Supporting Information

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MNAzyme-catalyzed nucleic acid detection enhanced by a cationic copolymer

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Figure S1. Metal ion dependency (square: 5 mM MnCl₂, circle: 25 mM MgCl₂) of MNAzyme activity in the absence of PLL-g-Dex at 2 nM target, 200 nM substrate, and 200 nM MNAzyme at 50°C.



B



Figure S2. (A) Sequences of parent DNAzymes of MNAzyme A (long arm) and B (short arm). (B) Temperature dependences of multiple-turnover reaction rate for the DNAzymes having long (square) and short (circle) substate-binding arms at [substrate]₀ = 200 nM, [DNAzyme] = 6.7 nM, and [MnCl₂] = 5 mM in the absence (hollow) or presence (solid, N/P=2) of PLL-g-Dex.

Table S1. Melting temperatures (*T*m) of duplexes having sequences of substrate- or target binding domains of MNAzymes A and B extimated at [substrate] = [MNAzyme] = [target] = 200 nM, [Na⁺] = 200 mM, [Mg²⁺] = 25 mM.

	<i>T</i> m / °C						
	5' MNAzyme bi	inding domain	3' MNAzyme binding domain				
	А	В	А	В			
Substrate	55.6 (10 bp)	27 (7 bp)	40.1 (11 bp)	23.3 (8 bp)			
Target	61.6 (15 bp)		64.0 (14 bp)				

The melting temperatures (*T*m) of duplexes with DNA sequences from the substrate and target binding domains were estimated by UINAMelt.¹



Figure S3 Figure 5B plotted with reduced y-axis.

The MNAzyme LOD determination

1. The repeated blank sample measurement ($\Delta I = I_{t=60} - I_{t=0}$)

Table S1. ΔI from blank MNAzyme reactions.

In	the	absence	of PLI	L-g-Dex
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		1	2	-	3	4		5	6		7	mea	n	σ
$\Delta I / a.u$	ı. 0.	.78	0.855	0.8	319	0.804	0.	756	0.855	0	.553	0.77	4	0.097
In the presence of PLL-g-Dex														
	1	2	3	4	5	6	7	8	9	10	11	12	mean	σ
ΔI / a.u.	2.698	2.138	2.852	1.59	2	2.11	2.167	1.861	2.356	1.98	1.54	2.16	2.121	0.37

200nM Substrate, 6.7 nM short partzymes, 5 mM Mn²⁺, 0.012 mg/ml (N/P=2) PLL-g-Dex, 37°C)

2. The slope of fluorescence intensity to target concentration.



Figure S4. MNAzyme reaction with different target concentration

200nM Substrate, 6.7 nM short partzymes, 5 mM Mn²⁺, 0 mg/ml (N/P =0) or 0.012 mg/ml (N/P=2) PLL-g-Dex, 37°C

	PLL-g-Dex (-)	PLL-g-Dex (+)
3σ	0.29	1.11
slope (a.u./pM)	0.0016	0.527
So LOD (= 3σ / slope) (pM)	181	2.1

Reference

(1) Markham, N. R.; Zuker, M., DINAMelt web server for nucleic acid melting prediction. Nucleic Acids Res., 2005, 33, W577-W581.