

Target-to-Signal Conversion and Spatial Enrichment Cascade Boosts

CRISPR/Cas12a Biosensing for Trace-Level Pathogen Detection

Yu Liu^{a, †}, Xudong Yue^{b, †}, Bowen Li^b, Haoyu Yang^a, Yu Wang^b, Shenguang Ge^{a, c}, Su Liu^{a, *}

^aSchool of Water Conservancy and Environment, University of Jinan, Jinan 250022, P.R. China

^bSchool of Biological Sciences and Technology, University of Jinan, Jinan 250022, P.R. China

^cJinan Engineering Research Center of Plant-Microbial Interaction, Jinan, 250022, P.R. China

† These authors contributed equally to this work

*Corresponding author: Su Liu, stu_lius@ujn.edu.cn

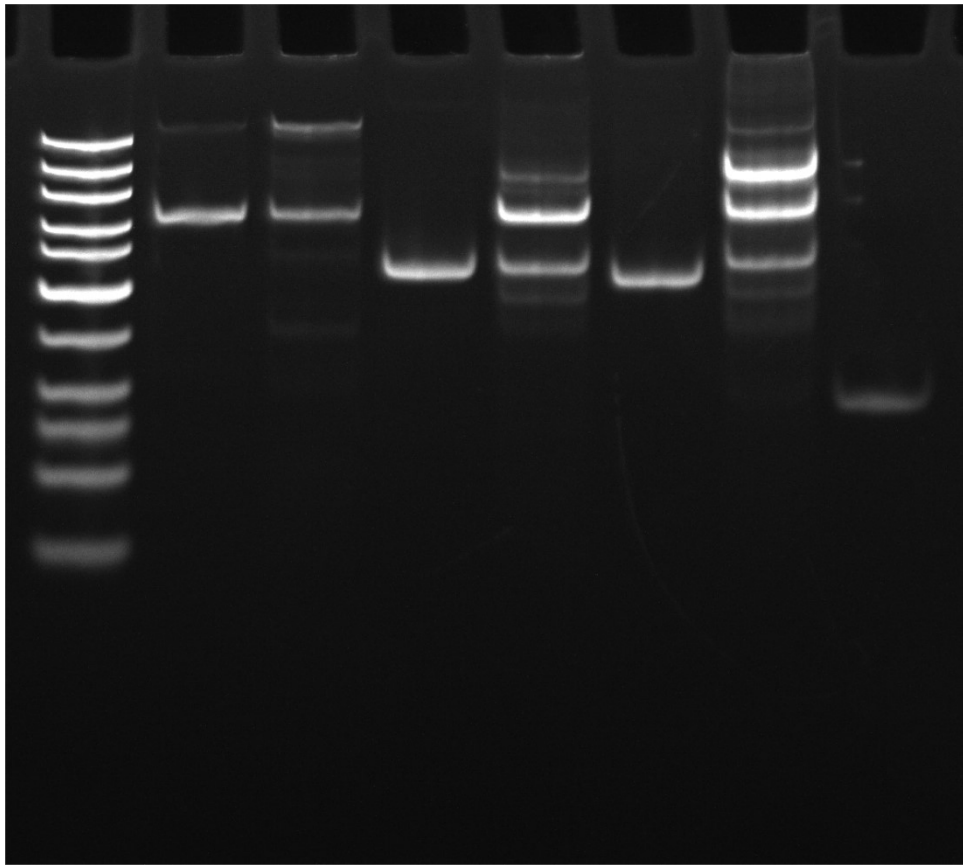


Fig. S1. The original image of Figure 2A in the paper

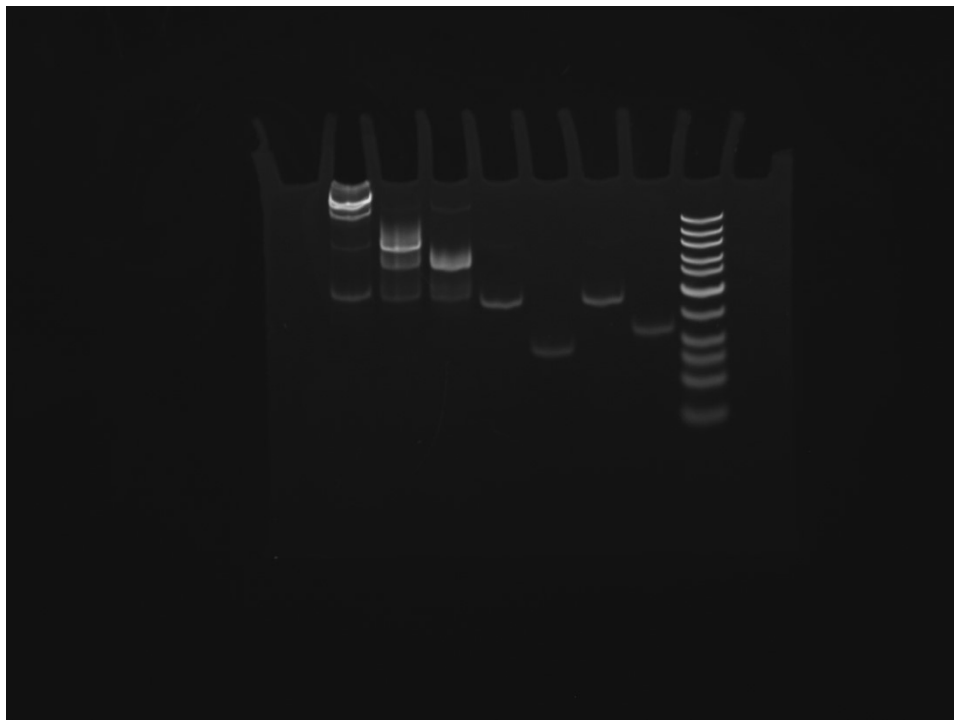


Fig. S2. The original image of Figure 2B in the paper

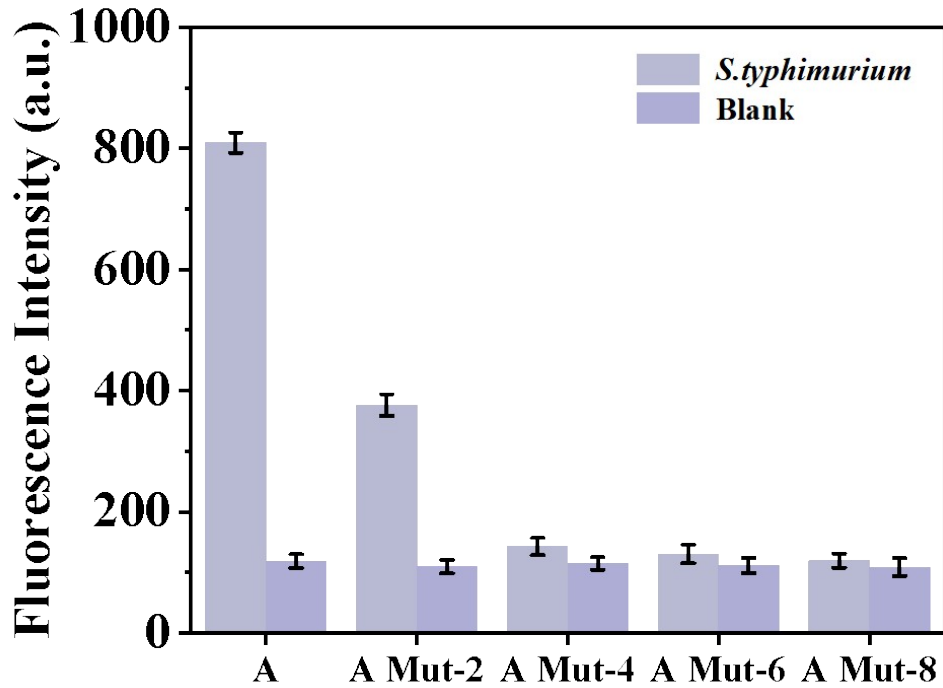


Fig. S3. Verification of mismatched sequences in Stand A

Table 1: The oligonucleotide sequences used in this study

DNA name	Sequence (5' to 3')
Apt	CTCCTCTGACTGTAACCACGGTGGTTTGATCACTATTGGGCCTTTGTGATGTCGGT AGT ^[40]
A	ACCGAGCCACTACCGACATCACAAAGGCCCTGACGAACGGTT ^[40]
AB	ACCGAGCCACTACCGACATCACAAAGGCCCTGACGAACGGTTG/rA/T AATGACGAACTCCTCCTTATGATTTGTGATGTCGGTAGT
DNAzyme	AACAATCAAACCGTTCGTCAGGGCCTTTGTGATGTCGGTAGTGGCTCGGACCCCTC <u>CGAGCCGGTCGTCGAAACTCTCTCGAACTATTTGATTGTT</u>
H1	ACTACCGACATCACAAATCATAAGGAGGAGTTCGTCATTAGGGCGGC CTTTGTGATGTCGGGCTGAGCTTTAAAGCAA
H2	CGGCCTTTGTTTCAGACTTAGGAACCGACATCACAAAGGCCGCC
H3	BHQ1-GGTTTTGCTTTAAAGCTCAGCCCGTT-FAM-CCTAAGTCTGAAC AAAGGCCGACTACCGACATCATCTGAGCTTTAAAG
crRNA	UUGCUIUAAAGCUGCUAGAUGUGAAUCAUCUIUAAUU

Table 2: Comparison of different S.typhimurium detection methods

Detection method	Limit of detection (LOD, cfu/mL)	Detection range (cfu/mL)	Assay time	References
Microfluidic sensor	63	$10^2 \sim 10^7$	1.5 h	1
Colorimetric	11	$10 \sim 10^4$	2.5 h	2
Fluorescence	82	$50 \sim 10^7$	3h50min	3
Electrochemical	100	$25 \sim 1.0 \times 10^5$	20 min	4
This study	6.26	$10 \sim 1 \times 10^5$	60 min	

References

- (1) Lin, X.; Li, C.; Meng, X.; Yu, W.; Duan, N.; Wang, Z.; Wu, S. CRISPR-Cas12a-mediated luminescence resonance energy transfer aptasensing platform for deoxynivalenol using gold nanoparticle-decorated Ti3C2Tx MXene as the enhanced quencher. *JOURNAL OF HAZARDOUS MATERIALS* **2022**, *433*, Article. DOI: 10.1016/j.jhazmat.2022.128750 .
- (2) Jiang, H.; Jiang, D.; Liu, X.; Yang, J. A self-driven PET chip-based imprinted electrochemical sensor for the fast detection of Salmonella. *SENSORS AND ACTUATORS B-CHEMICAL* **2021**, *349*, Article. DOI: 10.1016/j.snb.2021.130785.
- (3) Wang, H.; Wu, Q.; Zhou, M.; Li, C.; Yan, C.; Huang, L.; Qin, P. Development of a CRISPR/Cas9-integrated lateral flow strip for rapid and accurate detection of Salmonella. *FOOD CONTROL* **2022**, *142*, Article. DOI: 10.1016/j.foodcont.2022.109203.
- (4) Wang, L.; Qi, W.; Wang, M.; Jiang, F.; Ding, Y.; Xi, X.; Liao, M.; Li, Y.; Lin, J. A pipette-adapted biosensor for Salmonella detection. *BIOSENSORS & BIOELECTRONICS* **2022**, *218*, Article. DOI: 10.1016/j.bios.2022.114765.