

Supporting Information

A rapid immunoassay for dual-mode detection of HPV16 and HPV18 DNA

based on Au@PdPt nanoparticles

Huanxin Xiao;^a Weiguang Chen;^a Mingxia Lin;^a Shilin Jiang;^a Xiping Cui^{a,*} and

Suqing Zhao^{a,*}

^a Department of Pharmaceutical Engineering, School of Biomedical and Pharmaceutical Sciences, Guangdong University of Technology, Guangzhou 510006, People's Republic of China

*Corresponding Author:

Prof. Suqing Zhao

Tel.: +86 15820258676

E-mail address: sqzhao@gdut.edu.cn (S.-Q. Zhao)

Dr. Xiping Cui

Tel.: +86 18813758818

E-mail address: cuxiping1989@163.com (X.-P. Cui)

E-mail addresses for other authors:

hxq323723@163.com (H.-X. Xiao); weiguangchen2023@163.com;

lmxmingxia@163.com (M.-X. Lin); jiangshilin_22@163.com (S.-L. Jiang).

Figure and table captions

Table. S1 Sequence of standard DNA-RNA hybrid and RNA probe

Names	Sequences	
	sense (5'-3') DNA	antisense (5'-3') RNA
HPV16 RNA probe	/	Digoxin- CGACCCCUUAUUAUGGAAU CUUUGCUIUUUGUCC
HPV18 RNA probe	/	Biotin- CUAUACACCACAAUAAAUCU UUAAAUGCA
HPV 16 DNA- RNA-digoxin	GGACAAAAAGCAAAGATTCC ATAATATAAGGGGTCCG	Digoxin- CGACCCCUUAUUAUGGAAU CUUUGCUIUUUGUCC
HPV18 DNA-RNA- biotin	TGCATTAAAGATTTATTTGT GGTGTATAG	Biotin- CUAUACACCACAAUAAAUCU UUAAAUGCA

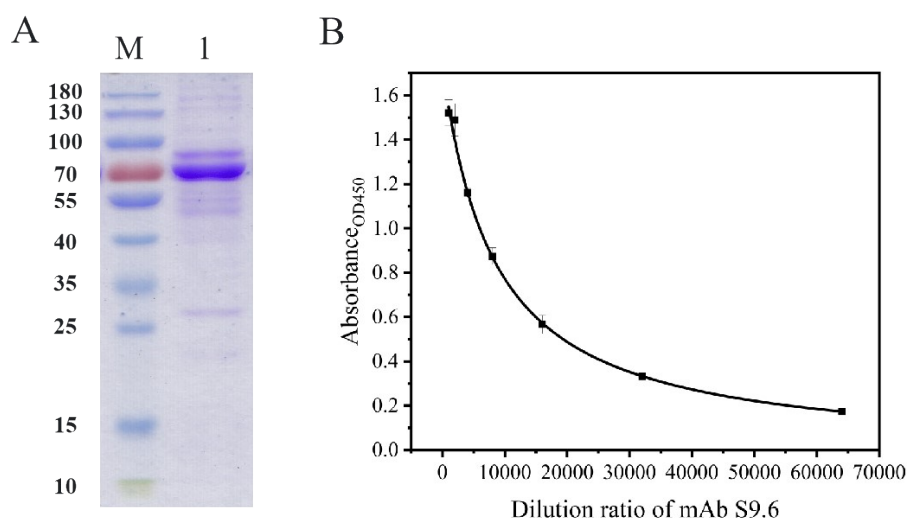


Fig. S1 Characterization of mAb S9.6. (A) SDS-PAGE char-acterization of mAb S9.6: M. Marker; lane 1. mAb S9.6; (B) ELISA characterization of mAb S9.6 Titer.

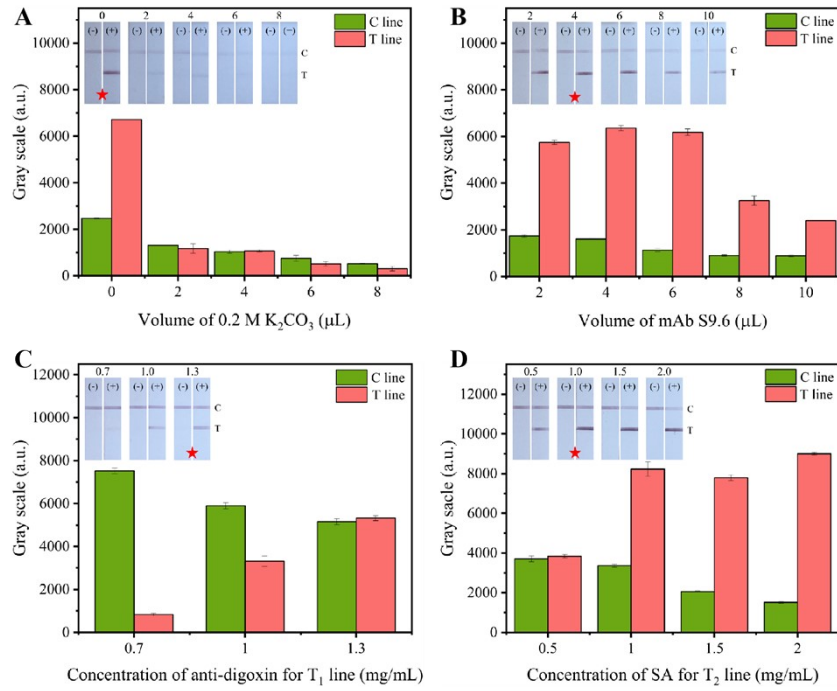


Fig. S2 Optimization of the AuNPs-LFIA. (A) Optimized volume of 0.2 M K_2CO_3 in the synthesis of AuNPs-mAb probe; (B) Optimized amount of mAb S9.6 in the synthesis of AuNPs-mAb probe; (C) Optimized concentration of T_1 line; (D) Optimized concentration of T_2 line.

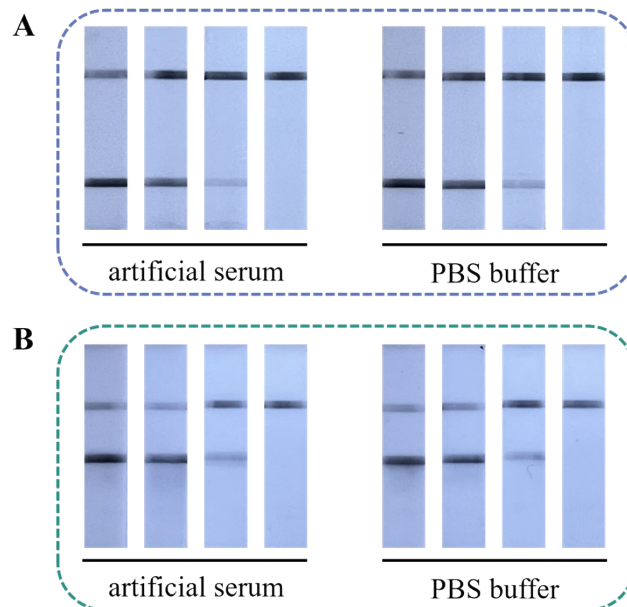


Fig. S3 Detection of HPV 16 DNA-RNA-digoxin (A) and HPV 18 DNA-RNA-biotin (B) in artificial serum and PBS buffer by Au@PdPt-LFIA.

Table. S2 Comparison with other studies of HPV detection

Method	Genotype	LOD	Assay duration	Reference
CuO-based LFSB ^a	16	1 nM	<20 min	1
ELISA ^b	16 E6/E7	0.92/0.42 pg/mL	>300 min	2
CRISPR/Cas12a based LFB ^c	16/18	0.9 copies/ μ L	~50 min	3
LAMP-LFD ^d	16/18	10/1 copies/reaction	45 min	4
tailed primer isothermal amplification and lateral flow detection assays	16/18/45	50/50/500 copies/reaction	<35 min	5
electrochemical DNA biosensor	16	0.23 copies/ μ L	75 min	6
MCLSA ^e	16	54 copies/tube	45 min	7
electrochemical resistive DNA biosensor	16	2.39 nM	>120 min	8
microchip electrophoresis	16/18	10 ² cells/mL	>130 min	9
WarmStart colorimetric LAMP ^f	16/18	100/10 copies/reaction	>80 min	10
PEC based on SiW12@CdS QDs ^g	16	0.8 nM	>120 min	11
DNA sandwich hybridization with AuNP probe	16/18	0.14 nM	>20 min	12
Paper-based colorimetric assay	16	1 nM	>30 min	13
AuNPs-LFIA ^h	16/18	0.23/0.20 nM	<15 min	This work
Au@PdPt-LFIA	16/18	0.05/0.02 nM	<15 min	This work

(a) LFSB: lateral flow strip biosensor; (b) ELISA: enzyme-linked immunosorbent assay; (c) LFB: lateral flow biosensor; (d) LFD: lateral flow dipstick; (e) MCLSA: multiple cross-linking spiral amplification; (f) LAMP: loop-mediated isothermal amplification; (g) QDs: quantum dots; (h) LFIA: lateral flow immunoassay.

References

- 1 Z. Yang, C. Yi, S. Lv, Y. Sheng, W. Wen, X. Zhang and S. Wang, *Sensors and Actuators B: Chemical*, 2019, **285**, 326-332.
- 2 S. Ding, S. Y. Qian, Y. Zhang, W. Wu, G. Lu, Y. Lu, X. Feng, L. Li and P. Shen, *Scientific reports*, 2015, **5**, 13686.
- 3 O. Mukama, T. Yuan, Z. He, Z. Li, J. d. D. Habimana, M. Hussain, W. Li, Z. Yi, Q. Liang and L. Zeng, *Sensors and Actuators B: Chemical*, 2020, **316**, 128119.
- 4 R. Kumvongpin, P. Jearanaikoon, C. Wilailuckana, N. Sae-Ung, P. Prasongdee, S. Daduang, M. Wongsena, P. Boonsiri, W. Kiatpathomchai, S. S. Swangvaree, A. Sandee and J. Daduang, *Mol Med Rep*, 2017, **15**, 3203-3209.
- 5 M. M. Chang, A. Ma, E. N. Novak, M. Barra, K. A. Kundrod, J. R. Montealegre, M. E. Scheurer, P. E. Castle, K. Schmeler and R. Richards-Kortum, *Scientific reports*, 2023, **13**, 20397.
- 6 P. Nakowong, P. Chatchawal, T. Chaibun, N. Boonapatcharoen, C. Promptmas, W. Buajeeb, S. Y. Lee, P. Jearanaikoon and B. Lertanantawong, *Talanta*, 2024, **269**, 125495.
- 7 D. Zhang, D. Liu, B. Liu and X. Ma, *Journal of microbiology and biotechnology*, 2021, **31**, 610-620.
- 8 J. R. Espinosa, M. Galván, A. S. Quiñones, J. L. Ayala, V. Ávila and S. M. Durón, *Molecules (Basel, Switzerland)*, 2021, **26**.
- 9 Z. Fan, X. Feng, W. Zhang, X. Zhang and J.-M. Lin, *Journal of Pharmaceutical Analysis*, 2020, **10**, 329-333.
- 10 M. Daskou, D. Tsakogiannis, T. G. Dimitriou, G. D. Amoutzias, D. Mossialos, C. Kottaridi, C. Gartzonika and P. Markoulatos, *Journal of virological methods*, 2019, **270**, 87-94.
- 11 Y. Cheng, C. Sun, Y. Chang, J. Wu, Z. Zhang, Y. Liu, S. Ge, Z. Li, X. Li, L. Sun and D. Zang, *Frontiers in bioengineering and biotechnology*, 2023, **11**, 1193052.
- 12 S. H. Chen, K. I. Lin, C. Y. Tang, S. L. Peng, Y. C. Chuang, Y. R. Lin, J. P. Wang and C. S. Lin, *IEEE transactions on nanobioscience*, 2009, **8**, 120-131.
- 13 S. Naorungroj, P. Teengam, T. Vilaivan and O. Chailapakul, *New Journal of Chemistry*, 2021, **45**, 6960-6967.