

Plasmonic Nanometal Surface Energy Transfer-based Dual Excitation Biosensing of Pathogens

Milad Torabfam,^a Hasan Kurt,^{b,c,d} Mustafa Kemal Bayazit,^e Meral Yüce^{e*}

^{a.} Sabanci University, Faculty of Engineering and Natural Sciences, 34956, Istanbul, Turkey.

^{b.} Istanbul Medipol University, School of Engineering and Natural Sciences, 34810, Beykoz, Istanbul, Turkey

^{c.} Istanbul Medipol University, Research Institute for Health Sciences and Technologies (SABITA), 34810, Beykoz, Istanbul, Turkey

^{d.} Nanosolar Plasmonics Ltd., 41400, Gebze, Kocaeli, Turkey

^{e.} Sabanci University, SUNUM Nanotechnology Research and Application Centre, 34956, Istanbul, Turkey.

***Corresponding authors:** meralyuce@sabanciuniv.edu

Supporting Information

Table S 1. NSET-based sensors for target detection in the literature.

Target Pathogen(s)	Donor	Acceptor	LOD	Ref.
DNA	SiO ₂ coated CdTe (CdTe/SiO ₂) core/shell nanoparticles	AuNPs	0.106 nmol/L	1
hepatitis C virus (HCV) RNA	Cy3 dye	AuNPs	300 fM	2
Cu ²⁺ and Hg ²⁺	Ring-open structured rhodamine spirolactam	AuNPs	----	3
Mercury(II)	DNA-conjugated QDs	AuNPs	1.2 ppb	4
Hg(II)	Rhodamine B (RhB) dye	AuNPs	2 ppt	5
Glutathione	5-aminofluorescein (FI-NH ₂)	AuNPs	----	6
C-Reactive protein	Fluorescein	AuNPs	----	7
Prostate specific antigen (PSA)	QD labeled PSA aptamer	Graphene oxide	0.05 fg mL ⁻¹	8
Hg ²⁺	S,N co-doped carbon dots (S,N-CDs)	AgNPs	0.51 nM	9
Heparin	Near-infrared fluorophore	AuNRs	6.7 ng/mL	10

Table S 2. Aptamers and complementary DNA sequences used in this study.

Target	Aptamers (5'-NH ₂ -C6)	cDNAs (5'-NH ₂ -C6)	Ref.
<i>Salmonella typhimurium</i> (ATCC® 14028™)	TATGGCGGCGTCACCCGACGGGGACTTGACATTATGACAG	ATAATGTCAAGTCCCCGTCGGG	11, 12
<i>Escherichia coli</i> O157:H7 (ATCC® 25922™)	CCGGACGCTTATGCCCTTGCCATCTACAGAGCAGGTGTGACGG	CCTGCTCTGTAGATGGCAAGGC	11, 12

Table S 3. The hydrodynamic size of the NPs utilized in this assay.

Samples	Hydrodynamic size (nm)
CdSe/ZnS core/shell QDs	25.93
EC-Aptamer-QDs	61.76
NaYF ₄ :Yb/Er UCNPs	41.8
ST-Aptamer-UCNPs	87.7
AuNR	55.7
EC-cDNA-AuNR	66.2
AuNU	91.3
ST-cDNA-AuNU	105.8

Table S 4. Zeta potential of the unmodified and aptamer capped NPs.

Samples	Zeta Potential (mV)
CdSe/ZnS core/shell QDs	-32.5
EC-Aptamer-QDs	-12.9
NaYF ₄ :Yb/Er UCNPs	-30.7
ST-Aptamer-UCNPs	-14.3
AuNR	-20.8
EC-cDNA-AuNR	-8.27
AuNU	-22.2
ST-cDNA-AuNU	-9.73

Table S 5. LOD values and calibration curve details for detection of *S. typhimurium* and *E. coli* using UCNP and QD-based aptasensor.

Bacteria	Linear Equation	R ²	Linear Range (CFU mL ⁻¹)	LOD (CFU mL ⁻¹)
<i>S. typhimurium</i>	y=76.15x+113.81	0.9913	10 ² -10 ⁶	7.55

Supporting Information

<i>E. coli</i>	$y=97.301x+128.96$	0.9743	10^2-10^6	4.94
----------------	--------------------	--------	-------------	------

Table S 6. LOD values and calibration curve parameters for simultaneous detection of *S. typhimurium* and *E. coli* using dual excitation luminescence nanoprobes.

Bacteria	Linear Equation	R ²	Linear Range (CFU mL ⁻¹)	LOD (CFU mL ⁻¹)
<i>S. typhimurium</i>	$y=40.54x+46.67$	0.994	10^2-10^5	9.313
<i>E. coli</i>	$y=64.36x+60.93$	0.972	10^3-10^5	7.38

Table S 7. Multiplexed sensing of *S. typhimurium* and *E. coli* in spiked lake water samples.

Sample	Spiked Concentration (CFU mL ⁻¹)		Measured Concentration (CFU mL ⁻¹)	
	<i>E. coli</i>	<i>S. typhimurium</i>	<i>E. coli</i>	<i>S. typhimurium</i>
Lake water 1	1.0×10^2	1.0×10^2	$(1.1462 \pm 0.13) \times 10^2$	$(0.9112 \pm 0.07) \times 10^2$
Lake water 2	1.0×10^3	1.0×10^3	$(1.0762 \pm 0.10) \times 10^3$	$(0.9860 \pm 0.12) \times 10^3$
Lake water 3	1.0×10^4	1.0×10^4	$(1.0288 \pm 0.11) \times 10^4$	$(1.113 \pm 0.09) \times 10^4$

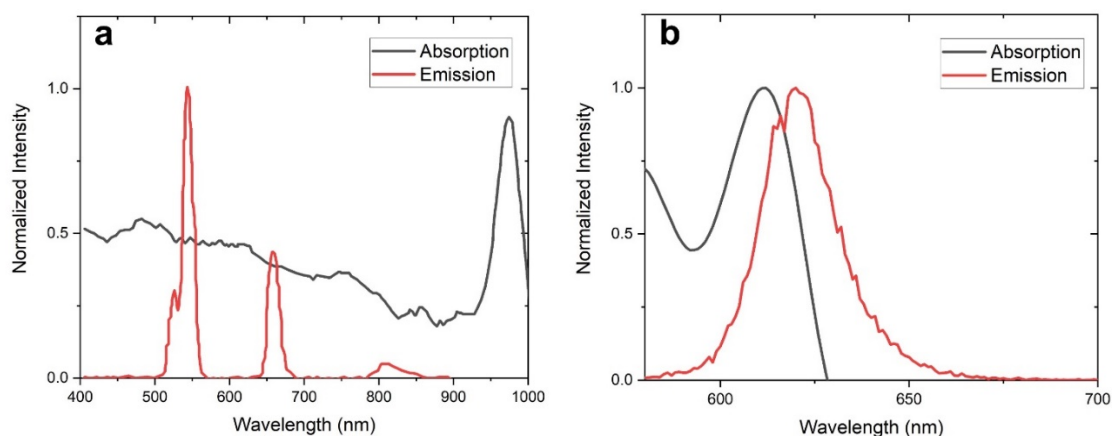
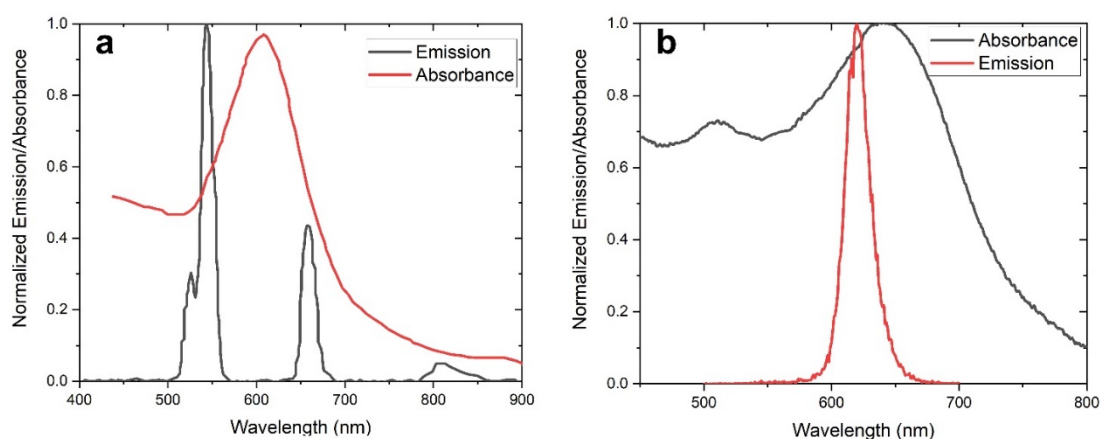


Figure S 1. Absorption and emission spectra related to a) UCNP, and b) QDs.

Figure S 2. Emission/absorption spectra of a) NaYF₄: Yb, Er UCNP/AuNU, and b) CdSe/ZnS core/shell QD/AuNR.

Supporting Information

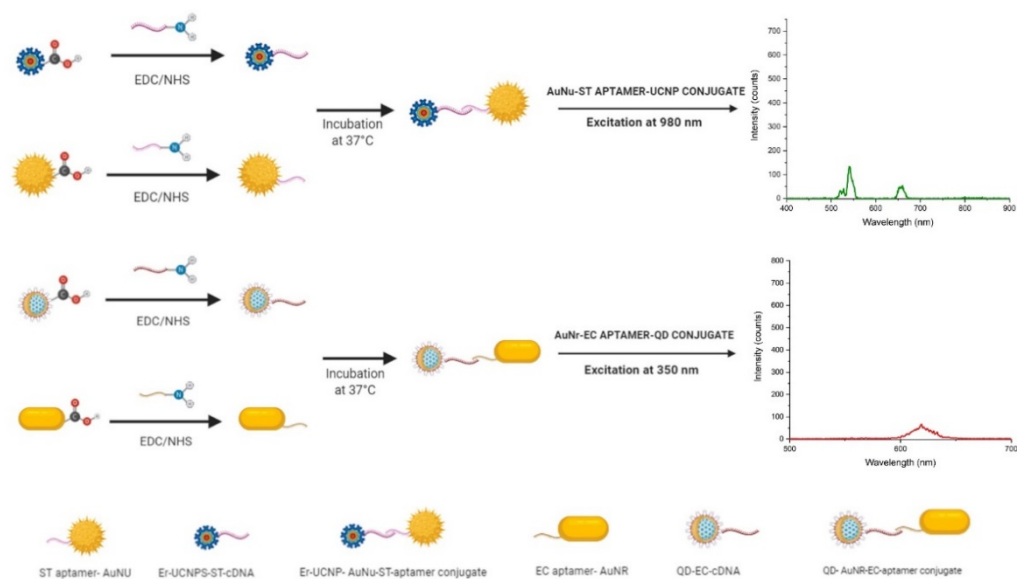


Figure S 3. Preparation of the AuNS-aptamer-cDNA-LNP conjugates.

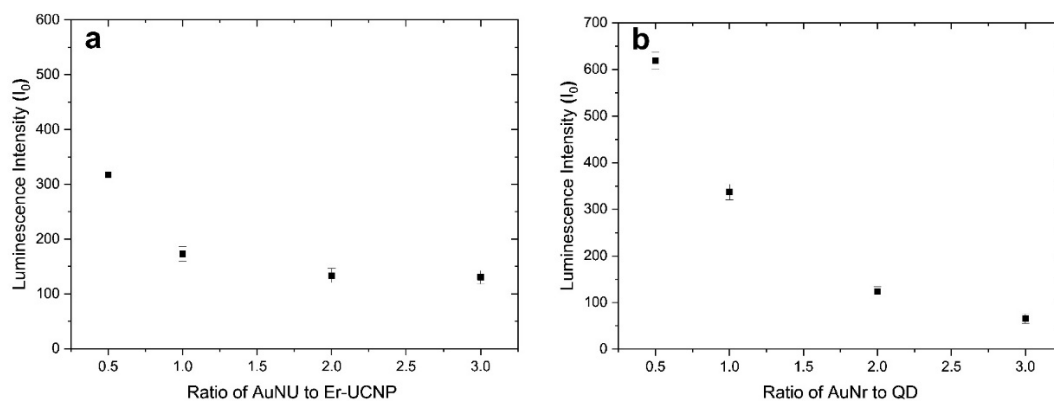
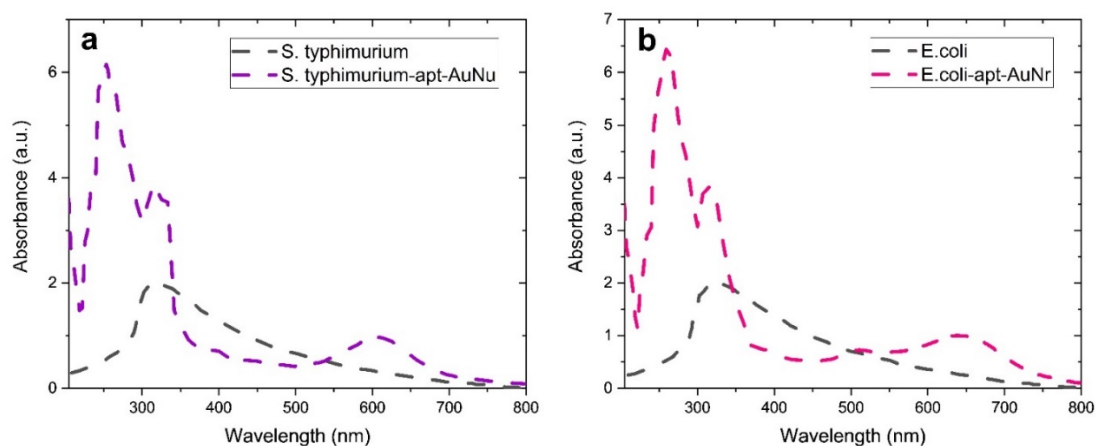


Figure S 4. The luminescence intensity of the a) UCNPs-AuNu conjugates at 545 nm b) QD-AuNR conjugates at 620 nm in terms of various initial acceptor/donor ratios in 1x PBS.

Figure S 5. UV-vis absorption spectra of a) *S. typhimurium* with and without related AuNu-aptamer, b) *E. coli* with and without AuNR-aptamer.

Supporting Information

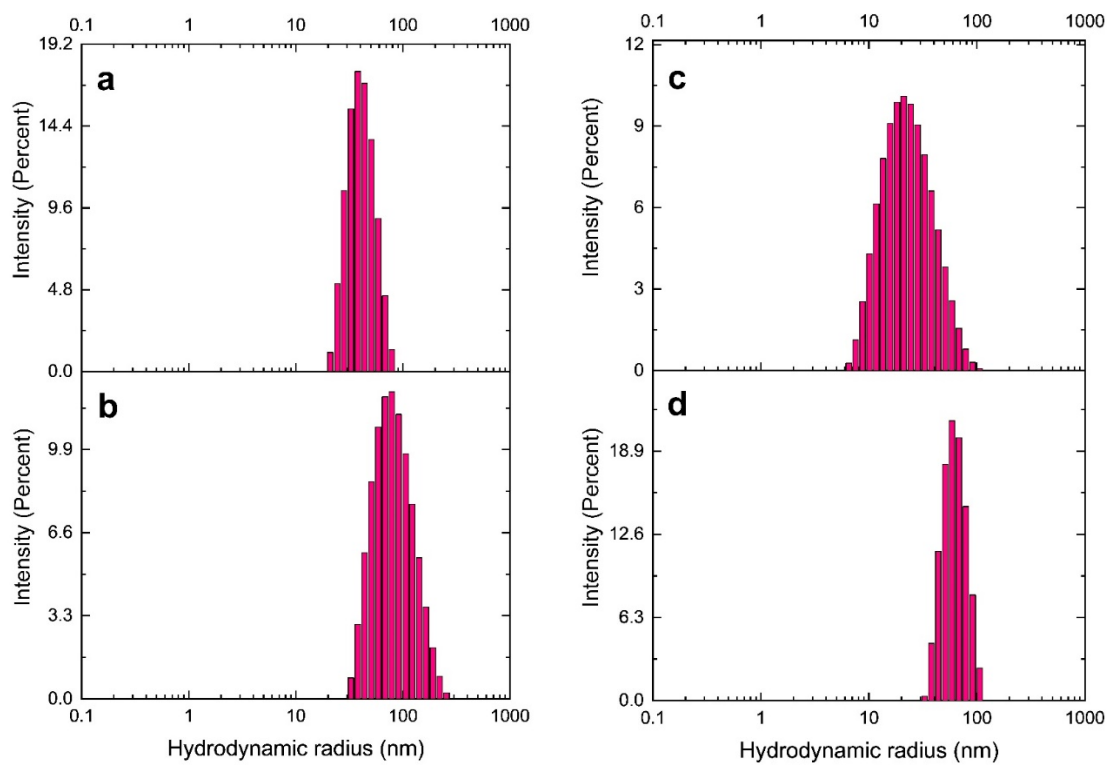


Figure S 6. Hydrodynamic size distribution of a) unmodified UCNP and b) ST cDNA-coupled UCNP, c) unlabeled QD, and d) EC cDNA-modified QDs.

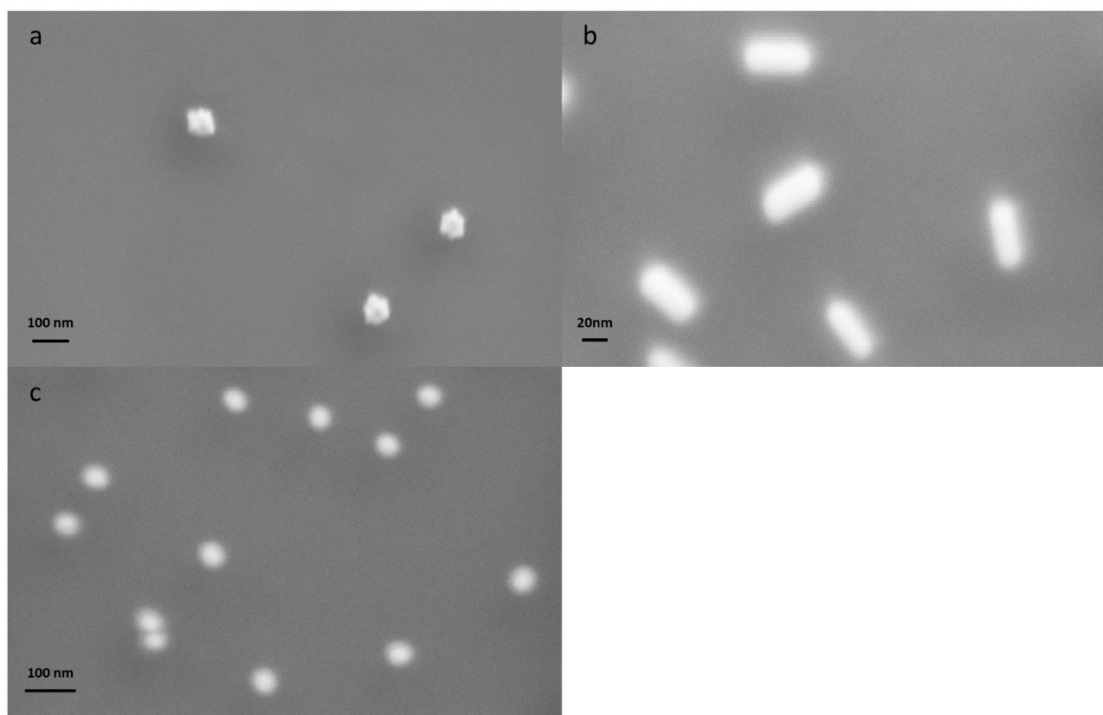


Figure S 7. SEM images of a) ST-aptamer-functionalized AuNU and b) EC-aptamer-functionalized AuNR, and c) ST-cDNA-modified UCNP.

Supporting Information

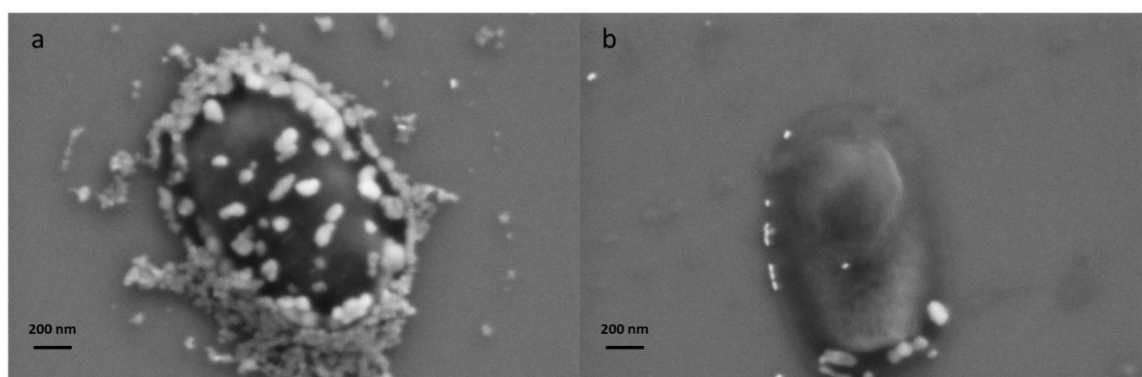


Figure S 8. SEM images of a) ST-AuNU nanoprobe with *S. typhimurium*, and b) EC-AuNR nanoprobe with *E. coli*.

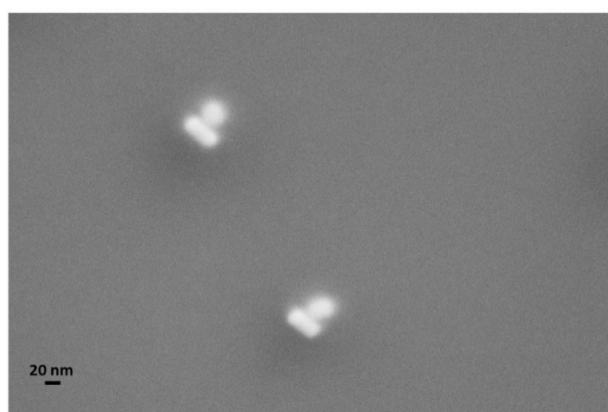


Figure S 9. SEM images of UCNP-AuNR conjugate.

Supporting Information

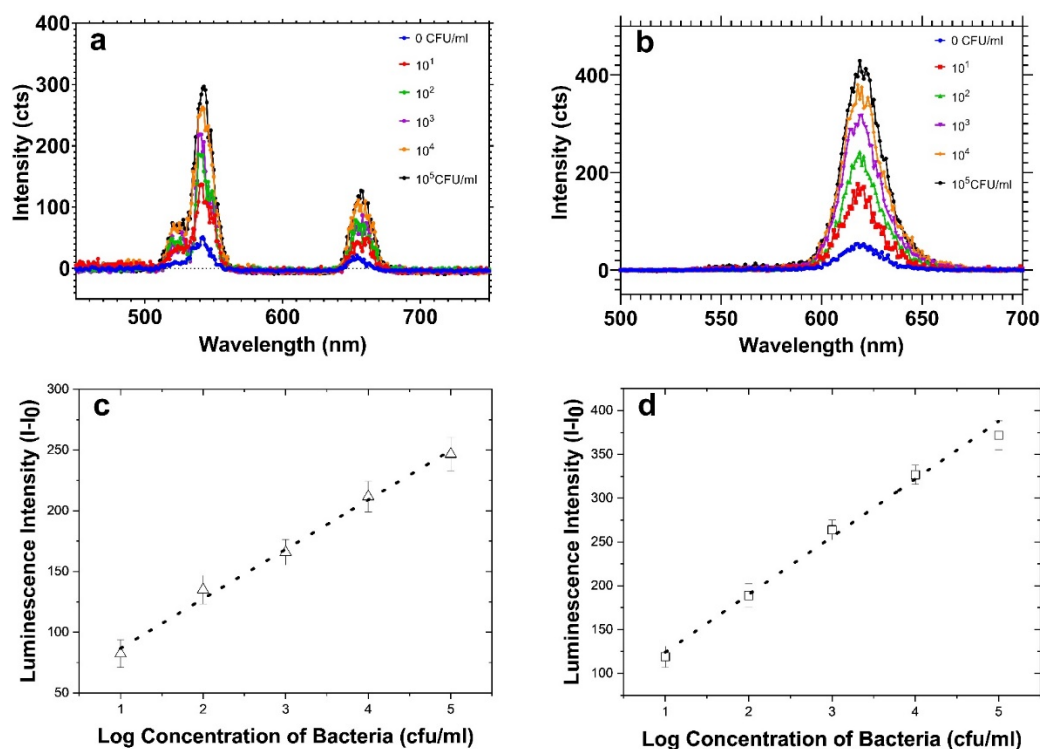


Figure S 10. The luminescence intensity increase of a) UCNP and b) QD nanoprobe versus the logarithmically increased concentrations of *S. typhimurium* and *E. coli*, respectively. The Calibration curves for multiplex sensing of c) *S. typhimurium* and d) *E. coli* are also provided.

Reference

1. W. Guo, Y. Wei, Z. Dai, G. Chen, Y. Chu and Y. Zhao, *Materials*, 2018, **11**.
2. J. Griffin, A. K. Singh, D. Senapati, P. Rhodes, K. Mitchell, B. Robinson, E. Yu and P. C. Ray, *Chemistry - A European Journal*, 2009, **15**, 342-351.
3. M. X. Gao, H. Y. Zou, Y. F. Li and C. Z. Huang, *Anal Chem*, 2017, **89**, 1808-1814.
4. M. Li, Q. Wang, X. Shi, L. A. Hornak and N. Wu, *Analytical Chemistry*, 2011, **83**, 7061-7065.
5. G. K. Darbha, A. Ray and P. C. Ray, *Acs Nano*, 2007, **1**, 208-214.
6. Q. Li, A. Sun, Y. Si, M. Chen and L. Wu, *Chemistry of Materials*, 2017, **29**, 6758-6765.
7. C. Bravin and V. Amendola, *Biosensors and Bioelectronics*, 2020, **169**, 112591.
8. B.-Y. Fang, C.-Y. Wang, C. Li, H.-B. Wang and Y.-D. Zhao, *Sensors and Actuators B: Chemical*, 2017, **244**, 928-933.
9. Z. Abolghasemi-Fakhri, T. Hallaj and M. Amjadi, *Luminescence*, 2021, DOI: 10.1002/bio.4040, 1-8.
10. J. J. Liu, D. Yuan, H. Z. Zhang, Y. D. Lu, N. Wang, H. Y. Zou and J. Wang, *Sensors and Actuators, B: Chemical*, 2018, **274**, 318-323.
11. H. Kurt, M. Yüce, B. Hussain and H. Budak, *Biosensors and Bioelectronics*, 2016, **81**, 280-286.
12. M. Yüce, H. Kurt, B. Hussain, C. W. Ow-Yang and H. Budak, *ChemistrySelect*, 2018, **3**, 5814-5823.