

Supporting Information

for

**Synthesis and optical properties of pyrrolidinyl
peptide nucleic acid carrying a clicked Nile red label**

Nattawut Yotapan¹, Chayan Charoenpakdee¹, Pawinee Wathanathavorn¹, Boonsong
Ditmangklo¹, Hans-Achim Wagenknecht*² and Tirayut Vilaivan*¹

Address: ¹Organic Synthesis Research Unit, Department of Chemistry, Faculty of
Science, Chulalongkorn University, Phayathai Road, Patumwan, Bangkok 10330,
Thailand and ²Institute of Organic Chemistry, Karlsruhe Institute of Technology
(KIT), Fritz-Haber-Weg 6, 76131 Karlsruhe, Germany

Email: Tirayut Vilaivan - vtirayut@chula.ac.th; Hans-Achim Wagenknecht -
wagenknecht@kit.edu

*Corresponding author

**NMR spectra, HPLC chromatogram, mass spectra and
additional spectroscopic data**

Content	Page
Figure S1: (a) ^1H NMR and (b) ^{13}C NMR spectra of propargyl Nile red 1	S3
Figure S2: HPLC chromatogram and MALDI-TOF mass spectrum of 10mer-Nr	S4
Figure S3: HPLC chromatogram and MALDI-TOF mass spectrum of 11merAA-Nr	S5
Figure S4: HPLC chromatogram and MALDI-TOF mass spectrum of 11merCC-Nr	S6
Figure S5: HPLC chromatogram and MALDI-TOF mass spectrum of 11merGG-Nr	S7
Figure S6: HPLC chromatogram and MALDI-TOF mass spectrum of 11merTT-Nr	S8
Figure S7: UV-vis and fluorescence spectra of 1 in MeCN-buffer	S9
Figure S8: UV-vis and fluorescence spectra of 10mer-Nr in MeCN-buffer	S9
Figure S9: UV-vis and fluorescence spectra of 10mer-Nr with various mismatched DNA	S10
Figure S10: UV-vis and fluorescence spectra of 10mer-Nr with various base-inserted DNA	S10
Figure S11: UV-vis and fluorescence spectra of 10mer-Nr with various base-inserted DNA (indirect)	S11
Figure S12: UV-vis and fluorescence spectra of 10mer-Nr with various base-inserted DNA (mismatched)	S11
Figure S13: UV-vis and fluorescence spectra of 11merAA-Nr with complementary and base-inserted DNA	S12
Figure S14: UV-vis and fluorescence spectra of 11merCC-Nr with complementary and base-inserted DNA	S12
Figure S15: UV-vis and fluorescence spectra of 11merGG-Nr with complementary and base-inserted DNA	S13
Figure S16: UV-vis and fluorescence spectra of 11merTT-Nr with complementary and base-inserted DNA	S13
Figure S17: UV-vis and fluorescence spectra of 10mer-Nr and its DNA hybrids before and after addition of β -cyclodextrin	S14
Figure S18: Photographs of 11merXX-Nr and its hybrids with various DNA under black light	S14

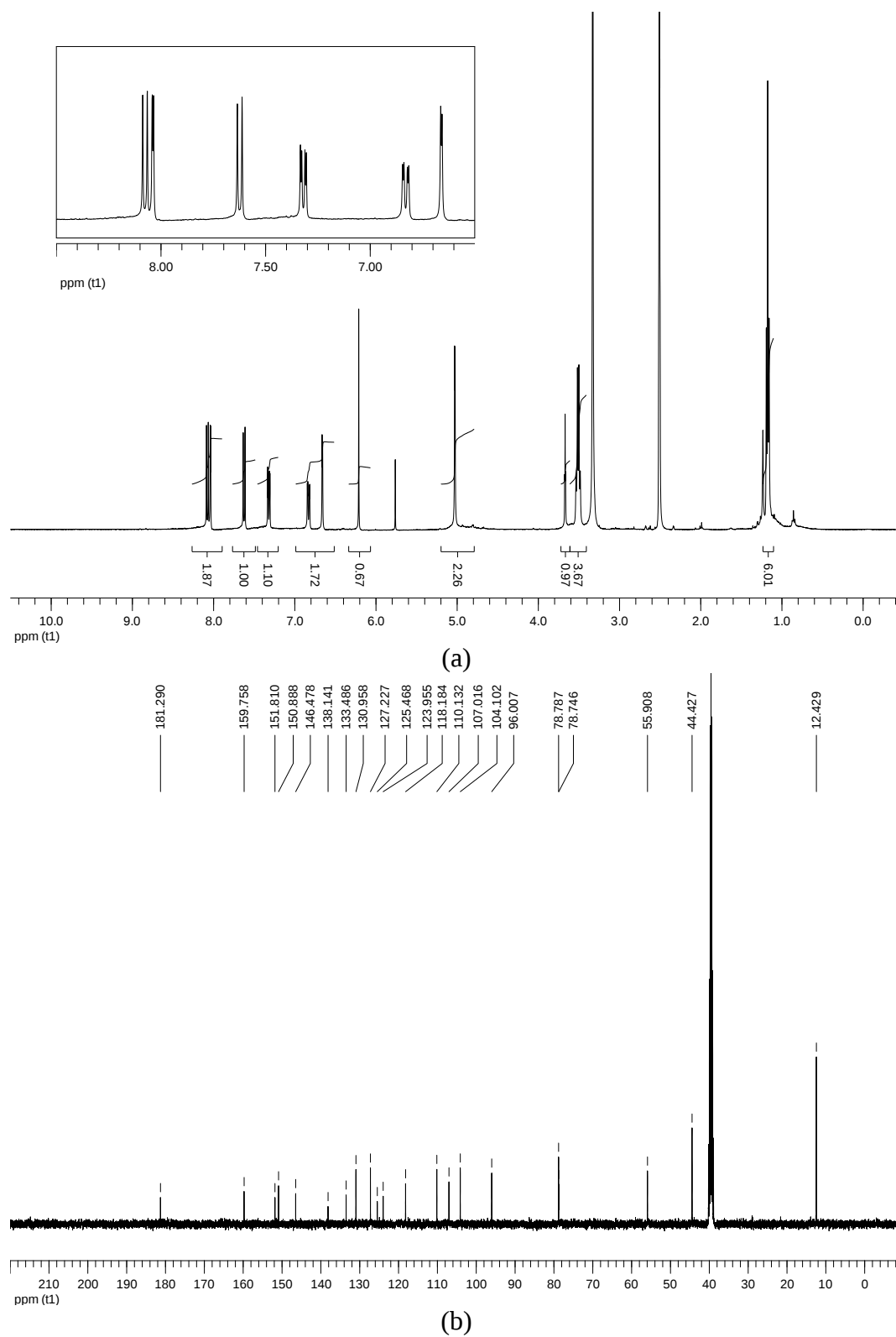
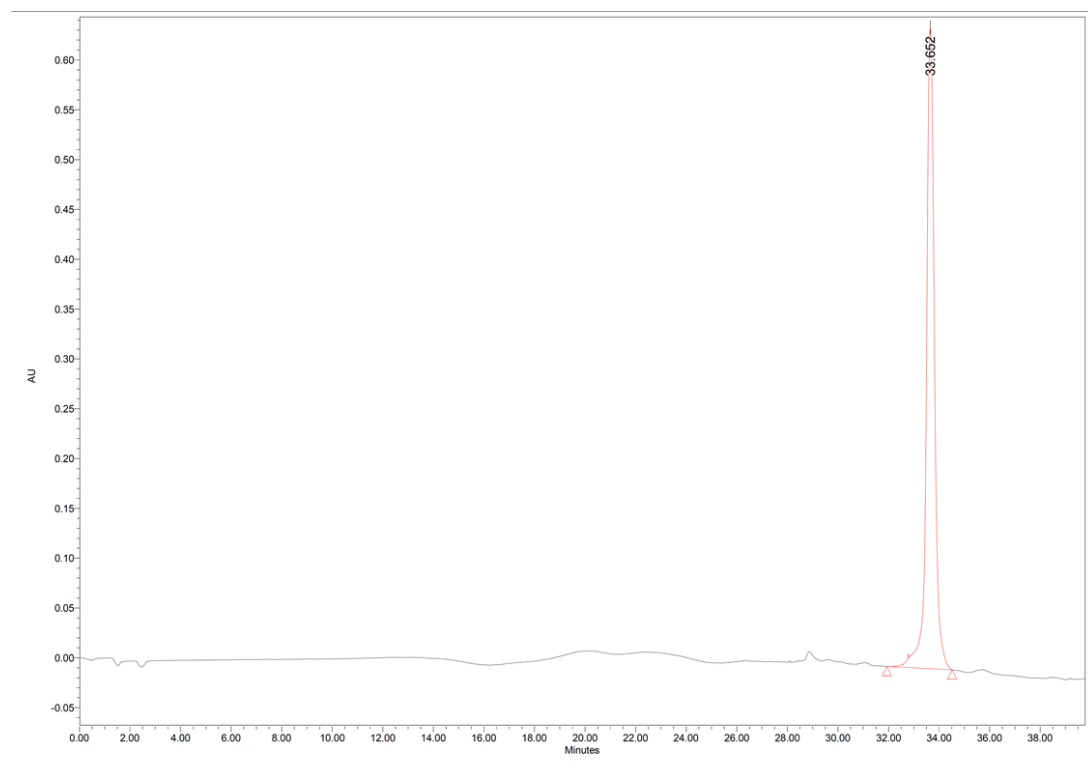
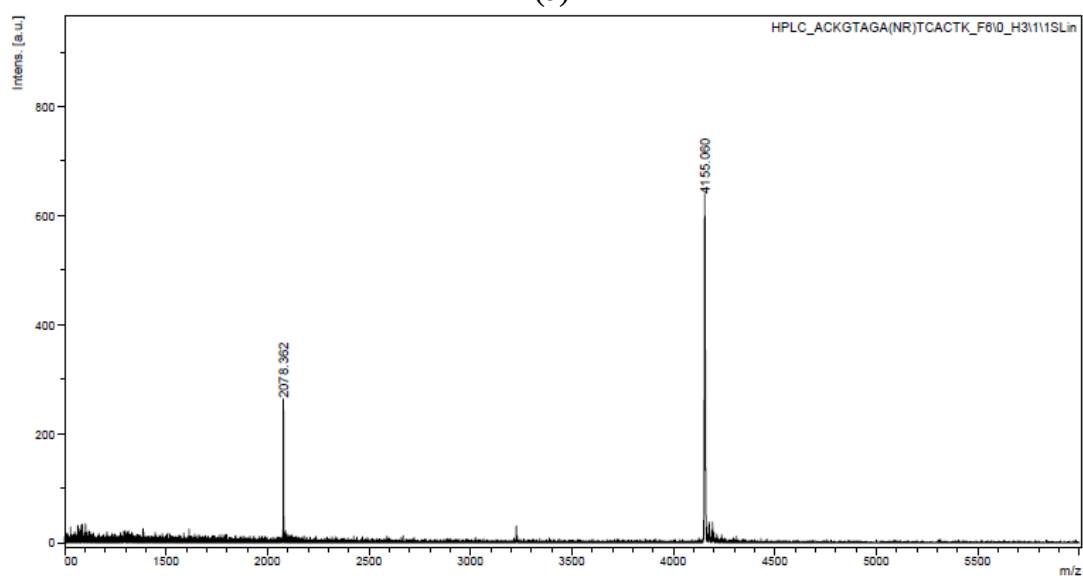


Figure S1: (a) ^1H NMR (CDCl₃, 400 MHz) and (b) ^{13}C NMR (CDCl₃, 100 MHz) spectra of propargyl nile red **1**

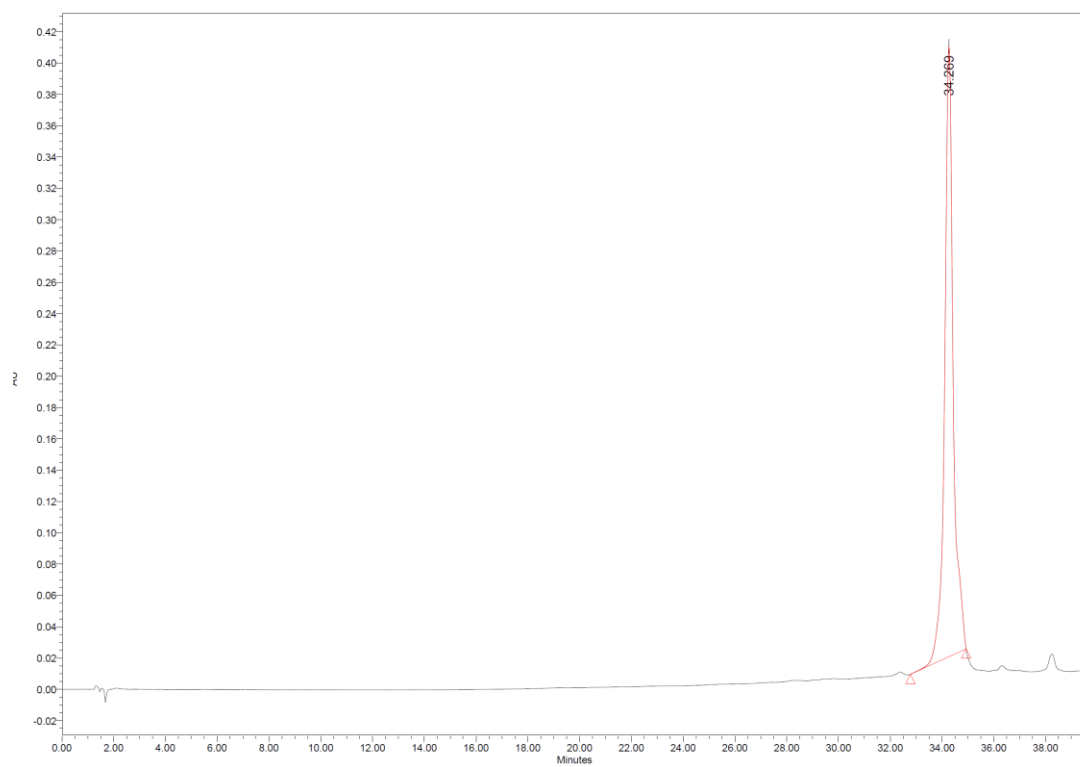


(a)

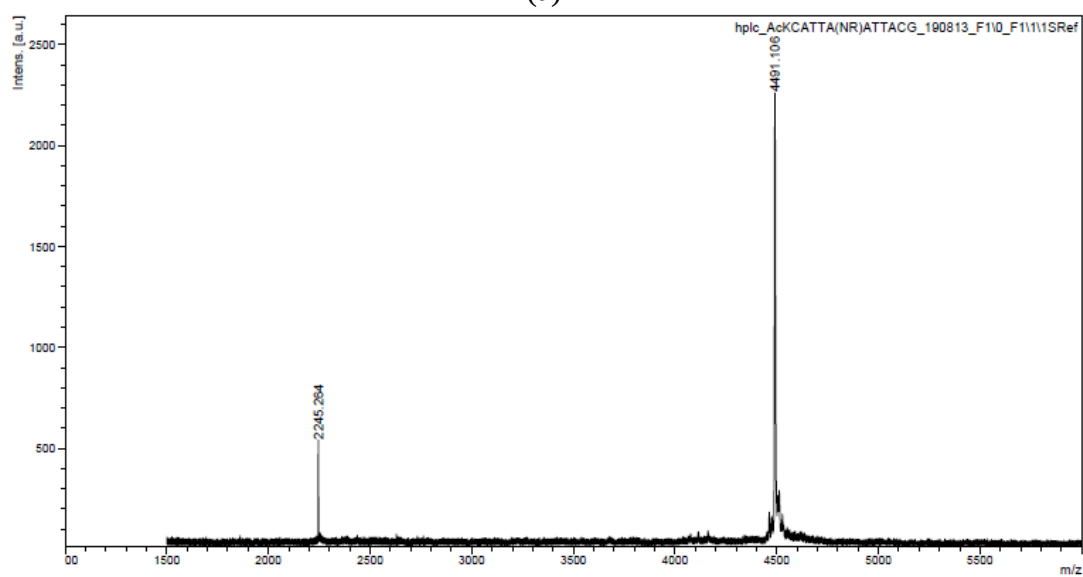


(b)

Figure S2: (a) Analytical HPLC chromatogram and (b) MALDI-TOF mass spectrum of **10mer-Nr** (calcd for $[M \cdot H]^+ = 4157.5$)

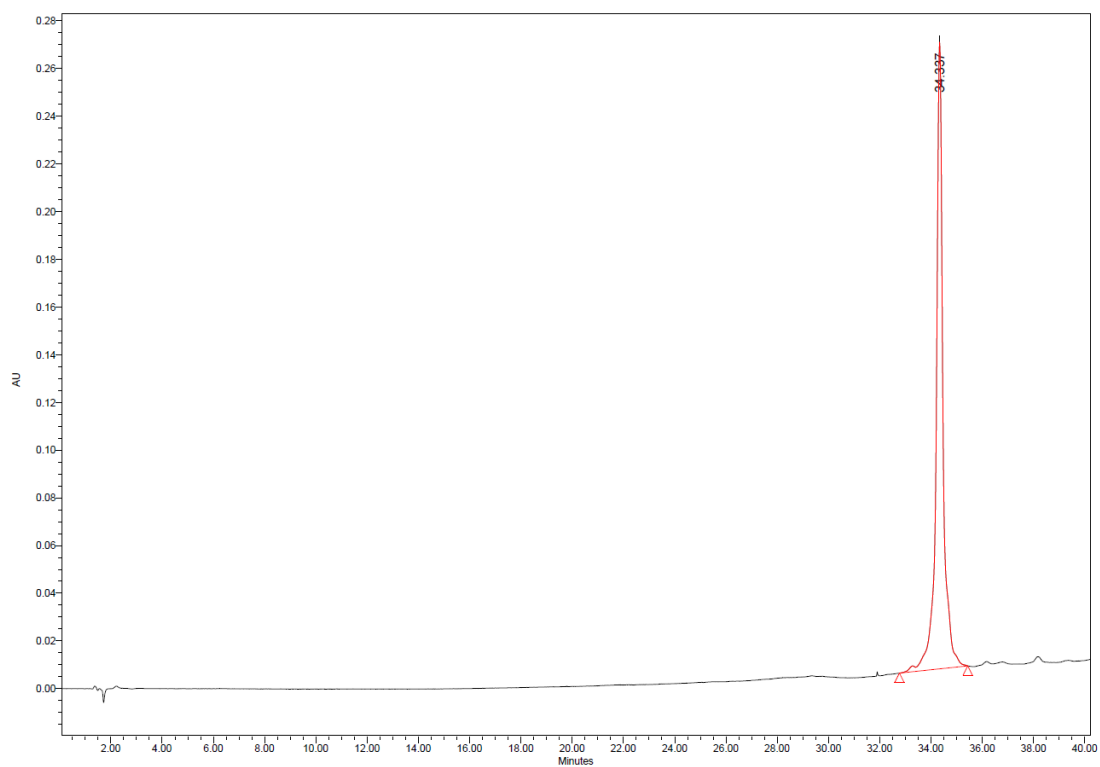


(a)

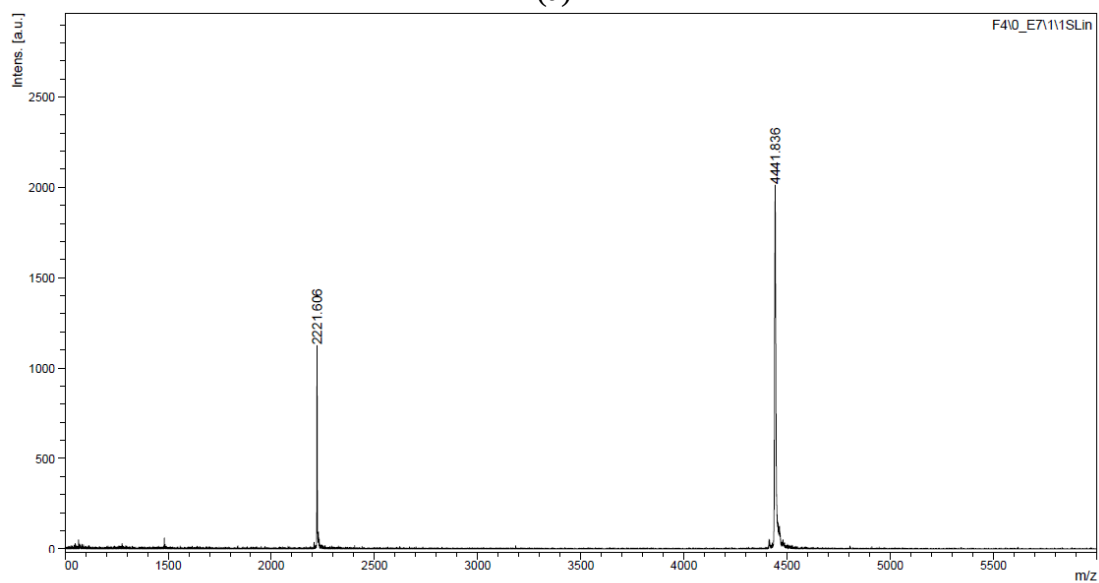


(b)

Figure S3: (a) Analytical HPLC chromatogram and (b) MALDI-TOF mass spectrum of **11merAA-Nr** (calcd for $[M \cdot H]^+ = 4491.9$)

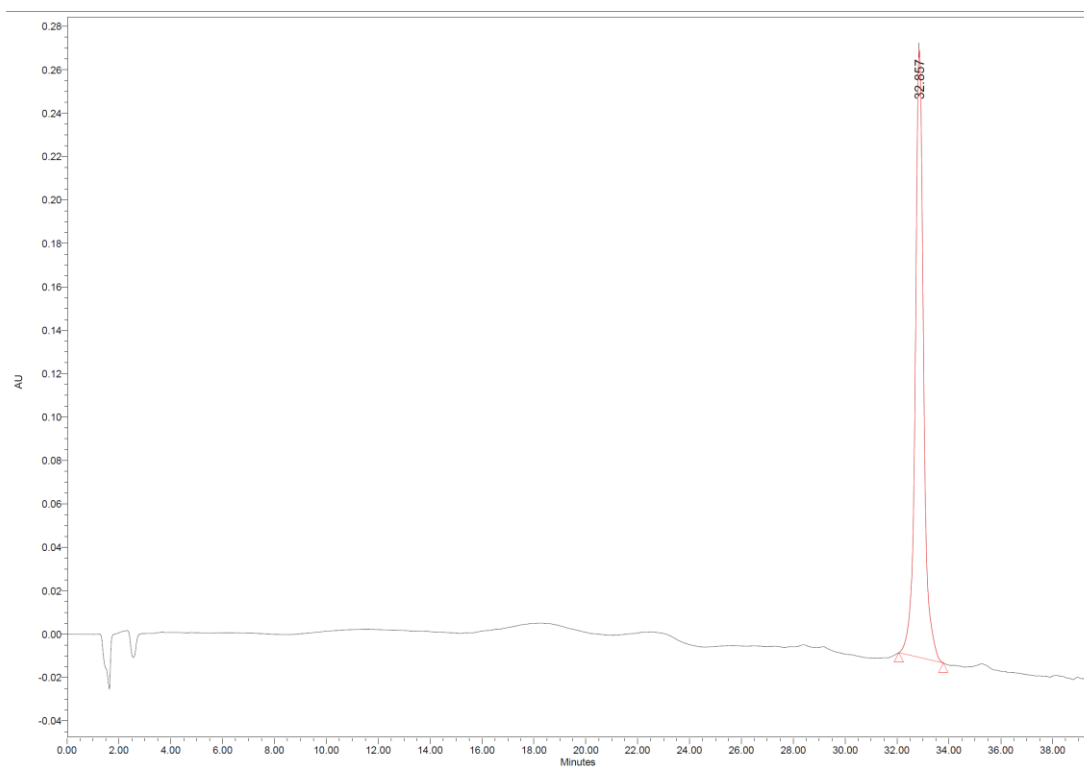


(a)

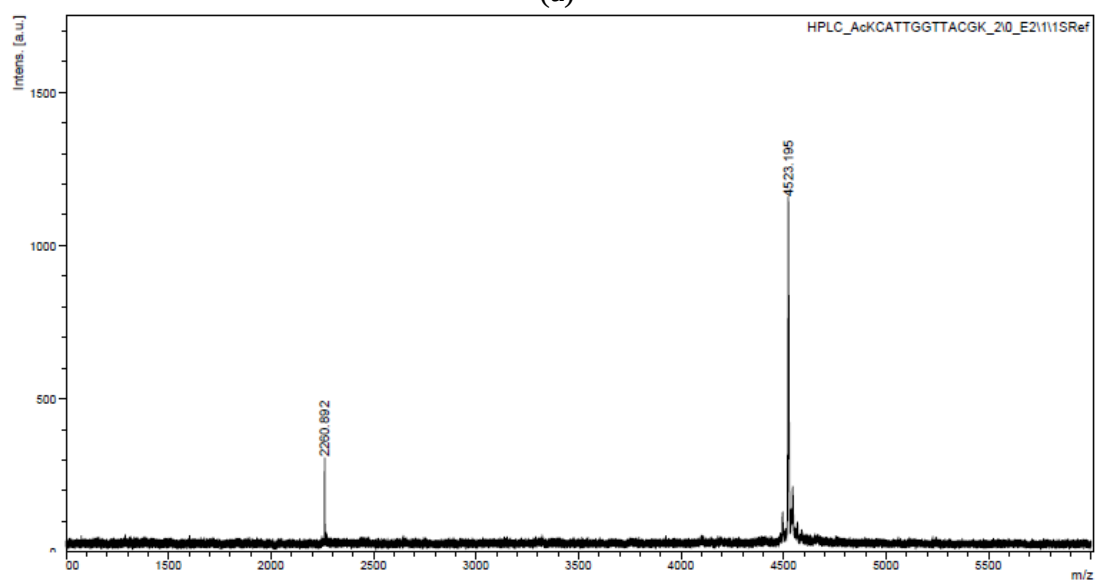


(b)

Figure S4: (a) Analytical HPLC chromatogram and (b) MALDI-TOF mass spectrum of **11merCC-Nr** (calcd for $[M \cdot H]^+ = 4443.9$)

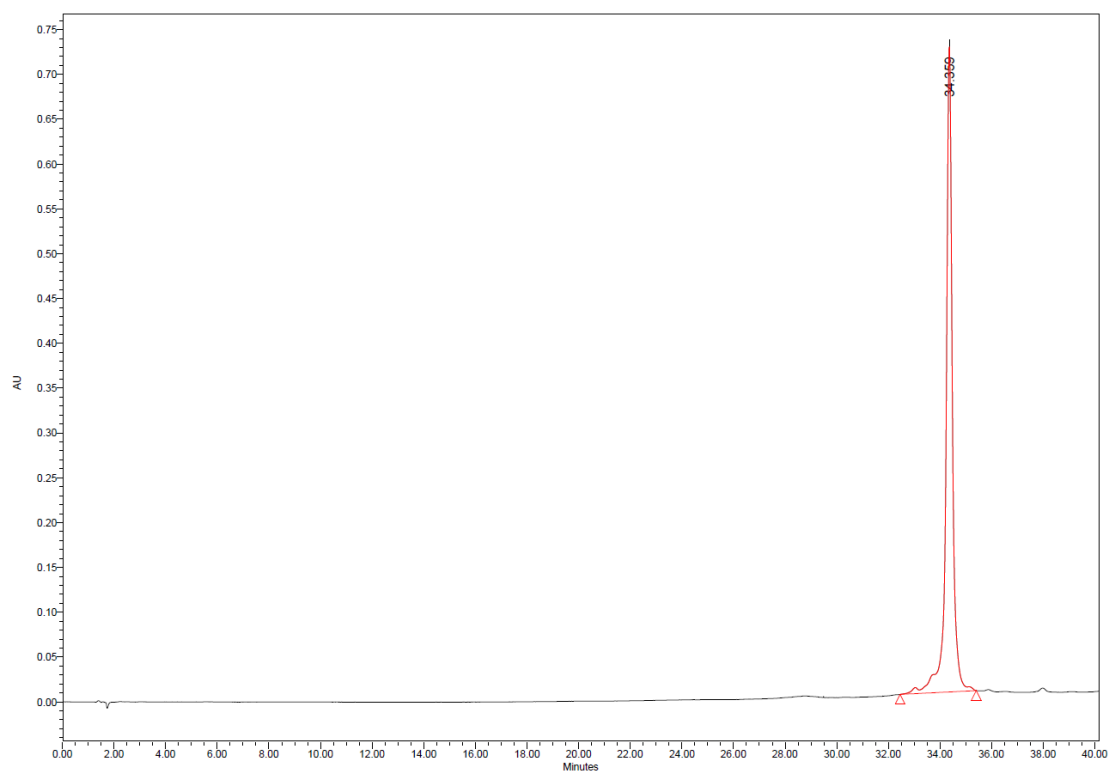


(a)

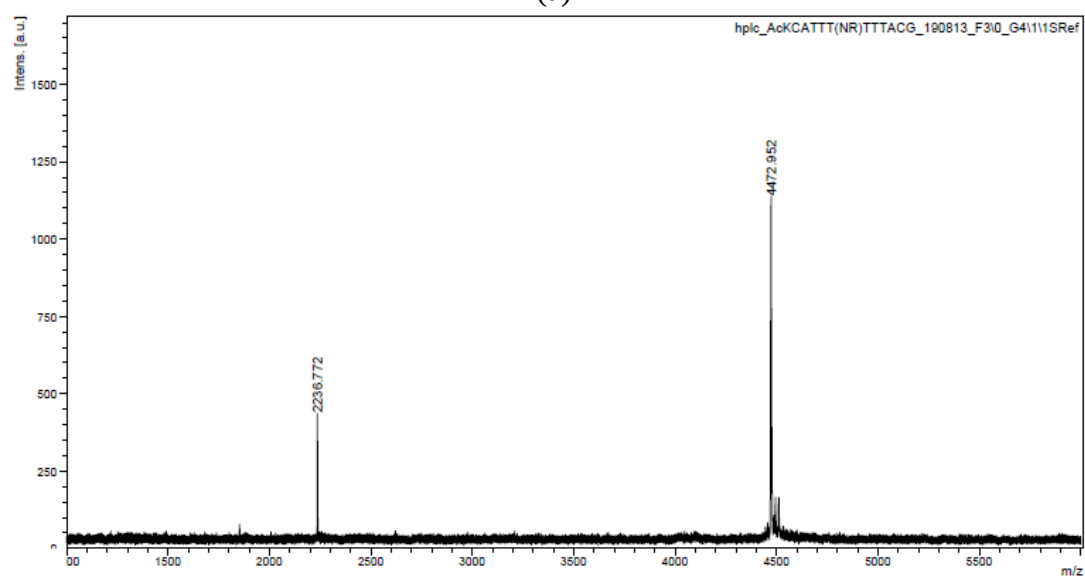


(b)

Figure S5: (a) Analytical HPLC chromatogram and (b) MALDI-TOF mass spectrum of **11merGG-Nr** (calcd for $[M \cdot H]^+ = 4523.9$)



(a)



(b)

Figure S6: (a) Analytical HPLC chromatogram and (b) MALDI-TOF mass spectrum of **11merTT-Nr** (calcd for $[M \cdot H]^+ = 4473.9$)

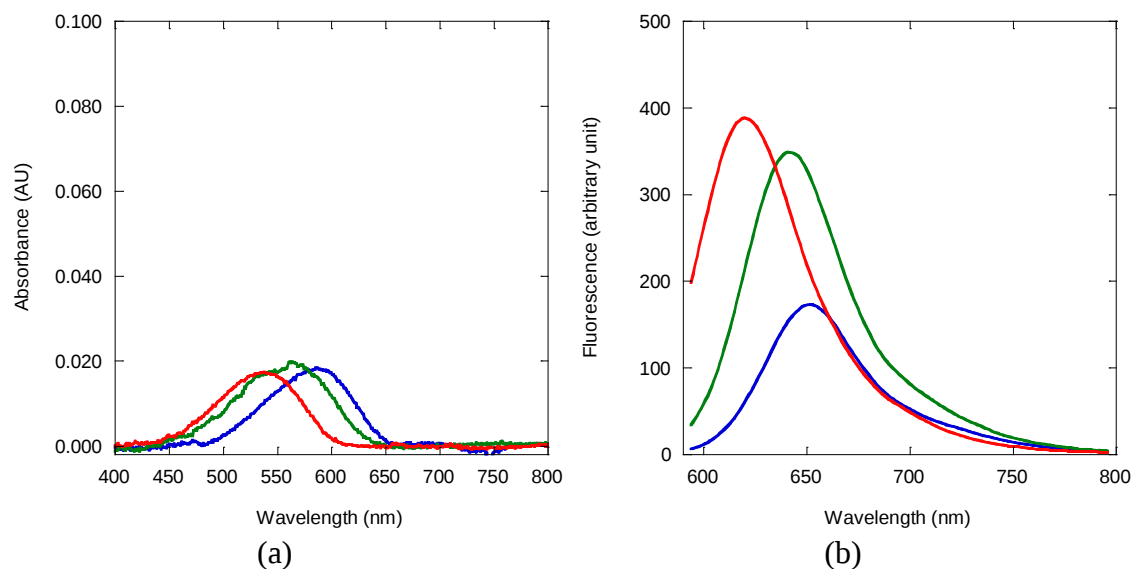


Figure S7: (a) UV-vis and (b) fluorescence spectra of **1** in MeCN-buffer (10 mM sodium phosphate, pH 7.0): 20% MeCN (blue), 50% MeCN (green), 100% MeCN (red). All spectra were measured at $[1] = 1.0 \mu\text{M}$, $\lambda_{\text{ex}} = 580 \text{ nm}$, 700 PMT.

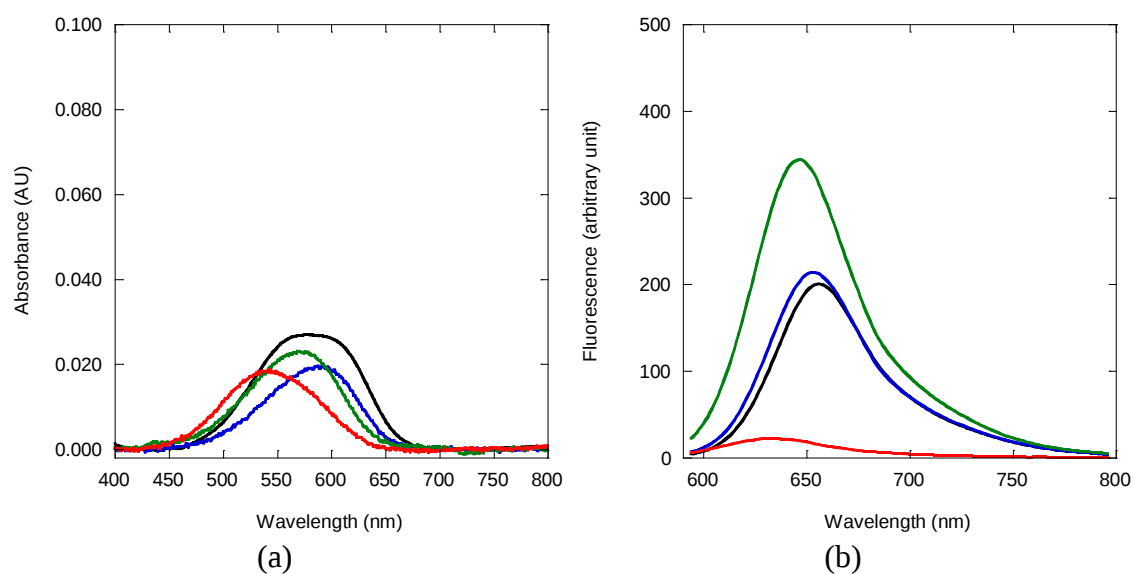


Figure S8: (a) UV-vis and (b) fluorescence spectra of **10mer-Nr** in MeCN-buffer (10 mM sodium phosphate, pH 7.0): 0% MeCN (black), 20% MeCN (blue), 50% MeCN (green), 100% MeCN (red). All spectra were measured at $[1] = 1.0 \mu\text{M}$, $\lambda_{\text{ex}} = 580 \text{ nm}$, 700 PMT.

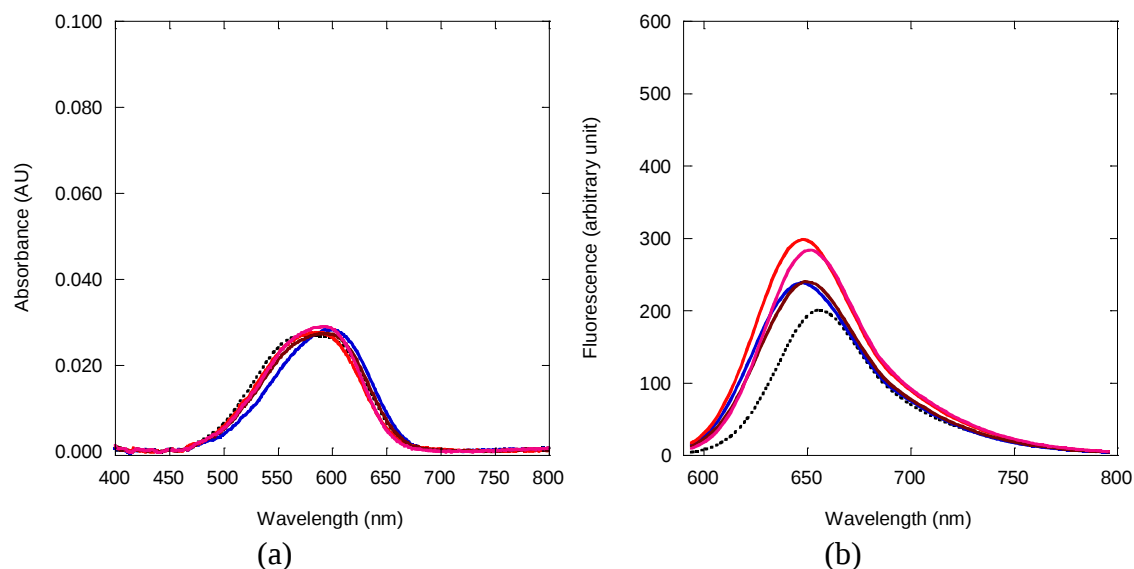


Figure S9: (a) UV-vis and (b) fluorescence spectra of **10mer-Nr**: single stranded (black dotted), with 5'-d(AGTGATCTAC)-3' (blue), with 5'-d(AGTGCTCTAC)-3' (red), with 5'-d(AGTCATCTAC)-3' (brown), with 5'-d(AGTGACCTAC)-3' (pink). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

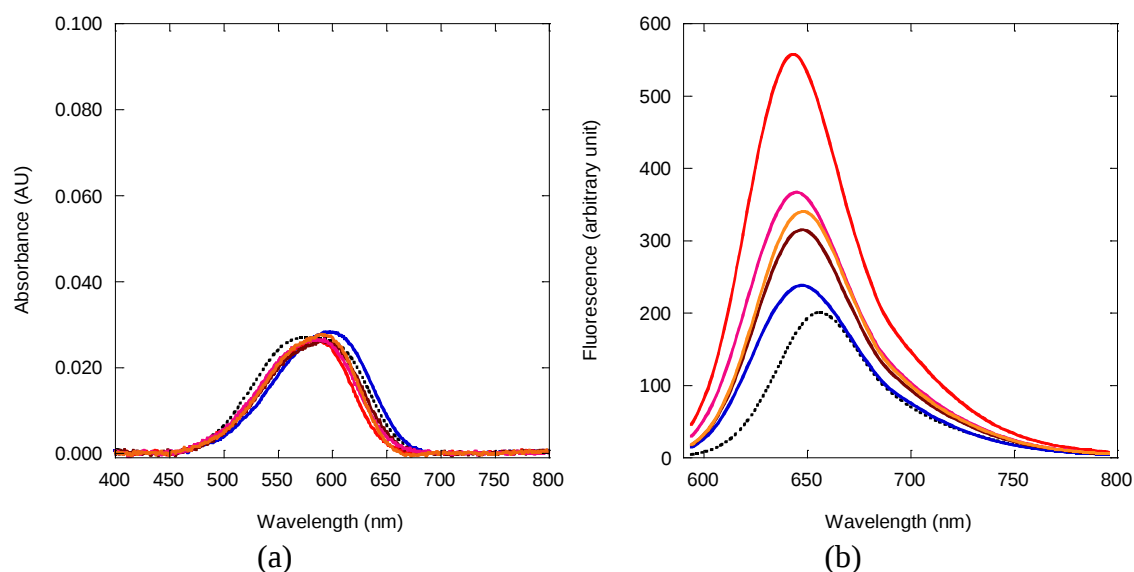


Figure S10: (a) UV-vis and (b) fluorescence spectra of **10mer-Nr**: single stranded (black dotted), with 5'-d(AGTGATCTAC)-3' (blue), with 5'-d(AGTGACTCTAC)-3' (red), with 5'-d(AGTGAATCTAC)-3' (brown), with 5'-d(AGTGAGTCTAC)-3' (pink), with 5'-d(AGTGATTCTAC)-3' (orange). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

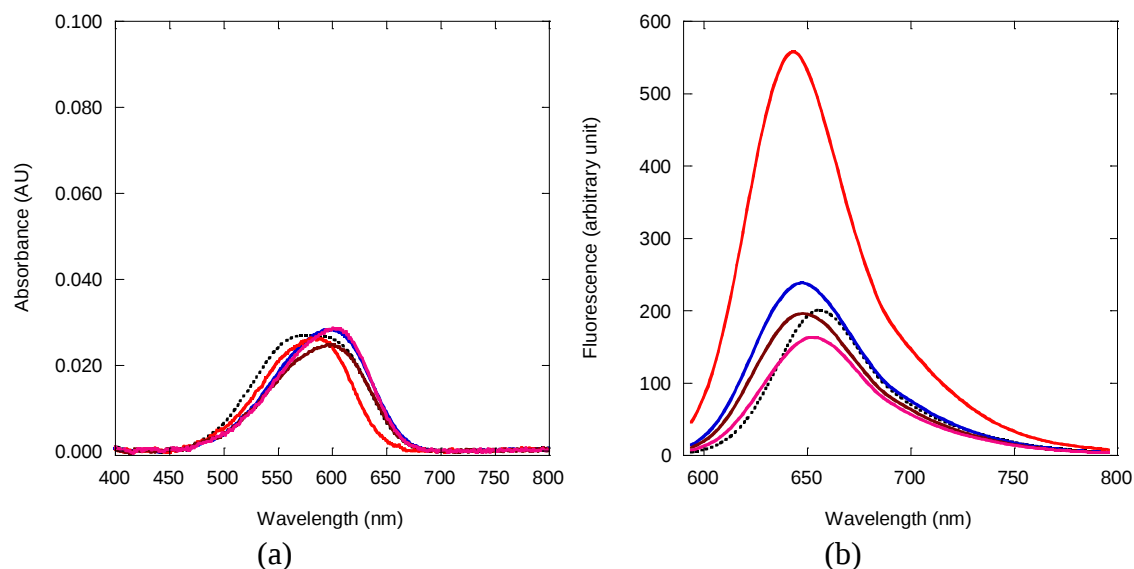


Figure S11: (a) UV-vis and (b) fluorescence spectra of **10mer-Nr**: single stranded (black dotted), with 5'-d(AGTGATCTAC)-3' (blue), with 5'-d(AGTGACTCTAC)-3' (red), with 5'-d(AGTCGATCTAC)-3' (brown), with 5'-d(AGTGATCCTAC)-3' (pink). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

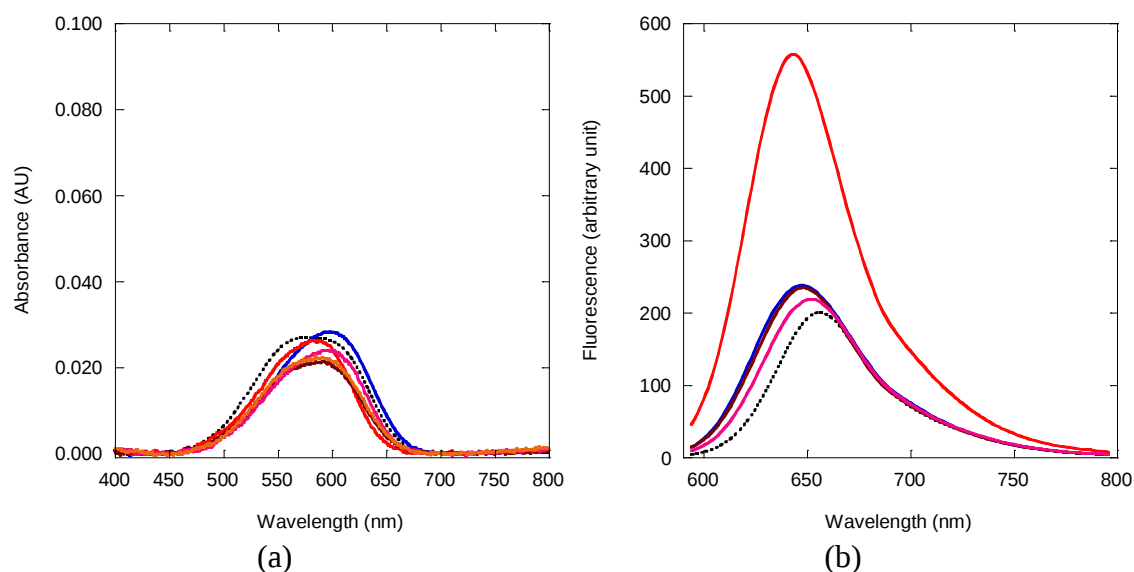


Figure S12: (a) UV-vis and (b) fluorescence spectra of **10mer-Nr**: single stranded (black dotted), with 5'-d(AGTGATCTAC)-3' (blue), with 5'-d(AGTGACTCTAC)-3' (red), with 5'-d(AGTG CCTCTAC)-3' (brown), with 5'-d(AGTGACCCTAC)-3' (pink), with 5'-d(AGTGACTCCAC)-3' (orange). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

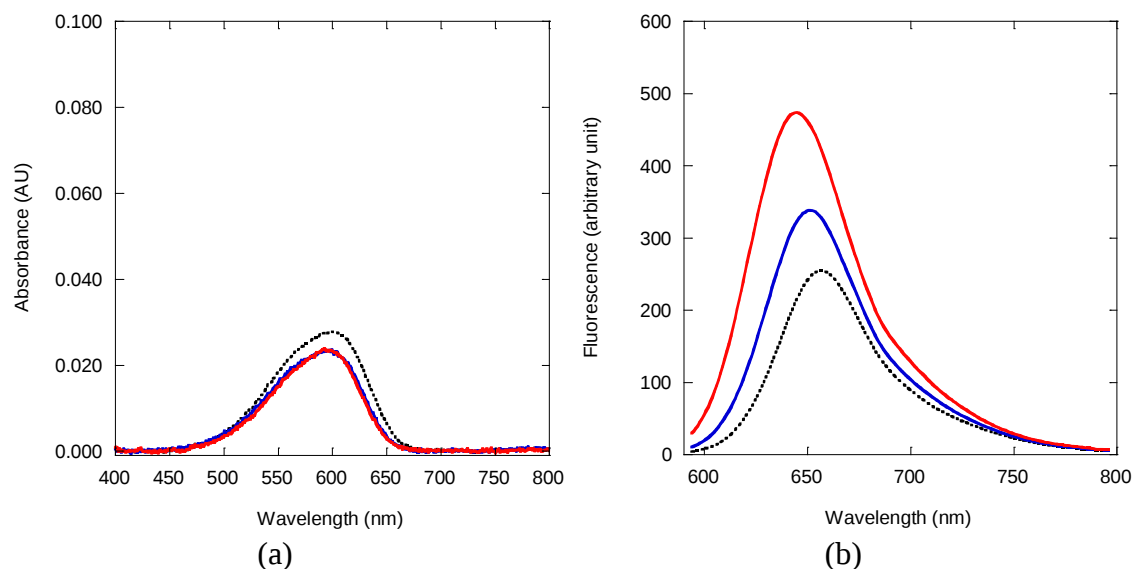


Figure S13: (a) UV-vis and (b) fluorescence spectra of **11merAA-Nr**: single stranded (black dotted), with 5'-d(CGTATTTTATG)-3' (blue) and with 5'-d(CGTATTCTTATG)-3' (red). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

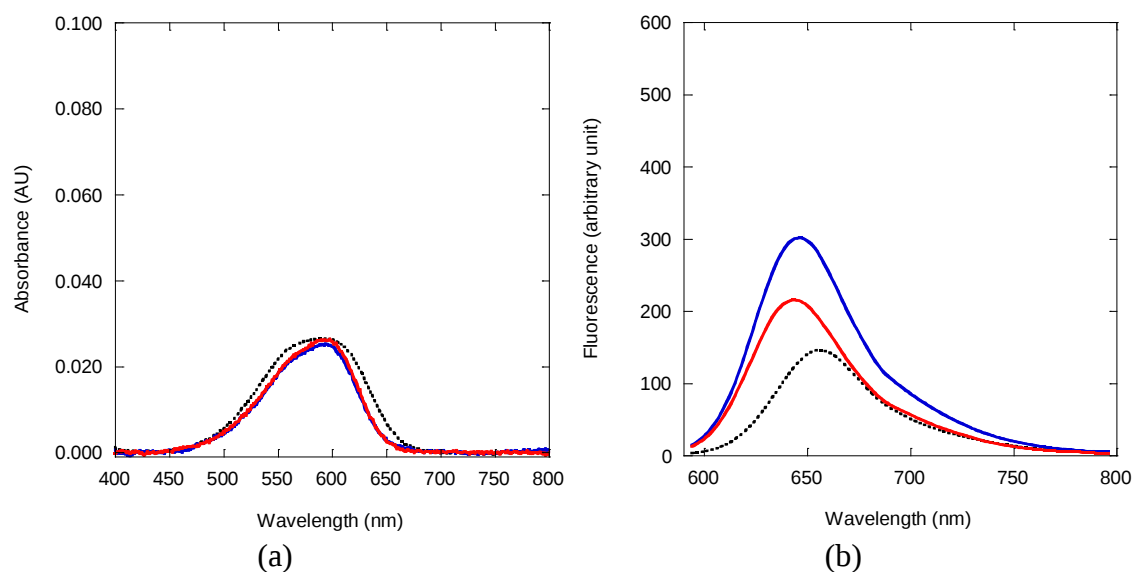


Figure S14: (a) UV-vis and (b) fluorescence spectra of **11merCC-Nr**: single stranded (black dotted), with 5'-d(CGTATAATATG)-3' (blue) and with 5'-d(CGTATACATATG)-3' (red). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

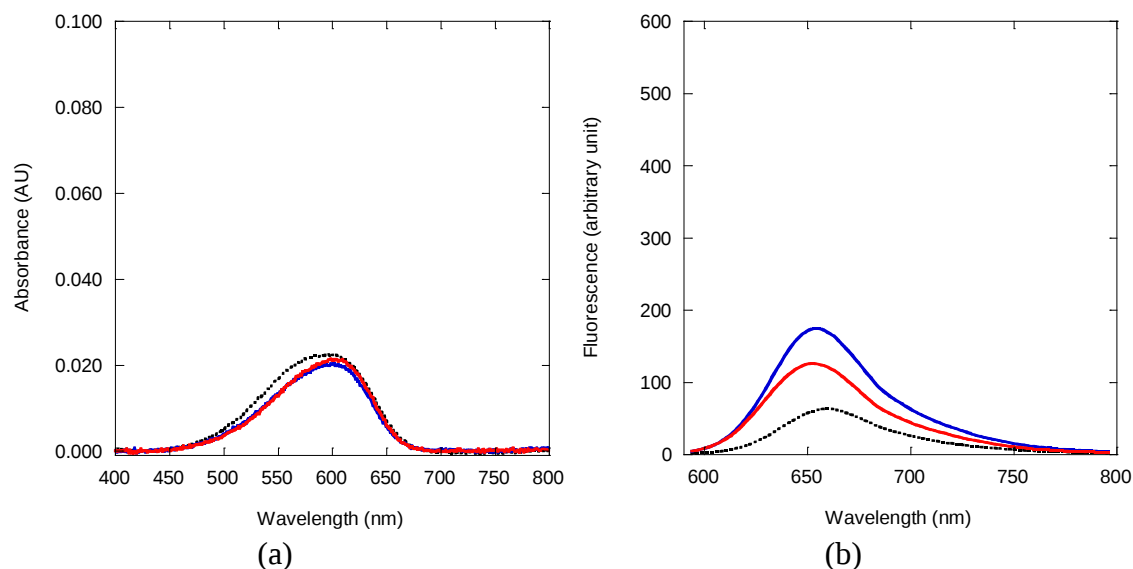


Figure S15: (a) UV-vis and (b) fluorescence spectra of **11merGG-Nr**: single stranded (black dotted), with 5'-d(CGTATCCTATG)-3' (blue) and with 5'-d(CGTATCCCTATG)-3' (red). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

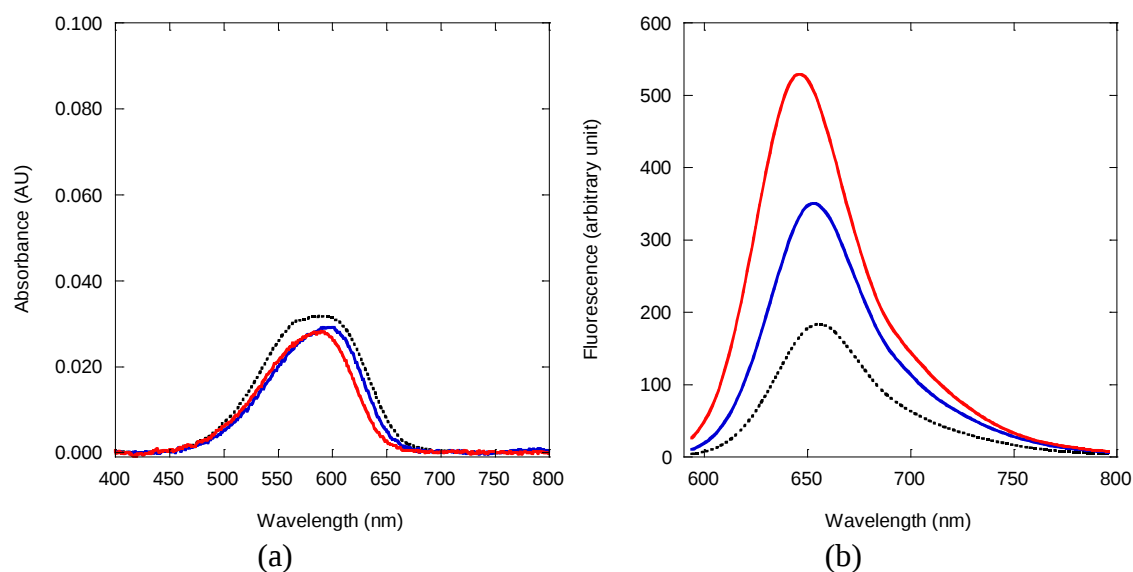


Figure S16: (a) UV-vis and (b) fluorescence spectra of **11merTT-Nr**: single stranded (black dotted), with 5'-d(CGTATAATATG)-3' (blue) and with 5'-d(CGTATACATATG)-3' (red). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

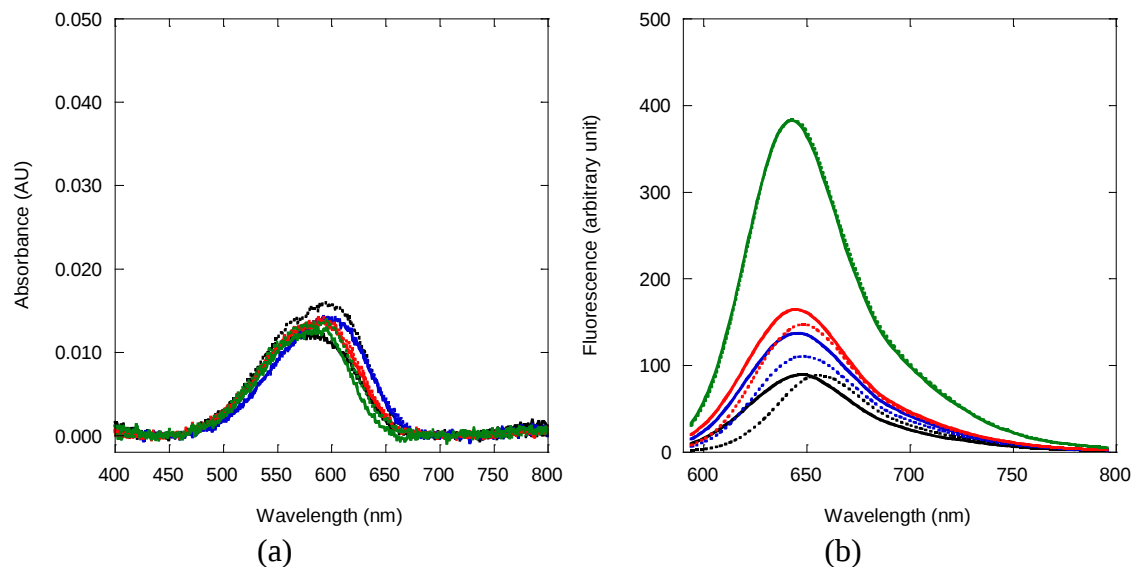


Figure S17: (a) UV-vis and (b) fluorescence spectra of **10mer-Nr** and its DNA hybrids before (---) and after (—) addition of β -cyclodextrin (10 mM): single stranded (black), with 5'-d(AGTGATCTAC)-3' (blue), 5'-d(AGTGCTCTAC)-3' (red), with 5'-d(AGTGACTCTAC)-3' (green). All spectra were measured in 10 mM sodium phosphate buffer pH 7.0, [PNA] = 1.0 μ M, [DNA] = 1.2 μ M, λ_{ex} = 580 nm, 700 PMT.

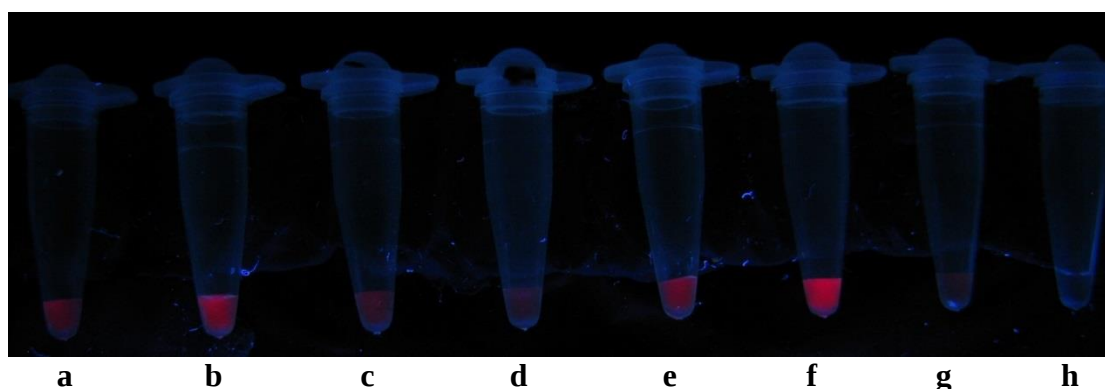


Figure S18: Photographs of **11merXX-Nr** hybrids with DNA under black light (405 nm): (a) **11merAA-Nr** and 5'-d(CGTATTTTATG)-3' (b) **11merAA-Nr** and 5'-d(CGTATTCTTATG)-3' (c) **11merCC-Nr** and 5'-d(CGTATGGTATG)-3' (d) **11merCC-Nr** and 5'-d(CGTATGCGTATG)-3' (e) **11merTT-Nr** and 5'-d(CGTATAATATG)-3' (f) **11merTT-Nr** and 5'-d(CGTATACATATG)-3' (g) **11merGG-Nr** and 5'-d(CGTATCCTATG)-3' (h) **11merGG-Nr** and 5'-d(CGTATCCCTATG)-3'; Conditions: 10 mM phosphate buffer pH 7.0, [PNA] = 10 μ M and [DNA] = 12 μ M