Supplementary Material

Metallacarborane complex boosts the rate of DNA oligonucleotide hydrolysis in the reaction catalyzed by snake venom phosphodiesterase

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Figure S1. Analytical RP HPLC profiles and MALDI-TOF or ESI-Q-TOF (*) mass spectra of **1**, **P1a**, **P1b**, **P1c**, **1a**, **1b**, **1c**, **2**, **P2a**, **P2b**, **2a, 2b, FL-1** and **FL-2.**
RP-HPLC conditions were as follow: the buffer A (0.1 M CH_3COONH_4) and buffer B (100% CH_3CN). The buffer B gradient: 0→2 min 0%; 2→25 min 0-48%; 25→30 min 48-60%; 30→35 min 60-0%; 35→38 min 0%.

Figure S2. PAGE analysis of the **1, 1a, 1b, 1c** and **2, 2a, 2b** oligonucleotides (20%/7 M urea)

Figure S3. svPDE - assisted hydrolysis of reference oligonucleotide **1** with and without magnesium ions, monitored by MALDI-TOF mass spectrometry.

Figure S4. Original MALDI-TOF MS spectra of hydrolysis of **1c** in the presence of svPDE (in triplicate).

Figure S5. MALDI-TOF mass spectrometry analysis of the 1:1:1 mixture of **1, 1a** and oligonucleotide **1** containing two FESAN clusters (synthetic data not shown). Intensity of peaks is given on the Figure.

Figure S6. Original MALDI-TOF MS spectra of the hydrolysis mixture of **1**+**1a** in the presence of svPDE (in triplicate).

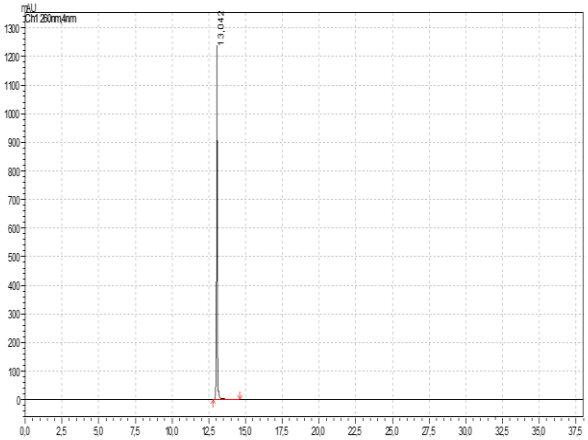
Figure S7. Figure S7. MALDI-TOF MS analysis of the reaction mixture of svPDE-assisted hydrolysis of unmodified oligonucleotides 1 and 2 (0.1 OD) in the presence of a free metallacarborane (FESAN, 182 nM) at 0-60 min.

Figure S8. MALDI-TOF MS analysis of the hydrolysis rate of oligonucleotide **1** in the svPDE-assisted reaction carried out in the presence of 0, 1, 2 or 3 equivalents of metallacarborane (in triplicate)
Figure S9. PAGE analysis of crude snake venom.
Figure S10. The protein samples capillary scan from microscale thermophoresis (the venom proteins fluorescently labeled with RED-NHS 2nd generation).
Figure S11. MST data for K_d determination of affinity of FESAN (ferra(III) bis(dicarbollide)) to **FL-1** (K_d=5.48±0.20 μM).

Figure S1. Analytical RP HPLC profiles and MALDI-TOF or ESI-Q-TOF (*) mass spectra of **1**, **P1a**, **P1b**, **P1c**, **1a**, **1b**, **1c**, **2**, **P2a**, **P2b**, **2a, 2b, FL-1** and **FL-2.**

RP-HPLC conditions were as follow: the buffer A (0.1 M CH_3COONH_4) and buffer B (100% CH_3CN). The buffer B gradient: 0→2 min 0%; 2→25 min 0-48%; 25→30 min 48-60%; 30→35 min 60-0%; 35→38 min 0%.

1 5'-d(TTT CTT TTC CTC C)-3'



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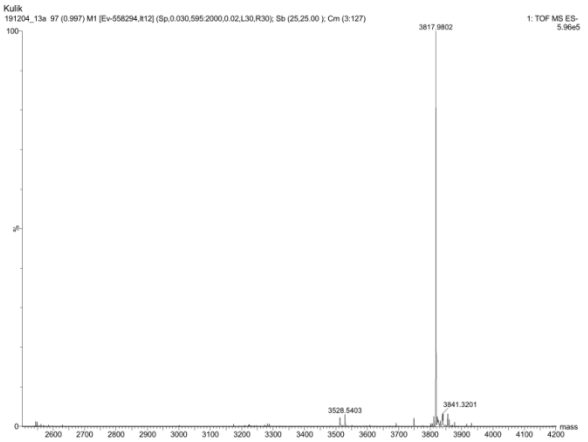
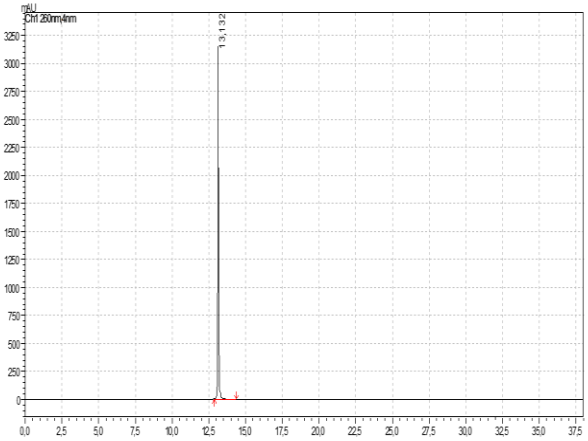


P1a 5'-d(**U_{Pr}**TT CTT TTC CTC C)-3'



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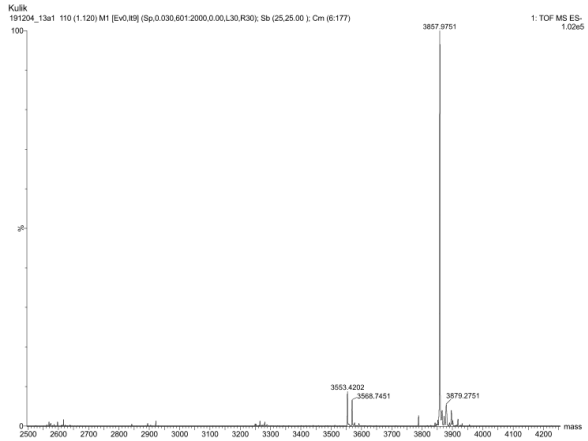
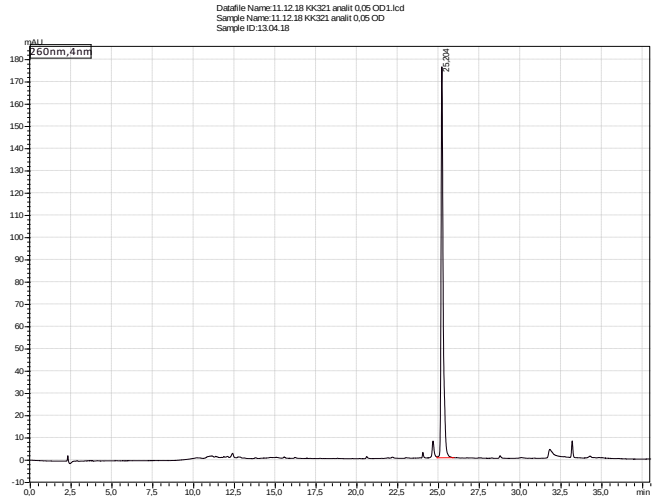
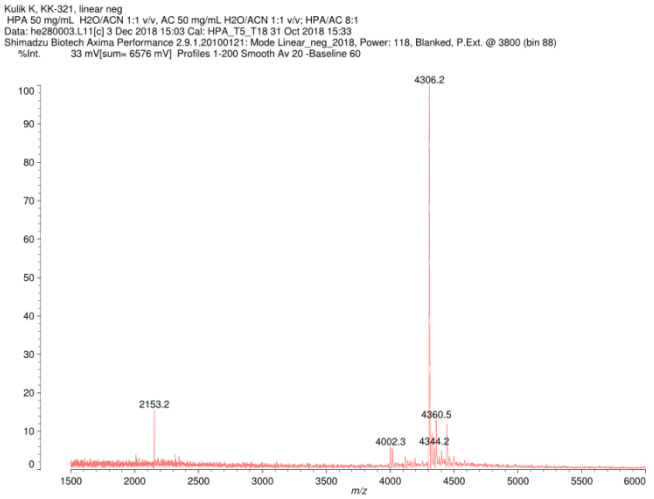
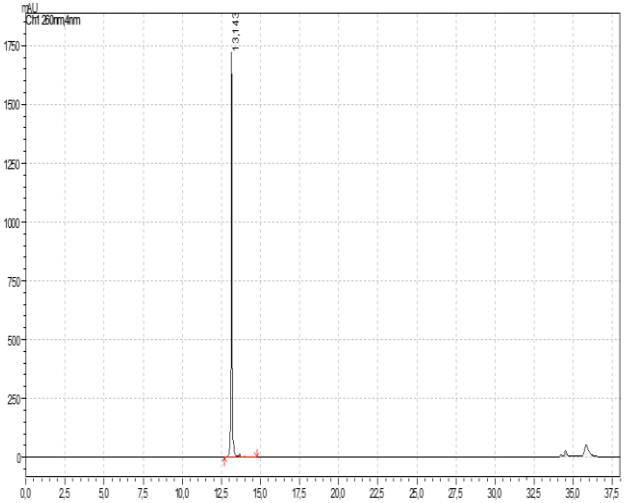


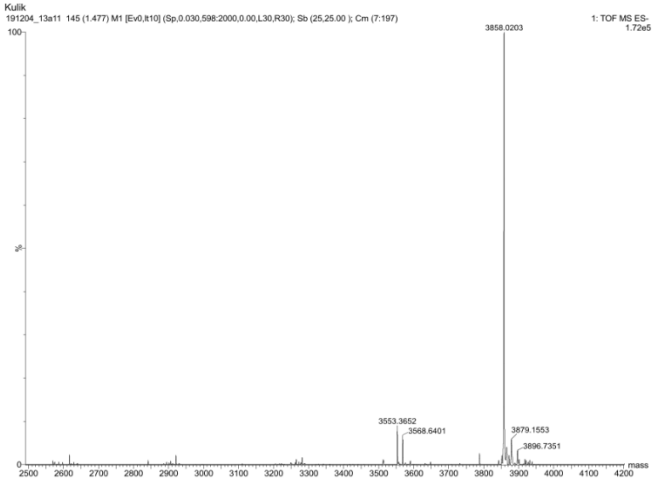
1a 5’-d(**U$_B$**TT CTT TTC CTC C)-3’



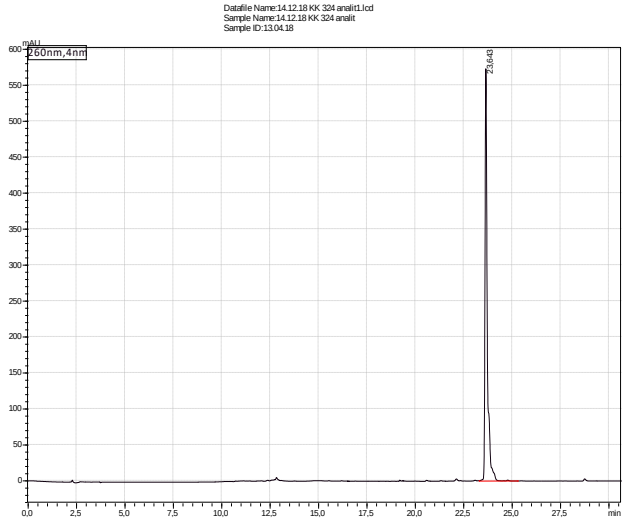


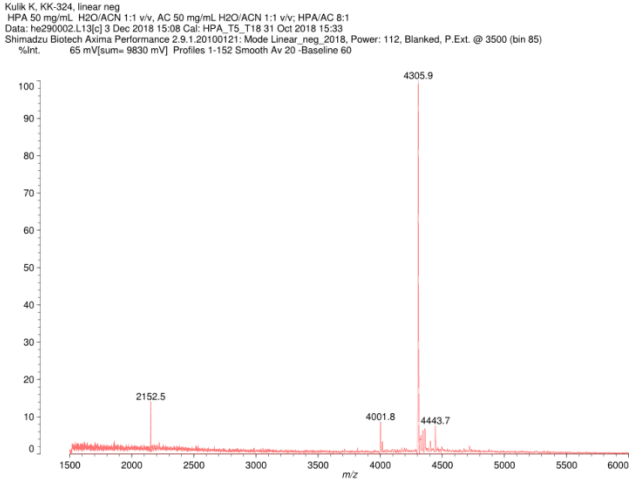
P1b 5’-d(TTT CTT TTC C**U$_{Pr}$**C C)-3’ (*)



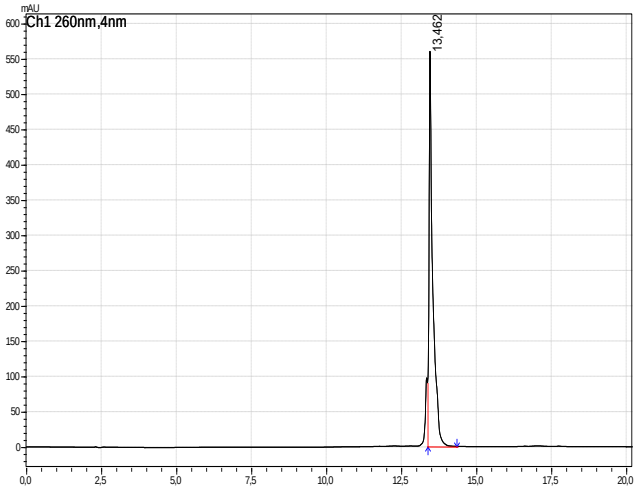


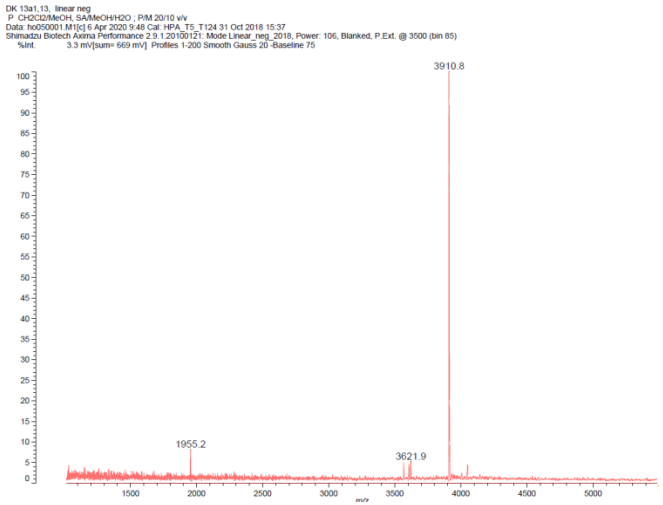
1b 5’-d(TTT CTT TTC C**U$_B$**C C)-3’





P1c 5’-d(**U$_{Pr}$**TT CTT TTC CTC **U$_{Pr}$**)-3’





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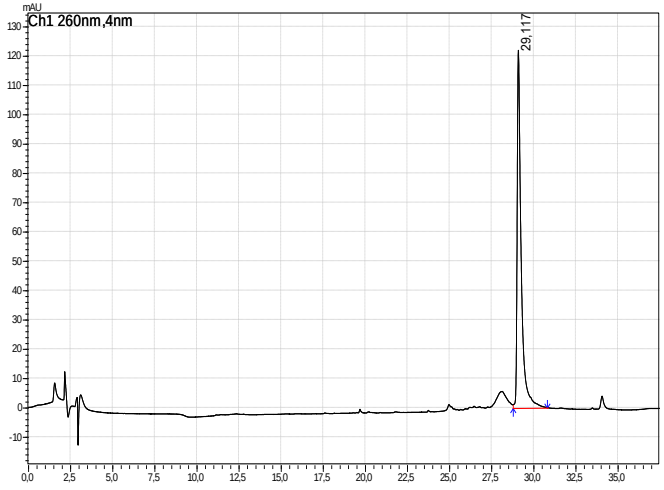
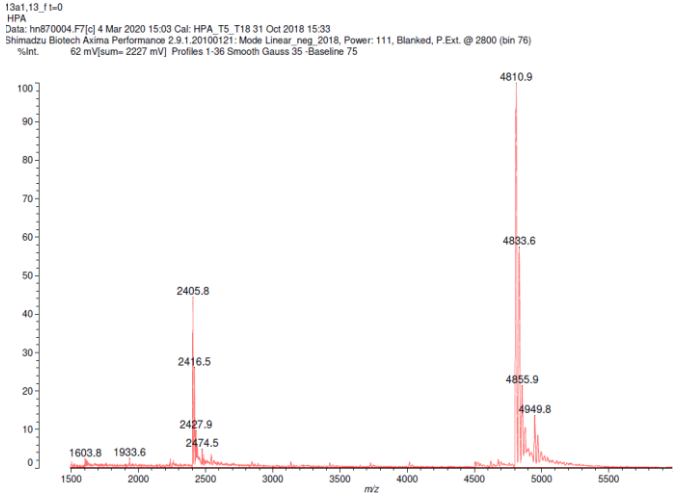
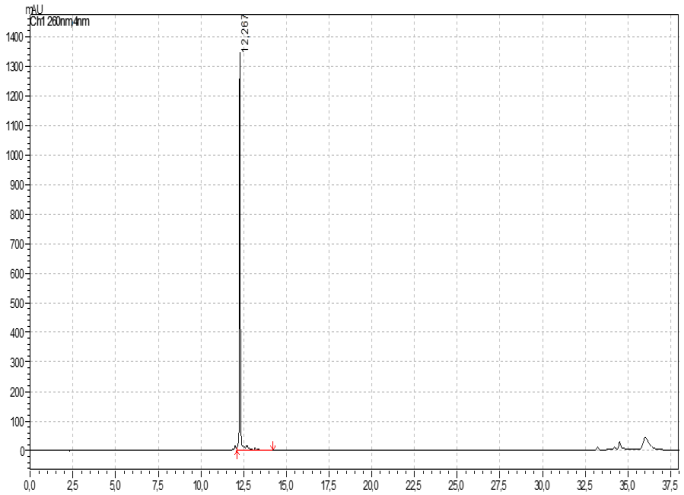
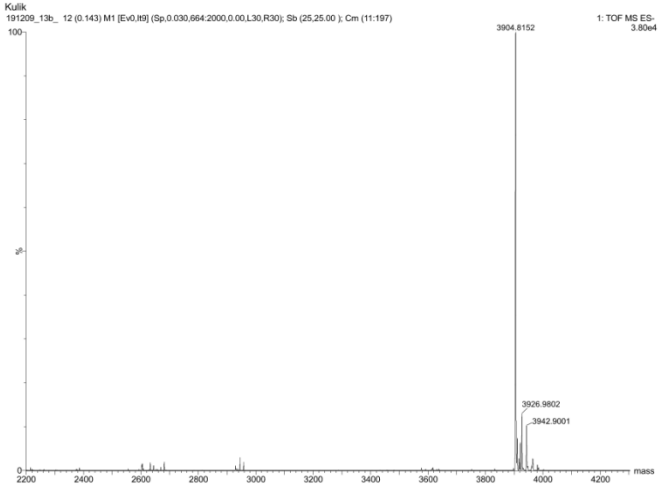


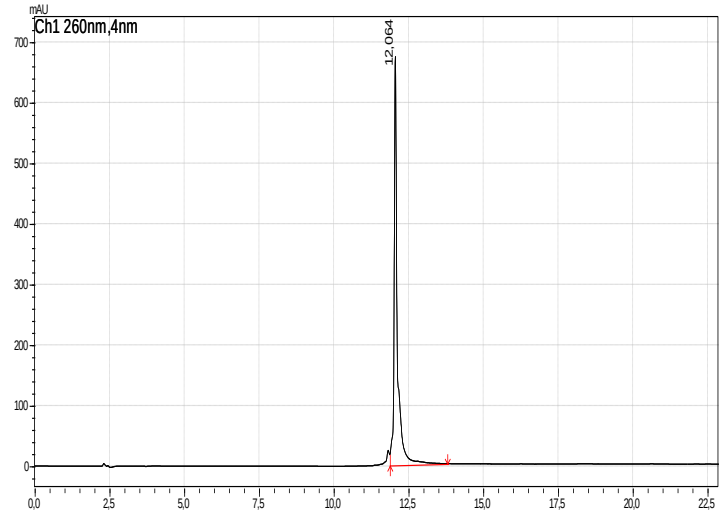


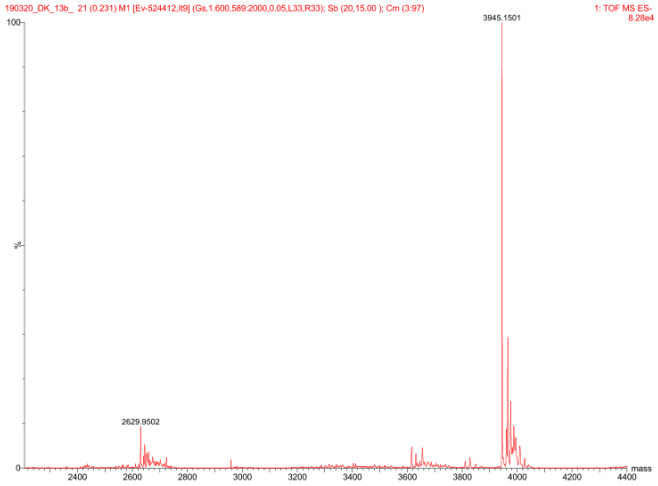
2 5’-d(CTC CAG AGC CCG A)-3’ (*)





P2a 5’-d(C**P**C CAG AGC CCGA)-3’ (*)





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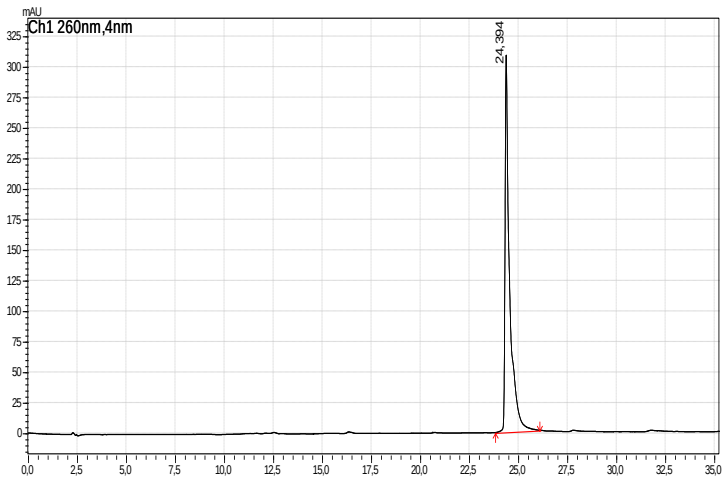

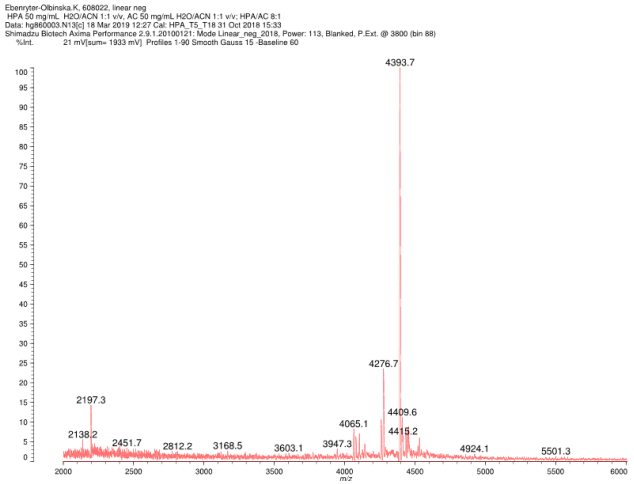

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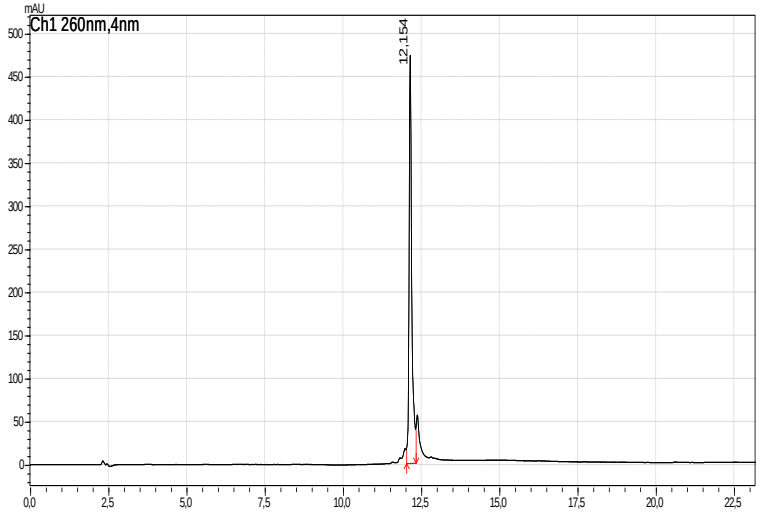
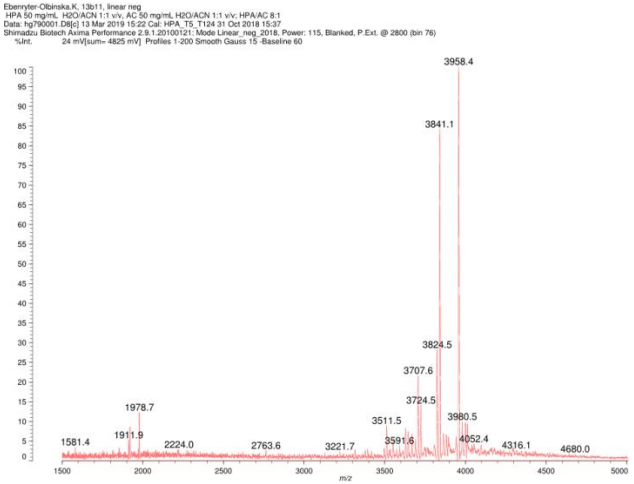
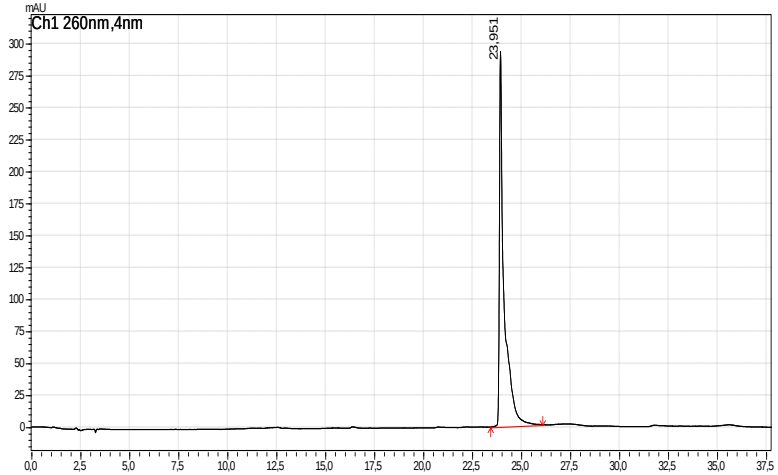
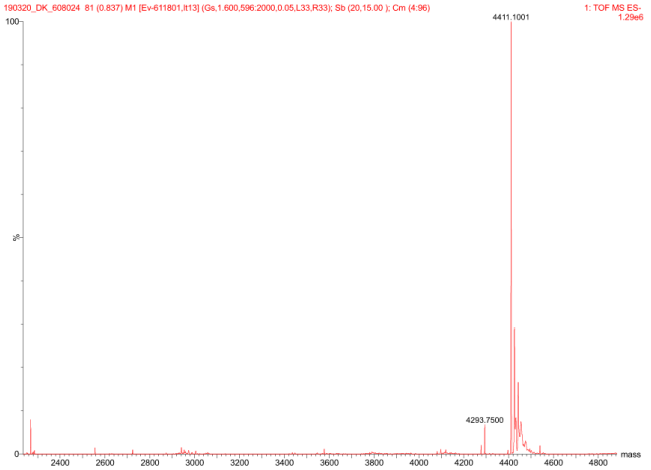


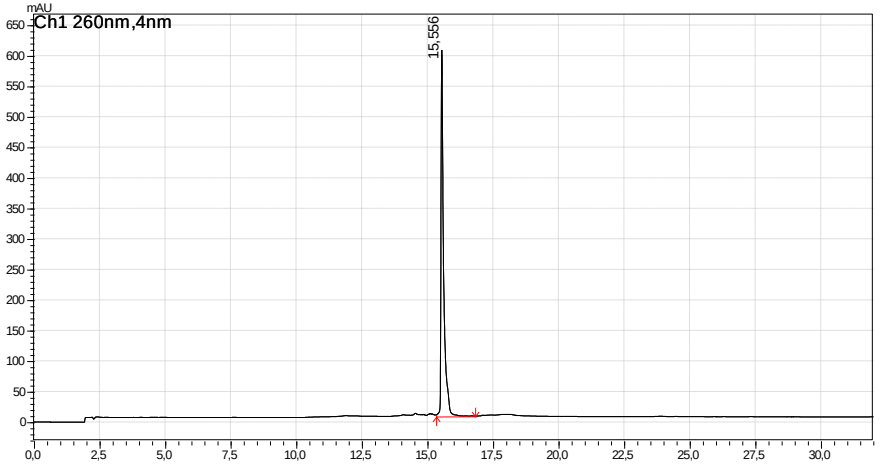


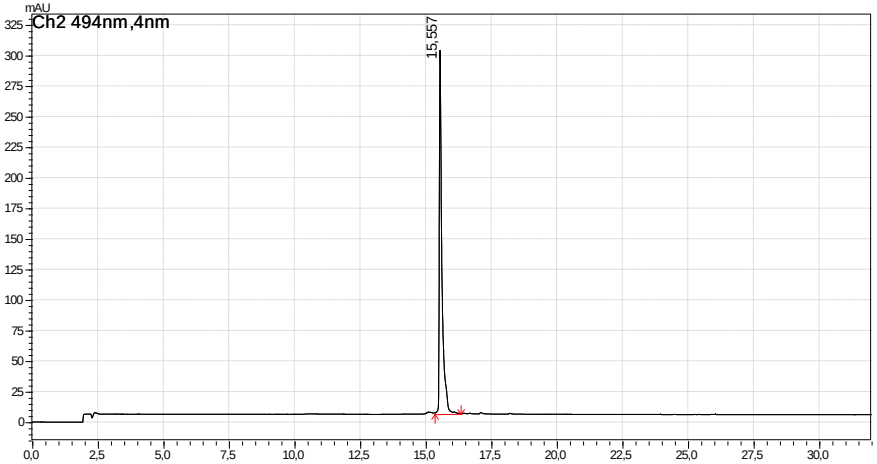
2b 5’-d(CTC CAG AGC C**B**GA)-3’ (*)





FL-1 5’-6FAM-d(TTT CTT TTC CTC C)-3’





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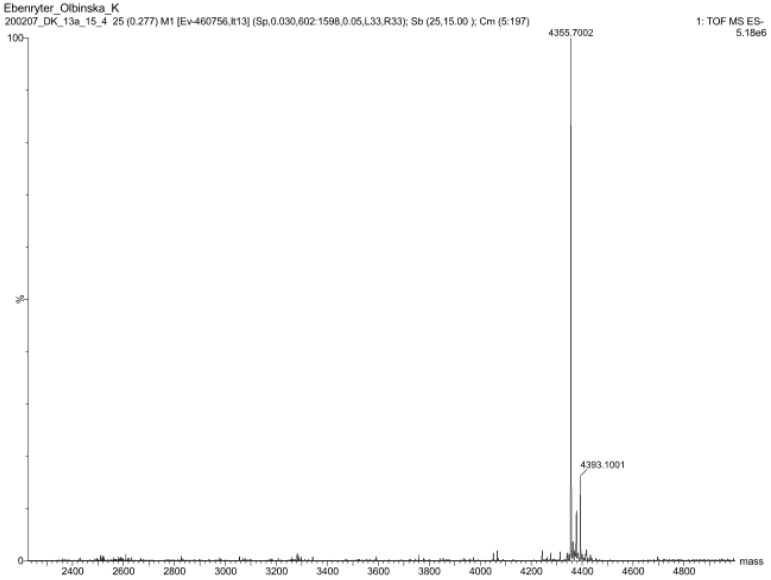
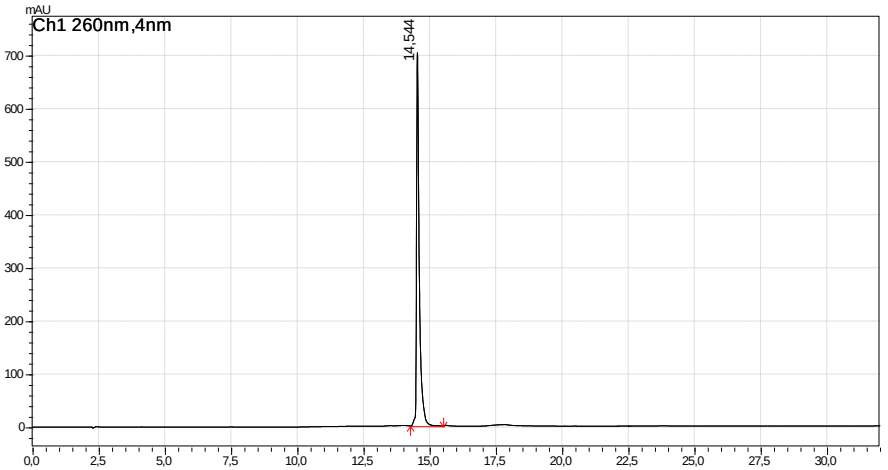
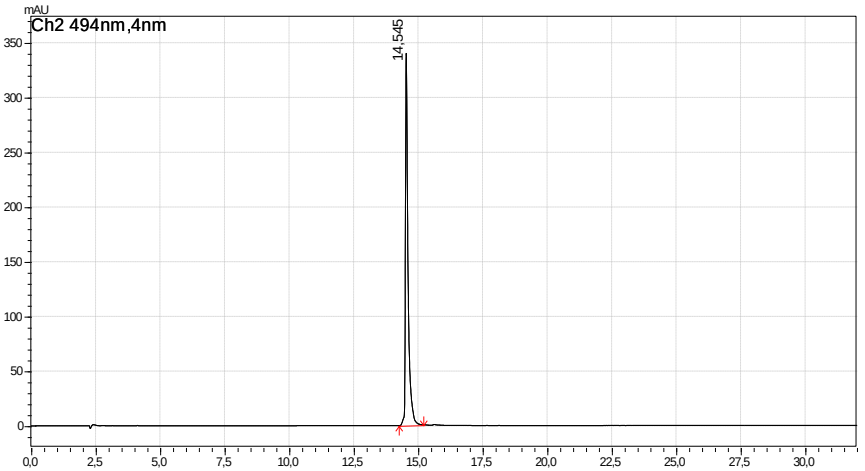

Ebenryter_Olbinska_K
200207_DK_13a_15_4 25 (0.277) M1 [Ev-460756,It13] (Sp,0.030,602:1598,0.05,L33,R33); Sb (25,15.00); Cm (5:197)
1: TOF MS ES-
5.18e6
4355.7002
4393.1001
100
%
0
2400
2600
2800
3000
3200
3400
3600
3800
4000
4200
4400
4600
4800
mass

FL-2 5’-6FAM-d(CTC CAG AGC CCG A)-3’





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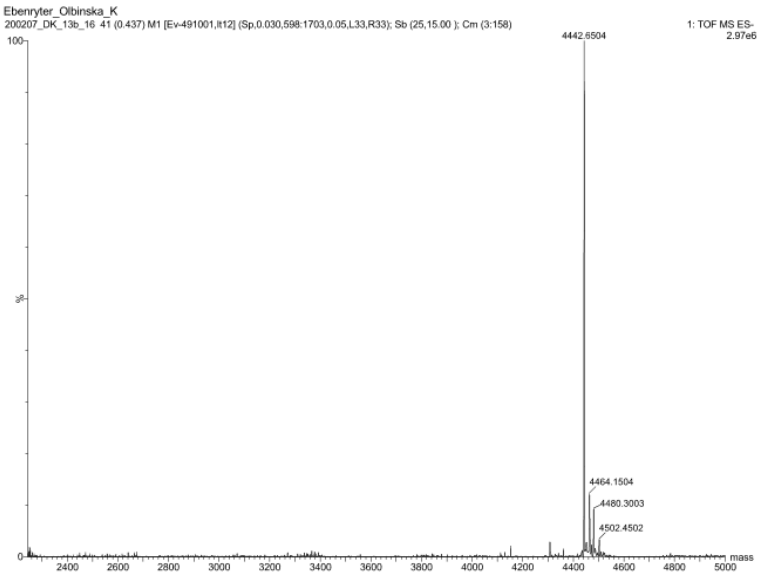


Figure S2. PAGE analysis of the **1, 1a, 1b, 1c** and **2, 2a, 2b** oligonucleotides (20%/7 M urea)

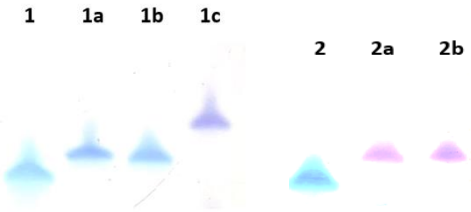


Figure S3. svPDE - assisted hydrolysis of reference oligonucleotide **1** with and without magnesium ions, monitored by MALDI-TOF mass spectrometry.

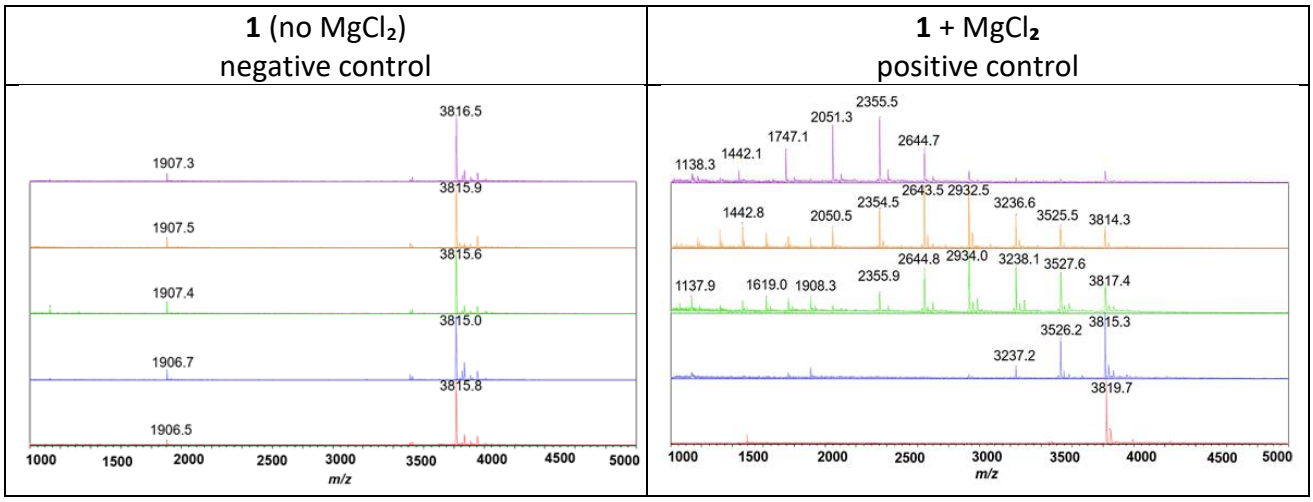
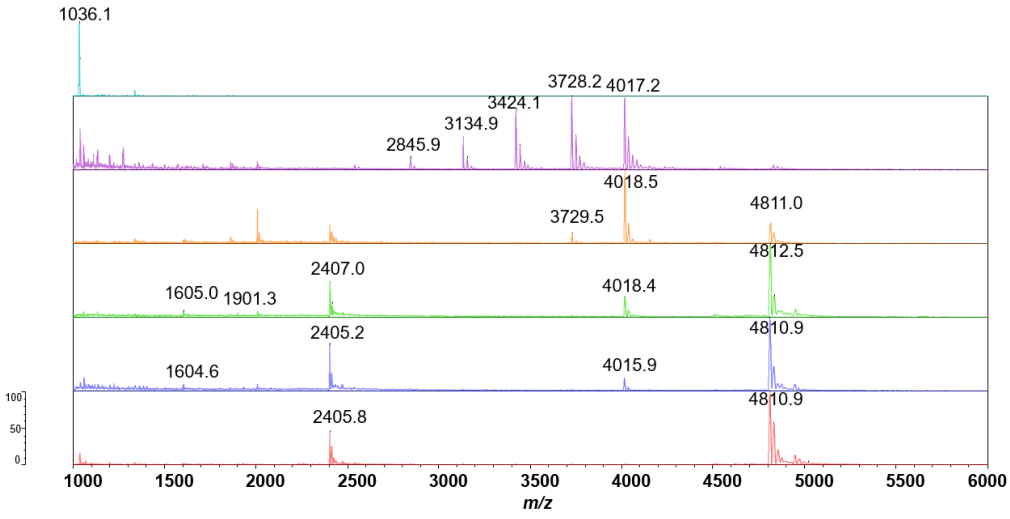
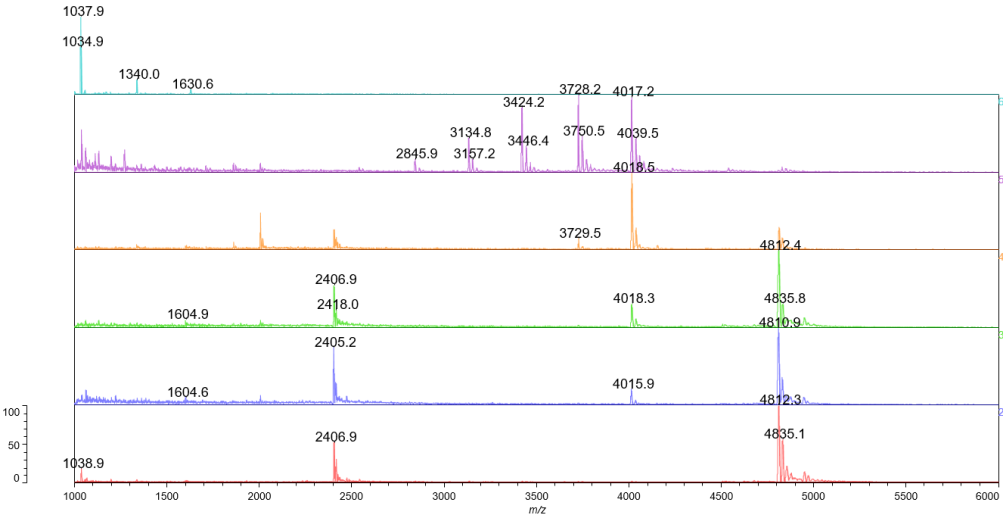


Figure S4. Original MALDI-TOF MS spectra of hydrolysis of **1c** in the presence of svPDE (in triplicate)

i) First experiment



ii) second experiment



iii) third experiment

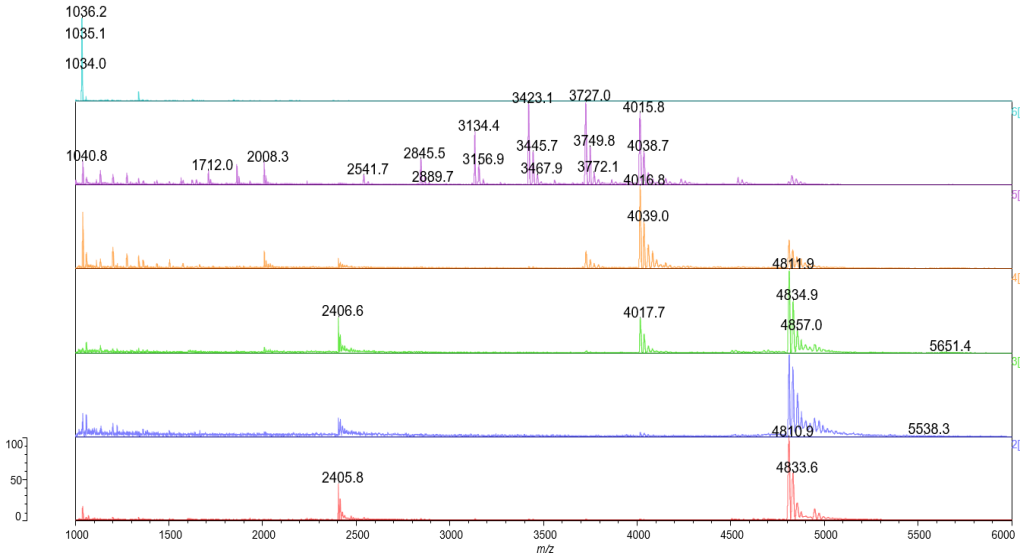


Figure S5. MALDI-TOF mass spectrometry analysis of the 1:1:1 mixture of **1, 1a** and oligonucleotide **1** containing two FESAN clusters (synthetic data not shown). Intensity of peaks is given on the Figure.

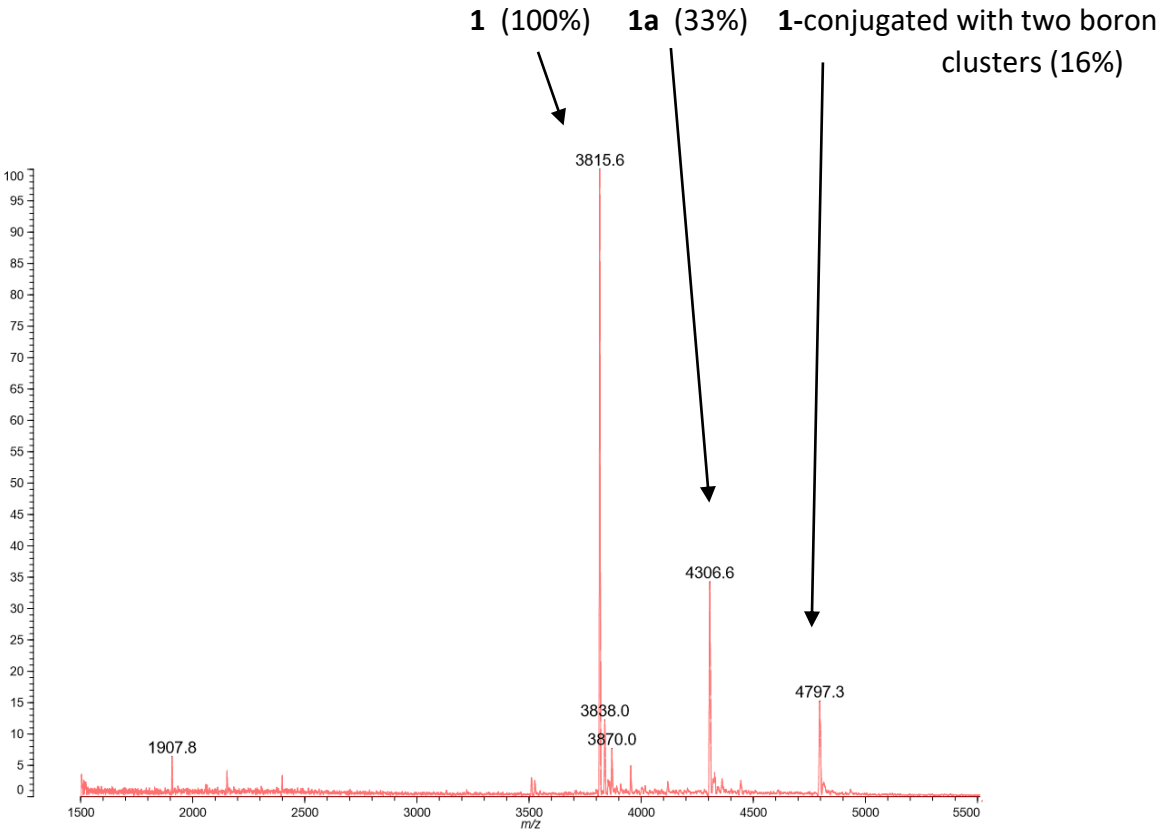
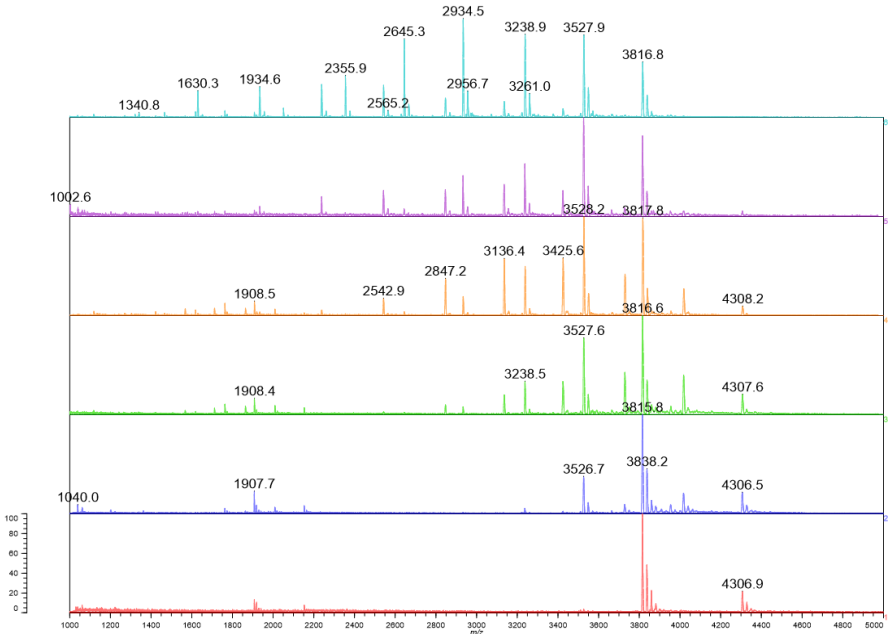
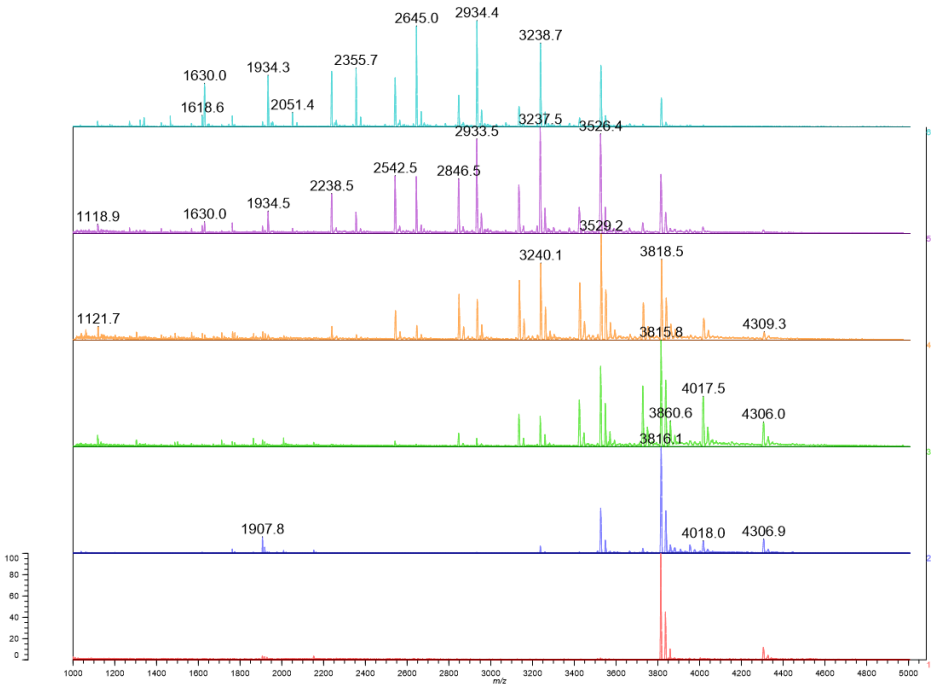


Figure S6. Figure S6. Original MALDI-TOF MS spectra of the hydrolysis mixture of **1**+**1a** in the presence of svPDE (in triplicate).

first experiment



i) second experiment



ii) third experiment

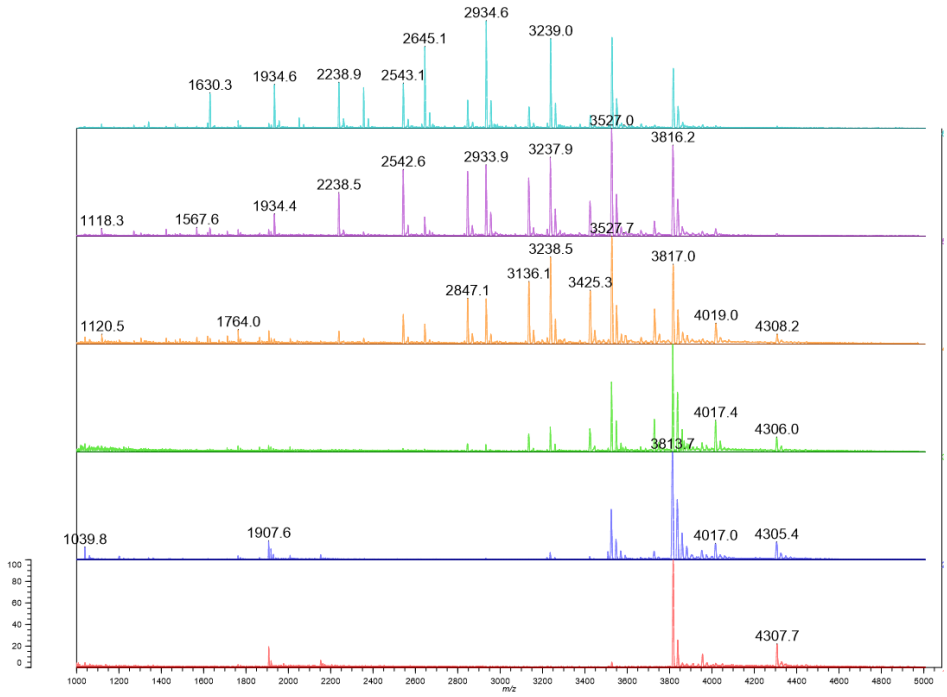


Figure S7. MALDI-TOF MS analysis of the reaction mixture of svPDE-assisted hydrolysis of unmodified oligonucleotides **1** and **2** (0.1 OD) in the presence of a free metallacarborane (FESAN, 182 nM) at 0-60 min.

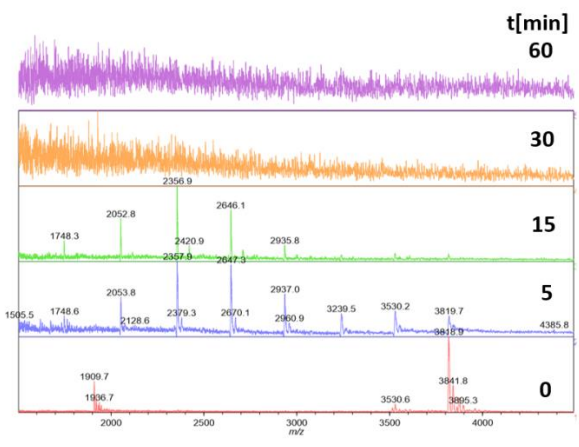
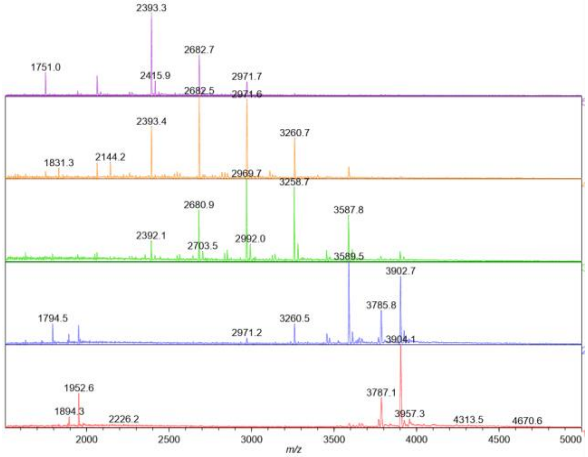
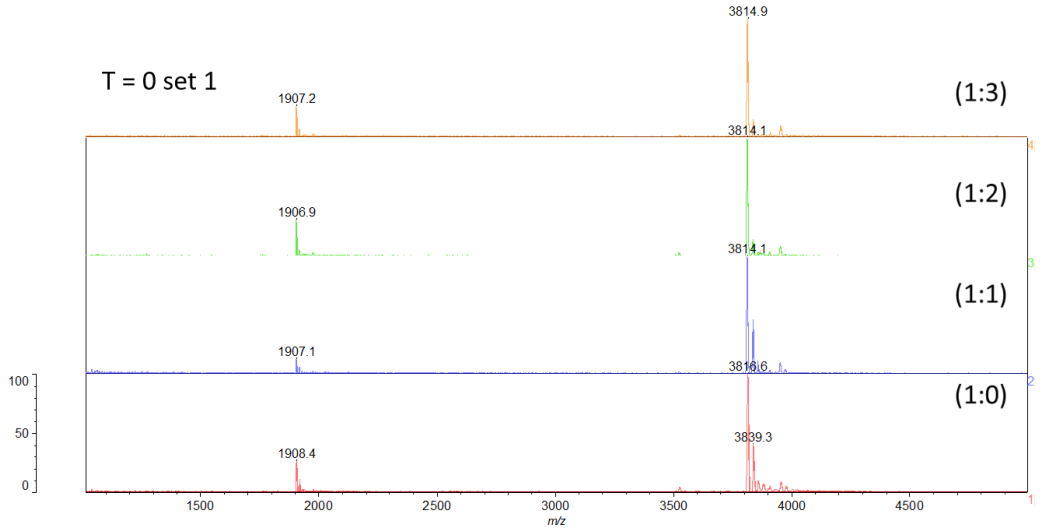
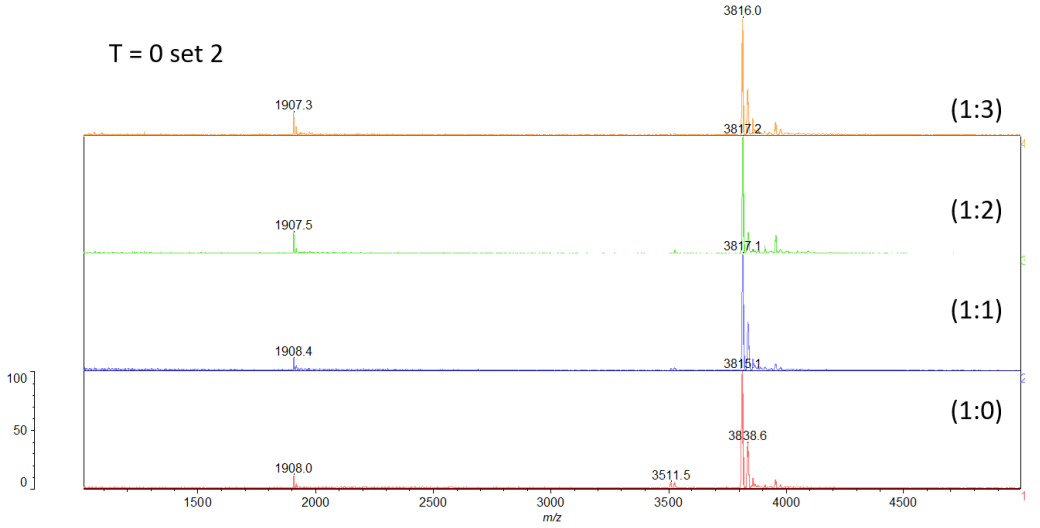
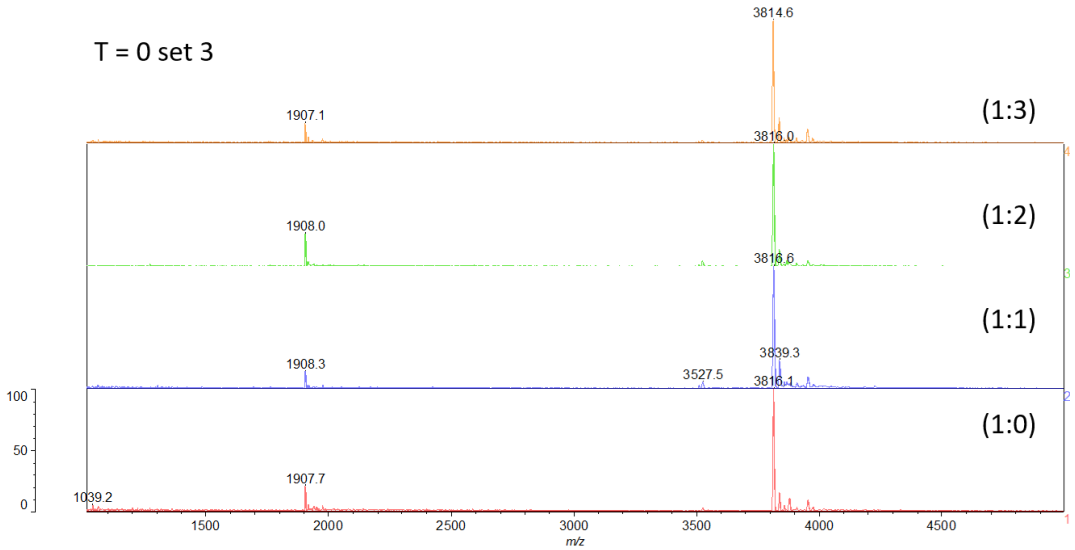
1 + FESAN	**2 + FESAN**
	

Figure S8. MALDI-TOF analysis of the hydrolysis rate of oligonucleotide **1** in the svPDE-assisted reaction carried out in the presence of 0, 1, 2 or 3 equivalents of metallacarborane (in triplicate)

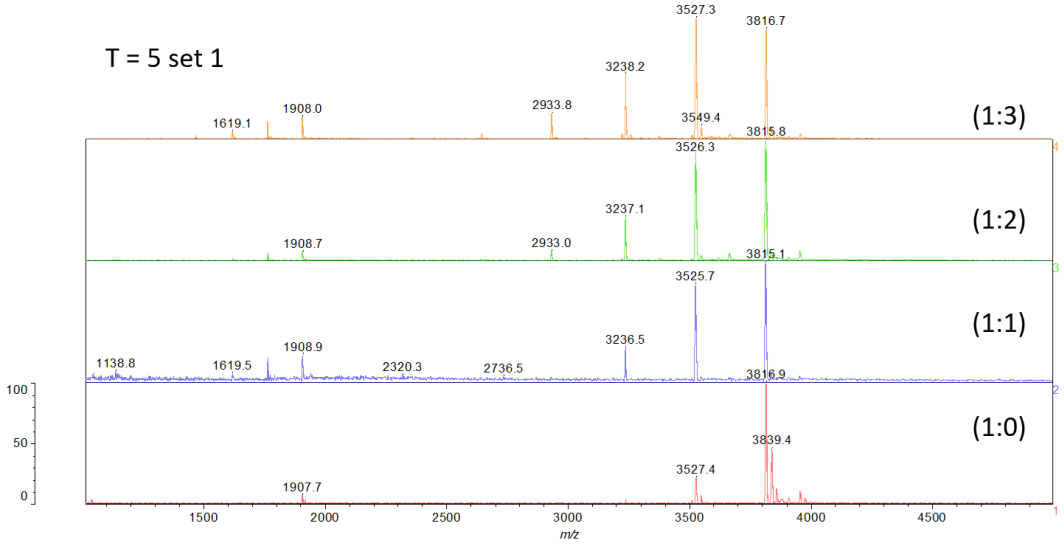
i) measurements at 0 min.

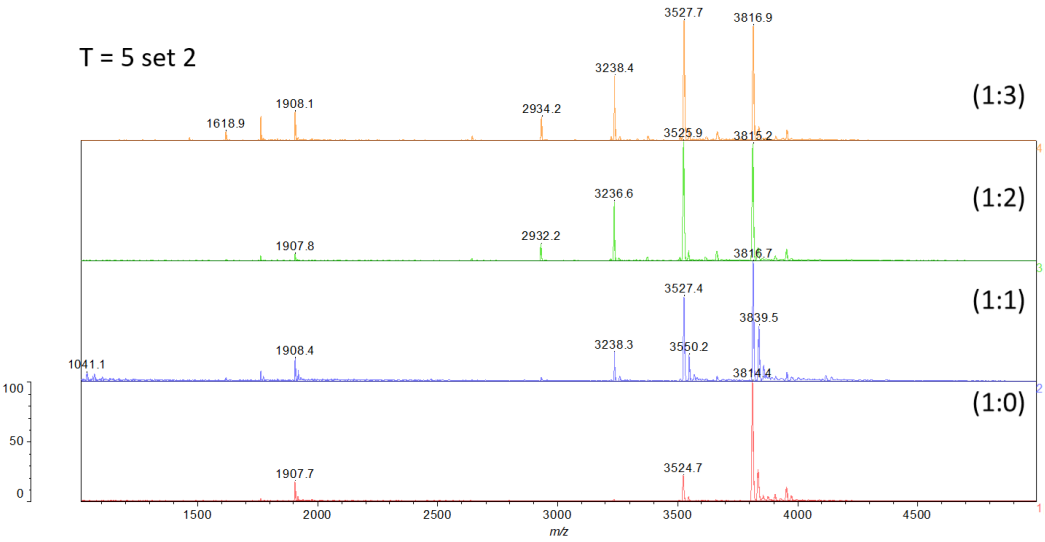


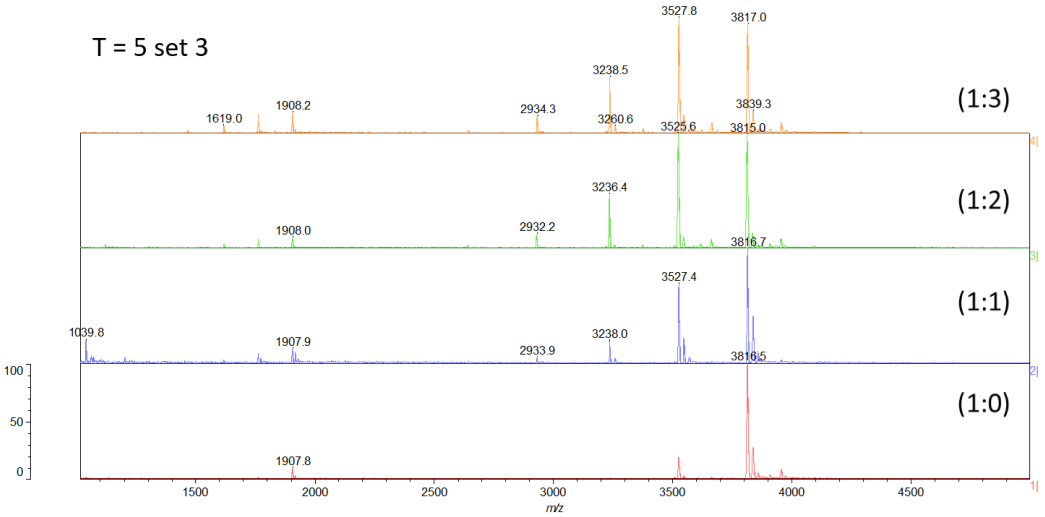




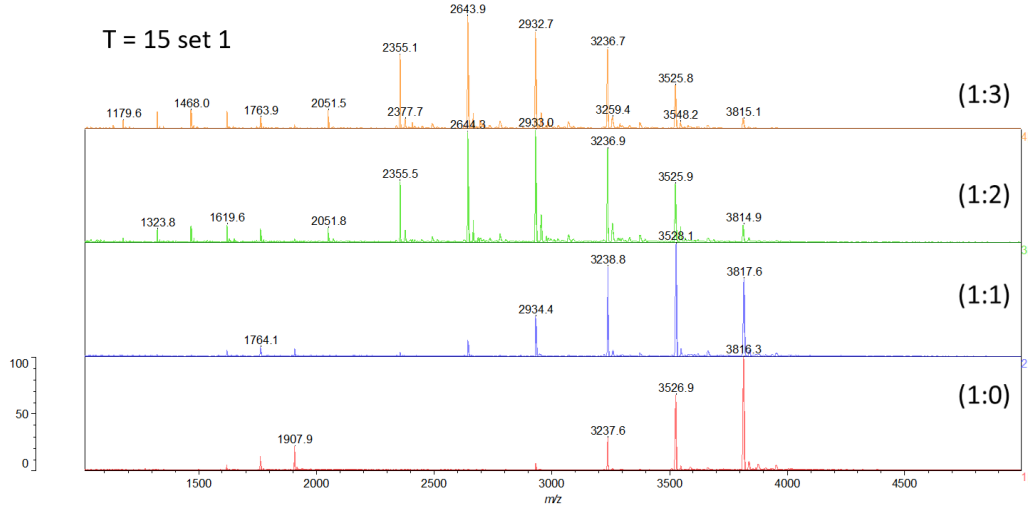
ii) Measurements at 5 min.

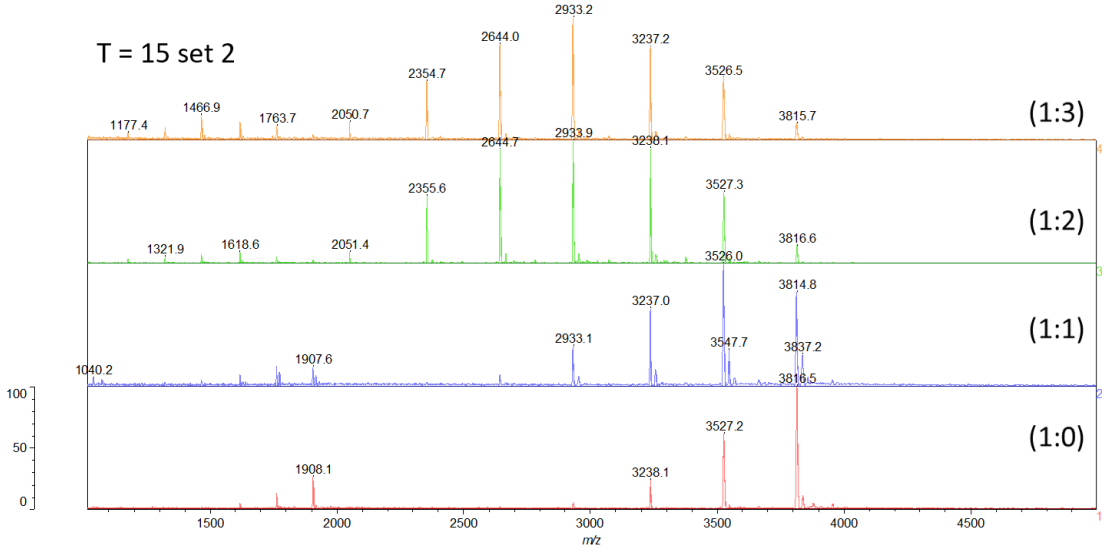






iii) Measurements at 15 min.





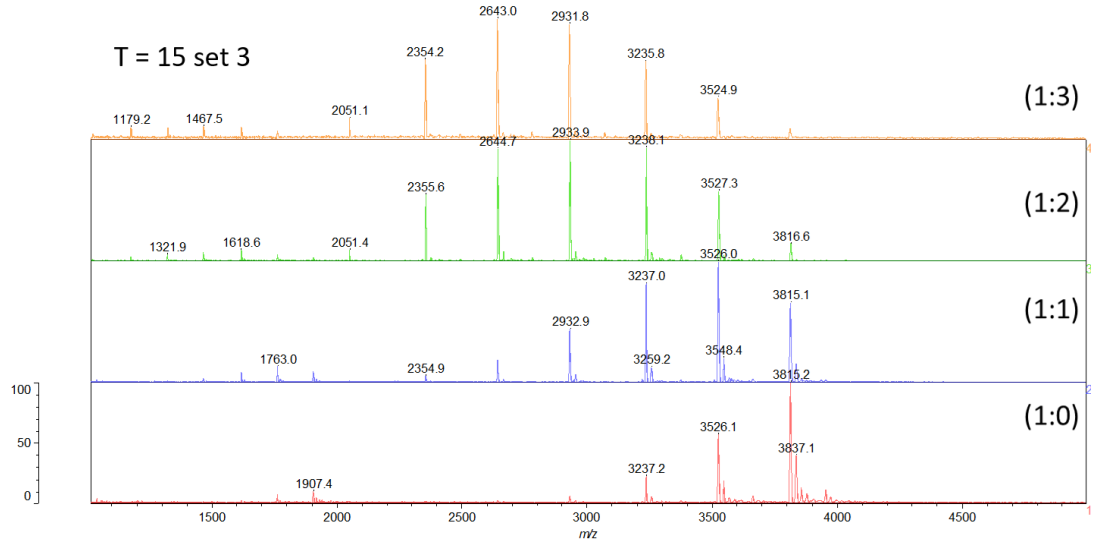


Figure S9. PAGE analysis of crude snake venom.
Lane 1 – molecular weight lader,
lane 2 – the sample of crude svPDE of MW 112 kDa (marked).

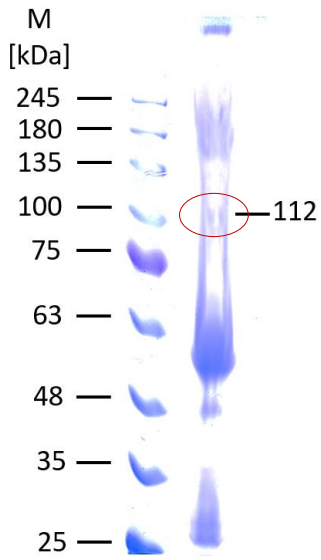


Figure S10. The protein samples capillary scan from microscale thermophoresis (the venom proteins fluorescently labeled with RED-NHS 2nd generation).

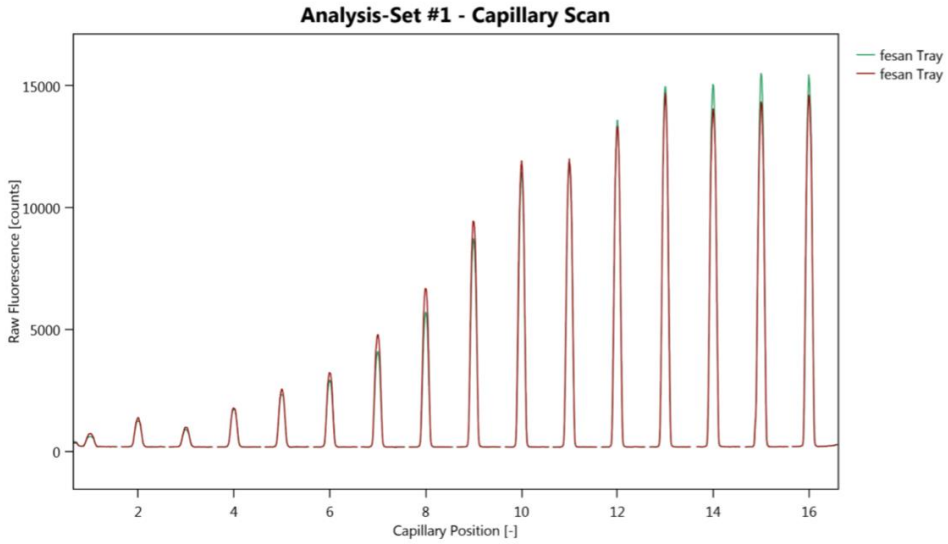


Figure S11. MST data for K_d determination of affinity of FESAN (ferra(III) bis(dicarbollide)) to **FL-1** (K_d=5.48±0.20 μM).

Sample Information	
Merge-Set Name:	Fesan Tray
Date of Measurement:	2020-02-07 11:51:19.022 → 2020-02-07 15:36:37.773
Capillary Type:	Monolith NT.115 Standard Treated Capillary
Target:	1-FL
TargetConcentration:	0,00000001
Ligand:	Fesan
Measurement Settings	
MST-Power:	Medium
Excitation-Power:	20%
Excitation type:	Green
Thermostat Setpoint:	22,0°C
Fit Results (Kd)	
Fit Model:	Kd
Bound	903,4445704
Unbound	~~923,7399589~~
Kd	5,4834E-06
TargetConc	~~0,00000001~~ 🔒
Standard Deviation:	0,207569137
Kd Confidence:	±4,34262E-06