

Supporting Information

Activation and Deactivation of DNAzyme and Antisense Function with Light for the Photochemical Regulation of Gene Expression in Mammalian Cells

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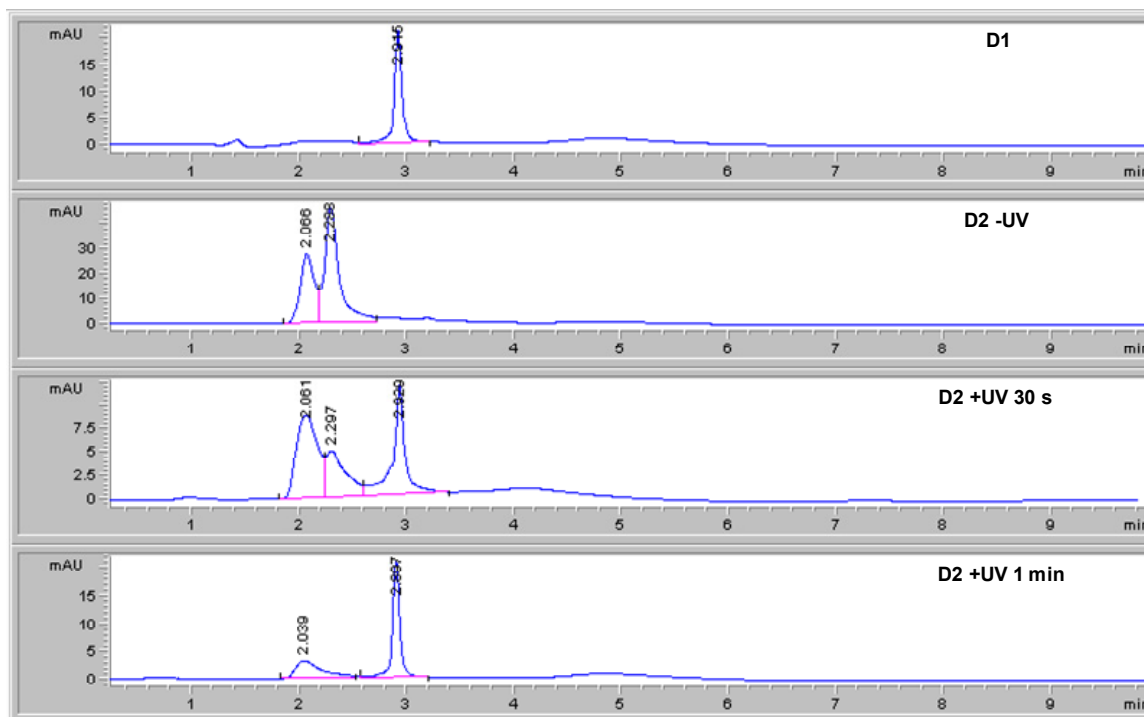


Figure S1. HPLC traces of DNAzymes **D1** and **D2** with and without irradiation. The non-caged DNAzyme (**D1**) has a retention time of 2.9 min, whereas **D2** possesses a caged thymidine, altering its retention time to 2.0/2.3 min. Upon irradiation at 365 nm for 1 min (25 W), **D2** is degraded to **D1** with 87% efficiency, as determined by peak integration.

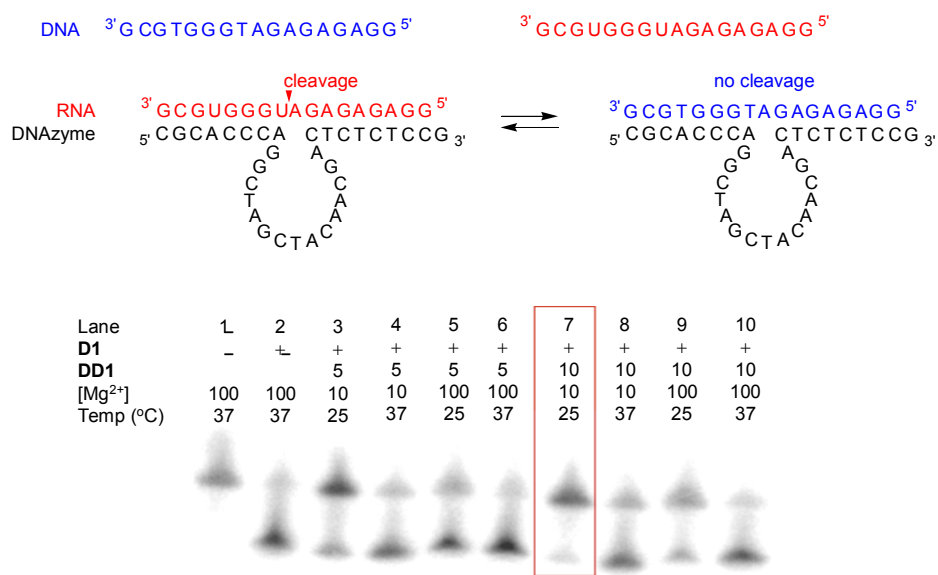


Figure S2. Optimization of decoy DNA complementary to the binding arms towards DNAzyme inhibition. Magnesium concentration (10 mM and 100 mM), DNAzyme decoy ratio (5:1 and 10:1), and temperature (25 °C and 37 °C) were altered to ascertain the best conditions for inhibition of DNAzyme activity. Ultimately, a 10:1 ratio of **DD1** to **D1** at 25 °C with 10 mM Mg²⁺ was determined to be the optimal condition for DNAzyme inhibition.

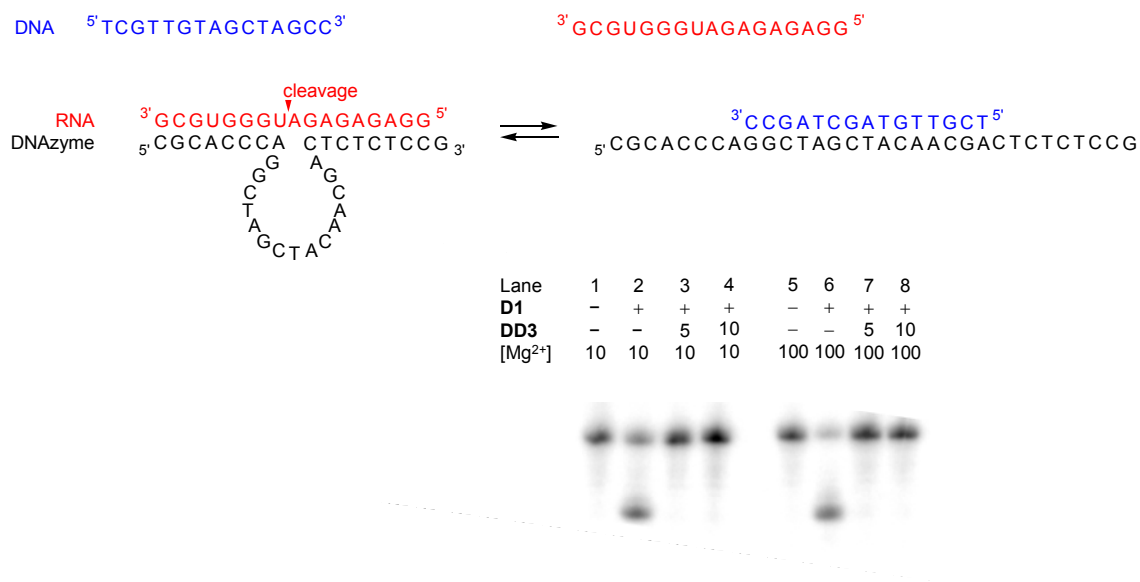


Figure S3. Optimization of decoy DNA complementary to the catalytic core towards DNAzyme inhibition. Magnesium concentration and DNAzyme decoy ratio were altered to ascertain the best conditions for inhibition of DNAzyme activity. Ultimately, a 5:1 ratio of **DD3** to **D1** with 10 mM Mg²⁺ was determined to be the optimal for DNAzyme inhibition.

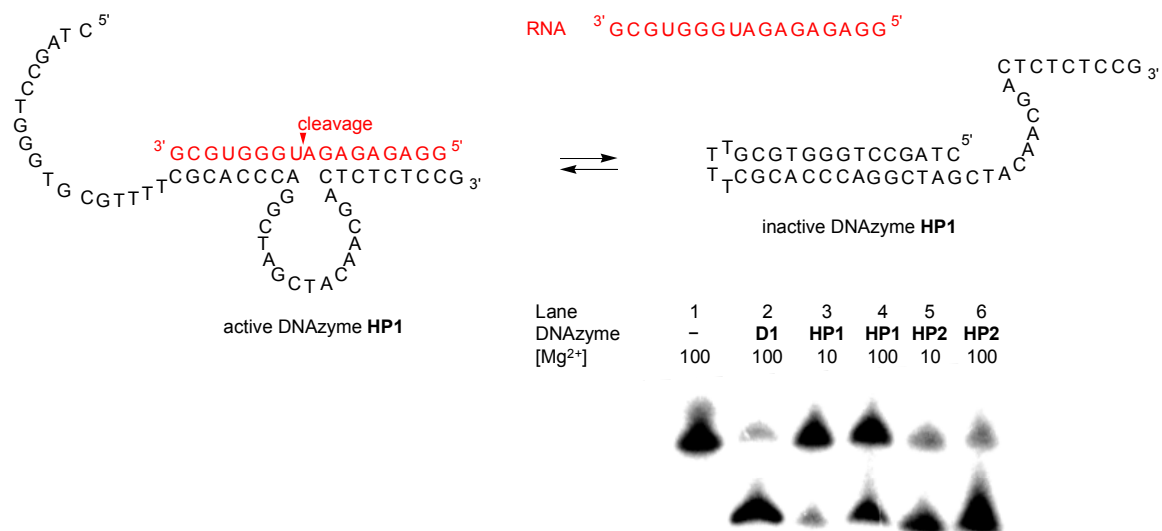


Figure S4. Optimization of hairpin formation towards DNAzyme inhibition. Magnesium concentrations and hairpin extension into the catalytic core were altered to ascertain the best conditions for inhibition of DNAzyme activity. Ultimately, **HP1** with 10 mM Mg²⁺ was determined to be the optimal for DNAzyme inhibition.

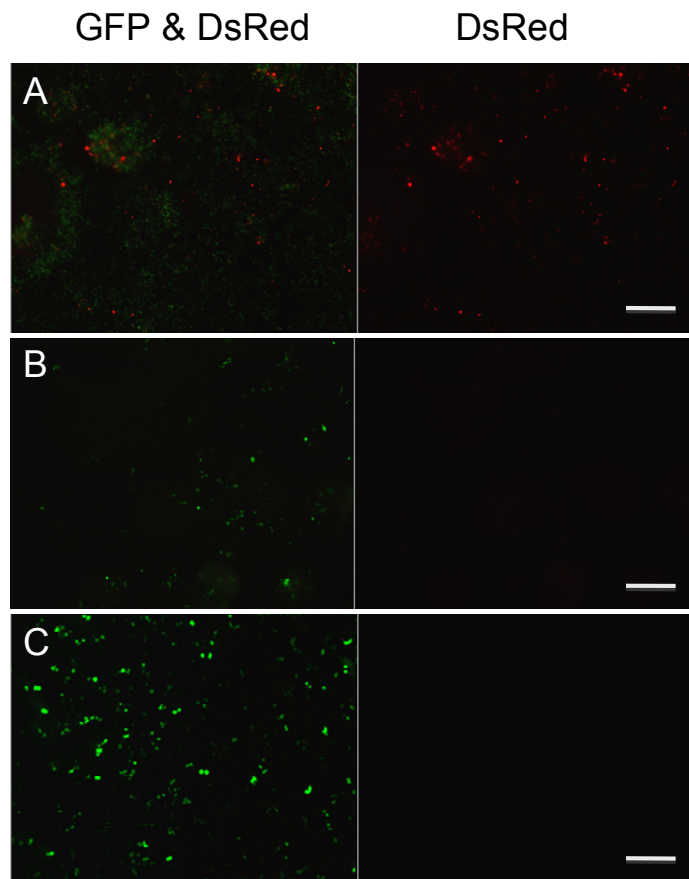


Figure S5. Determination of antisense activity *in vivo* using constructs containing either a mutated DNAzyme (**R3**) or a DNAzyme with the catalytic core removed (**R4**). A) Control DNA transfected into cells demonstrates no DsRed silencing. B) Construct **R3** containing a mutation in the catalytic core is still capable of silencing DsRed. C) Construct **R4** with the catalytic core removed again is still capable of silencing DsRed. These results suggest that catalytic DNAzyme activity is not required for *in vivo* silencing of genes, but rather may function via an antisense mechanism. Scale bar = 400 μm .