Spacial conservation studies of nucleobases in 10-23 DNAzyme by 2'-positioned isonucleotides and enantiomers for increased activity

Electronic Supplementary Information

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Table S1. MALDI-TOF of oligonucleotides with isonucleoside modification in catalytic core.

Name	Sequences (5'-3')	MW (found) [a]
DZ01	tgc tct cca GGC TAG CTA CAA CGA cct gca cct	9994.4
DZ-05-A_D	tgc tct cca GGC T***A_D***G CTA CAA CGA cct gca cct	9995.7
DZ-09-A_D	tgc tct cca GGC TAG CT***A_D*** CAA CGA cct gca cct	9995.8
DZ-11-A_D	tgc tct cca GGC TAG CTA C***A_D***A CGA cct gca cct	9997.2
DZ-12-A_D	tgc tct cca GGC TAG CTA CA***A_D*** CGA cct gca cct	9995.2
DZ-15-A_D	tgc tct cca GGC TAG CTA CAA CG***A_D*** cct gca cct	9995.4
DZ-05-A_L	tgc tct cca GGC T***A_L***G CTA CAA CGA cct gca cct	9995.6
DZ-09-A_L	tgc tct cca GGC TAG CT***A_L*** CAA CGA cct gca cct	9995.4
DZ-11-A_L	tgc tct cca GGC TAG CTA C***A_L***A CGA cct gca cct	9995.4
DZ-12-A_L	tgc tct cca GGC TAG CTA CA***A_L*** CGA cct gca cct	9995.4
DZ-15-A_L	tgc tct cca GGC TAG CTA CAA CGA_L cct gca cct	9995.1
DZ-04-T_D	tgc tct cca GGC ***T_D***AG CTA CAA CGA cct gca cct	9995.7
DZ-04-T_L	tgc tct cca GGC ***T_L***AG CTA CAA CGA cct gca cct	9995.8
DZ-08-T_D	tgc tct cca GGC TAG C***T_D***A CAA CGA cct gca cct	9995.6
DZ-08-T_L	tgc tct cca GGC TAG C***T_L***A CAA CGA cct gca cct	9994.7

[a] The calculated molecular weight of all the DNAzymes is 9994.4.

Table S2. MALDI-TOF of oligonucleotides with isonucleoside modification in recognition arms.

Name	Sequences (5'-3')	MW (found)[a]
DZ-07'-C_D	tgc tct ***C_D***ca GGC TAG CTA CAA CGA cct gca cct	9995.6
DZ-07'-C_L	tgc tct ***C_L***ca GGC TAG CTA CAA CGA cct gca cct	9995.8
DZ-08'-C_D	tgc tct c***C_D***a GGC TAG CTA CAA CGA cct gca cct	9995.3
DZ-08'-C_L	tgc tct c***C_L***a GGC TAG CTA CAA CGA cct gca cct	9995.3
DZ-09'-A_D	tgc tct cc***A_D*** GGC TAG CTA CAA CGA cct gca cct	9996.2
DZ-09'-A_L	tgc tct cc***A_L*** GGC TAG CTA CAA CGA cct gca cct	9995.8
DZ-10'-C_D	tgc tct cca GGC TAG CTA CAA CGA ***C_D***ct gca cct	9995.0
DZ-10'-C_L	tgc tct cca GGC TAG CTA CAA CGA ***C_L***ct gca cct	9995.3

DZ-11'-C_D	tgc tct cca GGC TAG CTA CAA CGA c$\boldsymbol{C_D}$t gca cct	9995.3
DZ-11'-C_L	tgc tct cca GGC TAG CTA CAA CGA cC_Lt gca cct	9994.1
DZ-12'-T_D	tgc tct cca GGC TAG CTA CAA CGA cc$\boldsymbol{T_D}$ gca cct	9995.3
DZ-12'-T_L	tgc tct cca GGC TAG CTA CAA CGA cc$\boldsymbol{T_L}$ gca cct	9994.7

Table S3. Thermal stability of modified DNAzyme-substrate complexes

Name	T_m (°C)	Name	T_m (°C)
DZ01	51.0	DZ-07'-C_D	48.4
DZ-05-A_D	51.0	DZ-07'-C_L	46.8
DZ-09-A_D	51.0	DZ-08'-C_D	46.9
DZ-11-A_D	50.0	DZ-08'-C_L	46.5
DZ-12-A_D	51.0	DZ-09'-A_D	48.1
DZ-15-A_D	51.0	DZ-09'-A_L	47.9
		DZ-10'-C_D	47.8
DZ-05-A_L	50.0	DZ-10'-C_L	48.4
DZ-09-A_L	50.0	DZ-11'-C_D	47.1
DZ-11-A_L	50.0	DZ-11'-C_L	47.1
DZ-12-A_L	51.0	DZ-12'-T_D	46.6
DZ-15-A_L	50.0	DZ-12'-T_L	44.2
DZ-04-T_D	51.0	DZ-08-T_D	50.5
DZ-04-T_L	51.0	DZ-08-T_L	50.4

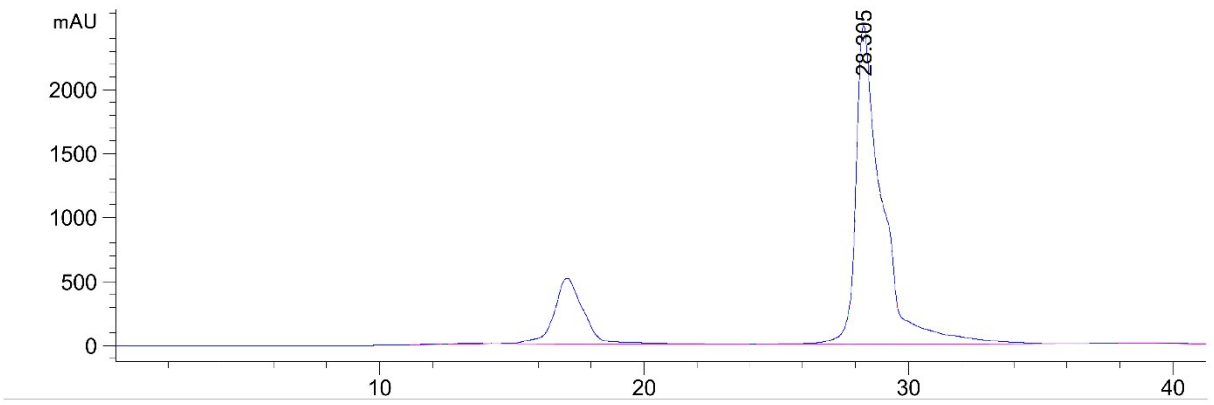

Figure S1. HPLC purification of oligonucleotides, DZ-08-$^{i}T_D$ as example. (Linear gradient using 5–60% acetonitrile–TEAB 100 mM in 40 min, Phenomenex Oligo-RP C18 250×10.0 mm 5μm, 40℃, 1.2 mL/min, 260 nm).

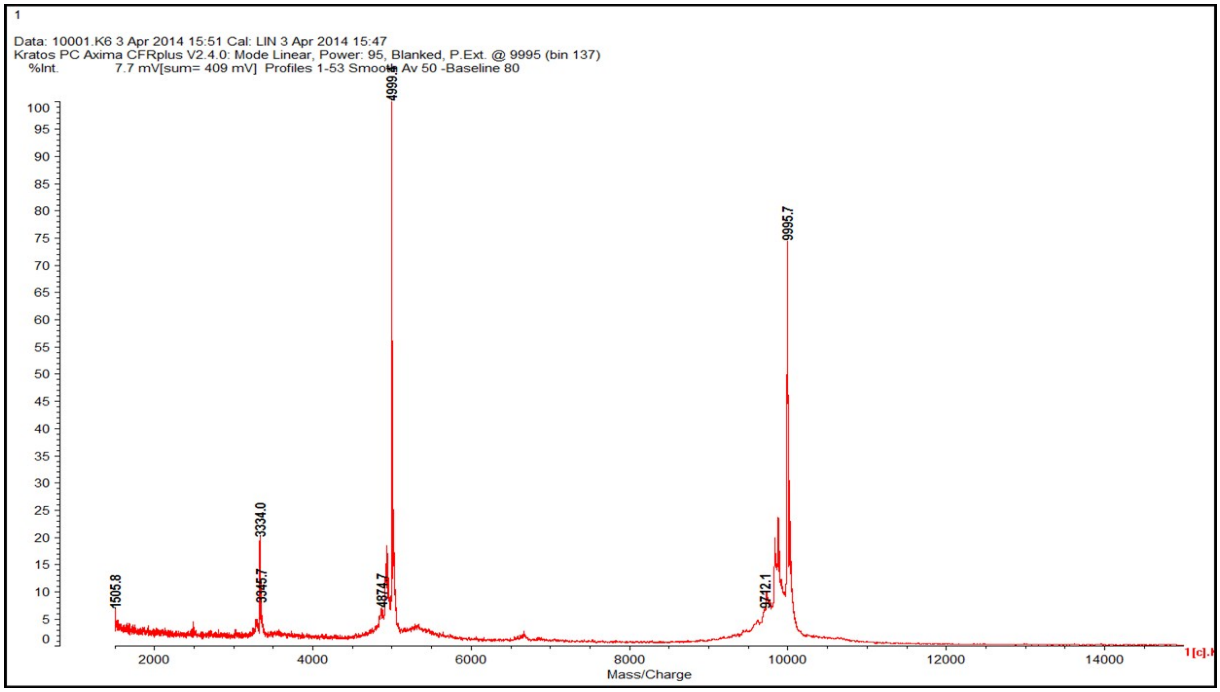

Figure S2. MALDI-TOF spectrum of sequence DZ-05-A_D.

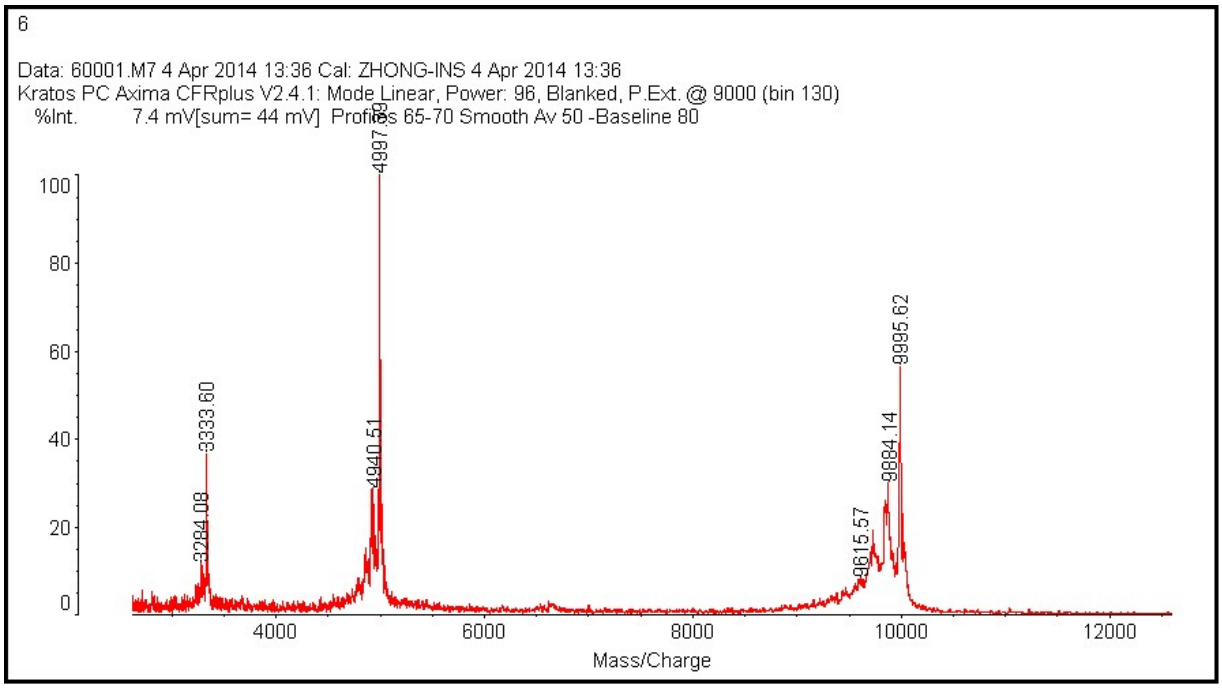

Figure S3. MALDI-TOF spectrum of sequence DZ-05-A_L.

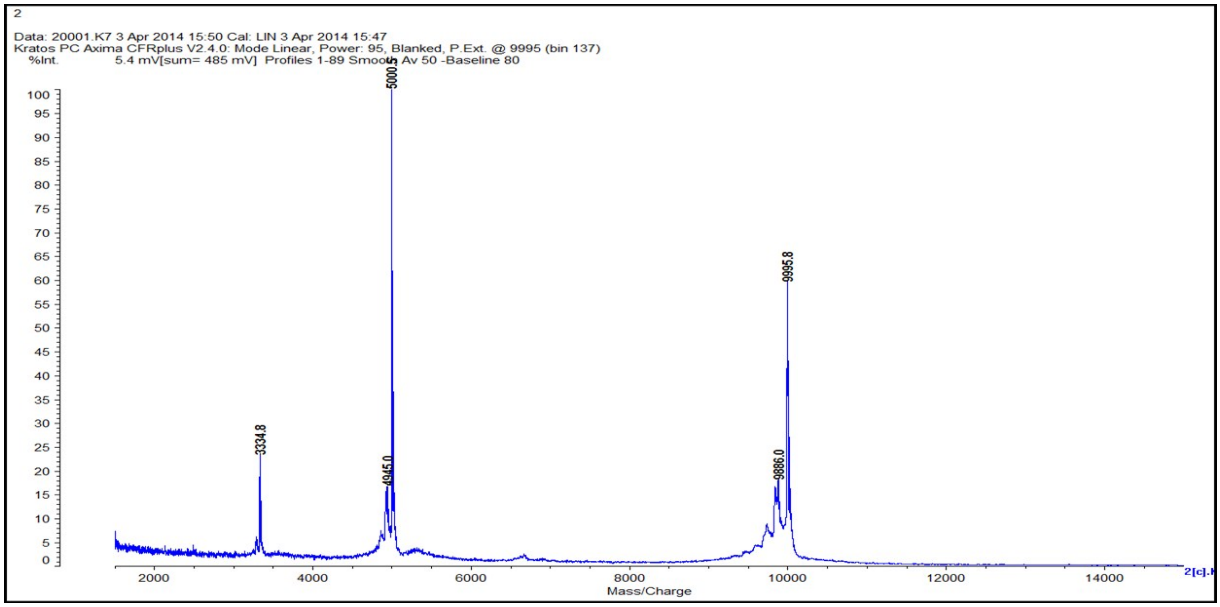

Figure S4. MALDI-TOF spectrum of sequence DZ-09-A_D.

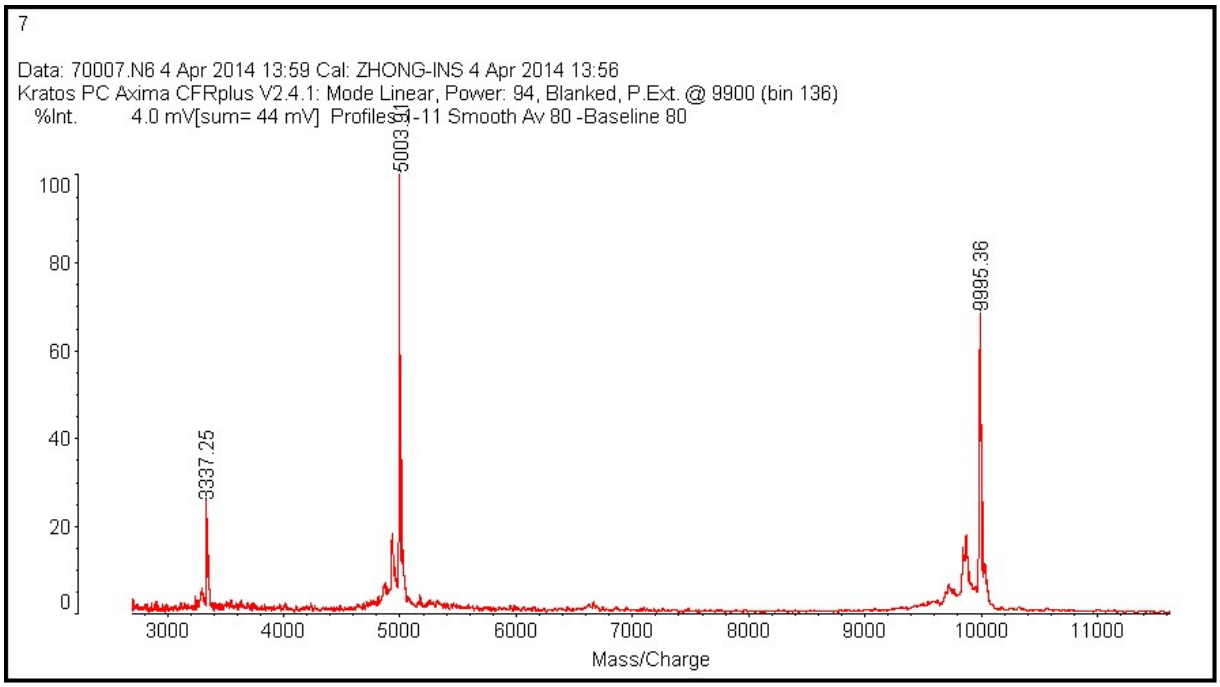

Figure S5. MALDI-TOF spectrum of sequence DZ-09-A_L.

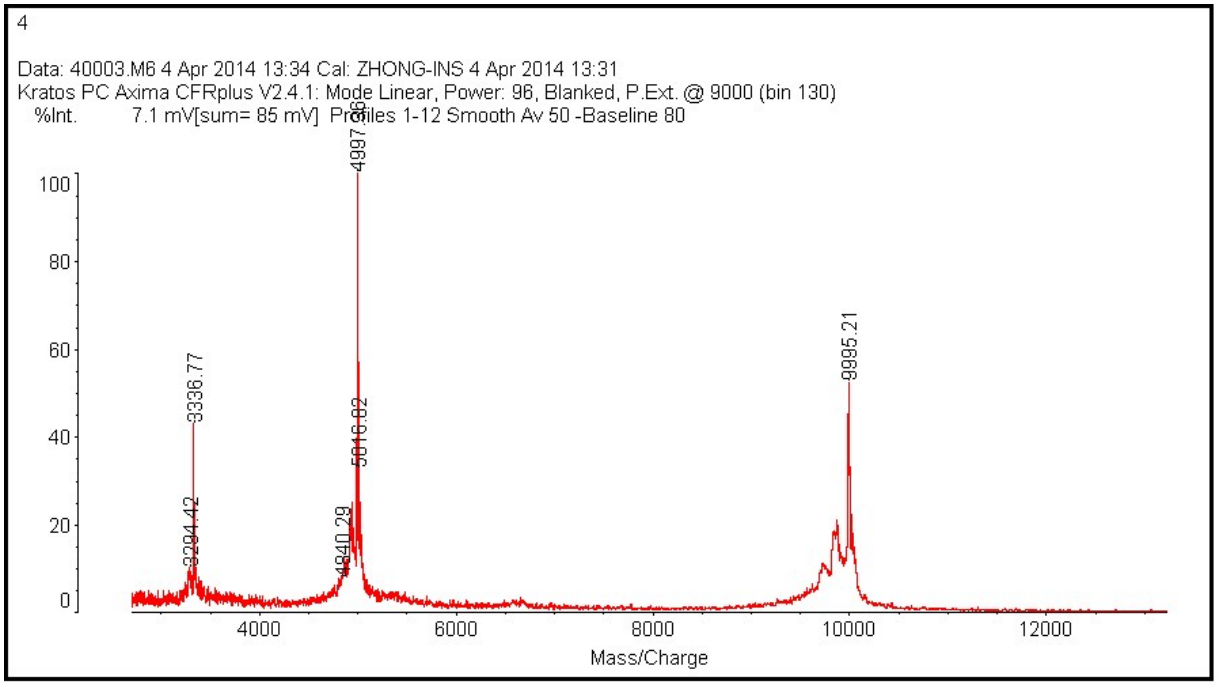

Figure S6. MALDI-TOF spectrum of sequence DZ-12-A_D.

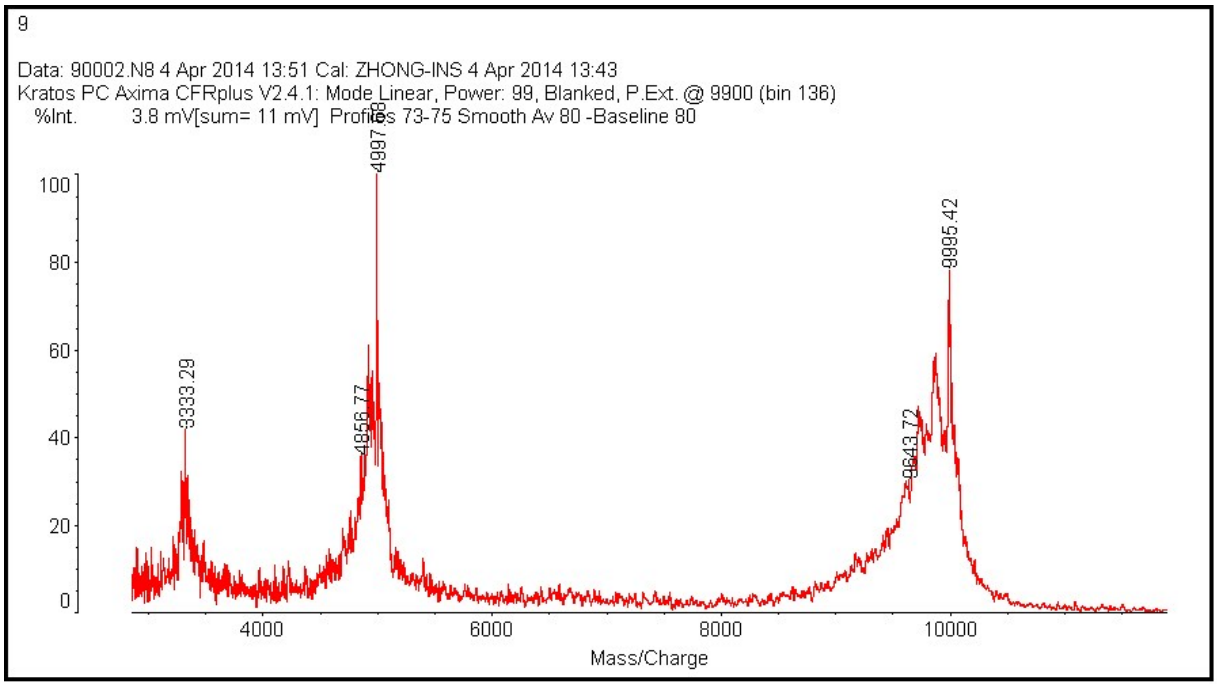

Figure S7. MALDI-TOF spectrum of sequence DZ-12-A_L.

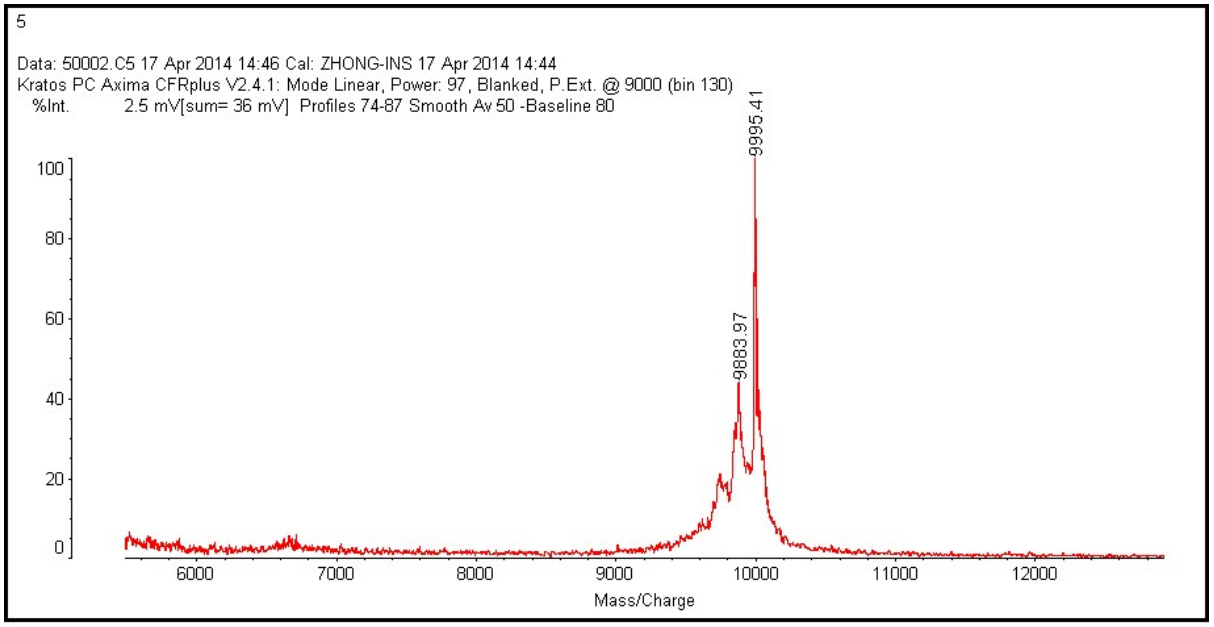

Figure S8. MALDI-TOF spectrum of sequence DZ-15-A_D.

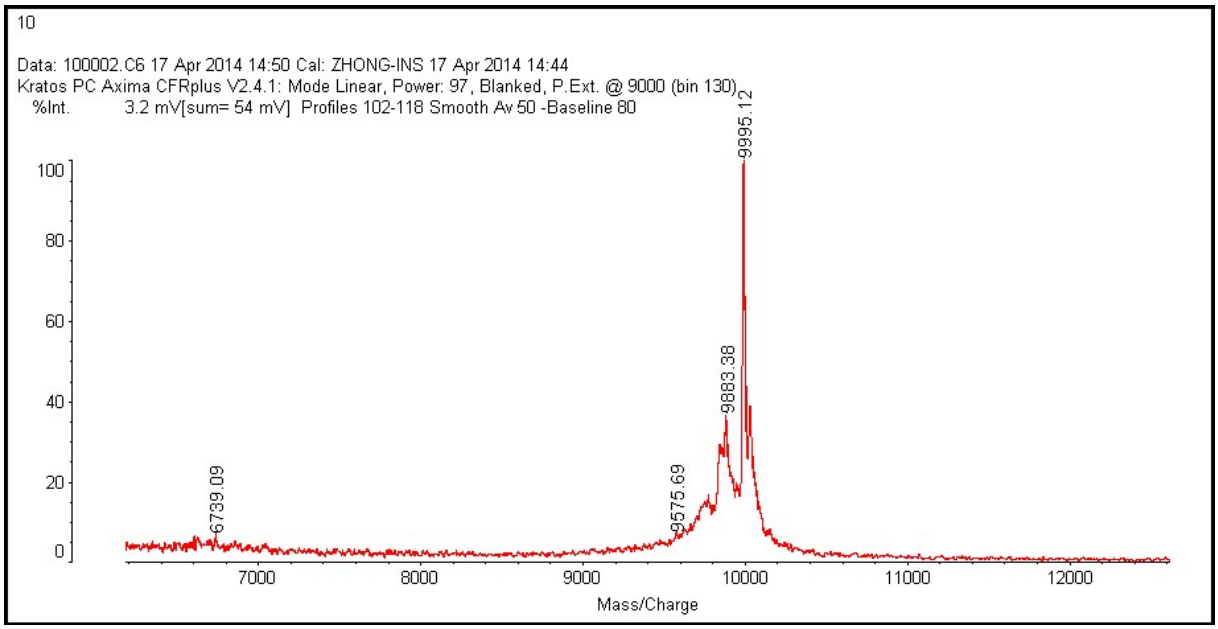

Figure S9. MALDI-TOF spectrum of sequence DZ-15-A_L.

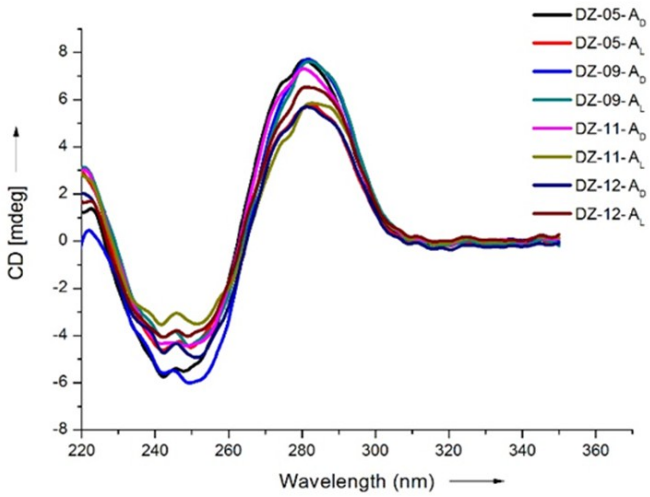

Figure S10. CD spectra of complexes of 10-23 DNAzyme with isonucleoside modification in catalytic core and its substrate.

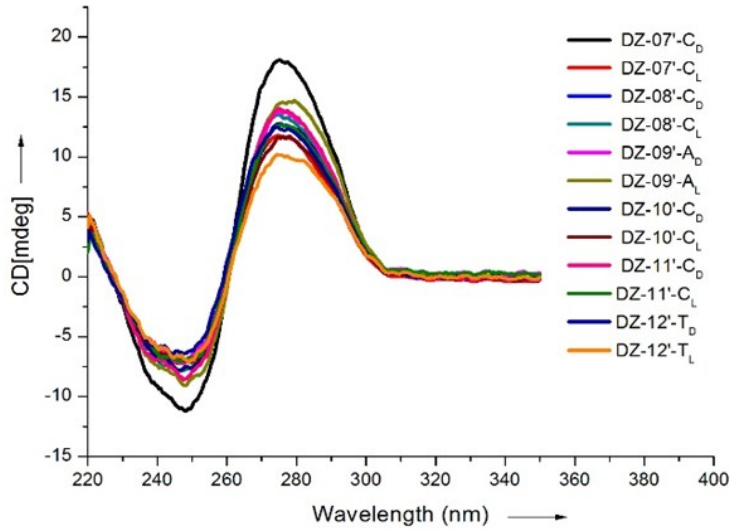

Figure S11. CD spectra of complexes of 10-23 DNAzyme with isonucleoside modification in recognition arm and its substrate.

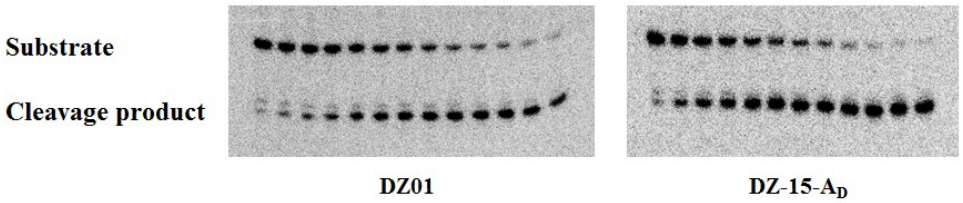

Figure S12. Denaturing PAGE (20%) of 10-23 DNAzyme cleavage reactions under single-turnover condition. Samples were set at 0, 10, 20, 30, 60, 90, 120, 180, 240, 360, 480 and 600 min.

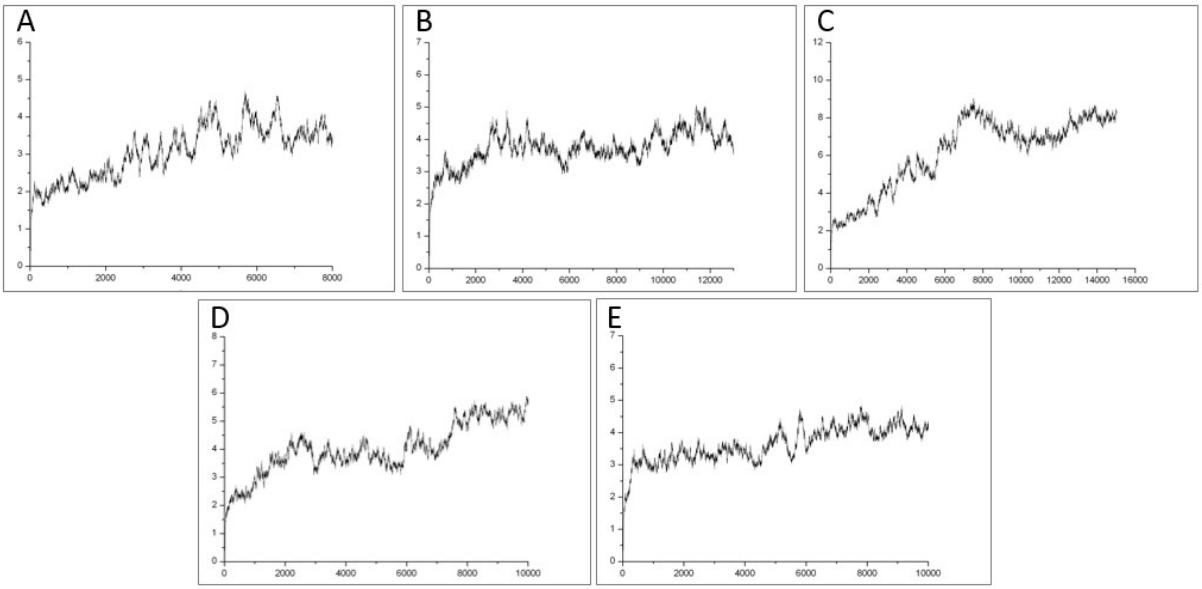

Figure S13. RMSD curve of native, **L-isoA** (B) and **D-isoA** (C) modified at the position 15 and **L-isoA** (B) and **D-isoA** (C) modified at the position 11 of 10-23DNAzyme and substrate complex. Modification at position 15 with **D-isoA** and position 11 with **L-isoA** bring more influence to the structure and consume relatively longer time for adjustment.

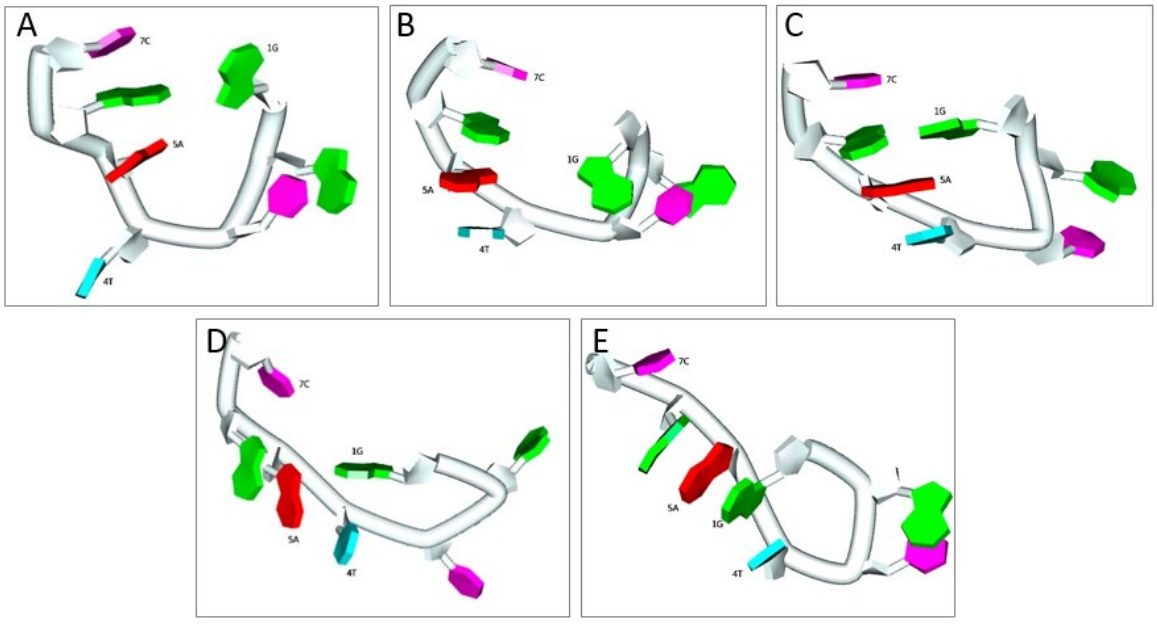

Figure S14. Conformation of the segment at the 5'-terminal of catalytic core. Compared to the native one (A), that segment of **L-isoA** (B) and **D-isoA** (C) modified at the position 15 of the DNAzyme core roughly maintain the same. However, modification at position 11 cause larger influence in both **L-isoA** (D) and **D-isoA** (E), the conformation and stack style of bases are quite different from the native one.