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Supporting Information

Two Pb^{2+}-specific DNAzymes with opposite trends in split-site-dependent activity

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Materials and Methods

1. Chemicals. The FAM-labeled substrate strand was purchased from Integrated DNA Technologies (IDT, Coralville, IA). The split enzyme strands are from Eurofins. Lead acetate was from Sigma-Aldrich. Pb^{2+} ions, urea, buffer and gel stock solutions were prepared with Milli-Q water. 2-[4-(2-hydroxyethyl)piperazin-1-yl]ethanesulfonic acid (HEPES) sodium salt, HEPES free acid and NaCl were from Mandel Scientific (Guelph, ON, Canada). 10× TBE and urea were purchased from Biobasic Inc. (Markham, ON, Canada).

2. Activity assays. For a typical gel-based activity assay, a final of 10 μM Pb^{2+} was incubated with 5 μL of 1 μM DNAzyme complex in buffer A (25 mM NaCl, 50 mM HEPES, pH 7.6). The DNAzyme complexes were prepared by mixing the FAM-labeled substrate and the enzyme with molar ratio 1:1.5 in buffer A. The mixture was annealed at 90 °C and gradually cooled down to room temperature. The enzymatic reactions were quenched with 8 M urea and run in 15% dPAGE at 120V for 80 min. The gel images were taken with Bio-Rad ChemiDoc MP imaging system. For kinetic experiments, a large volume of reaction mixture was prepared and aliquots were taken at designated time points into the quenching buffer.