OMTN, Volume *9*

Supplemental Information

The Bioactivity of D-/L-Isonucleoside- and 2′-Deoxyinosine-Incorporated Aptamer AS1411s Including DNA Replication/MicroRNA Expression

Xinmeng Fan, Lidan Sun, Kunfeng Li, Xiantao Yang, Baobin Cai, Yanfen Zhang, Yuejie Zhu, Yuan Ma, Zhu Guan, Yun Wu, Lihe Zhang, and Zhenjun Yang

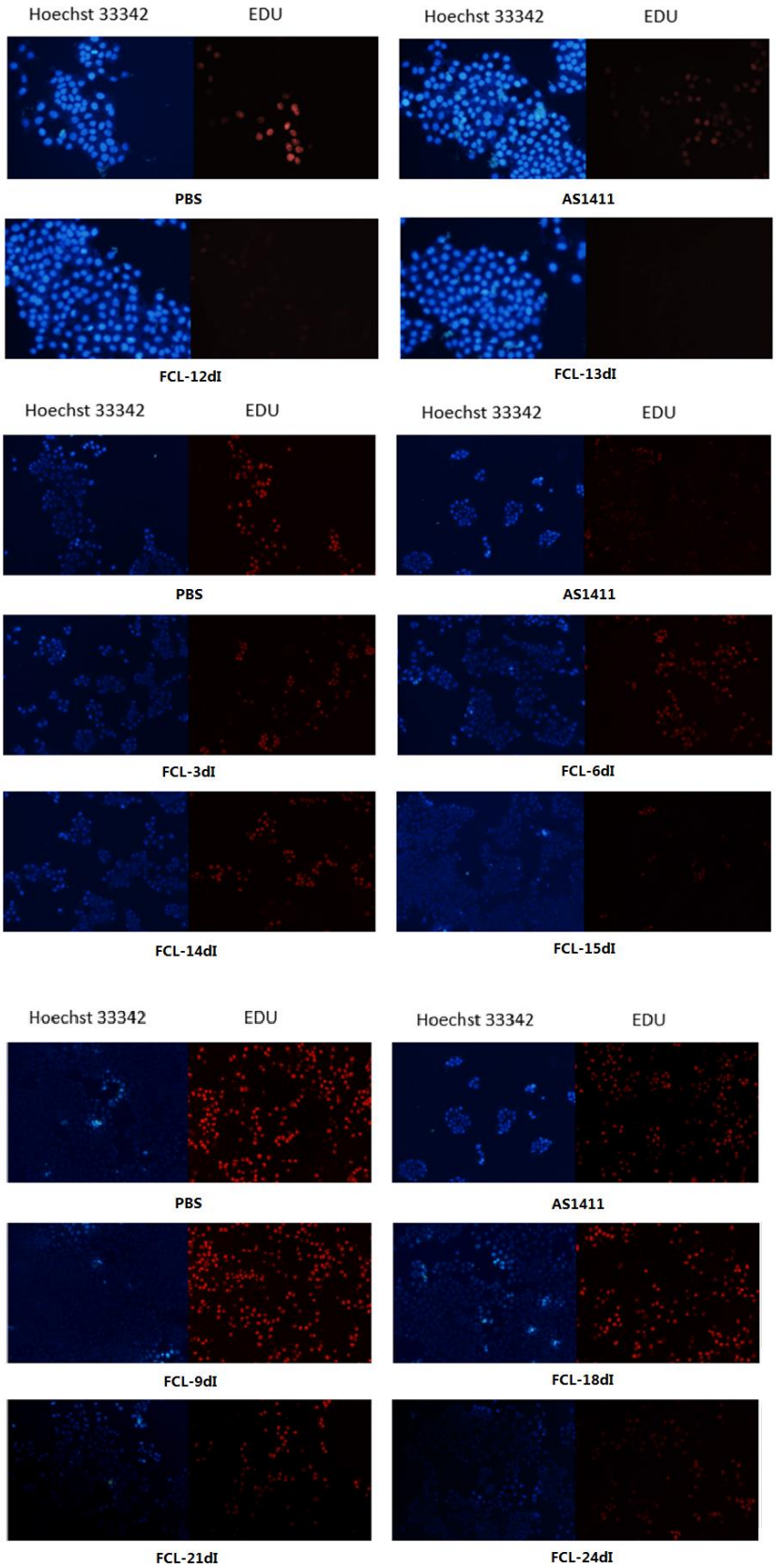


Figure S1. DNA synthesis in untreated MCF-7 cells (PBS as control) and cells treat with **AS1411** (control oligonucleotide) or **2′-dI** incorporated **AS1411** (active oligonucleotide). Cells are treated a final concentration of 18 μM for 72 h and then expose to 50 μM EdU for 2 h at 37 ℃.

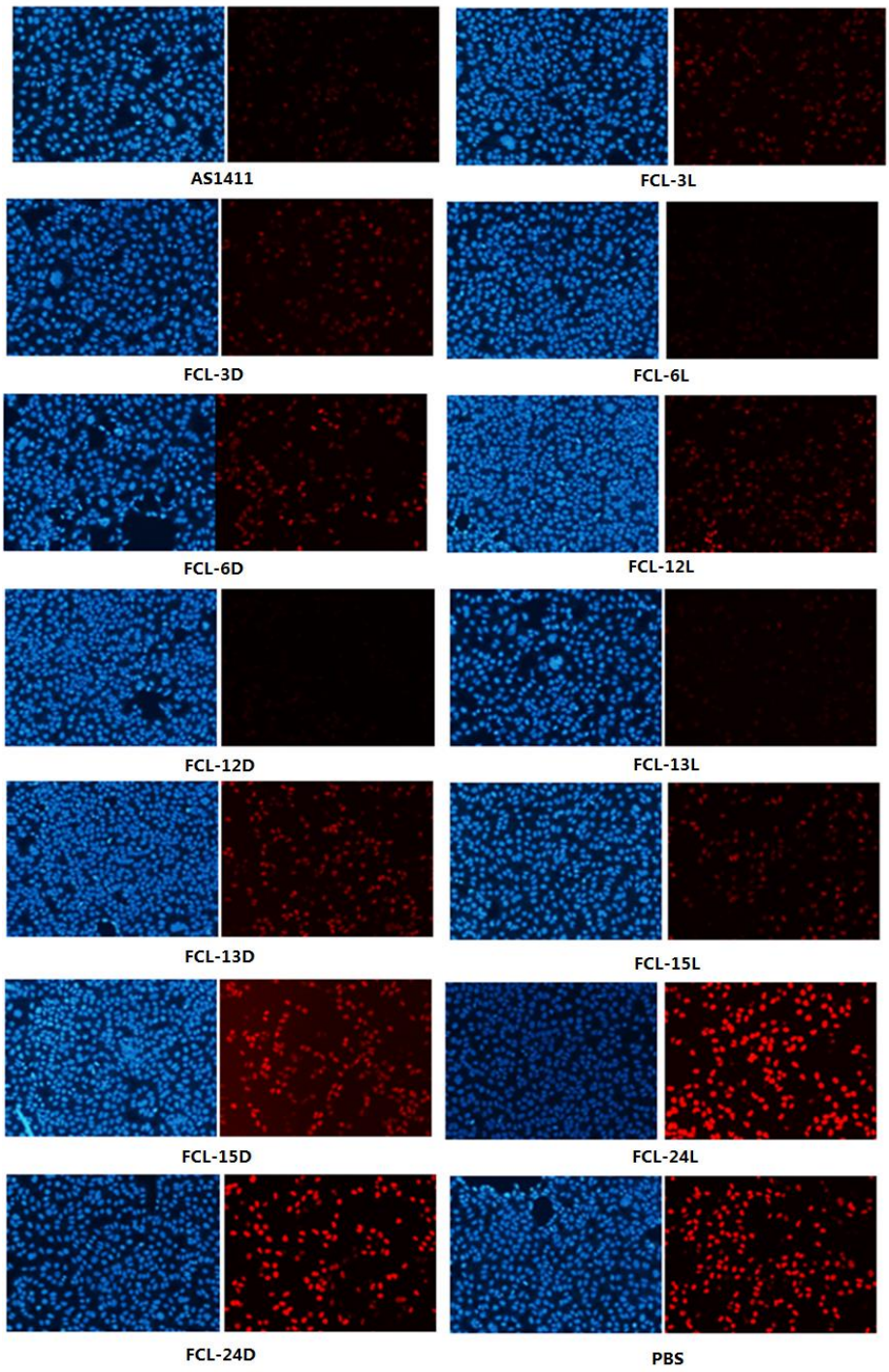


Figure S2. DNA synthesis in untreated MCF-7 cells (PBS as control) and MCF-7 cells treat with **AS1411** (control oligonucleotide) or unit **isoNA** modified **AS1411** (active oligonucleotide). Cells are treated a final concentration of 18 μM for 72 h and then expose to 50 μM EdU for 2 h at 37 ℃. DNA is stained with 5 μg/mL Hoechst 33342 (50 μL per well) for 30 min and images under a fluorescent microscope.

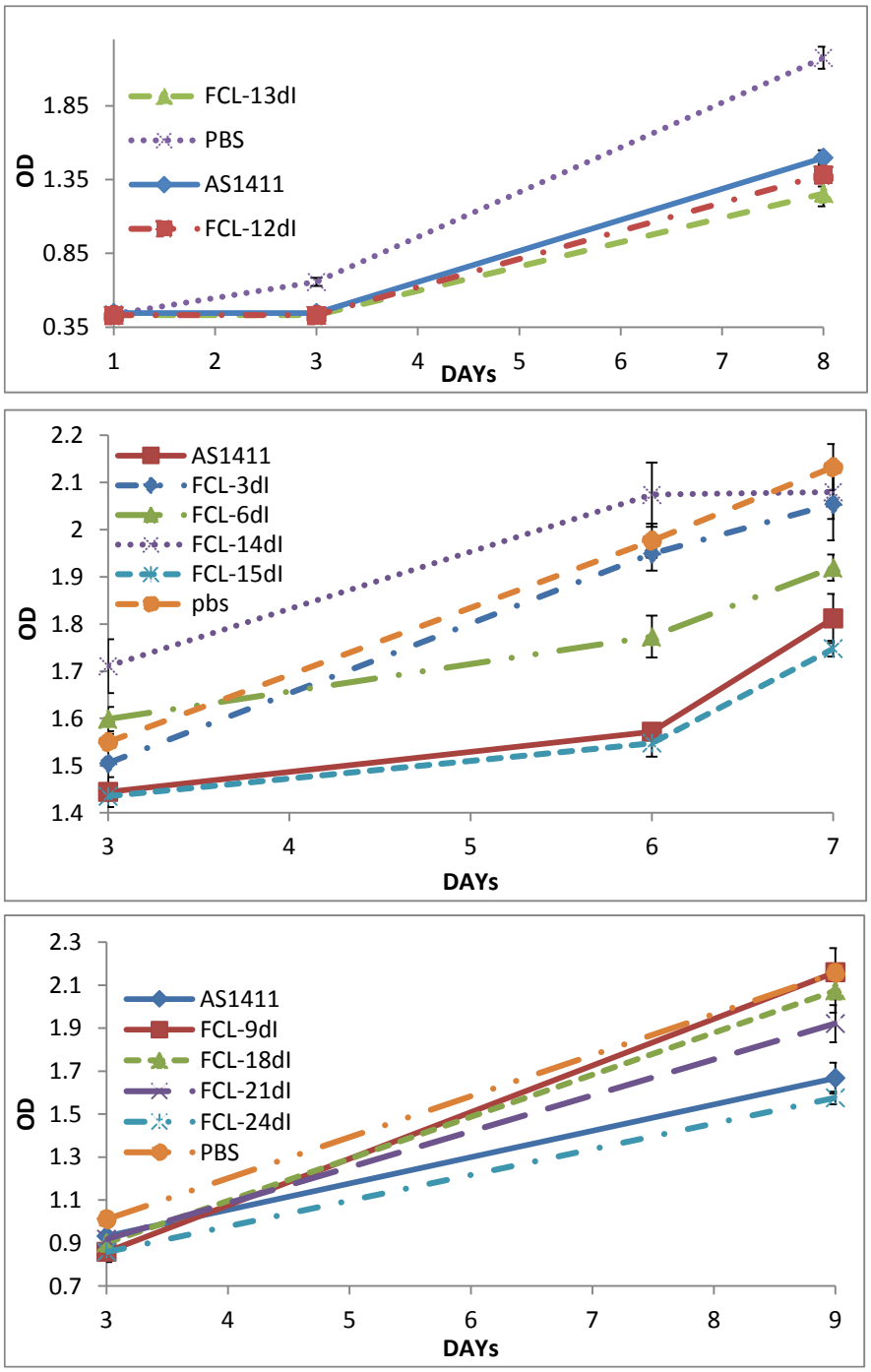


Figure S3. CCK-8 assays showing the growth of MCF-7 cells treated with **2′-dI** incorporated **AS1411** or PBS as a control. Oligonucleotides (or PBS as control) are added directly to the culture medium to give a final concentration of 15 μM (day 1). On days 2-4 further oligonucleotide equivalent to half the initial dose is added. The OD_{450} (nm) value is proportional to the number of viable cells in the sample.

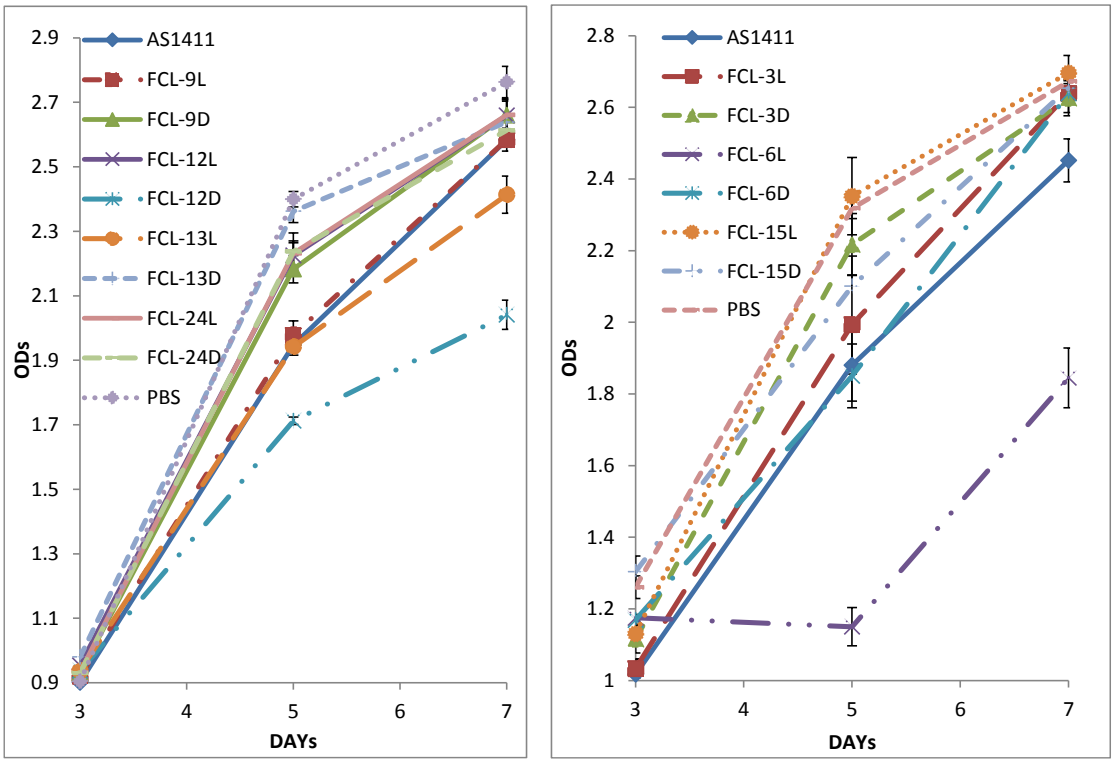


Figure S4. CCK-8 assays showing the growth of MCF-7 cells treated with unit D-/L-**isoT** modified **AS1411** or PBS as a control. Oligonucleotides (or PBS as control) are added directly to the culture medium to give a final concentration of 7.5 μM (day 1). On days 2-4 further oligonucleotide equivalent to half the initial dose is added. Cells are assayed using the cell counting kit-8 (CCK-8) (Dojindo Laboratorie, Japan) on 3, 5, 7 days after treatment. The OD_{450} nm value is proportional to the number of viable cells in the sample.

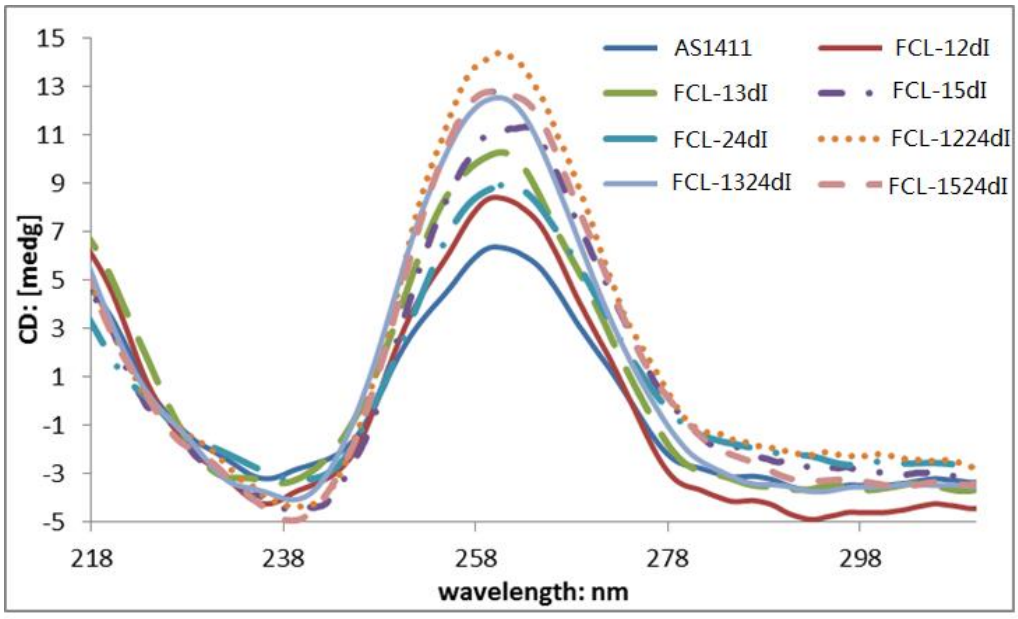
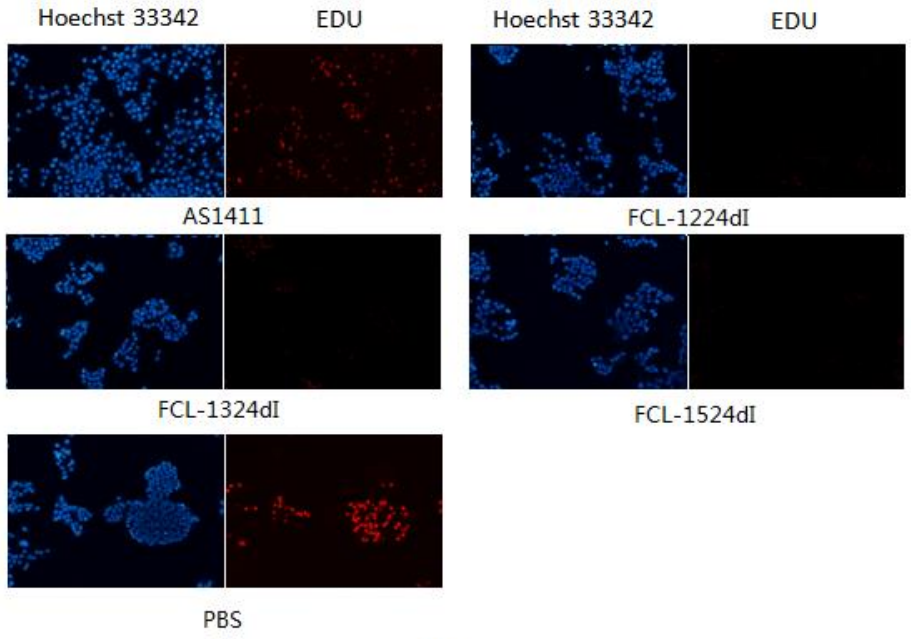
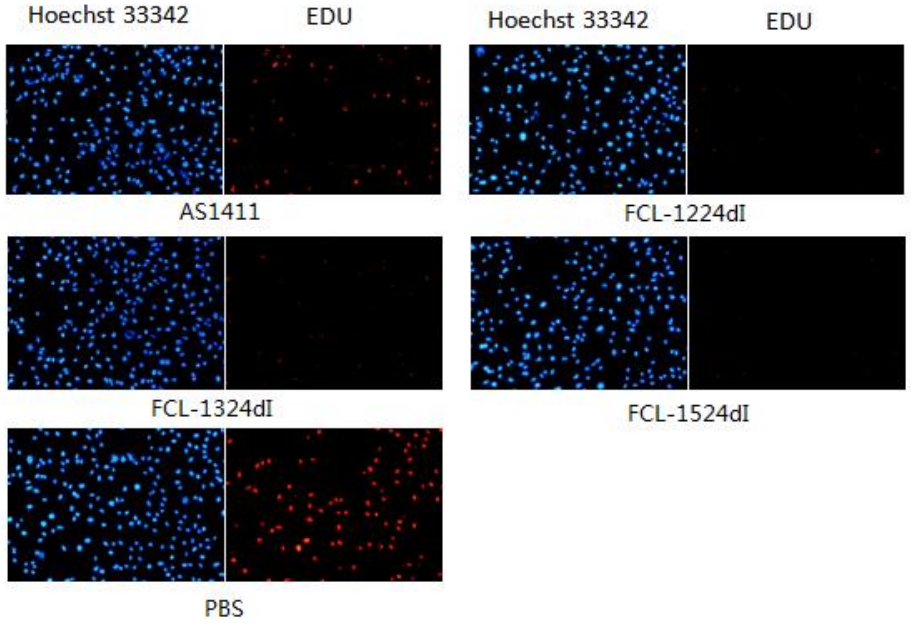


Figure S5. CD spectra of **2′-dI** modified **AS1411**. CD data is obtained with a 5 μM concentration in the presence of in 10 mM sodium phosphate buffer, pH 7.0, containing 0.1 M KCl. All aptamers are boiled for 5 min, and anneal at 60 °C for 50 h.



(A)



(B)

Figure S6. DNA synthesis in untreated 2 kind of cells (PBS as control) and cells treat with **AS1411** (control oligonucleotide) or **FCL-1224dI/ FCL-1324dI/ FCL-1524dI** (active oligonucleotide). Cells are treated a final concentration of 18 μM for 72 h and then expose to 50 μM EdU for 2 h at 37 ℃. DNA is stained with 5 μg/mL Hoechst 33342 (50 μL per well) for 30 min and images under a fluorescent microscope. (A) MCF-7 cells. (B) MDA-MB-231 cells.

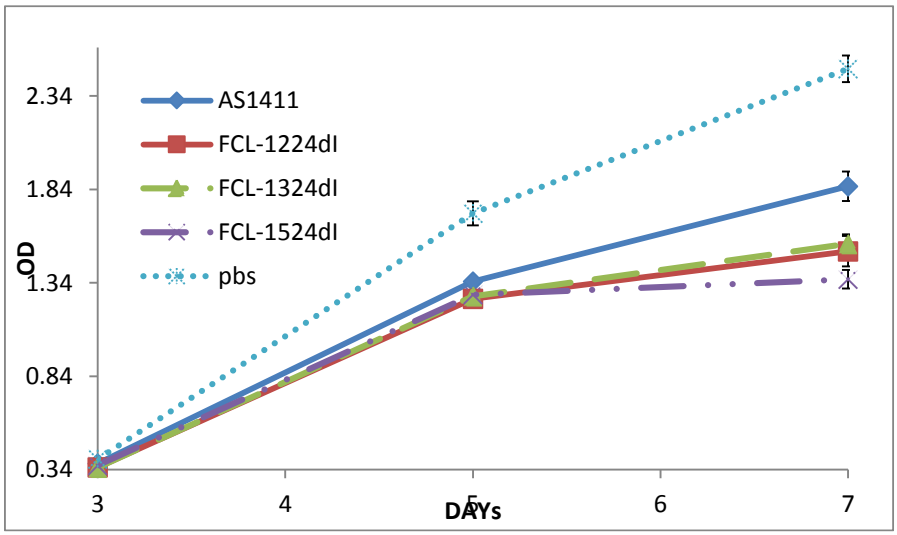


Figure S7. CCK-8 assays showing the growth of MCF-7 cells treated with **FCL-1224dI, FCL-1324dI, FCL-1524dI** or PBS as a control. Oligonucleotides (or PBS as control) are added directly to the culture medium to give a final concentration of 7.5 μM (day 1). On days 2-4 further oligonucleotide equivalent to half the initial dose is added. Cells are assayed using the cell counting kit-8 (CCK-8) (Dojindo Laboratorie, Japan) on 3, 5, 7 days after treatment. The OD_{450} nm value is proportional to the number of viable cells in the sample.

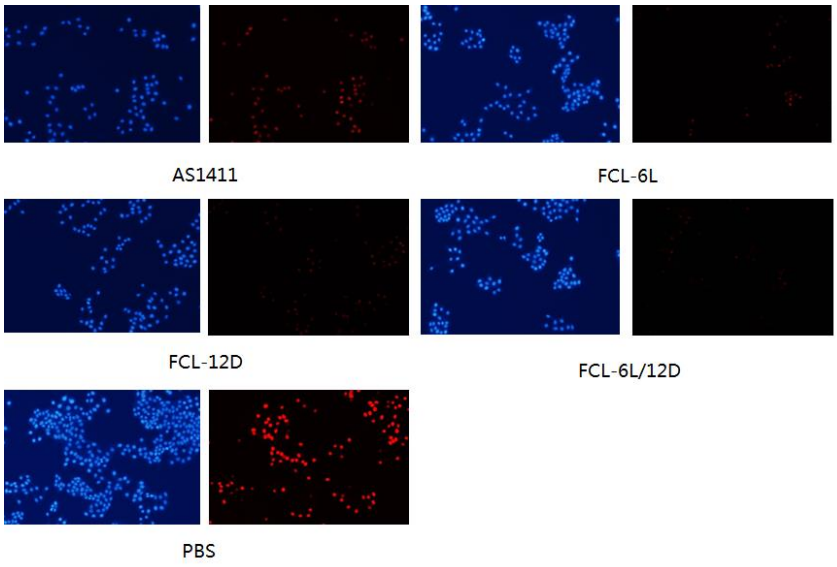


Figure S8. DNA synthesis in untreated MCF-7 cells (PBS as control) and cells treat with **AS1411** (control oligonucleotide) or **FCL-I** (active oligonucleotide). Cells are treated a final concentration of 18 μM for 72 h and then expose to 50 μM EdU for 2 h at 37 ℃. DNA is stained with 5 μg/mL Hoechst 33342 (50 μL per well) for 30 min and images under a fluorescent microscope.

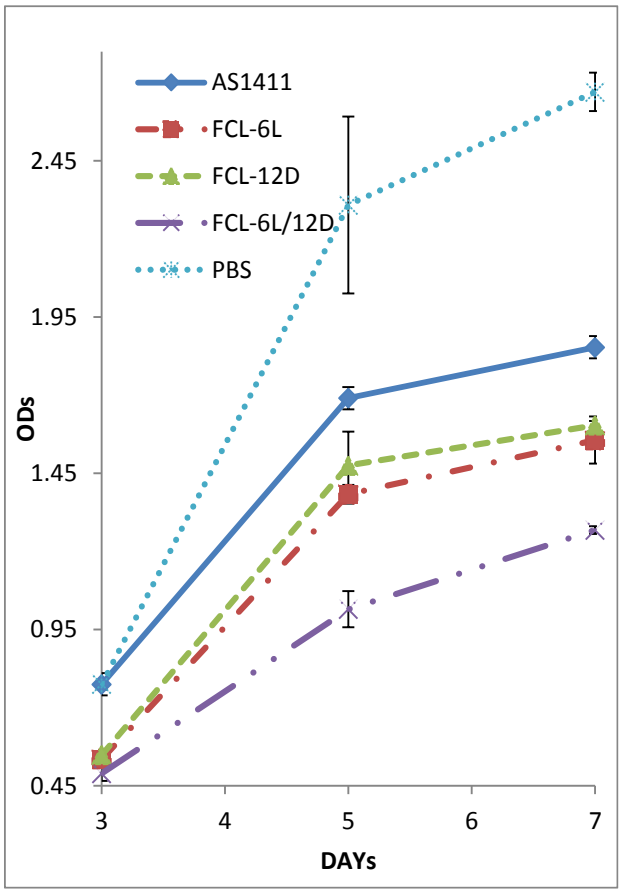
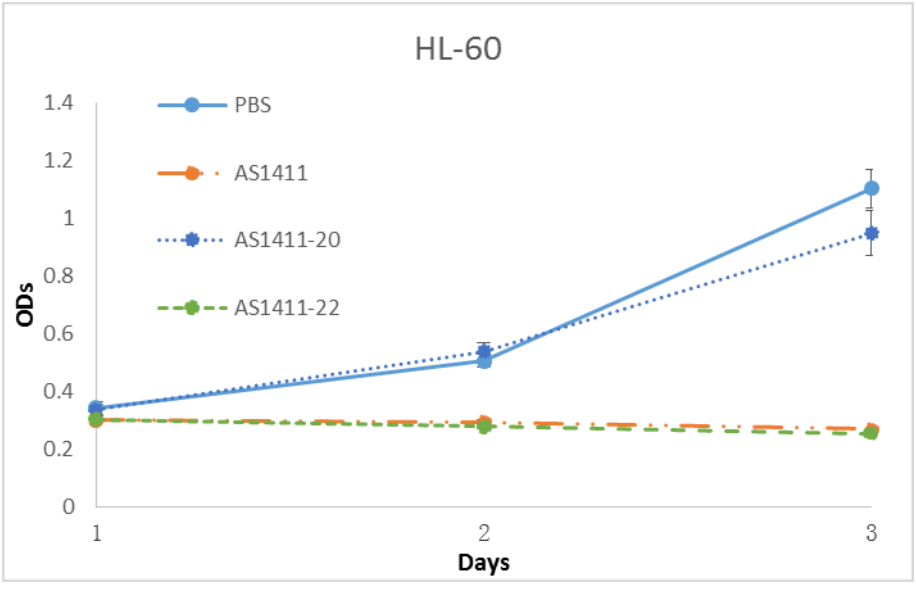


Figure S9. CCK-8 assays showing the growth of MCF-7 cells treated with **FCL-I** or PBS as a control. Oligonucleotides (or PBS as control) are added directly to the culture medium to give a final concentration of 7.5 μM (day 1). On days 2-4 further oligonucleotide equivalent to half the initial dose is added. Cells are assayed using the cell counting kit-8 (CCK-8) (Dojindo Laboratorie, Japan) on 3, 5, 7 days after treatment. The OD_{450} nm value is proportional to the number of viable cells in the sample.



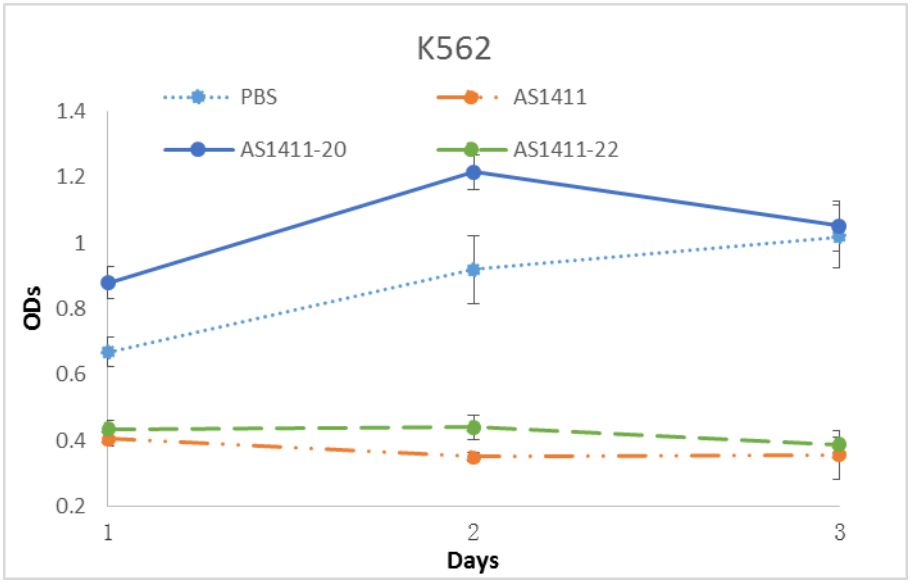


Figure S10. CCK-8 assays showing the growth of HL-60 cells and K562 cells treated with different truncated **AS1411** sequences or PBS as a control. oligonucleotides (or PBS as control) are added directly to the culture medium to give a final concentration of 7.5 μM (day 1). On days 2 and 3 further oligonucleotide equivalent to half the initial dose is added. Cells are assayed using the cell counting kit-8 (CCK-8) (Dojindo Laboratorie, Japan) on 1, 2, 3 days after treatment. The OD_{450} nm value is proportional to the number of viable cells in the sample.

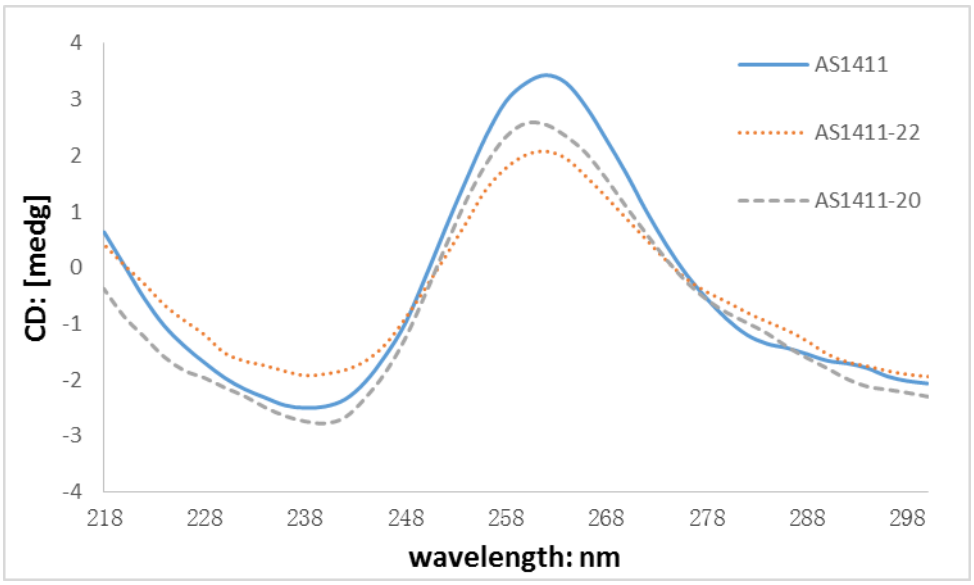


Figure S11. CD spectra of different truncated **AS1411** sequences. CD data is obtained with a 10 μM concentration without annealing.

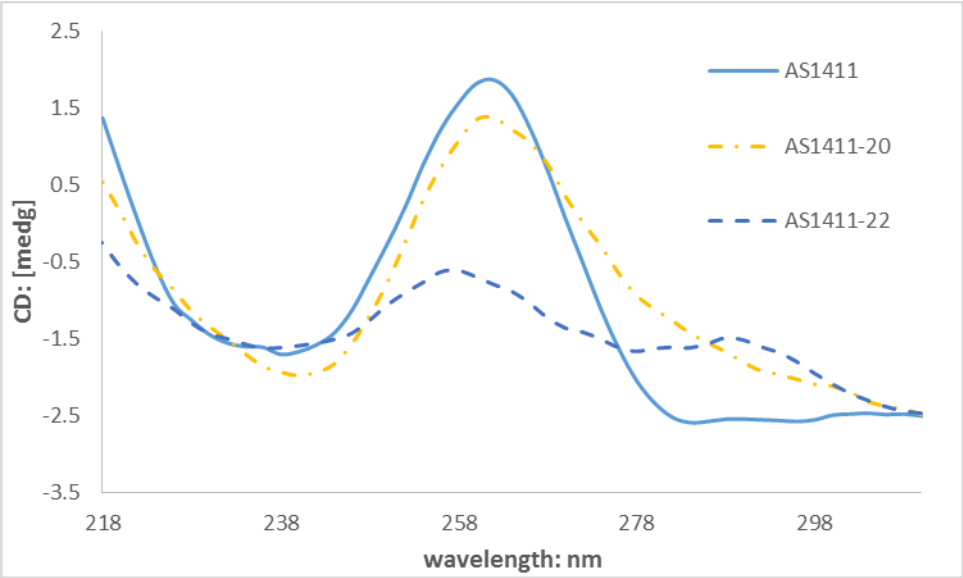


Figure S12. CD spectra of different truncated **AS1411** sequences. CD data is obtained with a 5 μM concentration in the presence of in 10 mM sodium phosphate buffer, pH 7.0, containing 0.1 M KCl. All aptamers are boiled for 5 min, and anneal at 60 ºC for 48 h.

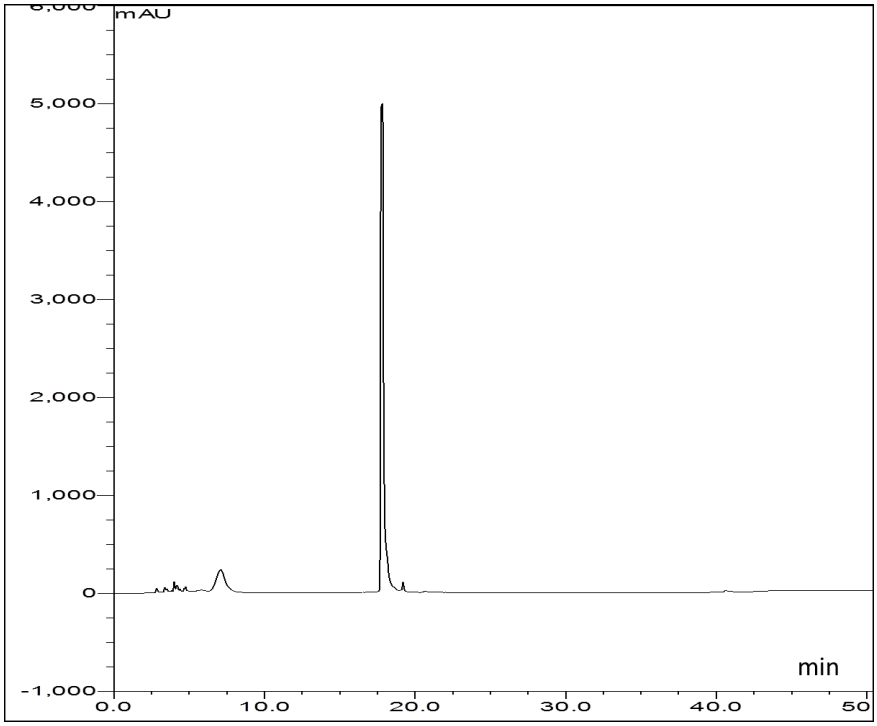


Figure S13. HPLC purification of oligonucleotides, AS1411 as example. (Linear gradient using 5-70% acetonitrile-TEAB 100 mM in 40 min, X-bridge C18 4.6 × 50 mm, 60 ºC, 1.5 mL/min, 260 nm).

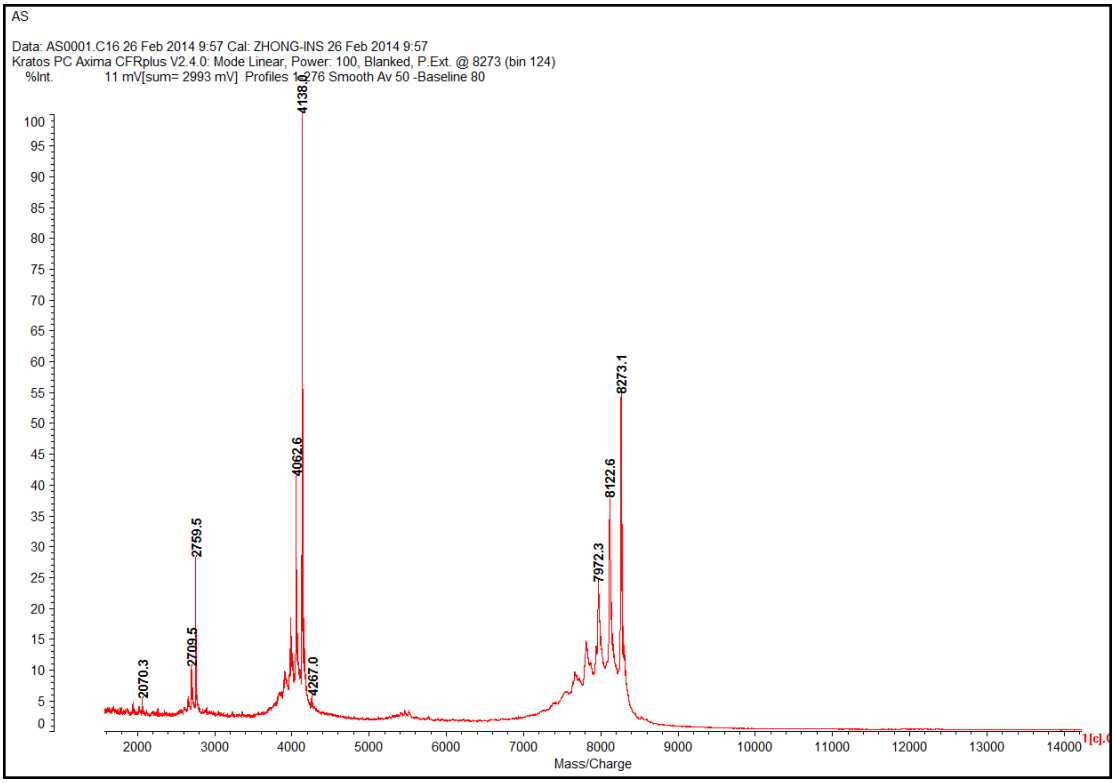


Figure S14. MALDI-TOF spectrum of sequence **AS1411**.

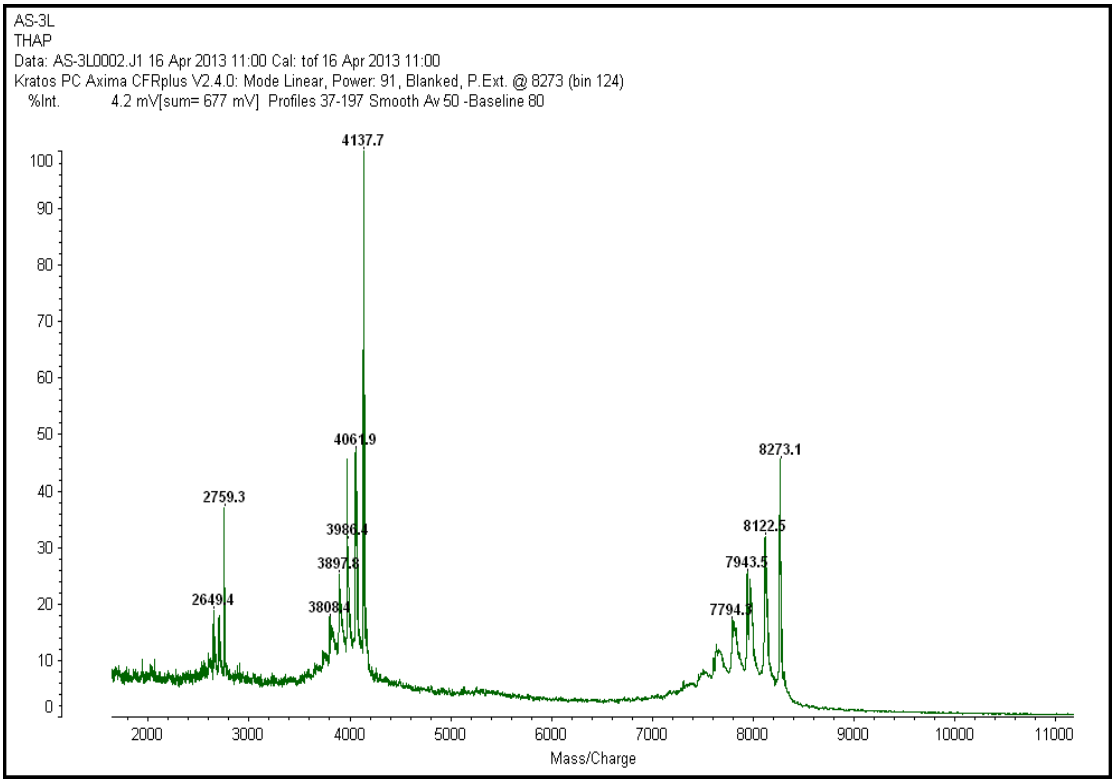


Figure S15. MALDI-TOF spectrum of sequence **AS1411-3L**.

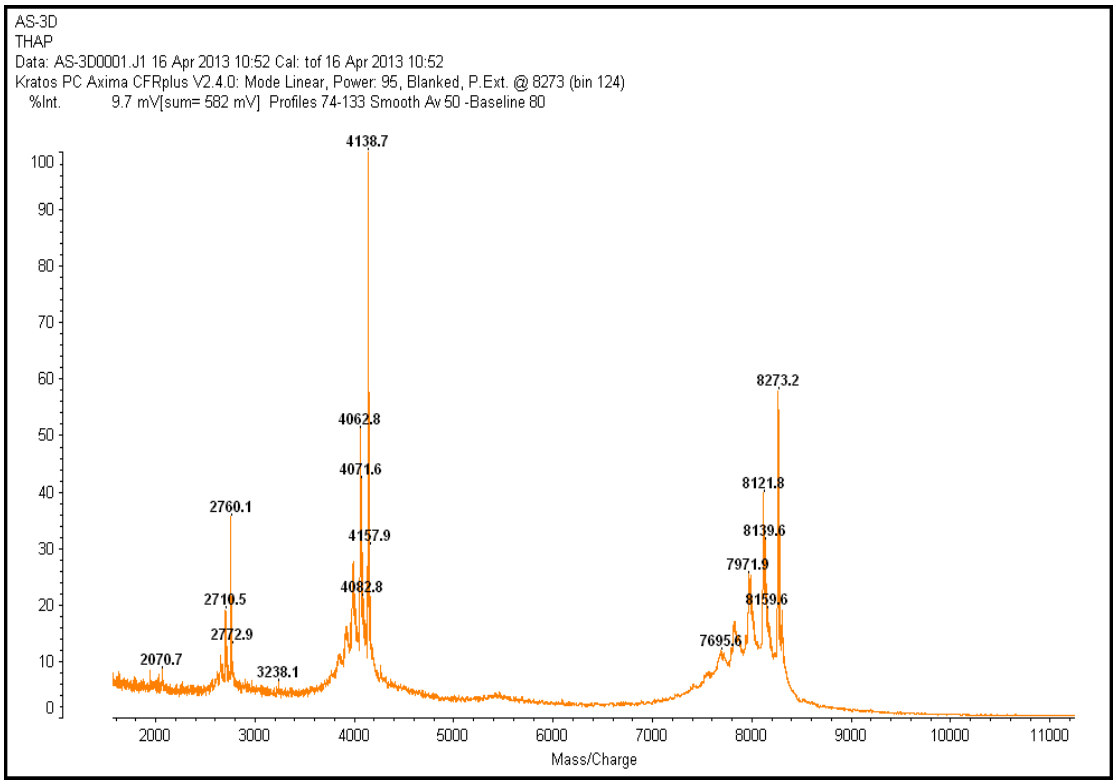


Figure S16. MALDI-TOF spectrum of sequence **AS1411-3D**.

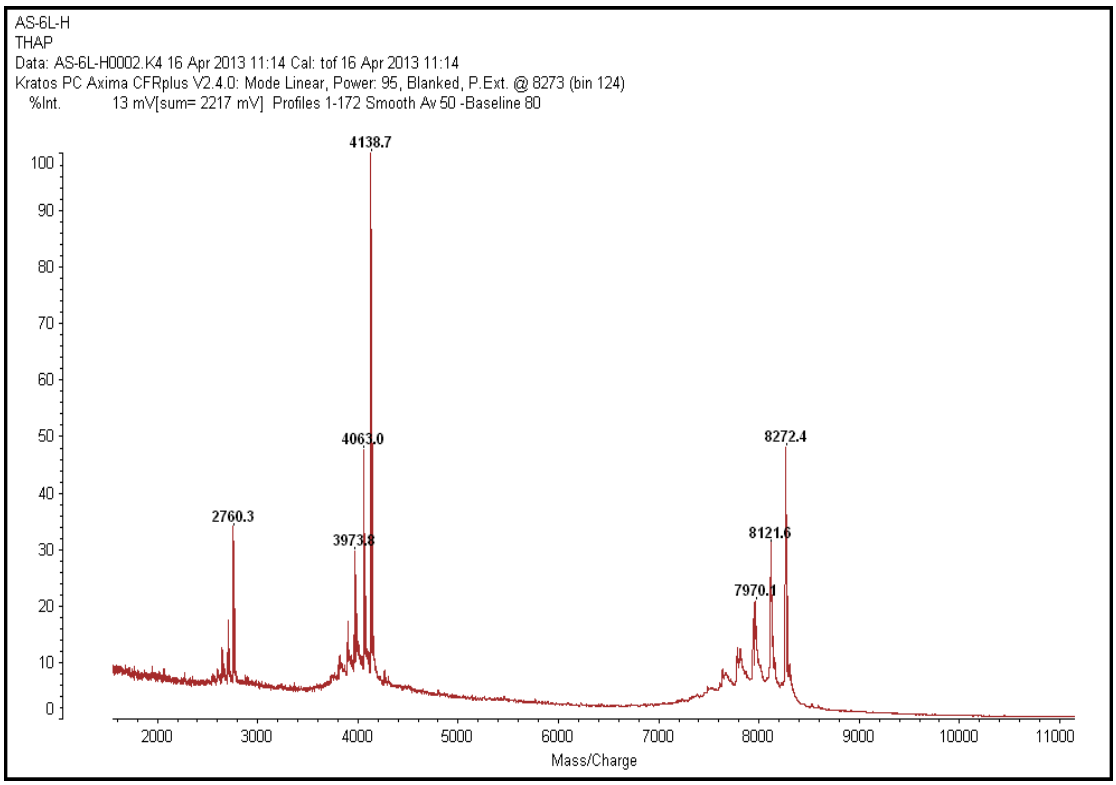


Figure S17. MALDI-TOF spectrum of sequence **AS1411-6L**.

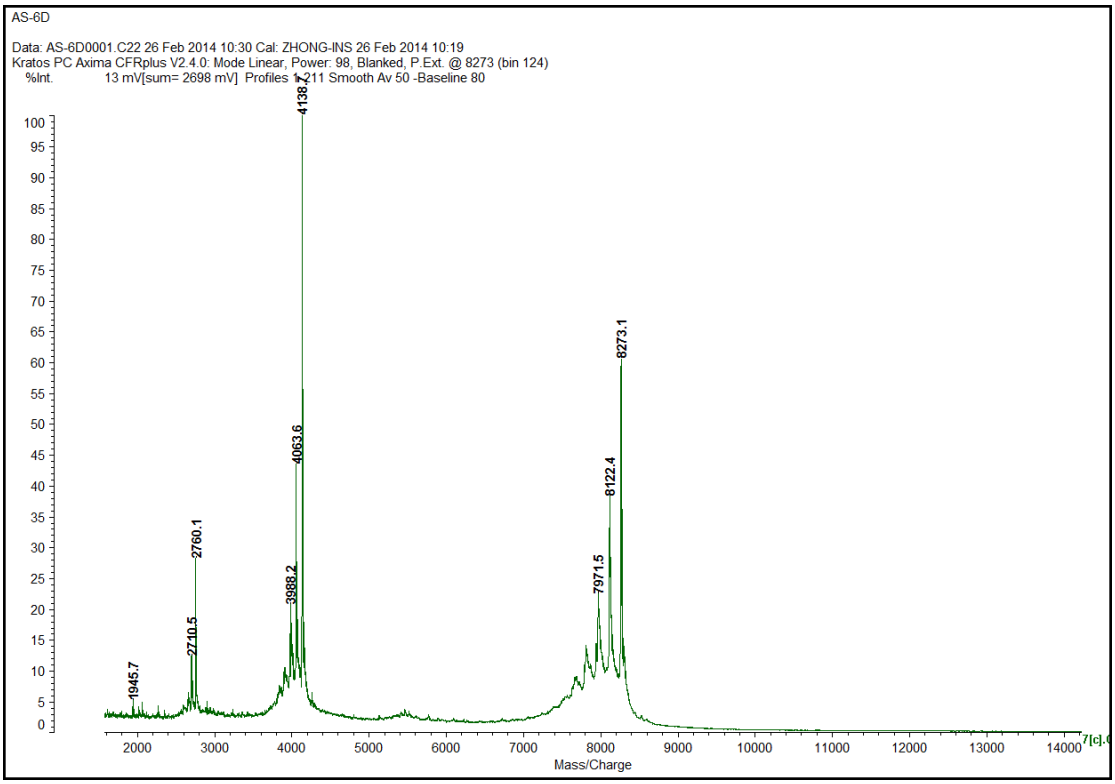


Figure S18. MALDI-TOF spectrum of sequence **AS1411-6D**.

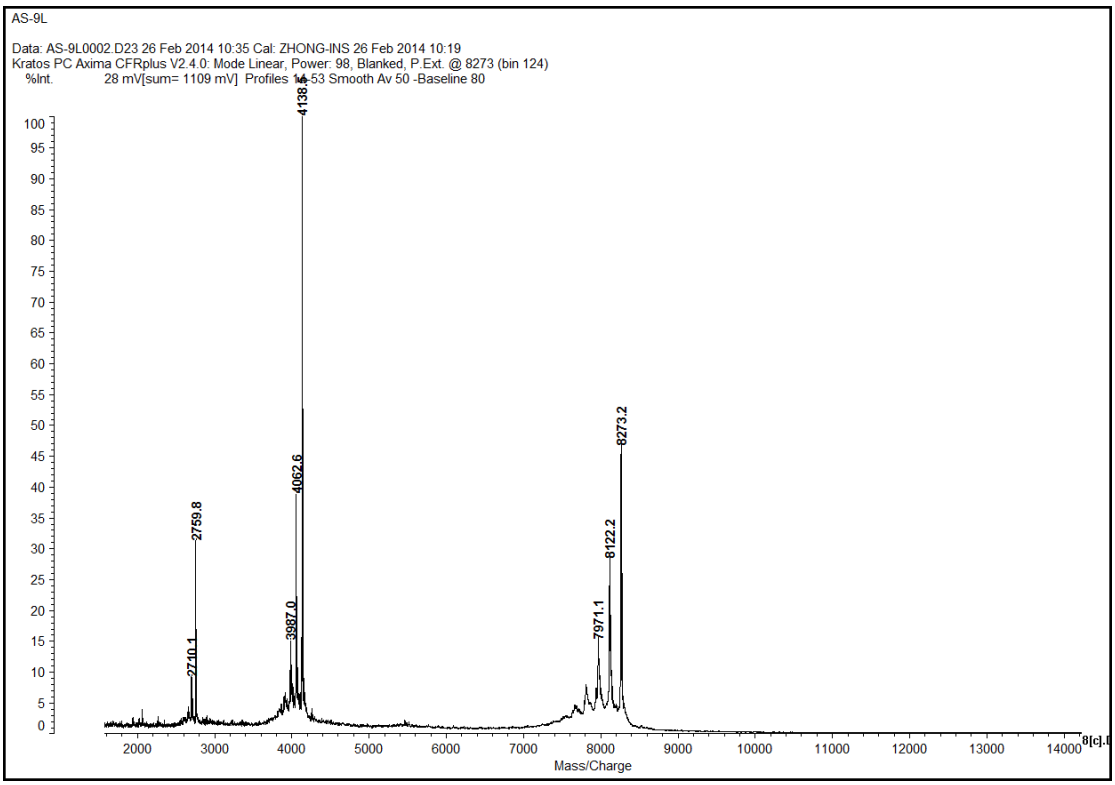


Figure S19. MALDI-TOF spectrum of sequence **AS1411-9$_L$**.

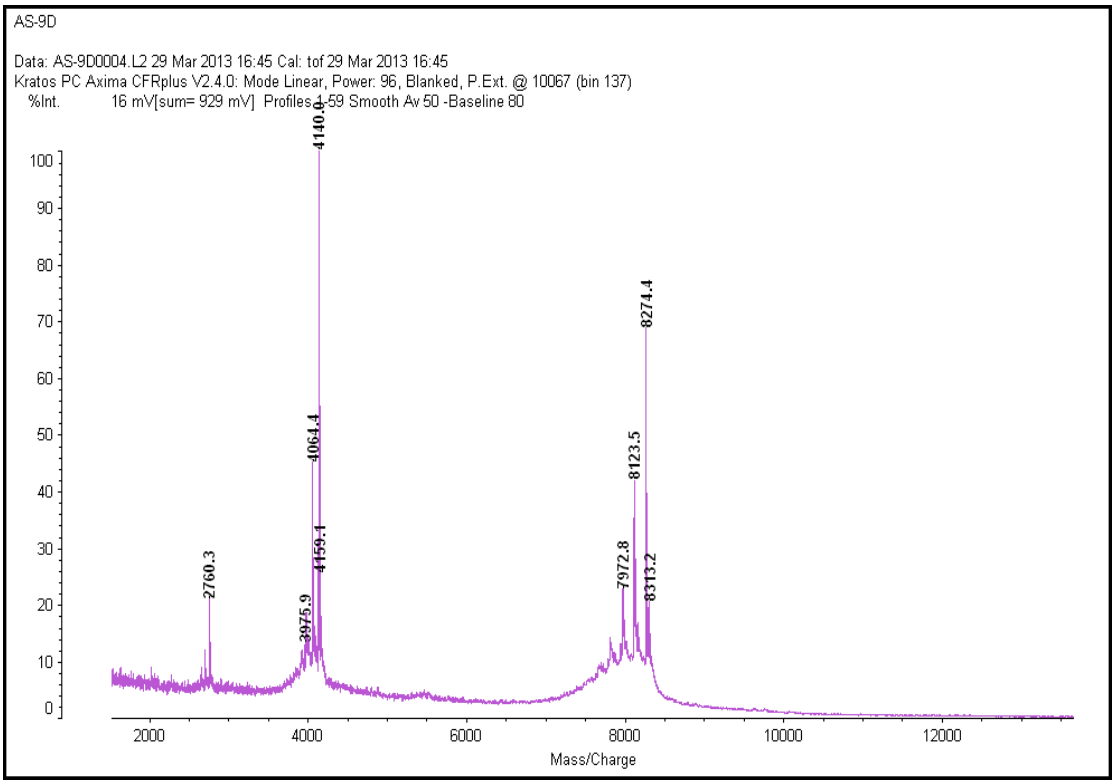


Figure S20. MALDI-TOF spectrum of sequence **AS1411-9$_D$**.

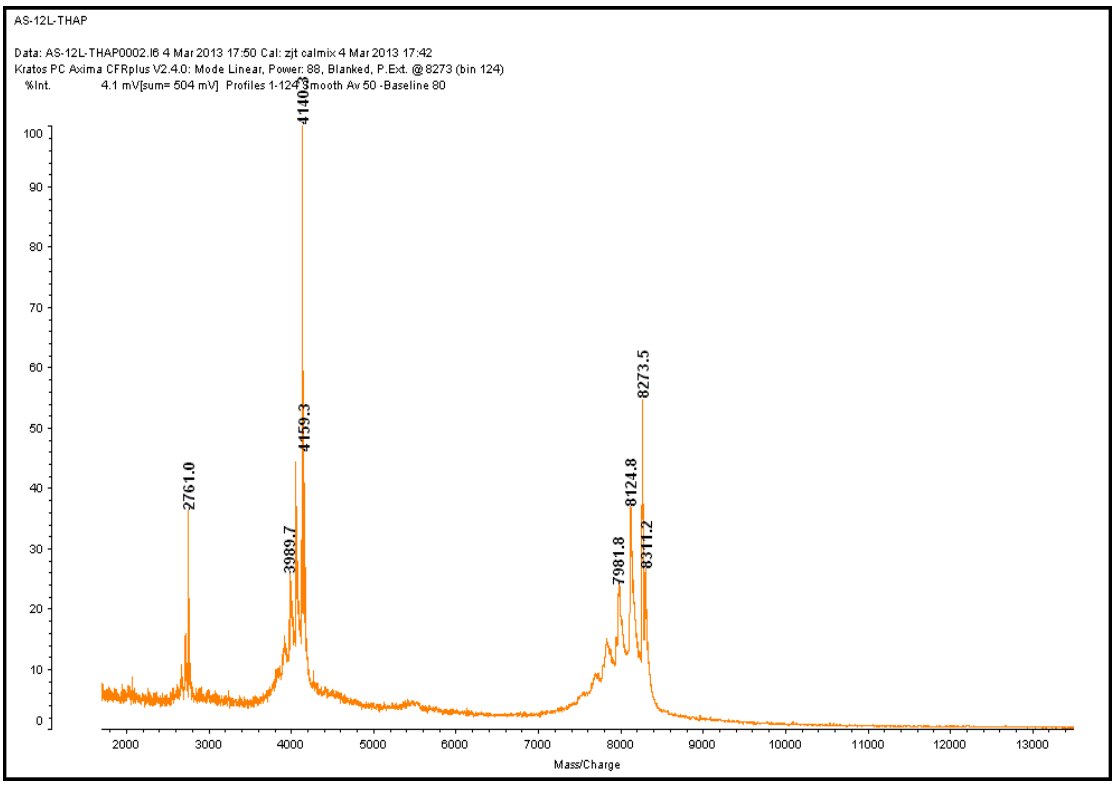


Figure S21. MALDI-TOF spectrum of sequence **AS1411-12$_L$**.

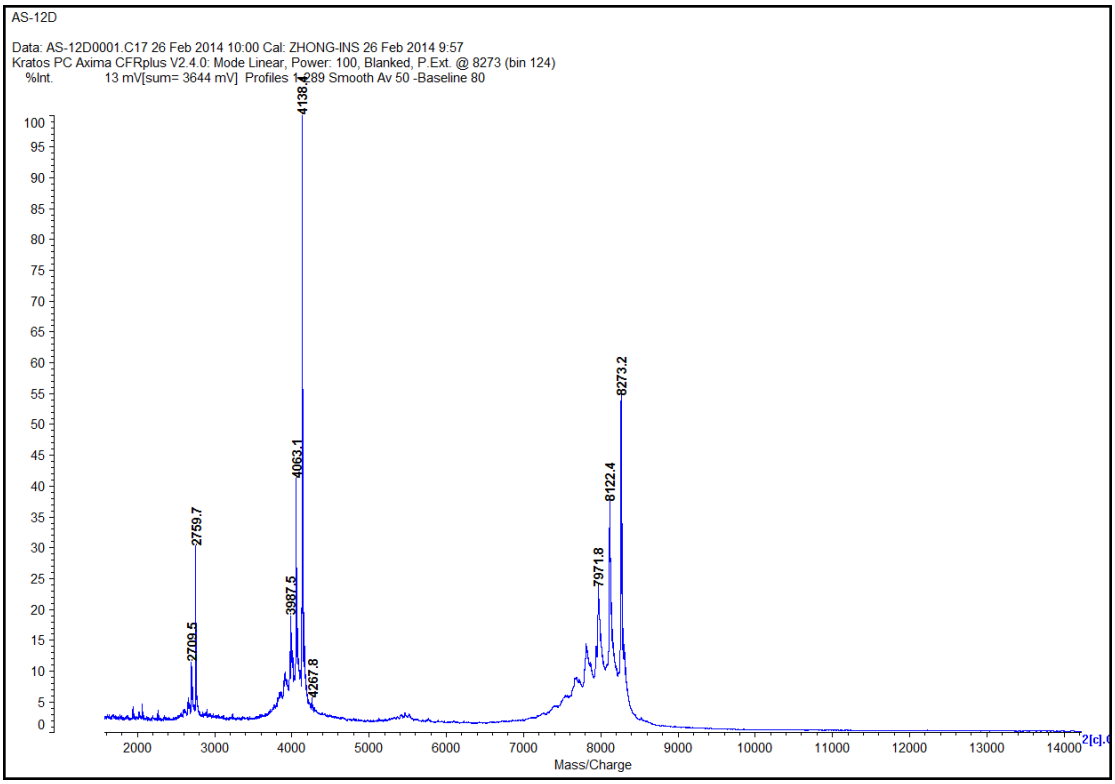


Figure S22. MALDI-TOF spectrum of sequence **AS1411-12$_D$**.

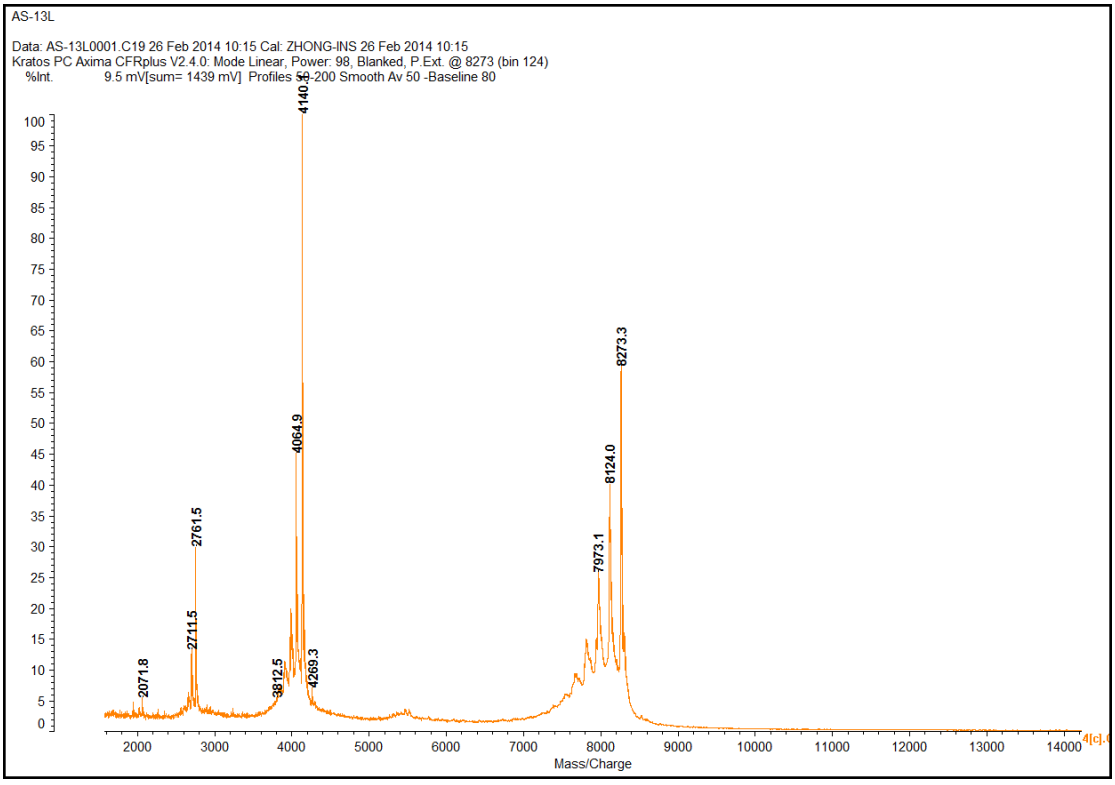


Figure S23. MALDI-TOF spectrum of sequence **AS1411-13$_L$**.

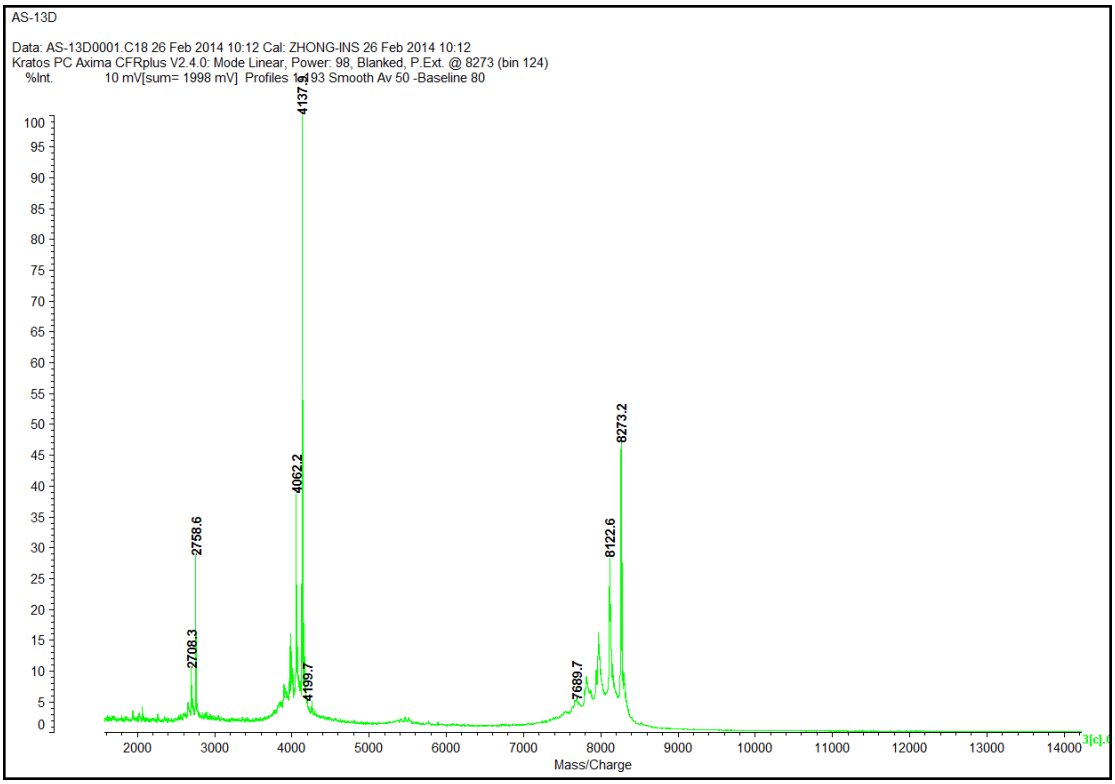


Figure S24. MALDI-TOF spectrum of sequence **AS1411-13$_D$**.

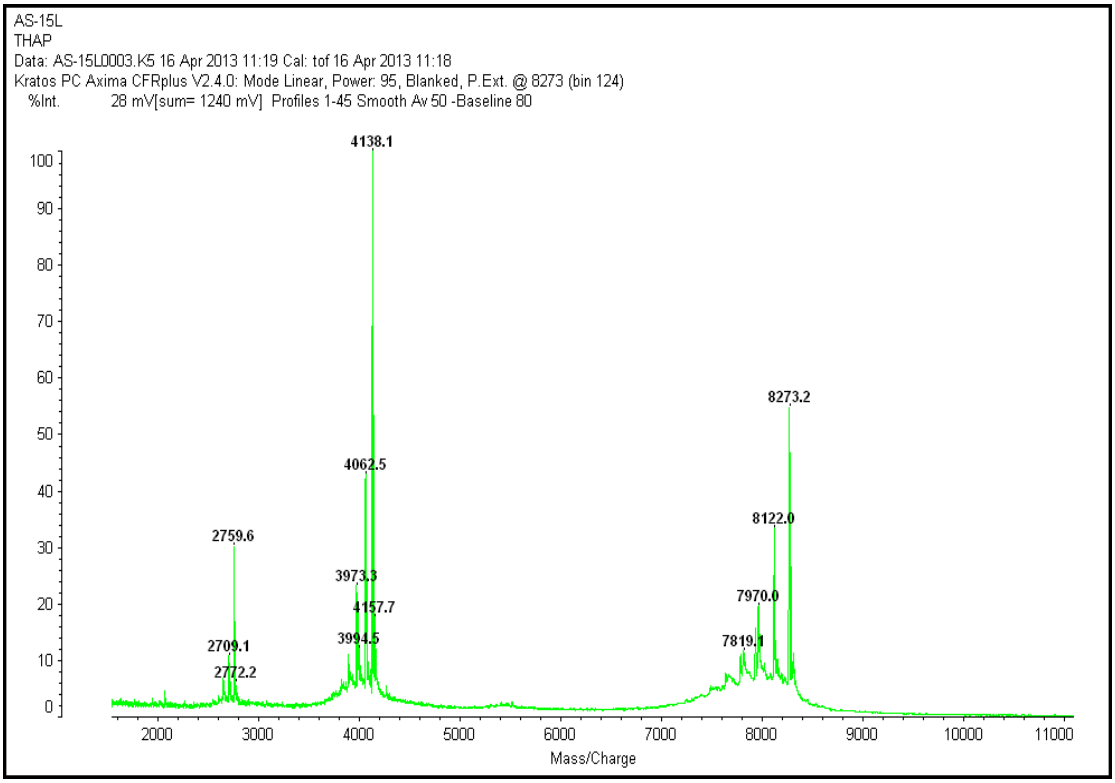


Figure S25. MALDI-TOF spectrum of sequence **AS1411-15$_L$**.

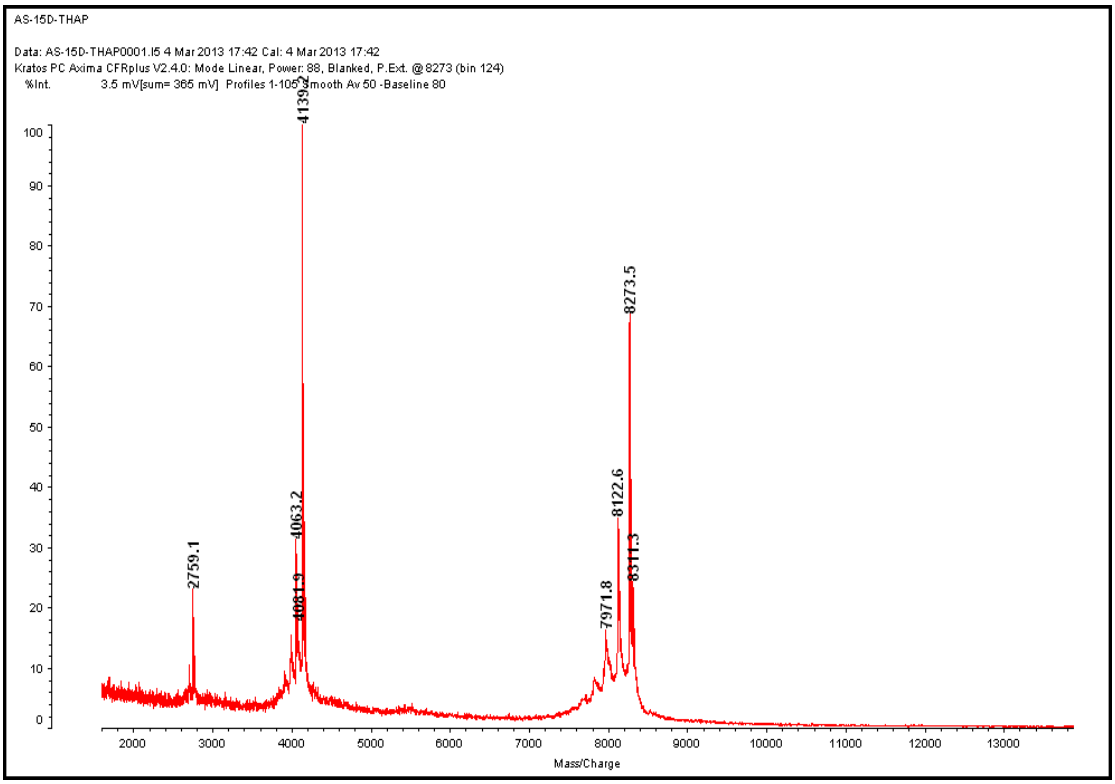


Figure S26. MALDI-TOF spectrum of sequence **AS1411-15$_D$**.

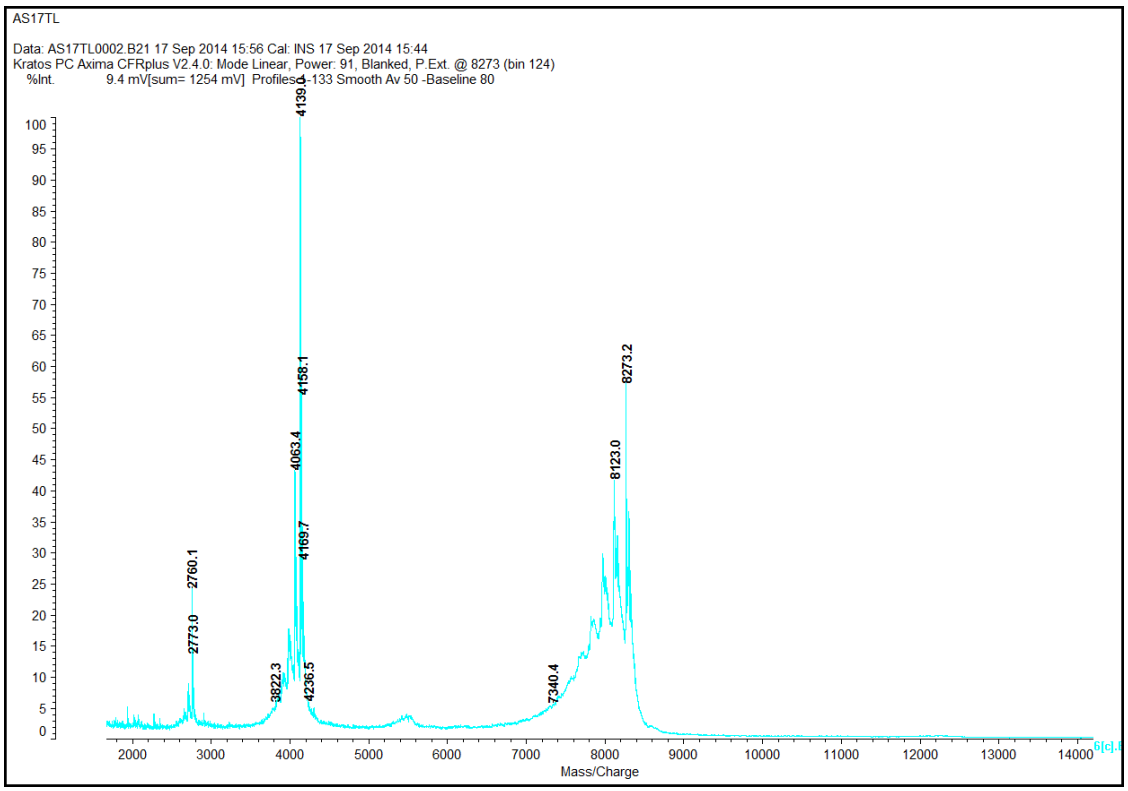


Figure S27. MALDI-TOF spectrum of sequence **AS1411-18$_L$**.

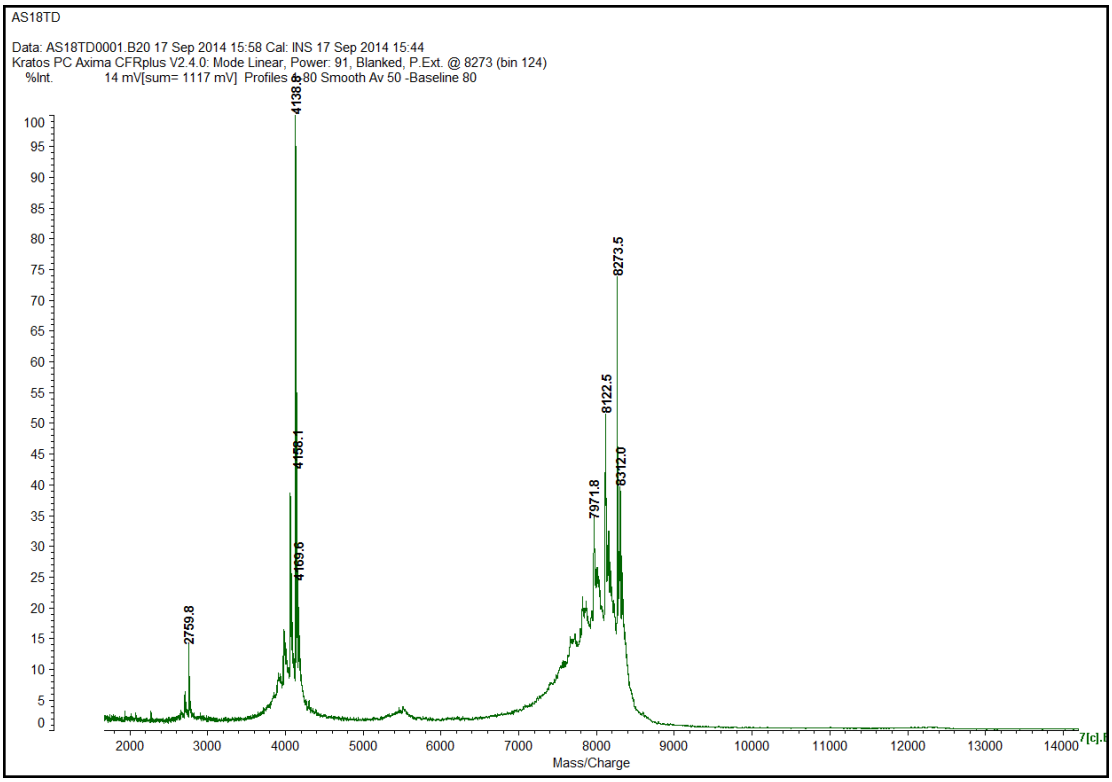


Figure S28. MALDI-TOF spectrum of sequence **AS1411-18$_D$**.

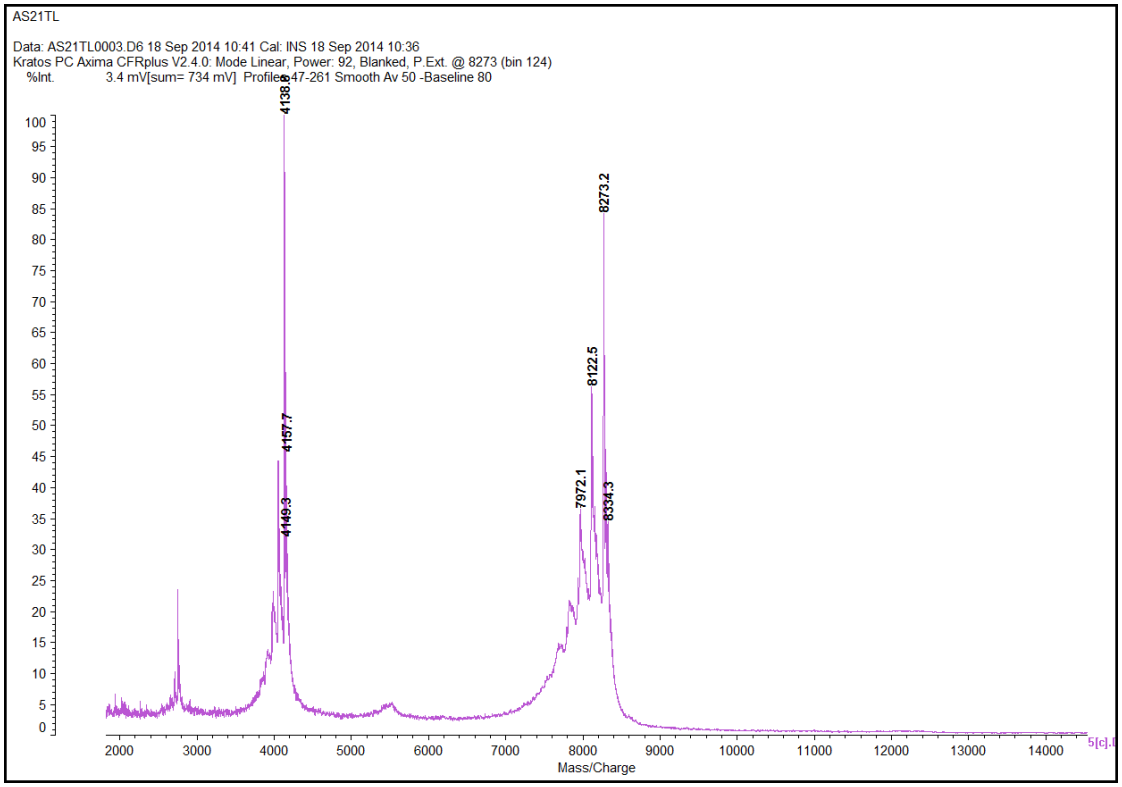


Figure S29. MALDI-TOF spectrum of sequence **AS1411-21$_L$**.

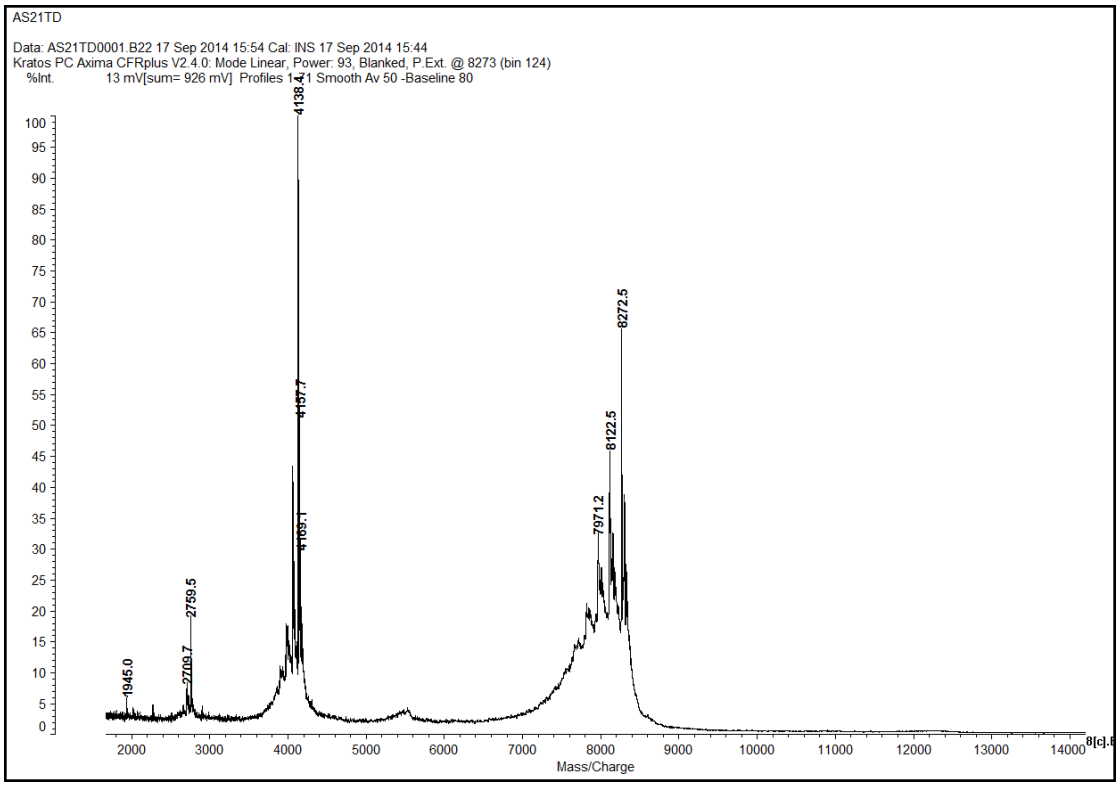


Figure S30. MALDI-TOF spectrum of sequence **AS1411-21$_D$**.

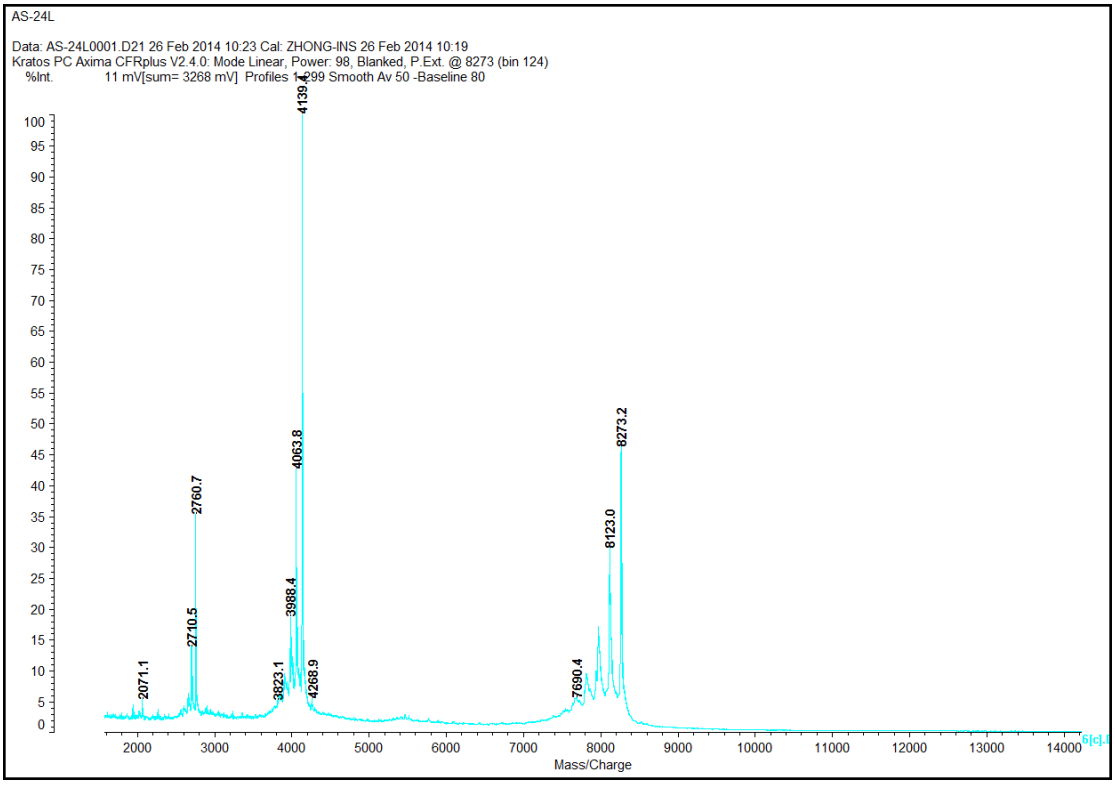


Figure S31. MALDI-TOF spectrum of sequence **AS1411-24$_L$**.

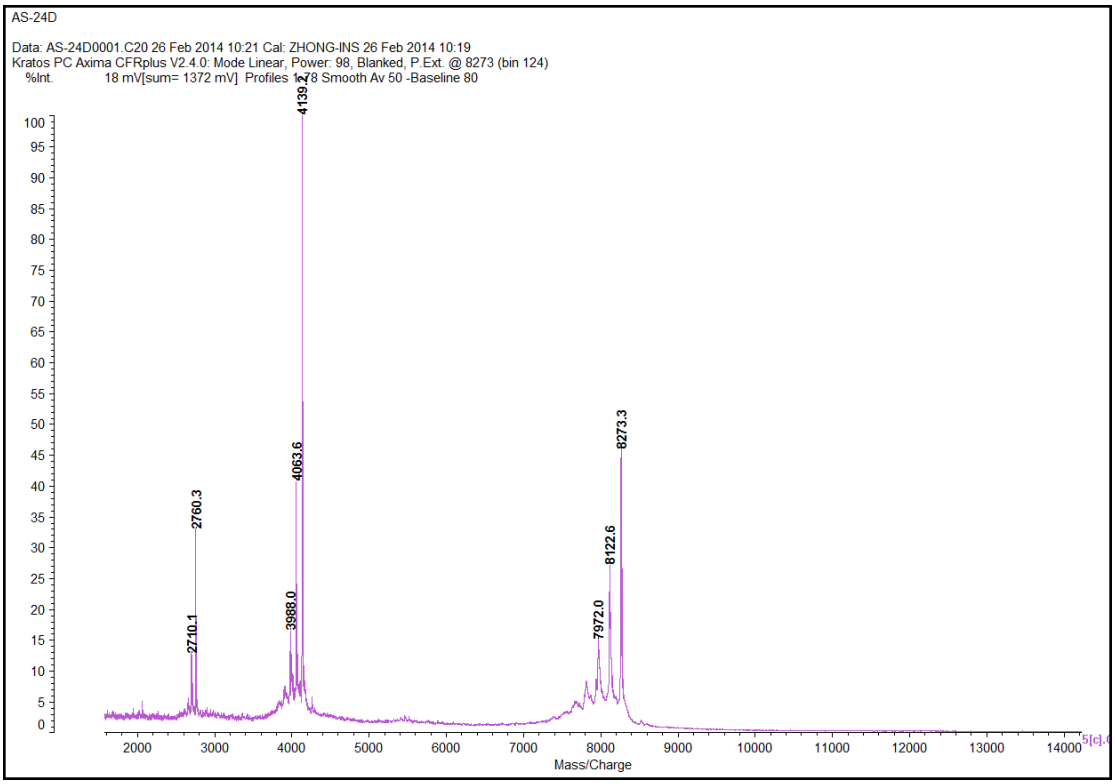


Figure S32. MALDI-TOF spectrum of sequence **AS1411-24$_D$**.

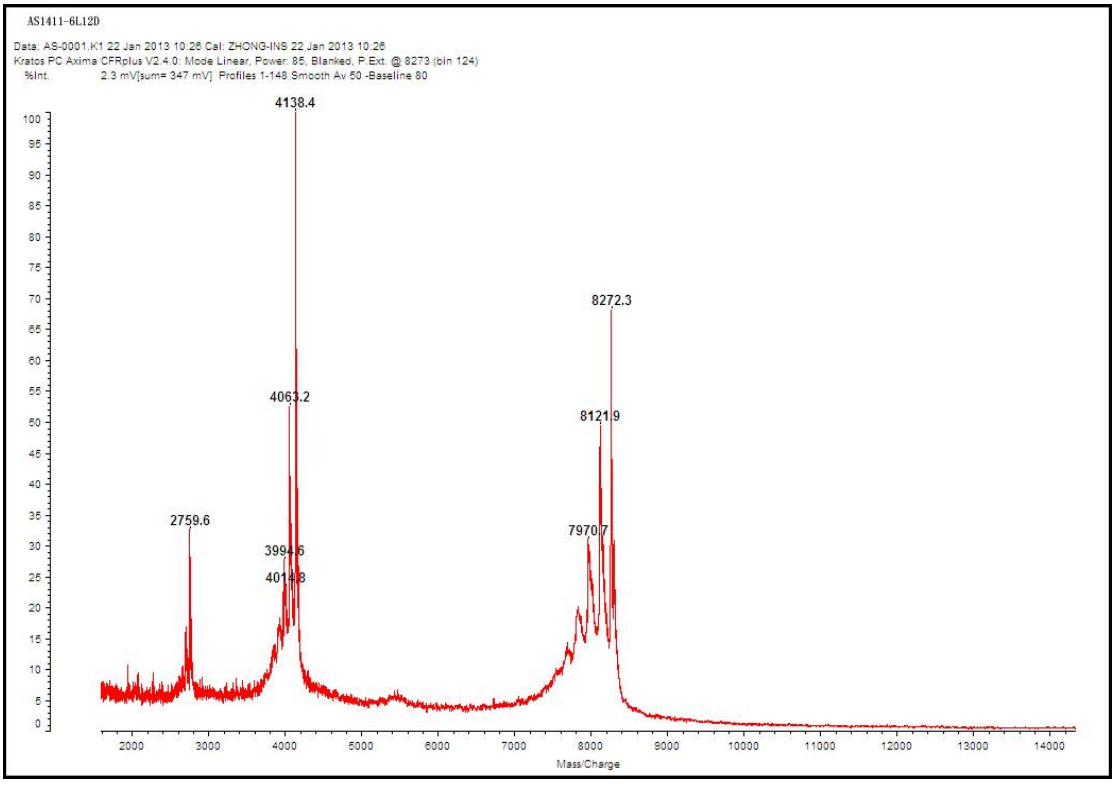


Figure S33. MALDI-TOF spectrum of sequence **AS1411-I (6$_L$/12$_D$)**.

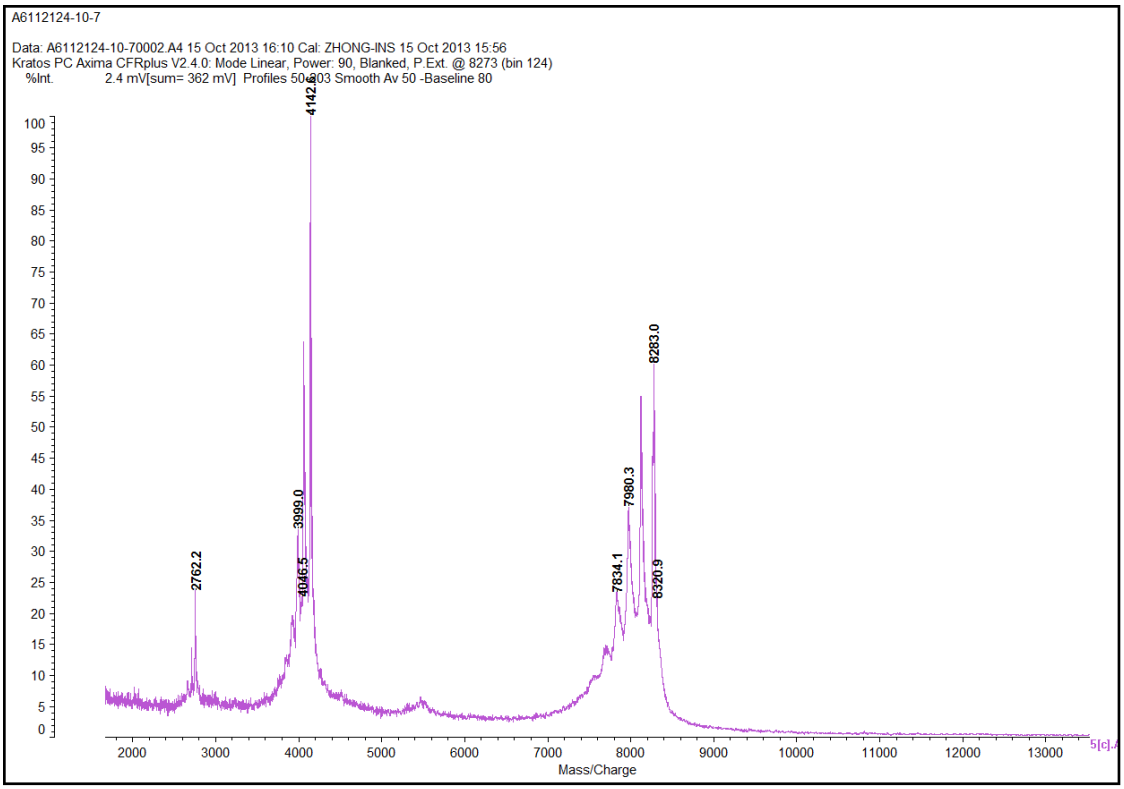


Figure S34. MALDI-TOF spectrum of sequence **AS1411-II ($6_L/12_D/24_{dI}$)**.