Supplementary Material (ESI) for Chemical Communications

Supporting information for the paper:

"pH-Triggered Mg^{2+}-dependent DNAzymes"

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Experimental Section

Material: 4 2-(N-Morpholino)ethanesulfonic acid sodium salt (MES), sodium chloride and magnesium chloride were purchased from Sigma-Aldrich . DNA oligonucleotides (**3**) and (**3b**) were purchased from Integrated DNA Technologies Inc, (Coralville, IA). The products are HPLC pure, and were characterized by mass spectrometry. All other oligonucleotide sequences were purchased from Sigma-Genosys. Ultrapure water from NANOpure Diamond (Barnstead) source was used in all of the experiments.

Instrumentation: Light emission measurements were performed using a Cary Eclipse Device (Varian inc). The fluorophore ROX was excited at 580 nm.

Assay: The pH-switchable Mg^{2+}-dependent DNAzyme system described in Scheme 1B was studied in a solution consisting of **1** (1μM) ,**2** (1μM) and **3** (1μM) in MES buffer (1 M NaCl, 20 mM $MgCl_2$ pH =7.2 or pH=5.8). The ON-OFF system was generated by adding NaOH (1M) in order to change the pH value from 5.8 to 7.2, and acetic acid (20%) in order to change the pH value from 7.2 to 5.8 into a system containing **1** (10μM) ,**2** (10μM) and **3** (5 μM) in MES buffer (1 M NaCl, 20 mM $MgCl_2$).

The pH-switchable Mg^{2+}-dependent DNAzyme system described in Scheme 1C was studied in a solution consisting of **3** (1μM) ,**4** (1μM), **5** (1μM) and **6** (0.9 μM) in MES buffer (1 M NaCl, 20 mM $MgCl_2$ pH =7.2 or pH=5.8). The ON-OFF system was generated by adding NaOH (1M) in order to change the pH value from 5.8 to 7.2, and 4 μL of acetic acid (20%) in order to change the pH value from 7.2 to 5.8 into a system containing **4** (1 μM), **5** (1 μM) **3** (1 μM) and **6** (0.9 μM) in MES buffer (1 M NaCl, 20 mM $MgCl_2$).

Nondenaturing polyacrylamide gel electrophoresis: Gels contained 30% polyacrylamide (acrylamide/bis-acrylamide). Tris-borate-EDTA (TBE) was used as the separation buffer and consisted of Tris base (89 Mm, pH=7.9), boric acid (89 mM) and EDTA (2 mM). Gels were run on a Hoefer SE 600 electrophoresis unit at 25°C (300 V, constant voltage) for 3 hours. After electrophoresis, the gels were stained with SYBR Gold nucleic acid gel stain (Invitrogen) and scanned.

The DNA sequences are providing in table S1:

Table S1: Different DNA sequences used to construct the pH switchable DNAzyme systems. [a],[b]

Number	Sequence
(1)	5’ CCC CTT TTC CCC T TA ATG C CA CCC ATG T TA GAG A 3’
(2)	5’ C TGC TCA GCG ATG CAT TAT CCC CTT TTC CCC 3’
(3)	5’ ROX-CONH$(CH_2)_6$-TCT CTA TrAG GAG CAG-Black Hole Quencher 2 3’
(3b)	5’ ATA CGC TTA TCG GCA CAT GAG A TCT CTA TrA G GAG CAG GAG TGA ACT G 3’
(4)	5’ GTG ATG TTA CAAT TGC CAC CCA TGT TAG AGA 3’
(5)	5’ CTG CTC AGC GAT GCA CAT CCC TAA CCC TAA CCC TAA CCC 3’
(6)	5’ TTA GTG TTA GTG ATG ATT GTA ACA TCA C 3’

a. The bases underlined are the two non-complementary bases (T-C).
b. Oligonucleotides were custom ordered from Sigma or Integrated DNA technology and are HPLC purified and characterized by mass-spectrometry.