

Supporting information

Supplementary experimental section, 2 Tables and 15 Figures are included

Treated Rape Pollen: A Metal-free Visible-Light-Driven Photocatalyst from Nature for Efficient Water Disinfection

Bo Wang,^a Zhifeng Jiang,^{*,b,c} and Jimmy C. Yu^{*,a}

^a Department of Chemistry, The Chinese University of Hong Kong, Shatin, New Territories, Hong Kong 999077, P. R. China.

^b Institute for Energy Research, Jiangsu University, Zhenjiang 212013, P. R. China.

^c School of Energy and Environment, City University of Hong Kong, Kowloon, Hong Kong 999077, P. R. China.

*Corresponding authors:

*J. C. Y; E-mail: jimyu@cuhk.edu.hk

*Z. J; E-mail: ntjiangzf@sina.com

Supplementary experimental section

Preparation of the other common waterborne bacteria

To prepare the other common waterborne bacteria for disinfection study, the other three bacterial strains including *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus pumilus* were inoculated in 50 mL LB broth (Lab M, Lancashire, UK) respectively. Then the bacteria were incubated at 37 °C in a shaking incubator for. After 16 h, 1 mL bacterial culture medium was collected and centrifuged by 13000 rpm for 5 min. The cell pellets were resuspended by 1 mL ultrapure water. To adjust all the bacterial concentration to 1×10^7 cell/mL, 100 uL of *P. aeruginosa*, 800 uL of *S. aureus* and 1000 uL of *B. pumilus* were added into 50 mL saline then mixed well, respectively.

Table S1. Comparison of the photocatalytic water disinfection performance between the TRP in this study and other recently reported photocatalysts.

Photocatalysts	Light source	Intensity (W/m ²)	Bacterial species	Disinfection performance	Ref.
TRP in this study (0.5 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1000	<i>E. coli</i> K-12 (1×10^7 CFU/mL)	~ 7 log in 4h	
HTCC (glucose) (0.2 g/L)	300W Xe lamp ($\lambda > 420$ nm)	2000	<i>E. coli</i> K-12 (1×10^7 CFU/mL)	no activity	[1]
I-HTCC (straw) (0.2 g/L)	300W Xe lamp ($\lambda > 420$ nm)	2000	<i>E. coli</i> K-12 (1×10^7 CFU/mL)	~ 2 log in 3h	[1]
I-HTCC (grass) (0.2 g/L)	300W Xe lamp ($\lambda > 420$ nm)	2000	<i>E. coli</i> K-12 (1×10^7 CFU/mL)	~ 4 log in 3h	[1]
I-HTCC (starch) (0.2 g/L)	300W Xe lamp ($\lambda > 420$ nm)	2000	<i>E. coli</i> K-12 (1×10^7 CFU/mL)	~ 6 log in 3h	[1]
g-C ₃ N ₄ (CN-12) (1.0 g/L)	300W Xe lamp ($\lambda > 400$ nm)	NA	<i>E. coli</i> K-12 (2.5×10^6 CFU/mL)	~ 1 log in 6h	[2]
g-C ₃ N ₄ (CN-128) (1.0 g/L)	300W Xe lamp ($\lambda > 400$ nm)	NA	<i>E. coli</i> K-12 (2.5×10^6 CFU/mL)	~ 6 log in 6h	[2]
g-C ₃ N ₄ bulk (0.1 g/L)	500W Xe lamp ($\lambda > 400$ nm)	1000	<i>E. coli</i> K-12 (2×10^7 CFU/mL)	~ 3 log in 4h	[3]
g-C ₃ N ₄ nanosheet (0.1 g/L)	500W Xe lamp ($\lambda > 400$ nm)	1000	<i>E. coli</i> K-12 (2×10^7 CFU/mL)	~ 5 log in 4h	[3]
g-C ₃ N ₄ single layer (0.1 g/L)	500W Xe lamp ($\lambda > 400$ nm)	1000	<i>E. coli</i> K-12 (2×10^7 CFU/mL)	~ 7 log in 4h	[3]
Red phosphorus* (0.1 g/L)	300W Xe lamp ($\lambda > 420$ nm)	1930	<i>E. coli</i> K-12 (2×10^7 CFU/mL)	~ 7 log in 2h	[4]
Elemental α -S ₈ (0.1 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1930	<i>E. coli</i> K-12 (2×10^6 CFU/mL)	~ 2 log in 4h	[5]
RGO/g-C ₃ N ₄ / α -S ₈ (0.1 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1930	<i>E. coli</i> K-12 (2×10^6 CFU/mL)	~ 7 log in 4h	[5]
g-C ₃ N ₄ /RGO/ α -S ₈ (0.1 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1930	<i>E. coli</i> K-12 (2×10^6 CFU/mL)	~ 4 log in 4h	[5]
g-C ₃ N ₄ wrapped α -S ₈ (0.1 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1930	<i>E. coli</i> K-12 (2×10^6 CFU/mL)	~ 3 log in 4h	[5]
RGO wrapped α -S ₈ (0.1 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1930	<i>E. coli</i> K-12 (2×10^6 CFU/mL)	~ 3 log in 4h	[5]
RGO/g-C ₃ N ₄ nanosheets (0.1 g/L)	300W Xe lamp ($\lambda > 400$ nm)	1930	<i>E. coli</i> K-12 (2×10^6 CFU/mL)	~ 3 log in 4h	[5]
TiO ₂ /g-C ₃ N ₄ (1.0 g/L)	300W Xe lamp ($\lambda > 420$ nm)	300	<i>E. coli</i> (1×10^7 CFU/mL)	~ 7 log in 3h	[6]
Bi ₂ MoO ₆ /g-C ₃ N ₄ ** nanosheets (0.1 g/L)	300W Xe lamp ($\lambda > 420$ nm)	NA	<i>E. coli</i> DH5 α (2.5×10^7 CFU/mL)	~ 6 log in 4h	[7]
Ag(0.5)/g-C ₃ N ₄ (0.1 g/L)	300W Xe lamp ($\lambda > 420$ nm)	NA	<i>E. coli</i> (1×10^7 CFU/mL)	~ 1 log in 1.5h	[8]
Ag(1)/g-C ₃ N ₄ (0.1 g/L)	300W Xe lamp ($\lambda > 420$ nm)	NA	<i>E. coli</i> (1×10^7 CFU/mL)	~ 3 log in 1.5h	[8]
Ag(2)/g-C ₃ N ₄ (0.1 g/L)	300W Xe lamp ($\lambda > 420$ nm)	NA	<i>E. coli</i> (1×10^7 CFU/mL)	~ 4 log in 1.5h	[8]

Bi ₂ O ₄ /g-C ₃ N ₄ (0.25:1) (0.4 g/L)	300W Xe lamp (λ > 400 nm)	1930	<i>E. coli</i> K-12 (1×10 ⁶ CFU/mL)	~ 5 log in 2h	[9]
TiO ₂ /graphene (1.0 g/L)	450W Xe lamp (visible light)	NA	<i>E. coli</i> PTCC 1330 (NA)	~ 1 log in 7h	[10]
ZnO/graphene (3.3 g/L)	UV light	NA	<i>E. coli</i> KCCM12119 (1×10 ⁵ CFU/mL)	~ 1 log in 3h	[11]
TiO ₂ /Ag ₃ PO ₄ /graphene (0.5 g/L)	350W Xe lamp (λ > 420 nm)	NA	<i>E. coli</i> (1×10 ⁶ CFU/mL)	~ 5 log in 4h	[12]
TRP in this study (0.5 g/L)	300W Xe lamp (λ > 400 nm)	1000	<i>S. aureus</i> (1×10 ⁷ CFU/mL)	~ 7 log in 2h	
g-C ₃ N ₄ /TiO ₂ /kaolinite	8W fluorescent lamp	NA	<i>S. aureus</i> (1×10 ⁷ CFU/mL)	~ 3 log in 5h	[13]
TiO ₂ /Ag ₃ PO ₄ /graphene (0.5 g/L)	350W Xe lamp (λ > 420 nm)	NA	<i>S. aureus</i> (1×10 ⁶ CFU/mL)	~ 5 log in 4h	[12]
Polythiophene/MnO ₂ (1 g/L)	Solar irradiation	NA	<i>S. aureus</i> (5×10 ³ CFU/mL)	~ 1 log in 5h	[14]
TRP in this study (0.5 g/L)	300W Xe lamp (λ > 400 nm)	1000	<i>B. pumilus</i> (1×10 ⁷ CFU/mL)	~ 3 log in 4h	
TiO ₂ /Ag ₃ PO ₄ /graphene (0.5 g/L)	350W Xe lamp (λ > 420 nm)	NA	<i>B. pumilus</i> (1×10 ⁶ CFU/mL)	~ 5 log in 4h	[12]
TRP in this study (0.5 g/L)	300W Xe lamp (λ > 400 nm)	1000	<i>P. aeruginosa</i> (1×10 ⁷ CFU/mL)	~ 7 log in 1h	
TiO ₂ /Ag ₃ PO ₄ /graphene (0.5 g/L)	350W Xe lamp (λ > 420 nm)	NA	<i>P. aeruginosa</i> (1×10 ⁶ CFU/mL)	~ 5 log in 4h	[12]
Ag-TiO ₂ film	UV lamp	NA	<i>P. aeruginosa</i> (1×10 ³ CFU/mL)	~ 3 log in 1h	[15]

NA: Not available

* Phosphate buffer solution (PBS, pH 7.0) is required in the experiment.

** Sacrificial agents are required

Table S2. Variation of the light intensity in the real solar experiment.

Time	Solar intensity (W m ⁻²)	UVA (μW cm ⁻²)	UVB (μW cm ⁻²)	UVC (μW cm ⁻²)
Day 1	11:00	620	675	685
	11:30	690	715	730
	12:00	750	780	795
	12:30	870	890	905
	13:00	900	930	950
	13:30	870	890	905
	14:00	850	870	895
	14:30	790	805	815
	15:00	700	720	730
Day 2	11:00	870	890	905
	11:30	960	985	995
	12:00	1020	1035	1050
	12:30	1070	1080	1100
	13:00	1100	1115	1125
	13:30	1020	1035	1050
	14:00	940	950	970
	14:30	890	905	920
	15:00	850	875	890
Day 3	11:00	770	795	805
	11:30	840	850	865
	12:00	940	950	970
	12:30	970	985	1000
	13:00	990	1105	1115
	13:30	910	920	935
	14:00	870	890	905
	14:30	830	845	855
	15:00	800	820	830

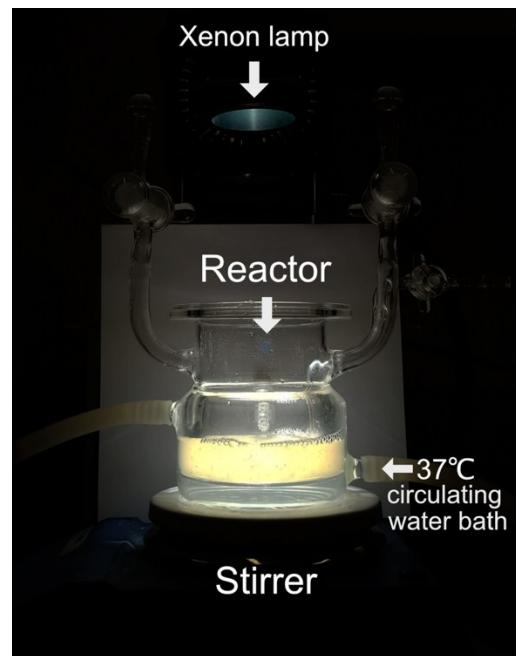


Figure S1. Setup of the photoreactor with temperature controller for photocatalytic disinfection study.



Figure S2. Setup of the practical application study under the simulated solar irradiation. No temperature controller is provided for the reaction.

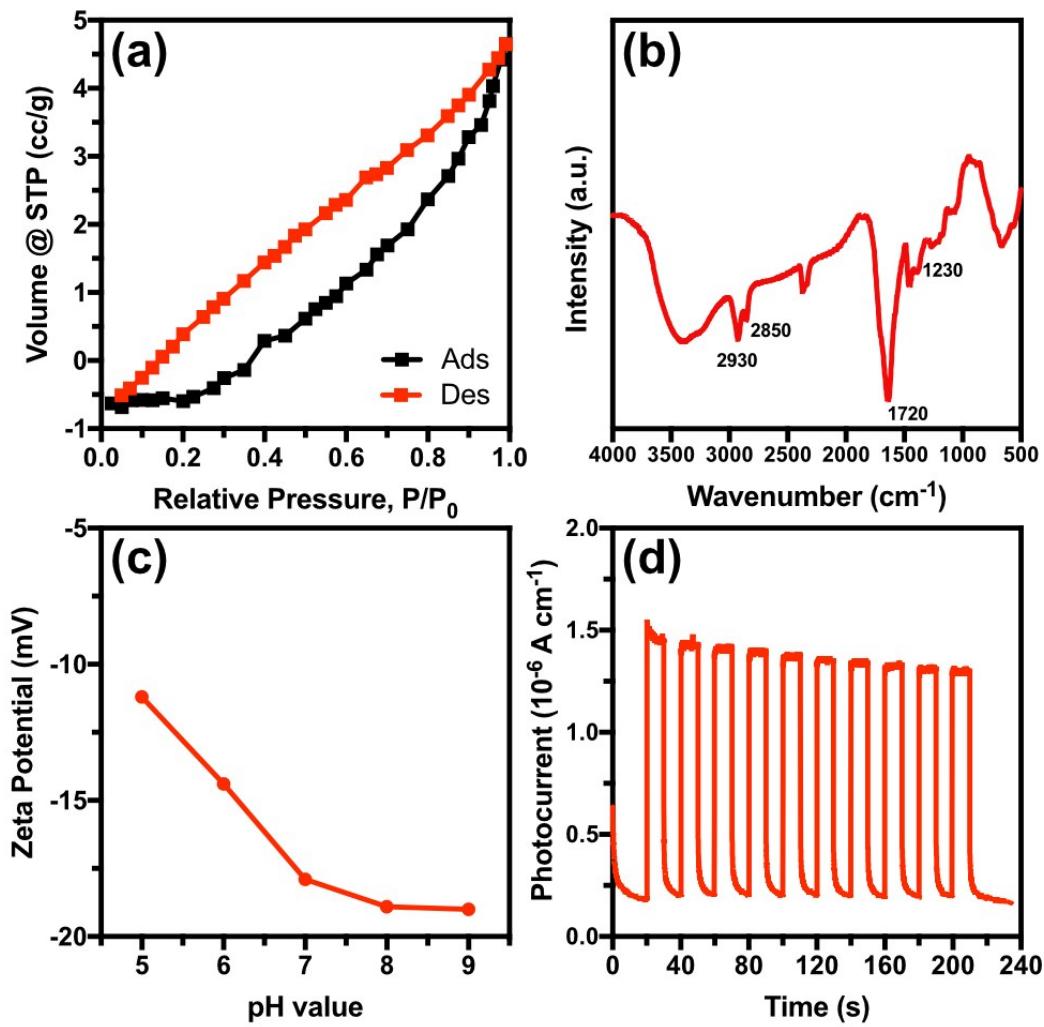


Figure S3. (a) BET specific surface area, (b) FTIR spectra, (c) Zeta-potential and (d) PEC characteristic of the TRP.

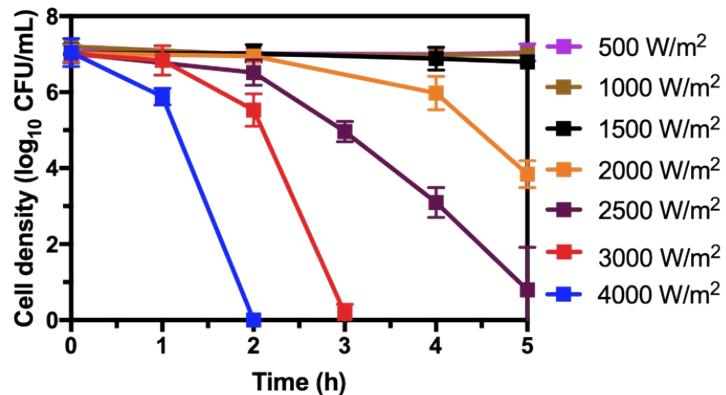


Figure S4. Viability of *E. coli* K-12 under the illumination of visible light with different intensities. The reaction temperature is controlled at 37 °C.

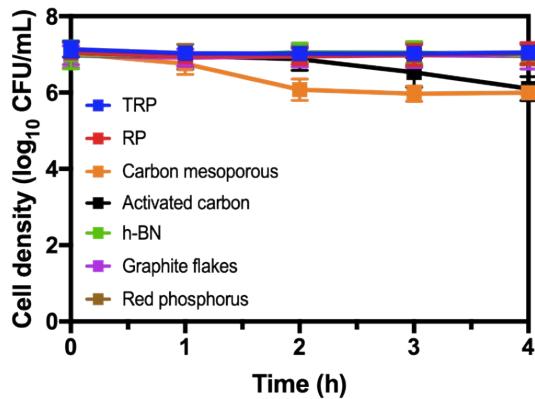


Figure S5. Viability of *E. coli* K-12 mixed with the TRP, RP, h-BN, graphite flakes, activated carbon, carbon mesoporous and red phosphorus in dark. The concentration of all the materials are 0.5 mg mL^{-1} . The reaction temperature is controlled at 37°C .

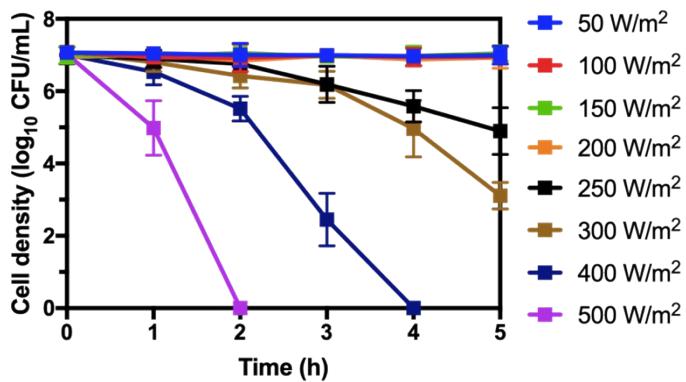


Figure S6. Viability of *E. coli* K-12 under the illumination of full spectrum (xenon lamp without filter) with different intensities. The reaction temperature is controlled at 37°C .

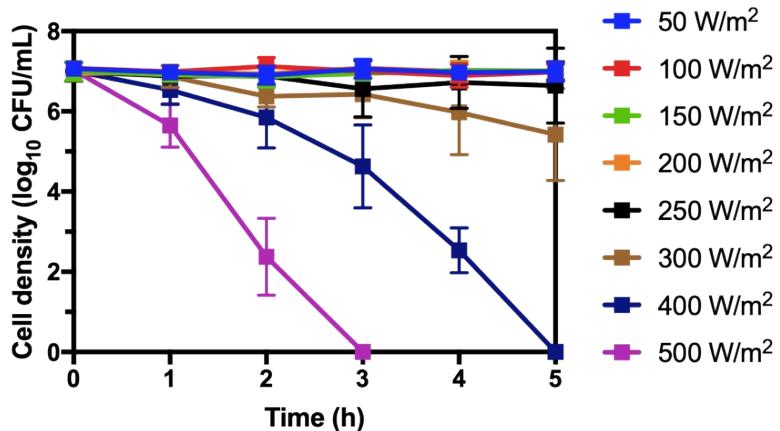


Figure S7. Viability of *E. coli* K-12 under the illumination of solar spectrum (xenon lamp with AM 1.5 filter) with different intensities. The reaction temperature is controlled at 37°C .

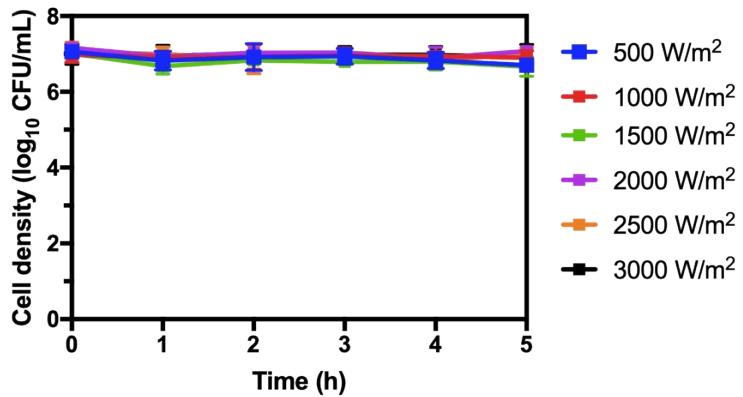


Figure S8. Viability of *E. coli* K-12 under the illumination of NIR spectrum (xenon lamp with NIR filter) with different intensities. The reaction temperature is controlled at 37 °C.

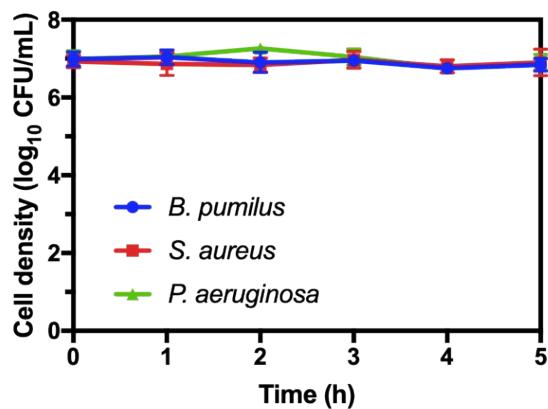


Figure S9. Viability of *B. pumilus*, *S. aureus* and *P. aeruginosa* mixed with the TRP in dark.

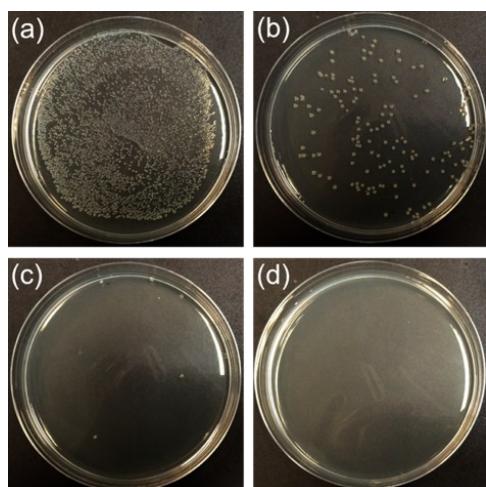


Figure S10. Spreading plate study of the *E. coli* K-12 collected at (a) 0 min, (b) 60 min (c) 120 min and (d) 240 min during the photocatalytic disinfection by TRP.

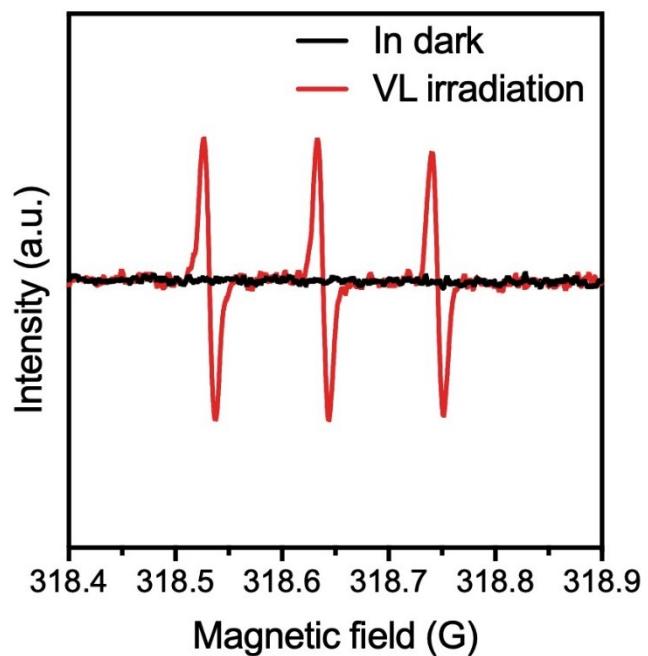


Figure S11. ESR analysis for ${}^1\text{O}_2$ generation by TRP under VL irradiation and in dark.

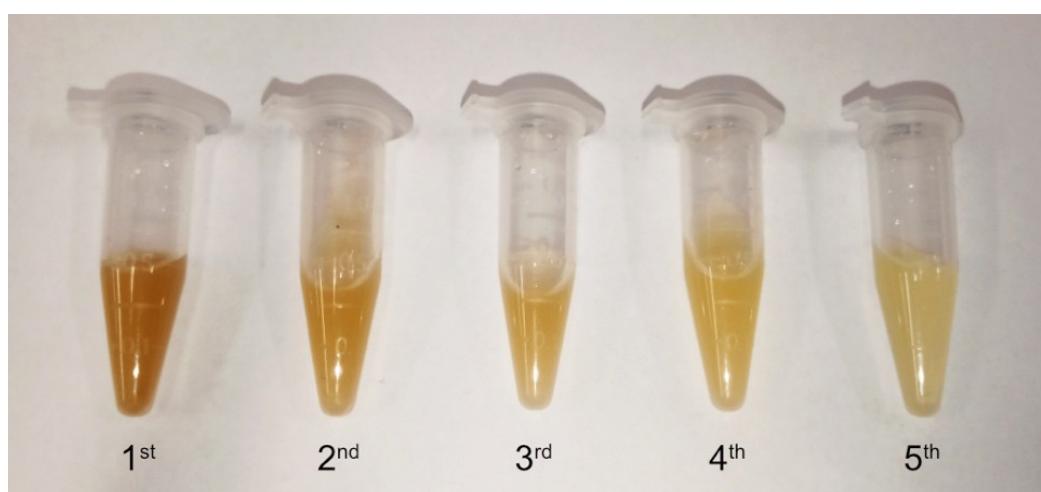


Figure S12. Color variation of the TRP disinfection system from 1st to 5th run under the irradiation of VL (1000 W m^{-2} provided by xenon lamp with UV cut-off filter).

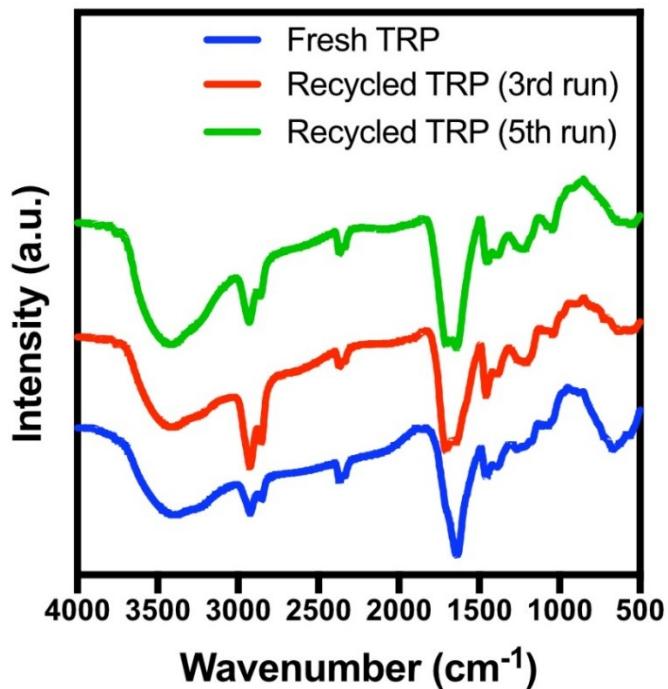


Figure S13. FTIR spectrum of the fresh TRP, recycled TRP from 3rd run and 5th run.

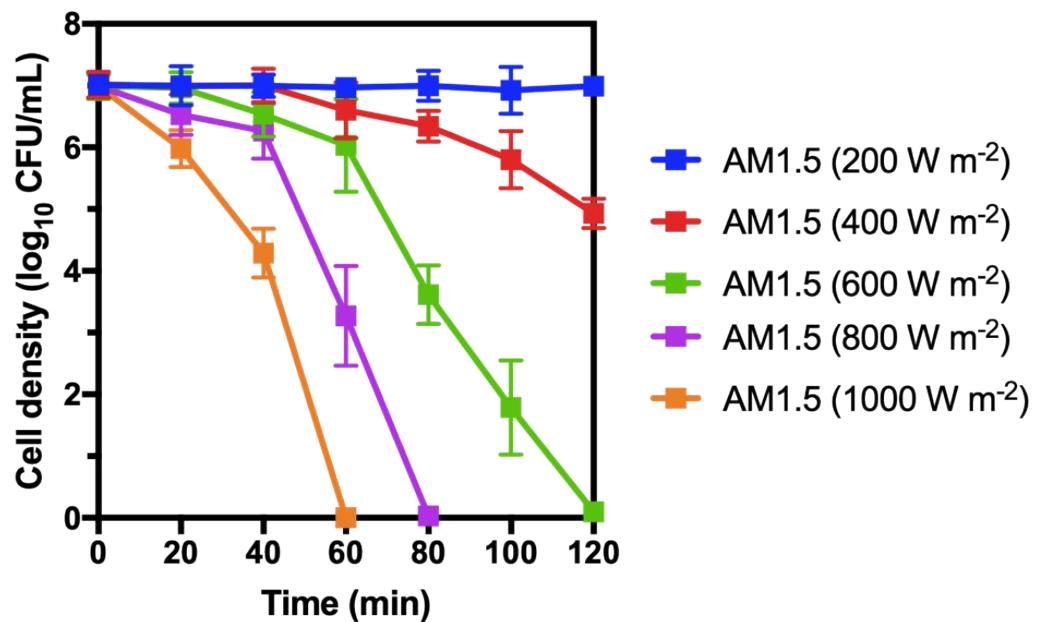


Figure S14. Control experiment of simulated solar disinfection (AM1.5 filter) of *E. coli* K-12 without temperature control.

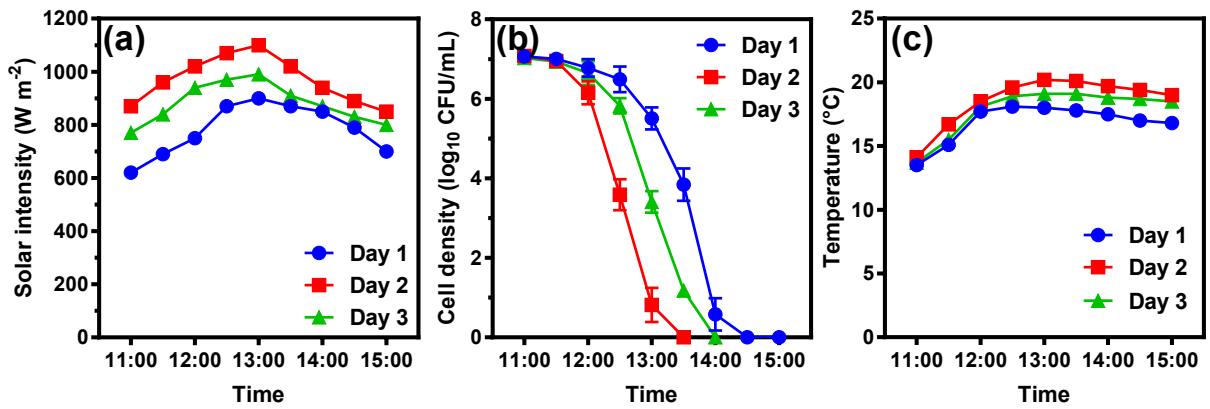


Figure S15. Photocatalytic disinfection of *E. coli* K-12 ($1 \times 10^7 \text{ CFU mL}^{-1}$) by the TRP (0.5 g/L) under real sunlight irradiation. (a) variation of solar intensity, (b) variation of living cell density, and (c) variation of the temperature.

Reference

- [1] Z. Hu, Z. Shen and J. C. Yu, Converting carbohydrates to carbon-based photocatalysts for environmental treatment, *Environ. Science Tech.*, 2017, **51**, 7076-7083.
- [2] J. Huang, W. Ho and X. Wang, Metal-free disinfection effects induced by graphitic carbon nitride polymers under visible light illumination, *Chem. Commun.*, 2014, **50**, 4338-4340.
- [3] H. Zhao, H. Yu, X. Quan, S. Chen, Y. Zhang, H. Zhao and H. Wang, Fabrication of atomic single layer graphitic-C₃N₄ and its high performance of photocatalytic disinfection under visible light irradiation, *Appl. Catal. B-Environ.*, 2014, **152**, 46-50.
- [4] D. Xia, Z. Shen, G. Huang, W. Wang, J. C. Yu and P. K. Wong, Red phosphorus: an earth-abundant elemental photocatalyst for “green” bacterial inactivation under visible light, *Environ. Sci. Technol.*, 2015, **49**, 6264-6273.
- [5] W. Wang, J. C. Yu, D. Xia, P. K. Wong and Y. Li, Graphene and g-C₃N₄ nanosheets cowrapped elemental α -sulfur as a novel metal-free heterojunction photocatalyst for bacterial inactivation under visible-light, *Environ. Sci. Technol.*, 2013, **47**, 8724-8732.
- [6] G. Li, X. Nie, J. Chen, Q. Jiang, T. An, P. K. Wong, H. Zhang, H. Zhao and H. Yamashita, Enhanced visible-light-driven photocatalytic inactivation of *Escherichia coli* using g-C₃N₄/TiO₂ hybrid photocatalyst synthesized using a hydrothermal-calcination approach, *Water Res.*, 2015, **86**, 17-24.
- [7] J. Li, Y. Yin, E. Liu, Y. Ma, J. Wan, J. Fan and X. Hu, In situ growing Bi₂MoO₆ on g-C₃N₄ nanosheets with enhanced photocatalytic hydrogen evolution and disinfection of bacteria under visible light irradiation, *J. Hazard. Mater.*, 2017, **321**, 183-192.
- [8] S. Ma, S. Zhan, Y. Jia, Q. Shi and Q. Zhou, Enhanced disinfection application of Ag-modified g-C₃N₄ composite under visible light, *Appl. Catal. B-Environ.*, 2016, **186**, 77-87.
- [9] D. Xia, W. Wang, R. Yin, Z. Jiang, T. An, G. Li, H. Zhao and P. K. Wong, Enhanced photocatalytic inactivation of *Escherichia coli* by a novel Z-scheme g-C₃N₄/m-Bi₂O₄ hybrid photocatalyst under visible light: The role of reactive oxygen species, *Appl. Catal. B-Environ.*, 2017, **214**, 23-33.
- [10] R. Rahimi, S. Zargari, A. Yousefi, M. Y. Berijani, A. Ghaffarinejad and A. Morsali, Visible light photocatalytic disinfection of *E. coli* with TiO₂-graphene nanocomposite sensitized with tetrakis (4-carboxyphenyl) porphyrin, *Appl. Surface Sci.*, 2015, **355**, 1098-1106.
- [11] T. Kavitha, A. I. Gopalan, K. P. Lee, and S. Y. Park, Glucose sensing, photocatalytic and antibacterial properties of graphene-ZnO nanoparticle hybrids, *Carbon*, 2012, **50**, 2994-3000.
- [12] X. Yang, J. Qin, Y. Jiang, R. Li, Y. Li, and H. Tang, Bifunctional TiO₂/Ag₃PO₄/graphene composites with superior visible light photocatalytic performance and synergistic inactivation of bacteria, *Rsc Advances*, 2014, **4**, 18627-18636.
- [13] C. Li, Z. Sun, W. Zhang, C. Yu and S. Zheng, Highly efficient g-C₃N₄/TiO₂/kaolinite composite with novel three-dimensional structure and enhanced visible light responding ability towards ciprofloxacin and *S. aureus*, *Appl. Catal. B-Environ.*, 2018, **220**, 272-282.
- [14] K. Shang, S. Ai, Q. Ma, T. Tang, H. Yin and H. Han, Effective photocatalytic disinfection of *E. coli* and *S. aureus* using polythiophene/MnO₂ nanocomposite photocatalyst under solar light irradiation,

Desalination, 2011, **278**, 173-178.

- [15] K. Ubonchonlakate, L. Sikong and F. Saito, Photocatalytic disinfection of *P. aeruginosa* bacterial Ag-doped TiO₂ film, *Procedia Eng.*, 2012, **32**, 656-662.