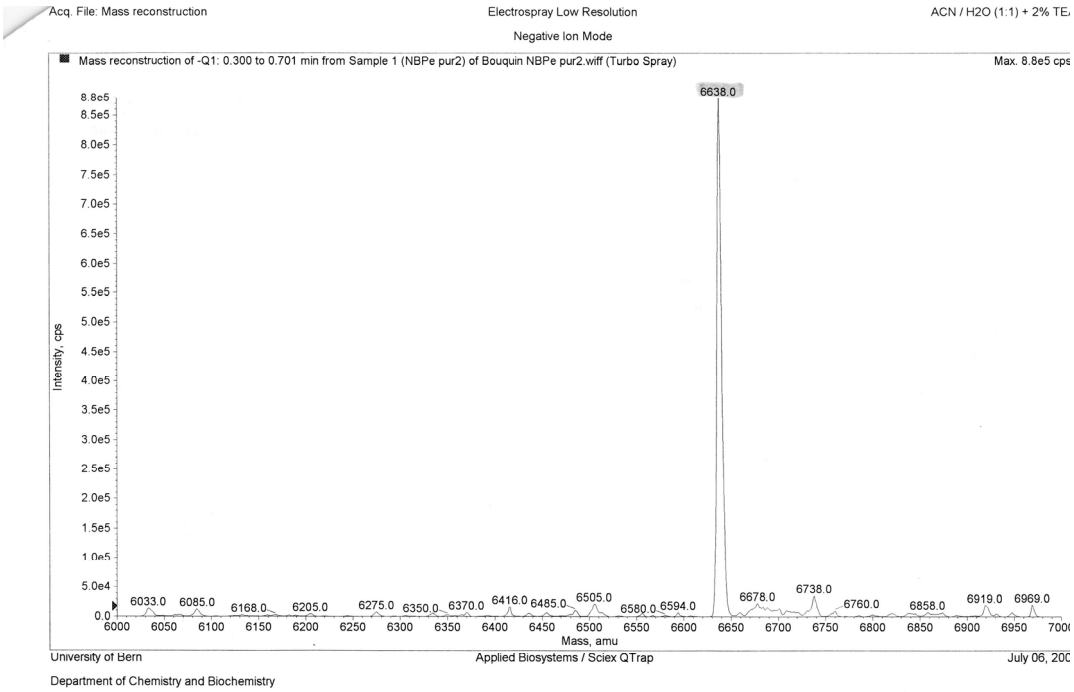
Conditions: 2.5 µM oligonucleotide concentration (each strand), 10 mM phosphate buffer (pH 7.4) and 100 mM NaCl (ex. slit 5 nm, em. slit 5 nm and detector with medium voltage at 600 V).

ESI mass spectra of oligonucleotides 3-6.

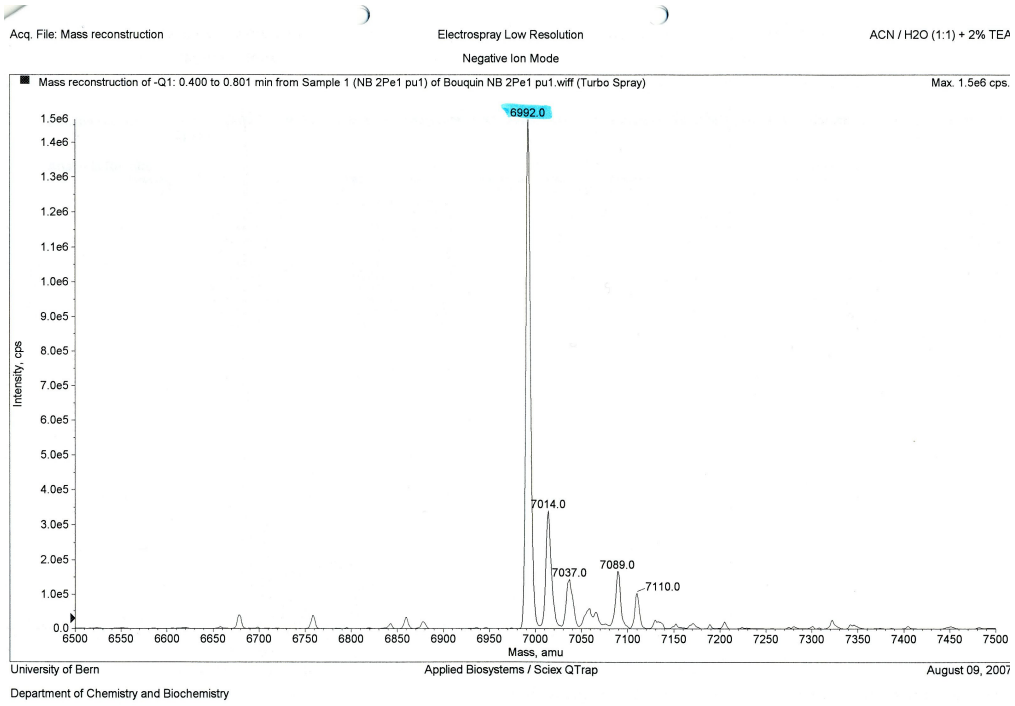
Oligo 3



Oligo 4



Oligo 5



Oligo 6