Recognition of HIV TAR RNA by triazole linked neomycin dimers

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Supporting Information

Appendices

1. Synthesis procedure (S1).

DPA 51. To a solution of 5”-azide-neomycin B (62 mg, 0.05 mmol) in dry toluene (5 mL), propargyl ether (2.35 mg, 0.025 mmol, 0.50 eq) was added followed by the addition of CuI (4.76 mg, 0.025 mmol) and DIPEA (6.46 mg, 0.05 mmol). The reaction mixture stirred at 90 °C for 18 h in the atmosphere of argon. The volatiles were removed under vacuum. Purification by flash column chromatography (0 to 10 % ethanol in CH_2Cl_2) afforded the desired product as a white solid (57.3 mg, 89%)

Deprotection of N-Boc protected neomycin dimers. To a solution of neomycin dimer (30 mg) in dioxane (3 mL), 4 M HCl/dioxane (1 mL) was added and the reaction started at room temperature. A white precipitate formed after 15 min. The reaction mixture was centrifuged and the solid was collected. The solid was washed with a solution of diethyl ether/hexane [3 × 5 mL each, 1:1 (v/v)]. The solid was then dissolved in water and lyophilized to afford a white solid (16.9 mg, 95%). ^{1}H NMR (500 MHz, D_2O): δ 8.02 (s, 2 H, triazole), 5.92 (d, *J* = 3.94 Hz, 2 H, H_{1II}), 5.28 (d, *J* = 3.16 Hz, 2 H, H_{1III}), 5.17 (s, 2 H, H_{1IV}), 4.41 (m, 2 H, H_{4III}), 4.34 (d, *J* = 5.20 Hz, 2 H, H_{2III}), 4.18 (d, *J* = 4.73 Hz, 2 H, H_{4IV}), 4.10 (d, *J* = 2.84 Hz, 2 H, H_{4I}), 3.96 (t, *J* = 9.62 Hz, 2 H, H_{6II}), 3.90 (t, *J* = 9.93 Hz, 2 H, H_{5I}), 3.76-3.86 (m, 8 H, H_{2IV}, H_{4II}, H_{5IV}, and H_{3III}), 3.70 (d, *J* = 1.89 Hz, 2 H, H_{6I}), 3.54 (t, *J* = 9.61 Hz, 2 H, H_{3II}), 3.45-3.49 (m, 4 H, H_{3I}, H_{5II}), 3.32-3.44 (m, 2 H, H_{1I}), 3.20-3.30 (m, 8 H, H_{5III} and propargyl ether protons), 2.32-2.39 (dt, *J1* = 3.63 Hz, *J2* = 4.57 Hz, 2 H, $H_{2Ieq.}$), 1.72-1.82 (q, *J* = 12.45 Hz, 2 H, $H_{2Iax.}$); MS MALDI-TOF m/z for $C_{52}H_{96}N_{18}O_{25}$ $[M+H_2O]^+$, calcd 1391.44, found 1392.66.

2. Characterization of DPA51 (S2).

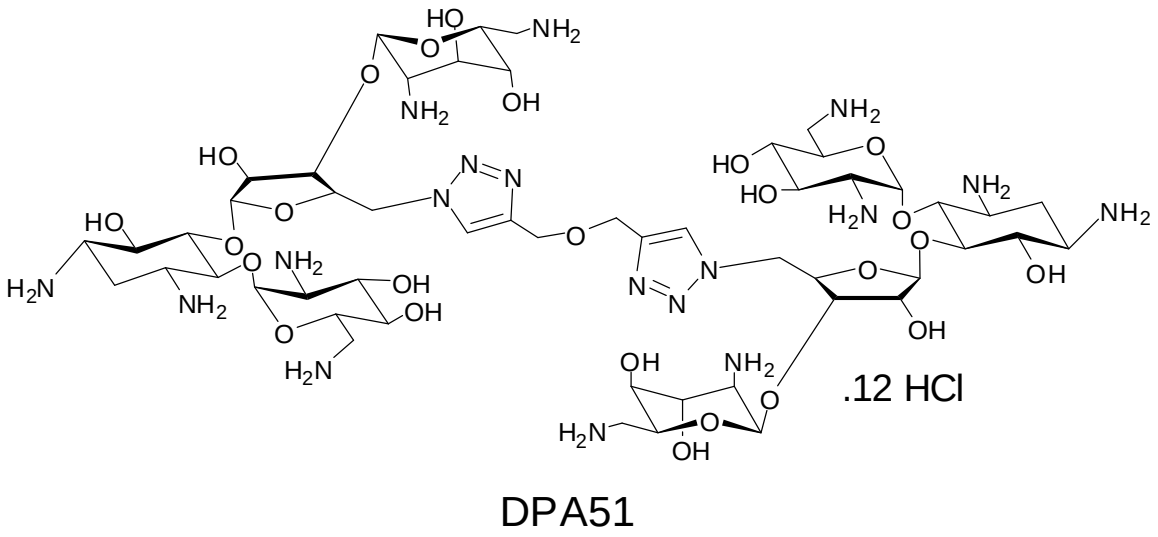

Figure S1. Chemical structure of DPA51

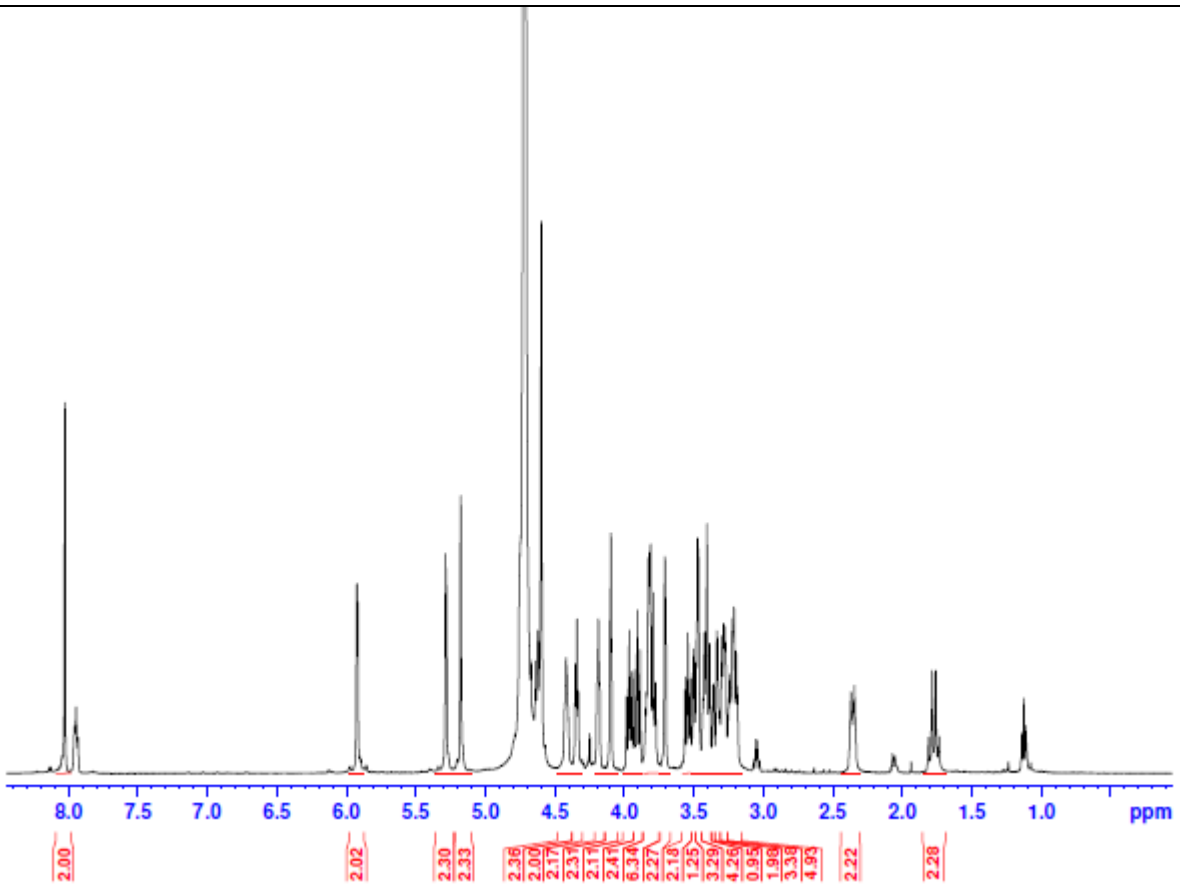

Figure S2. ^{1}H-NMR of DPA51

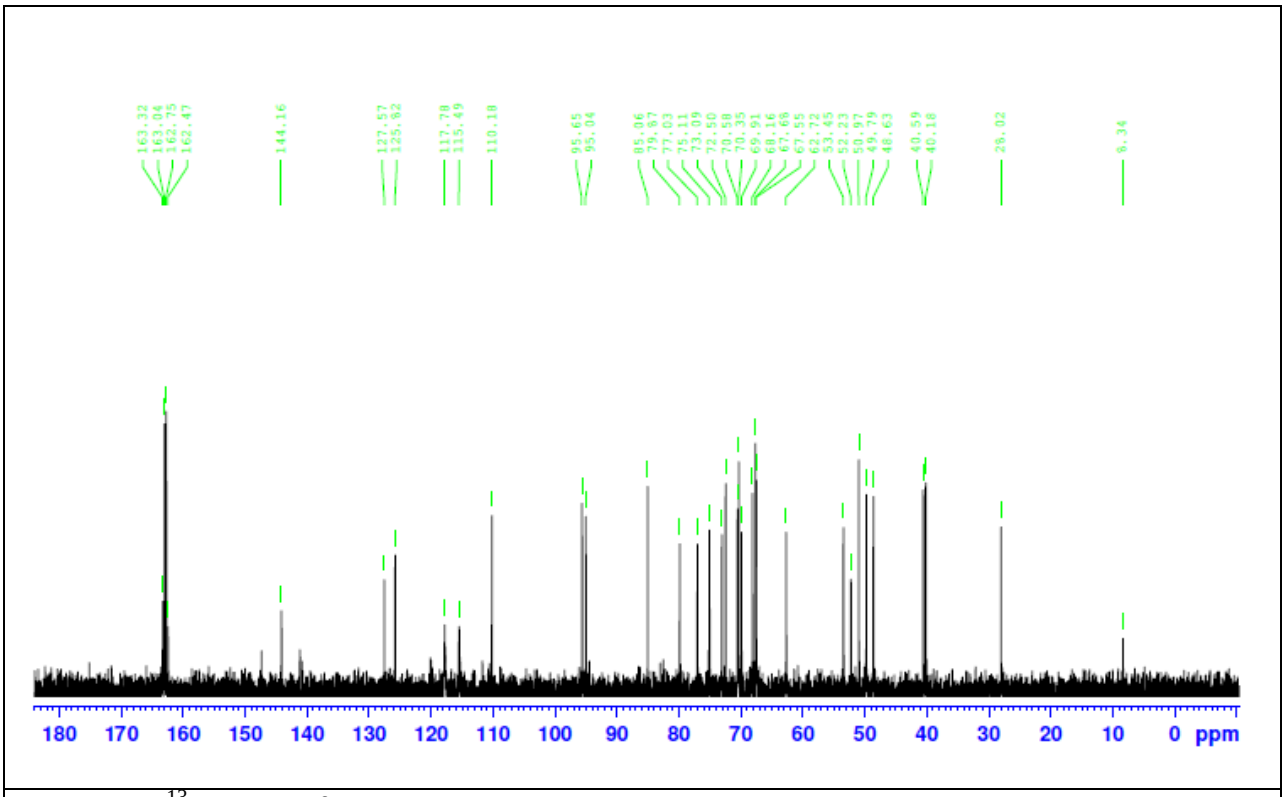

Figure S3. ^{13}C-NMR of DPA51

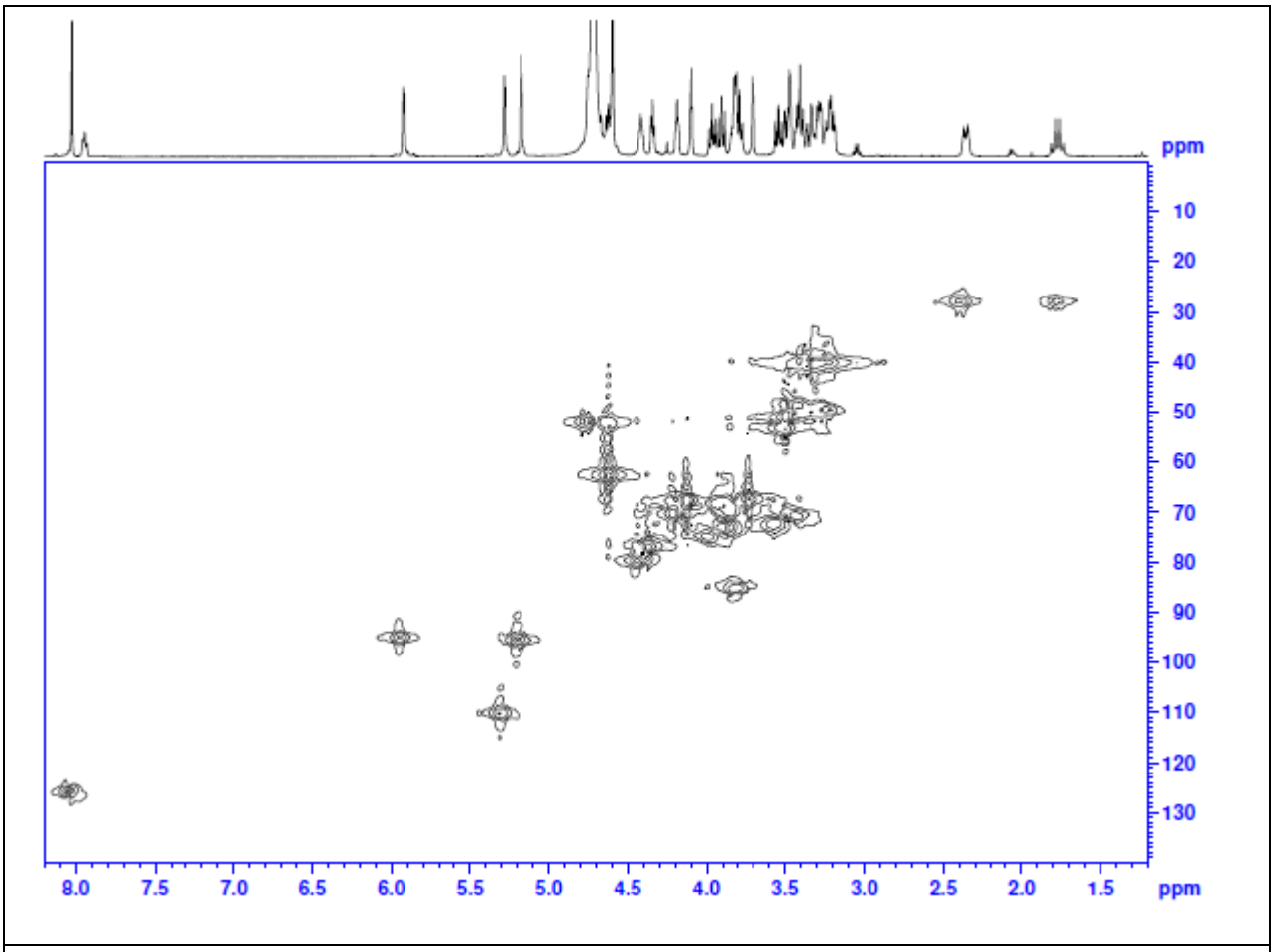

Figure S4. HMQC-NMR of DPA51

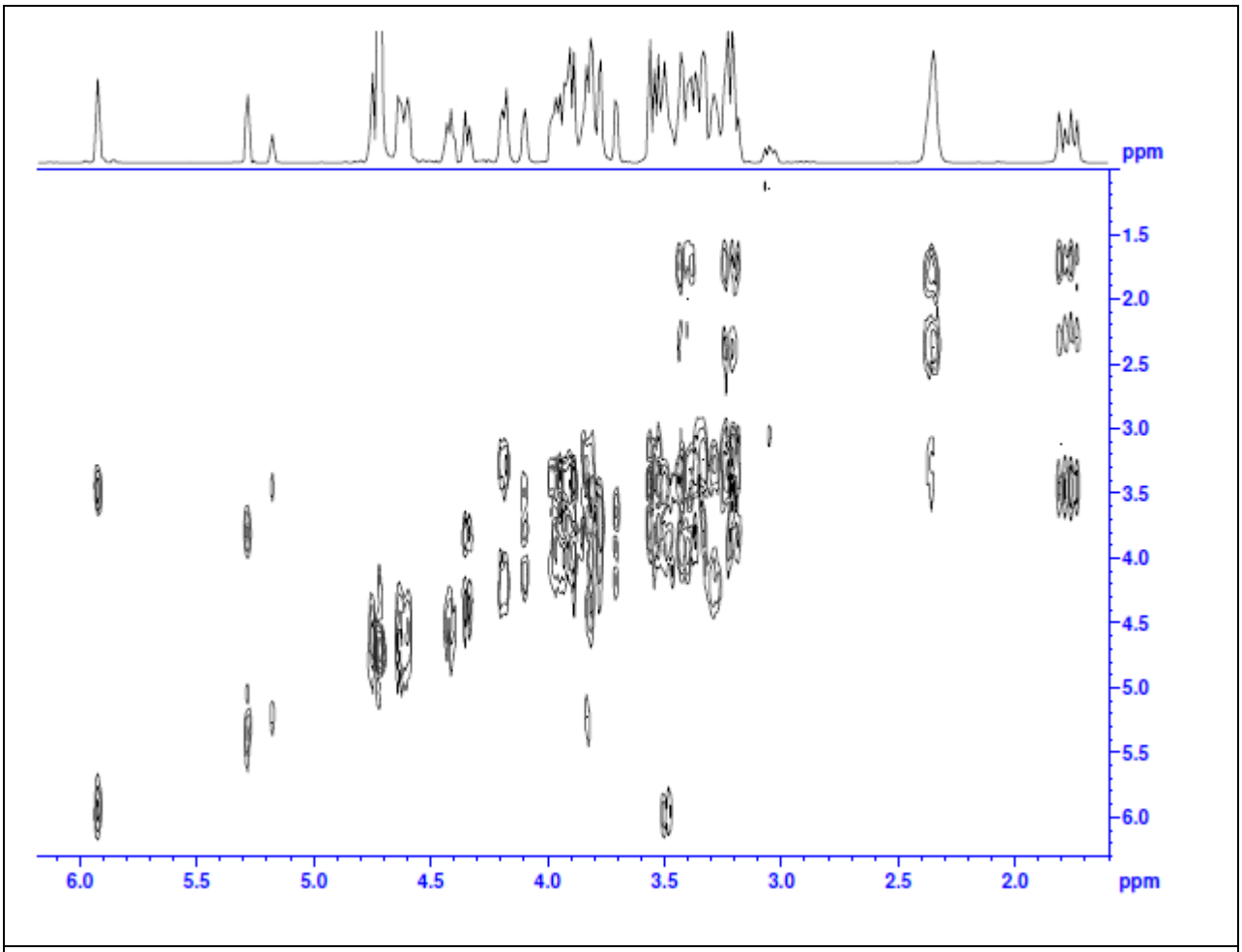

Figure S5. COSY-NMR of DPA51

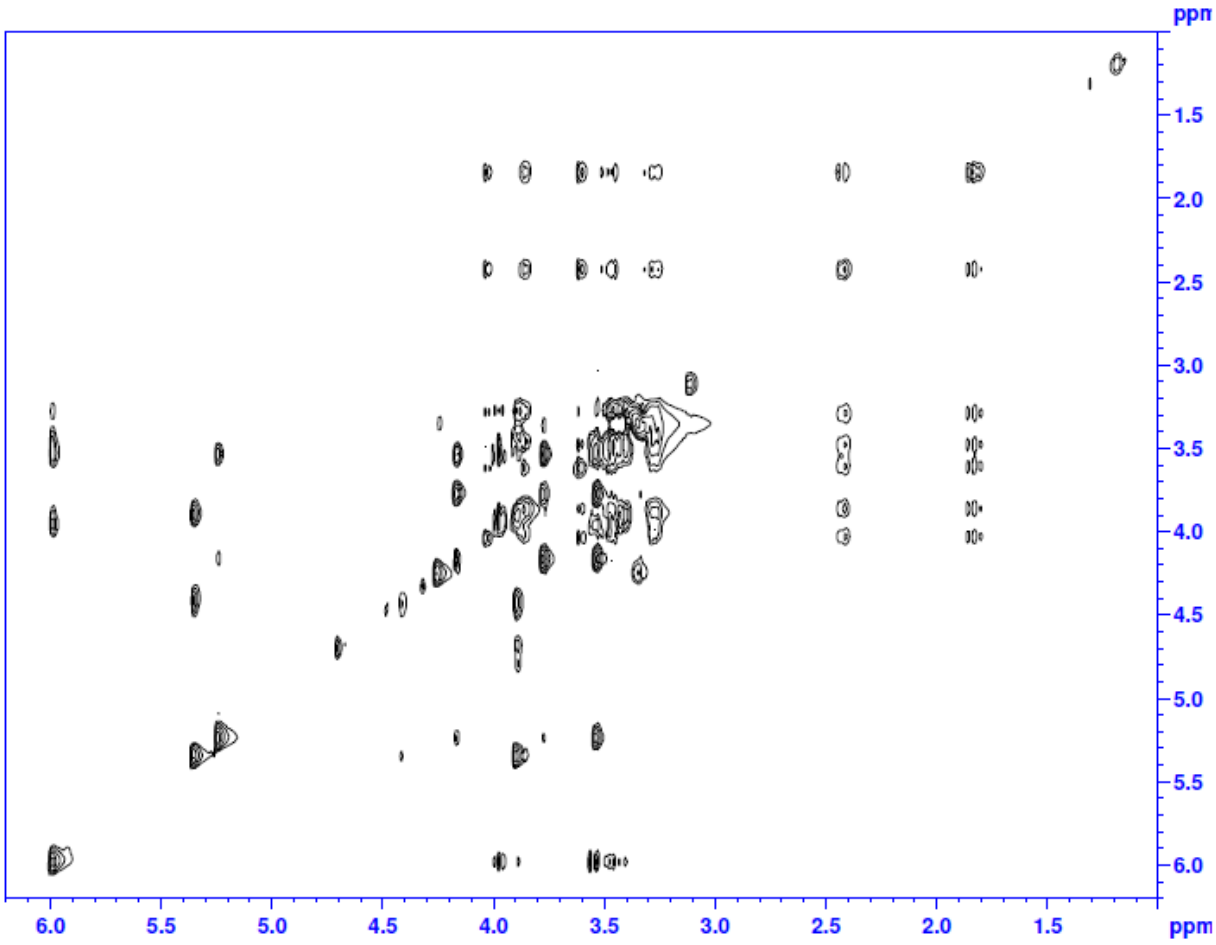

Figure S6. TOCSY-NMR of DPA51

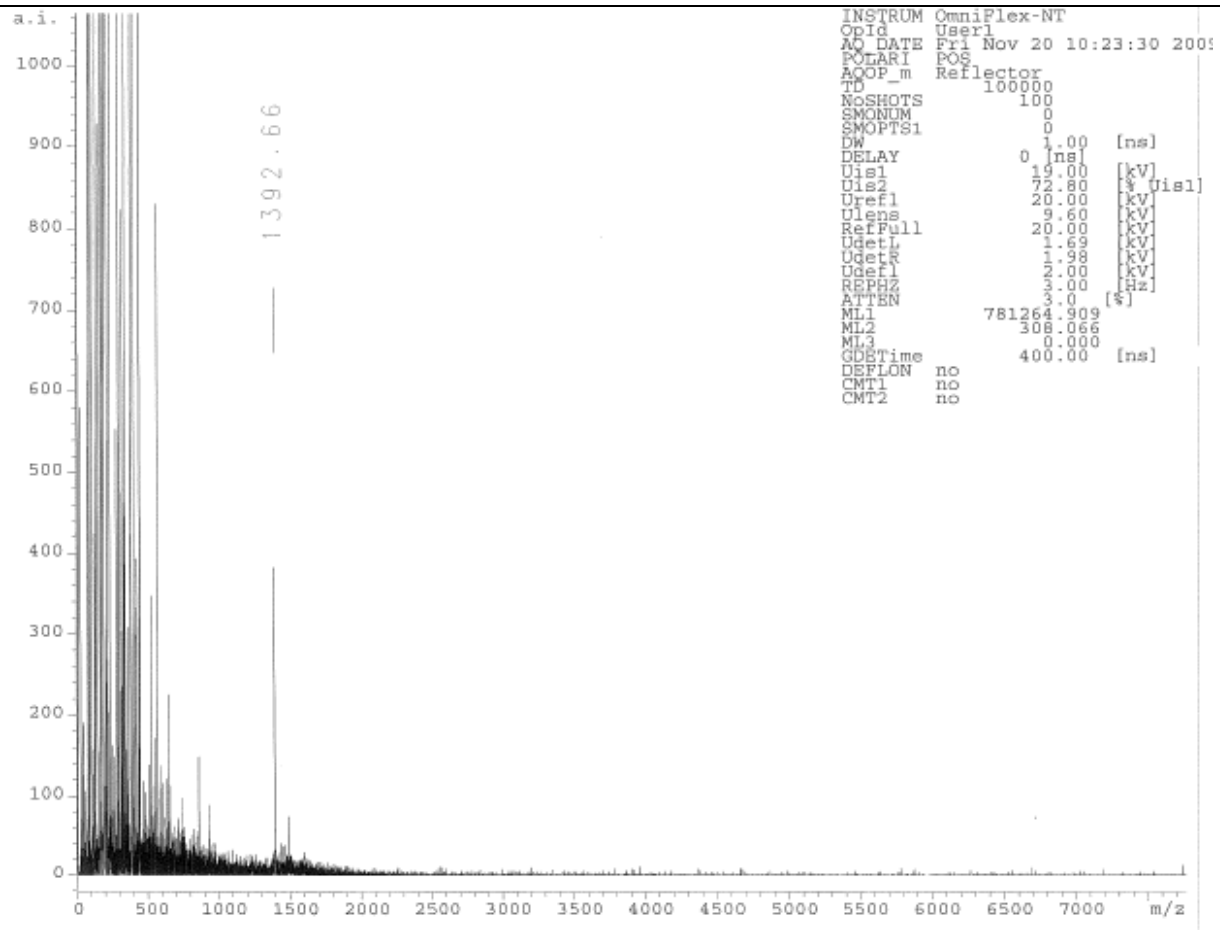

Figure S7. MALDI-TOF of DPA51

3. Conc. dependent UV thermal denaturation data for TAR RNA with DPA52

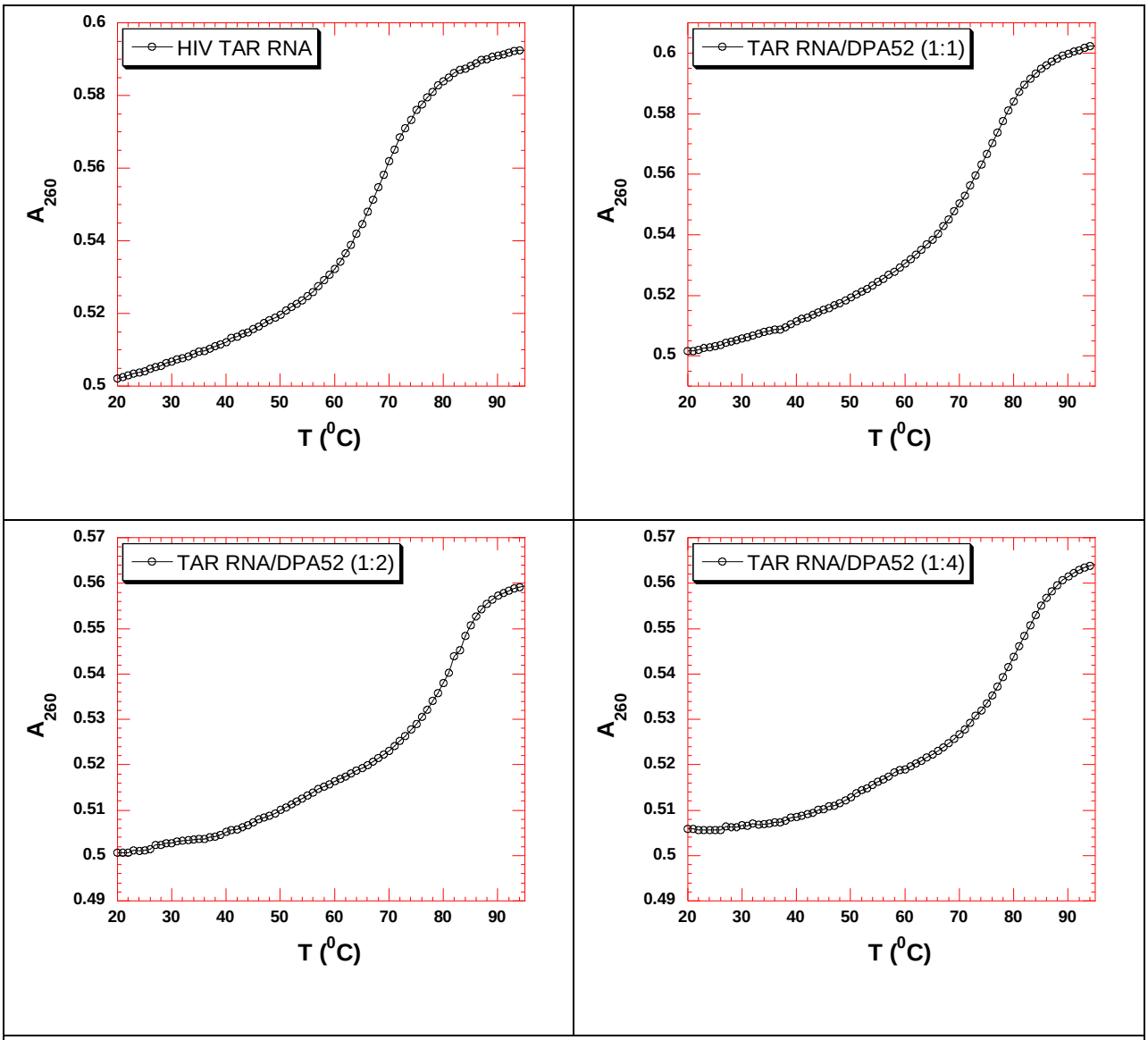

Figure S8. Conc. dependent UV thermal denaturation profile of HIV-1 TAR RNA with neomycin dimer. UV thermal denaturation data of HIV-1 TAR RNA at (A) 0, (B) 1 μM (1 mol. eq.), (C) 2 μM (2 mol. eq.), and (D) 4 μM (4 mol. eq.). Buffer conditions: 100 mM KCl, 10 mM SC, 0.5 mM EDTA, pH 6.8. HIV TAR RNA = 1 μM/strand. The heating rate was 0.3 °C.

Ratio (DPA52/TAR RNA)	T_m (°C)	ΔT_m(°C)
TAR RNA	68.9	-
1	78.2	9.3
2	80.4	11.5
4	81.3	12.4

Table S1. UV thermal denaturation data of HIV TAR RNA in the presence of increasing conc. of neomycin dimer (DPA52). Buffer conditions: 100 mM KCl, 10 mM SC, 0.5 mM EDTA, pH 6.8. HIV TAR RNA = 1 μM/strand. The heating rate was 0.3 °C.