

**Quaternization carbon dots with enhanced antimicrobial ability to Gram-negative bacteria
for the treatment of acute peritonitis caused by *E. coli***

Xintian Zhang^{a,#}, Pingping Wu^{a,#}, Xiaoli Hao^a, Jiamiao Liu^a, Zhengjun Huang^a, Shaohuang Weng^{a,*},
Weifeng Chen^b, Lingling Huang^{c,*}, Jianyong Huang^{b,*}

^a Department of Pharmaceutical Analysis, School of Pharmacy, Fujian Medical University, Fuzhou
350122, China

^b Department of Pharmacy, Fujian Medical University Union Hospital, Fuzhou, 350001, China

^c Department of Stomatology, The First Affiliated Hospital of Fujian Medical University, Fuzhou,
350005, China

Zhang and Wu contributed equally to this work.

Correspondence: shweng@fjmu.edu.cn (S. Weng), hjy8191@163.com (J. Huang), and
huanglings@163.com (L. Huang)

1. Experimental section

Identifying bacteria using mass spectrometry

Preparation of matrix solution: Take 100 μL of lysis solution 2 and buffer solution respectively, add them to the matrix, shake and mix well to fully dissolve the matrix, and set aside. The prepared matrix solution is sealed and stored at 2°C-8°C and can be used for 30 days.

Experimental operation: add 300 μL of deionized water to a 1.5mL centrifuge tube, take a sample (choose 1~2 single colonies from a medium-sized colony) in a centrifuge tube, and shake and mix well. Add 900 μL of absolute ethanol, mix well, centrifuge at room temperature for 2 to 4 minutes (8000rpm to 14800rpm), discard the supernatant, and centrifuge again to completely remove the supernatant. Dry the precipitate at 37°C to 40°C for 2 minutes to 5 minutes, until there is no obvious water mark on the surface of the precipitate. Add 10 μL of lysis solution 1, mix well, then add 10 μL equal volume of lysis solution 2, mix well, and centrifuge at room temperature for 2 to 4 minutes (8000rpm-14800rpm). Aspirate 1 μL of the supernatant, drop it on the sample target, and air dry until there is no obvious water trace on the sample point. Take 1 μL of matrix solution to cover the above-mentioned sample point, and air-dry until there is no

obvious water mark on the sample point. The sample target is placed into the mass spectrometer for identification.

Table S1. Pretreatment reagents for mass spectrometry samples

number	Solution name	100 tests/box
1	Lysate 1 (formic acid-containing solution)	1.0ml
2	Lysate 2 (acetonitrile-containing solution)	1.1ml
3	Buffer (solution containing trifluoroacetic acid)	0.3ml
4	Matrix (with alpha-cyano-4-hydroxycinnamic acid)	2.0mg

2. Figures and Figure captions

Table S2. MICs of QCDS against five samples of clinical drug-resistant bacteria.

Bacterial Strains	G ⁺ / G ⁻	MIC(μ g/mL)
Resistant <i>E. coli</i> -1	G ⁻	30
Resistant <i>E. coli</i> -2	G ⁻	30
Resistant <i>E. coli</i> -3	G ⁻	30
Resistant <i>E. coli</i> -4	G ⁻	30
Resistant <i>E. coli</i> -5	G ⁻	30

Table S3. The MIC of recent several carbon dots to *E. coli*.

Kinds of Materials	MIC value	Ref.
PC-CQDs	120 μ g/mL	1
QCQD	640 μ g/mL	2
qCQDs	50 μ g/mL	3
FA-CD	>2 mg/mL	4
P-doped CQDs	1.23 mg/mL	5
GCD	50 μ g/mL	6
CNDs	192 μ g/mL	7
F-CDs	64 μ g/mL	8
QCDS	30 μ g/mL	This study

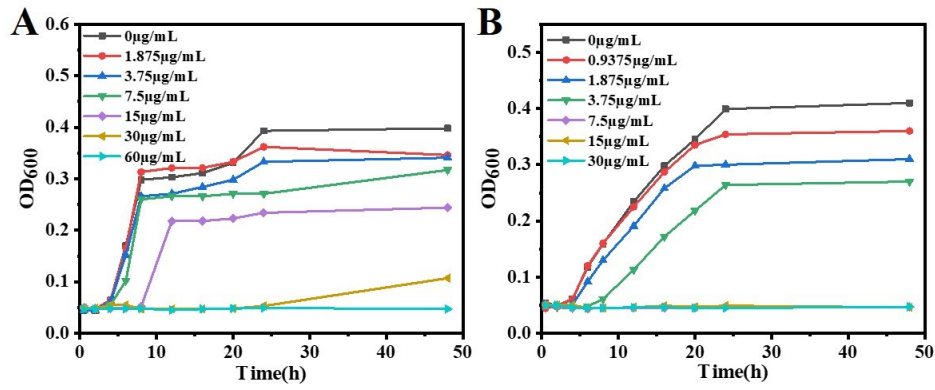


Figure S1. Bacterial inhibition curves by QCDs. (A) *E. coli*. (B) *S. aureus*.

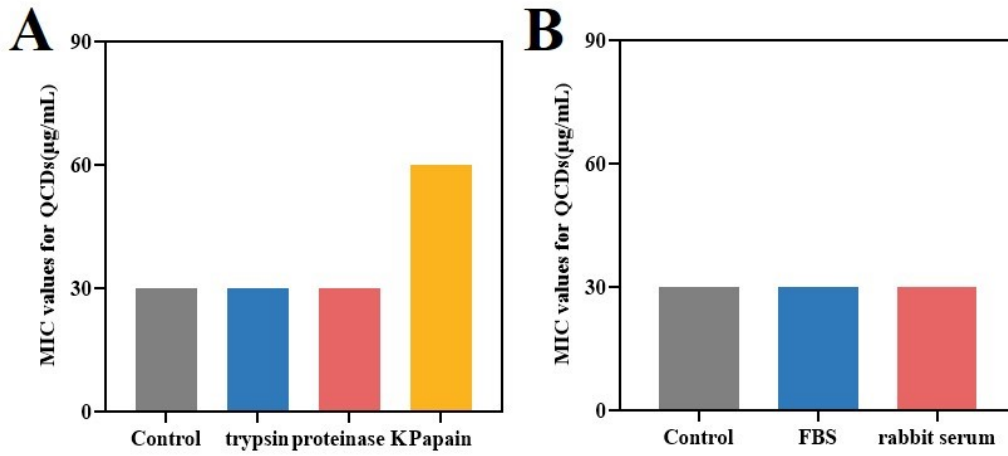


Figure S2. Stability of antimicrobial activity of QCDs in different conditions against *E. coli*. (A) Various protein. (B) Various serum.

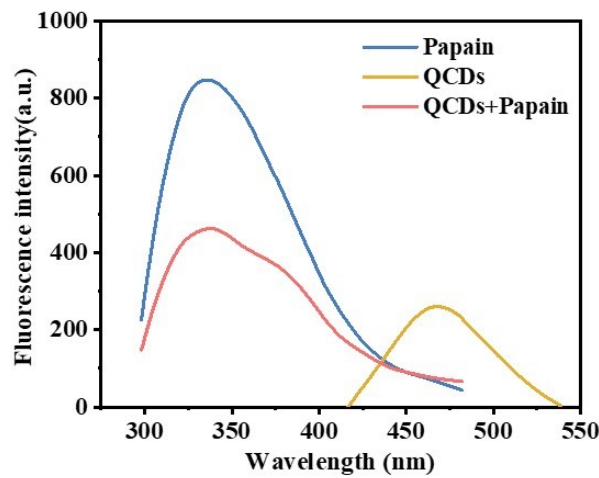


Figure S3. Fluorescence spectra of papain, QCDs and the interaction of QCDs and papain.

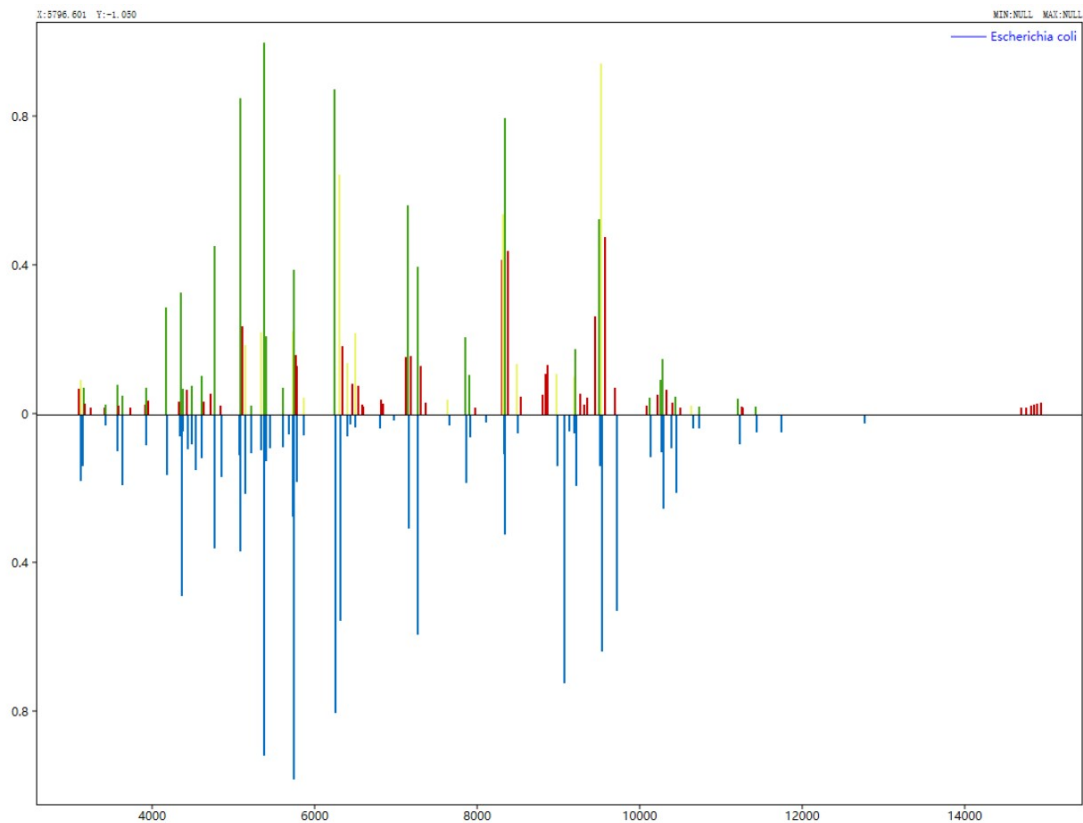


Figure S4. The MS identification result of the colonies on the petri dish of the rat in the negative control group.

References

- [1] X. L. Hao, L. L. Huang, C. F. Zhao, S. N. Chen, W. J. Lin, Y. N. Lin, L. R. Zhang, A. A. Sun, C. F. Miao, X. H. Lin, M. Chen and S. H. Weng, *Mater. Sci. Eng. C*, 2021, **123**, 111971.
- [2] C. F. Zhao, L. N. Wu, X. W. Wang, S. H. Weng, Z. P. Ruan, Q. C. Liu, L. Q. Lin and X. H. Lin, *Carbon*, 2020, **163**, 70-84.
- [3] C. F. Zhao, X. W. Wang, L. Y. Yu, L. N. Wu, X. L. Hao, Q. C. Liu, L. Q. Lin, Z. J. Huang, Z. P. Ruan, S. H. Weng, A. L. Liu and X. H. Lin, *Acta Biomater*, 2022, **138**, 528-544.
- [4] M. Yu, X. Guo, H. Lu, P. Li, R. Huang, C. Xu, X. Gong, Y. Xiao and X. Xing, *Carbon*, 2022, **199**, 395-406.

- [5] S. Chai, L. Zhou, S. Pei, Z. Zhu and B. Chen, *Micromachines (Basel)*, 2021, **12**, 1116.
- [6] Z.Y. Wang, L.A. Sheng, X.X. Yang, J.D. Sun, Y.L. Ye, S.X. Geng, D.L. Ning, J.Y. Zheng, M.H. Fan, Y.Z. Zhang and X.L. Sun, *Sustainable Materials and Technologies*, 2023, **36**, e00584.
- [7] F. Du, S. Shuang, Z. Guo, X. Gong, C. Dong, M. Xian and Z. Yang, *Talanta*, 2020, **206**, 120243.
- [8] J. Liang, W. Li, J. Chen, X. Huang, Y. Liu, X. Zhang, W. Shu, B. Lei and H. Zhang, *ACS Appl Bio Mater*, 2021, **4**, 6937-6945.