Electronic Supplementary Material (ESI) for ChemComm.

Supporting materials

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1. Details of the experimental procedures

Materials and Methods. Oligonucleotides were custom-made by Integrated DNA Technologies, Inc. (Coralville, IA). The concentration of the oligonucleotides in stock solutions was determined based on optical density at 260 nm, which was measured using a Perkin Elmer Lambda 35 UV/VIS Spectrometer (San Jose, CA). Fluorescent spectra were taken on a Perkin-Elmer LS-55 Luminescence Spectrometer (San Jose, CA) with a Hamamatsu Xenon lamp. Experiments were performed at the excitation wavelength of 485 nm and emission scan of 500-550 nm.

Fluorescent assay. Samples containing 200 nM **Mz F s1** and 10 nM **5iAND-Dz$_a$,** 10 nM **5iAND-Dz$_b$**, and 10 nM input strands in 50 mM HEPES, 50 mM $MgCl_2$, 20 mM KCl, 120 mM NaCl, 0.03% Triton X-100, 1% DMSO at pH 7.4, were incubated at 30 ^oC for 30 min. Fluorescence spectra of the samples were recorded on a PerkinElmer (San Jose, CA) LS-55 luminescence spectrometer with a Hamamatsu xenon lamp (excitation at 485 nm; emission 517 nm).

Native polyacrylamide gel electrophoresis (PAGE). Samples containing 10 nM **5iAND-Dz$_a$,** 10 nM **5iAND-Dz$_b$**, 200 nM **Mz F s1 mimic** and 10 nM input strands in 50 mM $MgCl_2$, 130 mM Tris, 45 mM boric acid and 25 mM EDTA, 50 mM $MgCl_2$, pH 8.3 were incubated 30 min at 30^{o}C. Twenty microliter of the samples were mixed with 5 μL of the gel loading buffer containing 50% glycerol, 0.02% bromophenol blue, 0.02% xylene cyanol. The samples were analyzed in 10% native polyacrylamide gel containing 130 mM Tris, 45 mM boric acid and 25 mM EDTA, pH 8.3, 50 mM $MgCl_2$. The electrophoresis was carried out at 45 min at constant voltage of 100 V. The gels were photographed using a UGENEUS Imaging System (Syngene, MD) after staining with SYBR Gold (Invitrogen, OR).

2. Table 1. Oligonucleotides used in this study.

Name	Sequence	Conc. nM	Purification*
Mz F s1	5'-AAG GT(dT-FAM) TCC TCg uCC CTG GGC A(BHQ-1) -3'	200	HPLC
Mz F s1 mimic	AAG GTTTCC TCGTCC CTG GGC A	200	SD
5iAND$_a$	TCA TCA TTA CCA GGC AGT ATT A TG CCA GGG A *GG CTA GCT* C TGA TAA GCT ACC CAC AAA TTC GGT TCT ACA GGG TAGCTTATC	10	SD
5iAND$_b$	TTGATGG CAA ACA CCA TTG TCA CAC TCC A TCA ACA TCA GT A CAA CGA *GAG GAA A*CCAC AAG ATC GGA TCT ACG GGT TTCC	10	SD
I1 (miR-200B)	5'-T AAT ACT GCC TGG TAA TGA TGA	10	SD
I2 (miR-99a)	5'-AAC CCG TAG ATC CGA TCT TGT G	10	SD
I3 (miR 10b)	5'-AGC TAC CCT GTA GAA CCG AAT TTG TGG G	10	SD
I4 (miR122)	5'-TGG AGT GTG ACA ATG GTG TTT G	10	SD
I5 (miR-21)	5'-T AGC TTA TCA GAC TGA TGT TGA	10	SD

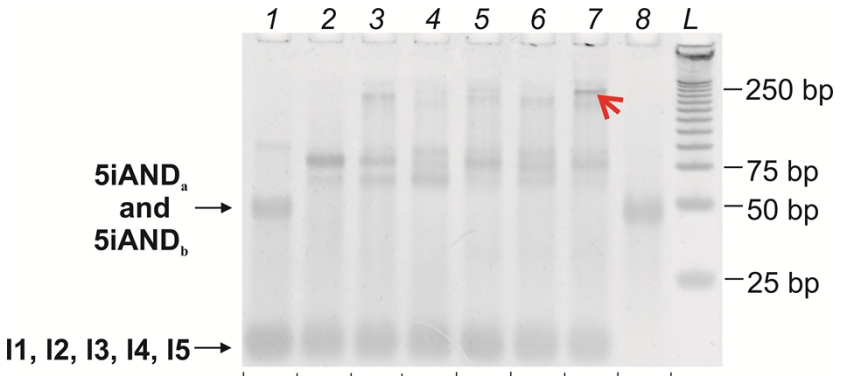
*SD, standard desalting; low case letters are ribonucleotides; dT-FAM fluorescein-conjugated deoxythymidine monophosphate residue; BHQ1, black hole quencher 1. Fragments of Dz core are in italic.

3. Figure S1. The truth table for 5-input AND gate

I1	I2	I3	I4	I5	Output	Bar # in Fig 2
0	0	0	0	0	0	1
1	0	0	0	0	0	2
0	1	0	0	0	0	3
0	0	1	0	0	0	4
0	0	0	1	0	0	5
0	0	0	0	1	0	6
1	1	0	0	0	0	7
1	0	1	0	0	0	8
1	0	0	1	0	0	9
1	0	0	0	1	0	10
0	1	1	0	0	0	11
0	1	0	1	0	0	12
0	1	0	0	1	0	13
0	0	1	1	0	0	14
0	0	1	0	1	0	15
0	0	0	1	1	0	16
1	1	1	0	0	0	17
1	1	0	1	0	0	18
1	1	0	0	1	0	19
1	0	1	1	0	0	20
1	0	0	1	1	0	21
1	0	1	0	1	0	22
0	1	1	1	0	0	23
0	1	1	0	1	0	24
0	1	0	1	1	0	25
0	0	1	1	1	0	26
1	1	1	1	0	0	27
1	1	1	0	1	0	28
1	1	0	1	1	0	29
1	0	1	1	1	0	30
0	1	1	1	1	0	31
1	1	1	1	1	1	32

Figure S1. **5iAND** gate truth table. The high output is produced only in the presence of all 5 inputs (last row).

4. Polyacrylamide gel electrophoresis (PAGE) analysis of the reaction mixtures containing different combinations of input oligonucleotides



5iAND$_a$	+	+	+	+	+	+	+	+
5iAND$_b$	+	+	+	+	+	+	+	+
Mz F s1 mimic	+	+	+	+	+	+	+	
I1		+	+	+	+		+	
I2		+	+	+		+	+	
I3		+	+		+	+	+	
I4		+		+	+	+	+	
I5			+	+	+	+	+	

Figure S2. Analysis of **5iAND** in polyacrylamide gel in the presence of different input combinations. The samples contained oligonucleotides as indicated on for each lane, which corresponds to bars 1, 27-32 in Figure 2b. Lane L contained 25 bp DNA ladder (Promega). The positions of **5iAND$_a$, 5iAND$_b$** and the oligonucleotide inputs are indicated. The full complex (322 nucleotides) shown in Fig 2A is indicated by the red arrow. The complex migrates roughly at 250 bp DNA markers.

In order to confirm the formation of the complex shown in Figure 2a, we analyzed **5iAND** gate by polyacrylamide gel electrophoresis (Figure S2). The gate was analyzed in the absence of all inputs (lane 1); in the presence of all possible combinations of 4 inputs (lanes 2-6, bars 27-31 in Figure 2b) and in the presence of all 5 inputs (Lane 7, last bar in Figure 2b).

It is of note that in the absence of the bridging input **I5** the formation of high molecular weight complexes (migrating above 100 bp marker) is negligible (lane 2), which correlates with the low fluorescent response of the gate (bar 27 in Figure 2B). The low mobility complex formed in the presence of all 5 inputs (red arrow in lane 7) corresponds to the structure depicted in Figure 2a. The formation of this complex observed in gel corresponds to the high fluorescent response of the **5iAND** (Figure 2B, last bar). At the same time, the presence of the complex is also visible in the absence of **I2** (lane 5), which correlates with the relatively high fluorescent response for this sample (Fig. 2B). Overall the data of PAGE analysis correlates with the fluorescent results and supports the hypothesis of the formation of catalytically active complex shown in Figure 2a in the presence of all 5 inputs.