Supplementary Information (SI) for Analyst.
This journal is © The Royal Society of Chemistry 2024

Construction of DNA walker nanomachine aptasensor for the simultaneous detection of dual-cancer biomarkers

Songbai Tian[a,c,&], Jiao Zheng[a,b,&], Xinghu Ji[b], Fuxiang Zhou[a], Zhike He[a,b]*

[a]Department of Radiation and Medical Oncology, Zhongnan Hospital of Wuhan University, Hubei Key Laboratory of Tumor Biological Behaviors, Hubei Province Cancer Clinical Study Center, Zhongnan Hospital of Wuhan University, 430071 Wuhan, China

[b]College of Chemistry and Molecular Sciences, Wuhan University, 430072, P. R. China

[c]School of Basic Medical Sciences, Hubei University of Medicine, 442000 Shiyan, China.

[&]These authors contributed equally to this work and should be considered co-first authors.

E-mail: zhkhe@whu.edu.cn

Table of content

Experiments section.

Characterization of AuNPs. As a proof-of-concept demonstration, we selected CEA aptamer and MUC-1 aptamer because both have been shown to be highly selective for CEA and MUC-1, cancer biomarkers can be used for risk assessment, diagnosis, prognosis in the early detection of cancer. To achieve excellent analytical performance for the DNA walker nanomachine aptasensor, an appropriate gold nanoparticle (AuNP) is needed for optimal walking efficiency. As shown in Figure S1, 16.5 ± 1.94 nm of AuNP was synthesized and the particles disperse extremely well.

Optimization the concentration of EXO I. To optimize the performance of the designed DNA walker nanomachine aptasensor system, when we tested the effect of concentration of EXO I, we observed that the fluorescence of FAM signal at 520 nm was increased with the increment of concentration of EXO I (Figure S3). After the addition of 25 U to this aptasensor, a strong fluorescence was measured and kept stable, demonstrating that 20 U EXO I could be opted for the following study.

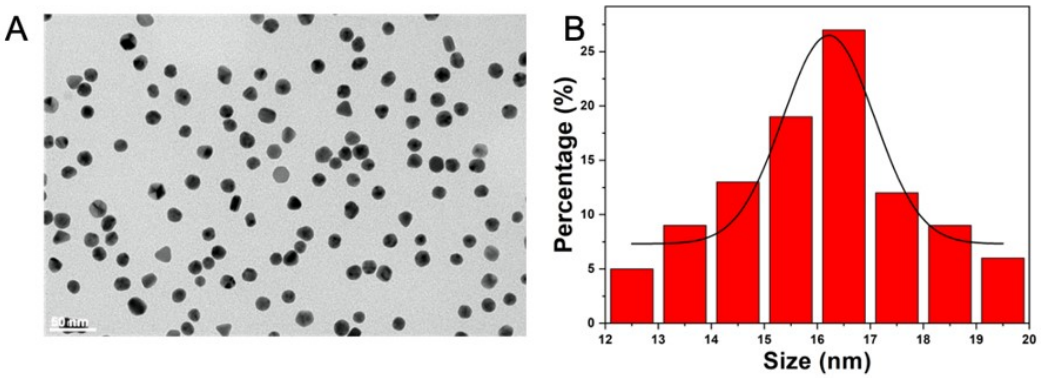

Figure S1. Characterization of AuNPs. (A) TEM images of the synthesized AuNPs. (B) The size distribution of AuNPs.

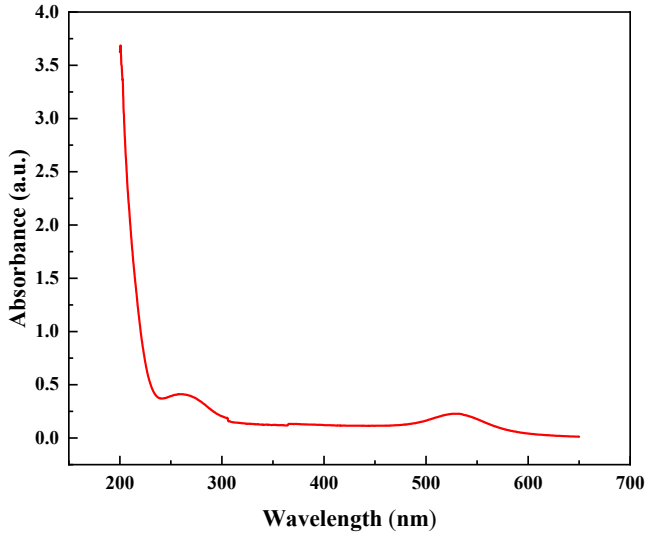

Figure S2 UV−vis absorption spectra of AuNPs-DNA.

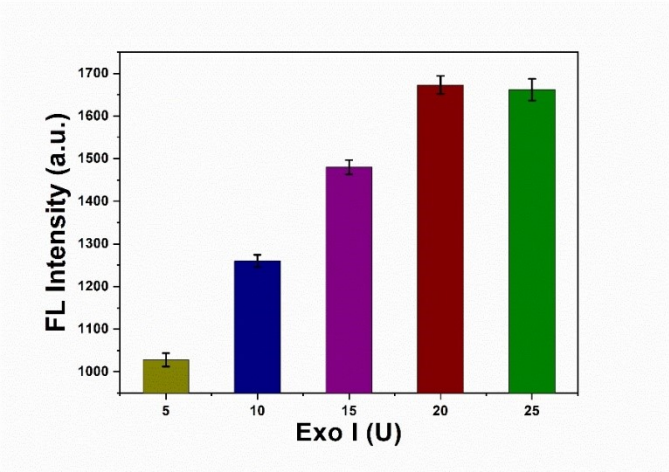

Figure S3. Effect of the concentration of Exo I for aptasensor.

Table S1. Comparison of the reported chemosensors for MUC-1 and CEA.

Target	Dynamic range (ng/mL)	Limit of detection (ng/mL)	Refs.
MUC-1	500–5.0× 10^4	62.5	[1]
	0.10-100	0.53	[2]
	1.0-100	0.43	[3]
	0.167-16.67	0.08	This work
CEA	5-40	3.4	[4]
	4-25	0.19	[5]
	1-110	0.3	[6]
	0.167-3.34	0.1	This work

Notes and References.

1. J. Tian, T. Huang and J. Lu, *Analytical Methods*, 2016, **8**, 2375-2382.
2. K. Kuntamung, J. Jakmunee and K. Ounnunkad, *Journal of Materials Chemistry B*, 2021, **9**, 6576-6585.
3. P. Song, M.-L. Wang, Y.-X. Duan, A.-J. Wang, Y. Xue, L.-P. Mei and J.-J. Feng, *Microchimica Acta*, 2023, **190**, 85.
4. Q.-L. Wang, H.-F. Cui, X. Song, S.-F. Fan, L.-L. Chen, M.-M. Li and Z.-Y. Li, *Sensors and Actuators B: Chemical*, 2018, **260**, 48-54.
5. L. Yang, M. Cui, Y. Zhang, L. Jiang, H. Liu and Z. Liu, *Sensors and Actuators B: Chemical*, 2022, **350**, 130857.
6. J. Qian, C. Wang, X. Pan and S. Liu, *Analytica Chimica Acta*, 2013, **763**, 43-49.