Electronic Supplementary Material (ESI) for Sensors & Diagnostics.

Supporting information for

A sensitive isothermal fluorescence biosensor for microRNAs detection coupling Primer Exchange Reaction with Catalytic Hairpin Assembly

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Table S1. Sequences of DNA and RNA probes used.

Name	Sequence（from 5’ to 3’）
miR-200a	UAA CAC UGU CUG GUA ACG AUG U
Primer **a**	CAT CAT CAT
Self-gated Hairpin	GGT GCA TCA TCA TAC ATC GTT ACC AGA CAG TGT TAA CAT CAT CAT GGG CCT TTT GGC CCA TGA TGA TGT ATG ATG ATG CAC C
Hairpin-2	GGCATCATCATACATCGTTACCAGACAGTGTTA ACATCATCATGGGCCTTTTGGCCCATGATGATG TATGATGATGCC
Hairpin-4	CCCTCCCATCATCATACATCGTTACCAGACAGT GTTAACATCATCATGGGCCTTTTGGCCCATGAT GATGTATGATGATGGGAGGG
PER-CHA-H1	BHQ1-ATG TAT GAT GAT GTA TGA TGA TGT TCC AAT CAC AAC ACA TCA TCA TAC ATC ATC-FAM
PER-CHA-H2	GTA TGA TGA TGT GTT GTG ATT GGA ATC ATC ATA CAT TCC AAT CAC AAC ACA TCA
PER-CHA-H3	GTT GTG ATT GGA ATG TAT GAT GAT ACA TCA TCA TAC ATC ATC ATA CAT TCC AAT
miR-200b	CAU CUU ACU GGG CAG CAU UGG A
miR-200c	CGU CUU ACC CAG CAG UGU UUG G
miR-429	UAA UAC UGU CUG GUA AAA CCG U
miR-141	CAU CUU CCA GUA CAG UGU UGG A

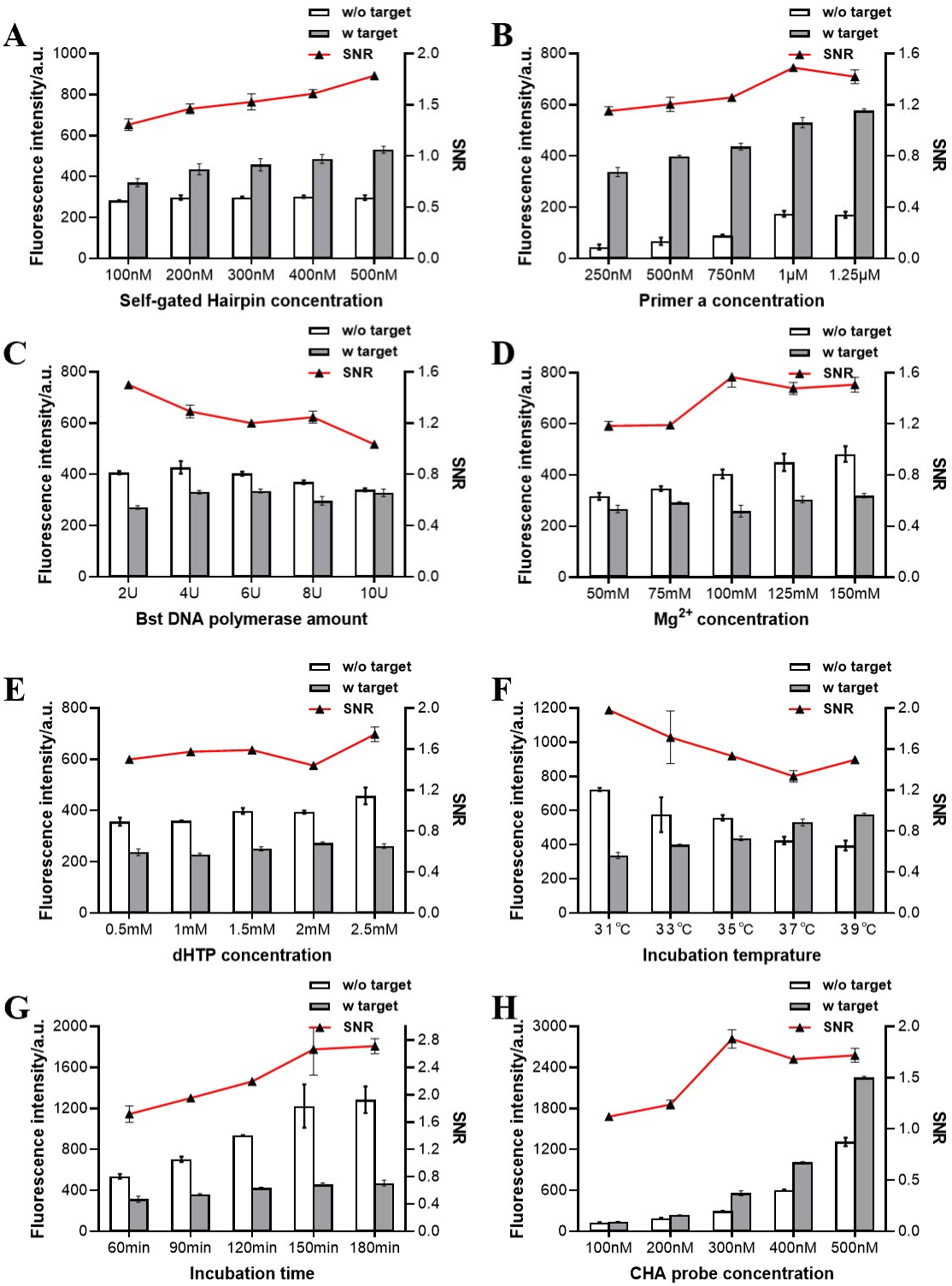

Figure S1. Optimization of the experimental parameters. Fluorescence intensity and SNR results of various parameters, including (A) Self-gated Hairpin concentration, (B) Primer a concentration, (C) Bst DNA polymerase amount, (D) Mg2+ concentration, (E) dHTP concentration, (F) the incubation temperature, (G) the incubation time, and (H) the CHA probes concentration. The concentration of target miR-200a was 1 nM. Error bars: the standard deviation of triplicate independent measurements.

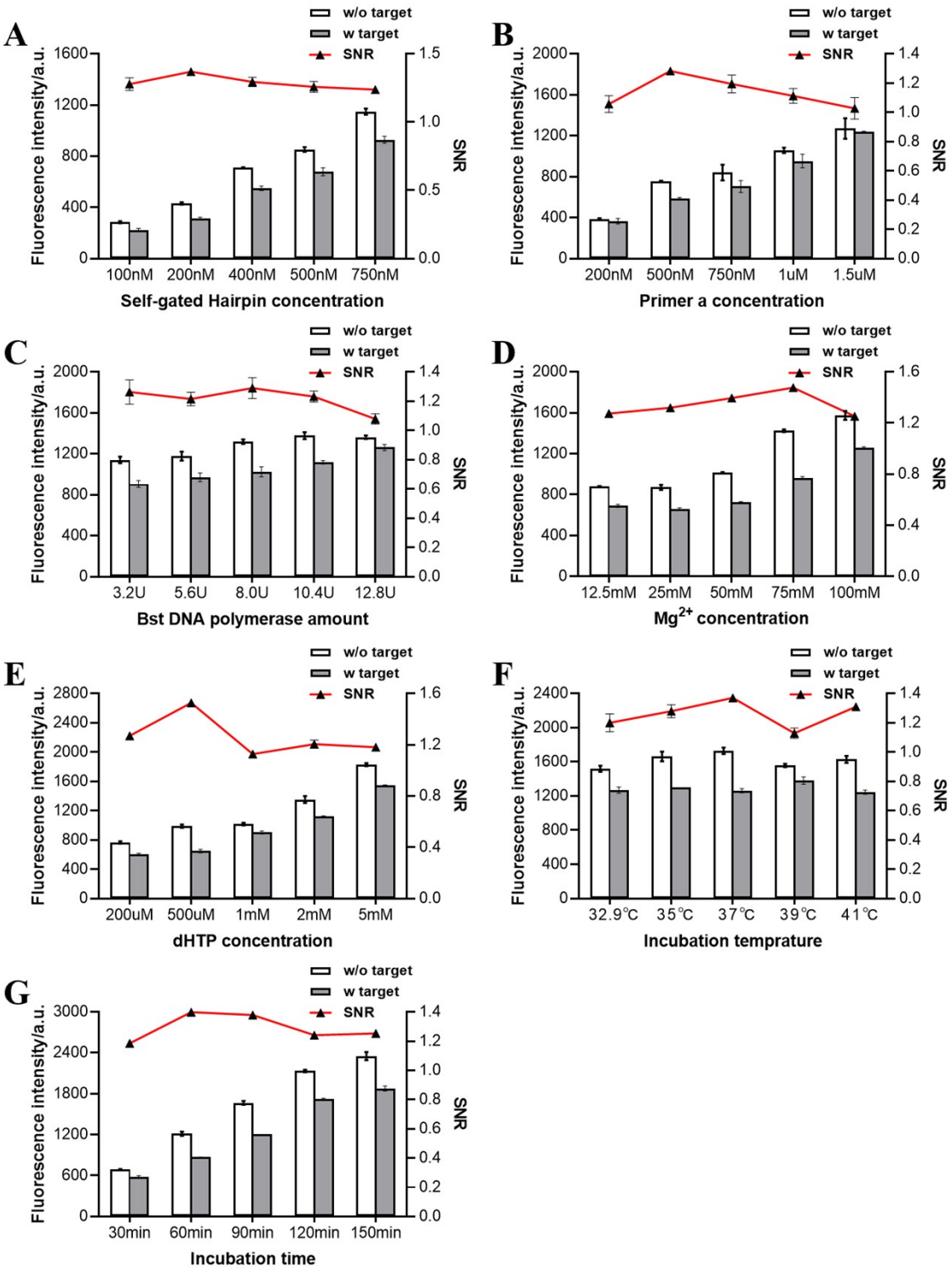

Figure S2. Optimization of the experimental parameters of PER. Fluorescence intensity with and without target of various parameters, including (A) Self-gated Hairpin concentration, (B) Primer a concentration, (C) Bst DNA polymerase amount, (D) Mg^{2+} concentration, (E) dHTP concentration, (F) the incubation temperature and (G) the incubation time. The concentration of target miR-200a was 1 nM. Error bars: the standard deviation of triplicate independent measurements.

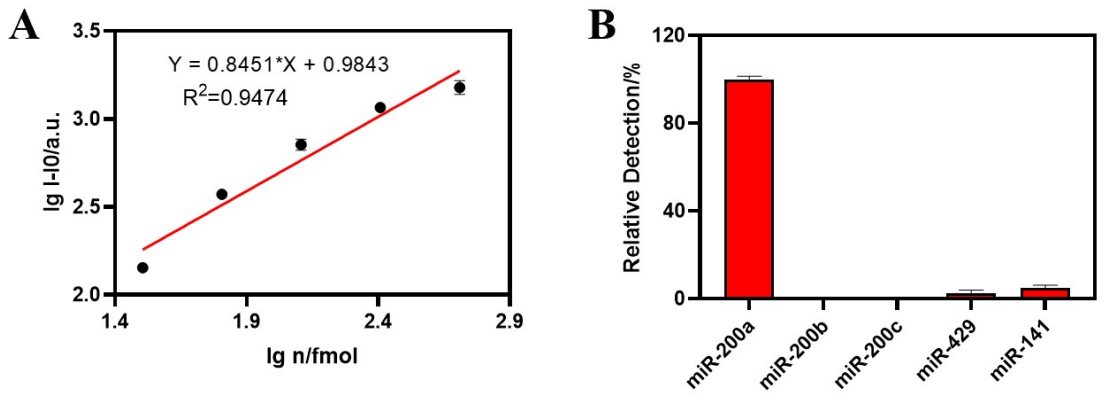

Figure S3. Analytical performance of the proposed biosensor. The Linear correlation between logarithmic fluorescence intensity and logarithmic concentration of PER (A). Selectivity of PER strategy. (B). Error bars: the standard deviation of triplicate independent measurements.

Table S2. Comparison of previously reported CHA method for miRNA detection

Target	Method	Reaction time	Linear range	LOD	Ref.
miR-21	Catalytic-hairpin-assembly assisted DNA tetrahedron nanoprobe	8 h	0.1-10 nM	120 pM	[1]
miR-21	Duplex-specific nuclease and catalytic hairpin assembly	1 h	10 fM-100 pM	5.4 fM	[2]
miR-21	Catalytic hairpin assembly coupled with enzymatic repairing amplification	2h	100 fM-1 nM	50 fM	[3]
miRNA	Rolling circle amplification-based DNA machine coupling catalytic hairpin assembly with DNAzyme formation	75 min	1 fM-1 pM	0.68 fM	[4]
miR-155	Cascaded catalytic hairpin assembly	1 h	10 pM-1000 pM	6.9 pM	[5]
miR-21	DNA nanowire based localized catalytic hairpin assembly	3 h	0-8 nM	2.0 pM	[6]
miR-let-7a	Catalytic hairpin assembly and spherical nucleic acid	2 h	0.1-100 pM	53.7 fM	[7]
miR-200a	Primer exchange reaction coupling with catalytic hairpin assembly	1 h	250 pM-10 nM	14.35 pM	this work

Table S3. Recovery detection of miR-200a in 20% human serum (n=3)

miRNA	Added (fmol)	Detected (fmol)	Recovery (%)	RSD (%)
miR-200a	128	130.7	102.1	5.72
	256	266.4	104.1	1.82
	512	436.2	85.2	1.22

References

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