Electronic Supplementary Material (ESI) for Environmental Science: Nano.

Bacillus subtilis Causes Dissolution of Ceria Nanoparticles at the Nano-Bio Interface

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Synthesis of Rod-ceria. Rod-Ceria was synthesized by hydrothermal method.[1] Briefly, 0.694g of Ce(NO3)3·6H2O (purity > 99.9%) and 7.68g NaOH were dissolved in 4 mL and 28 mL of deionized (DI) water, respectively. These two solutions were mixed in a Teflon-lined autoclave, and kept stirring for 2 minutes. Then, the autoclave was transferred into a temperature-controlled electric oven, and subjected to hydrothermal treatment at temperatures of 100 °C for 24 h. After cooling to room temperature, fresh white precipitates were separated by centrifugation, and washed with DI water and ethanol several times.

Characteristics of Rod-ceria. The crystal structure, size, and shape of the synthesized ceria nanoparticles were investigated using powder X-ray diffraction (XRD), scanning electron microscopy (SEM), and transmission electron microscopy (TEM). The hydrodynamic size and zeta potential of rod-ceria in DI water were characterized by dynamic light scattering (DLS) with a Zetasizer Nano ZS instrument (Malvern Instruments, UK). The XANES spectra of the as-synthesized rod-ceria was collected on the 1W1B beamline of the Beijing Synchrotron Radiation Facility (BSRF, China).

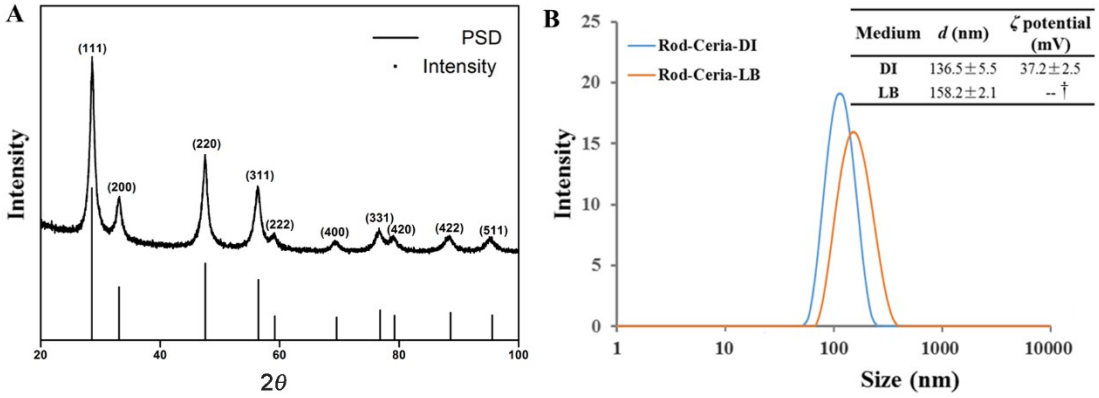

Fig. S1. The physicochemical properties of rod-ceria. A) The XRD patterns of rod-ceria (JCPDS card no. 34-0394). B) Size distribution and zeta potential of rod-ceria in the deionized water (DI) and LB medium were measured by dynamic light scattering assay; †: the zeta potential of rod-ceria in LB culture medium could not be determined due to the high sample conductivity.

TEM and SEM.

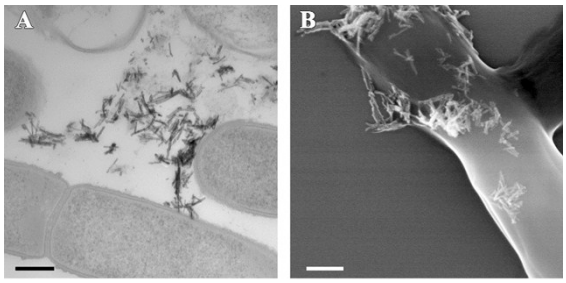

Fig. S2. The TEM and SEM images of *B. subtilis* and *E. coli* incubated with 100 mg/L rod-ceria in LB medium for 48 h. TEM (A) and SEM (B) images of *B. subtilis*. The scale bars are 200 nm.

Ce Speciation by LCF Combined XANES Analysis.

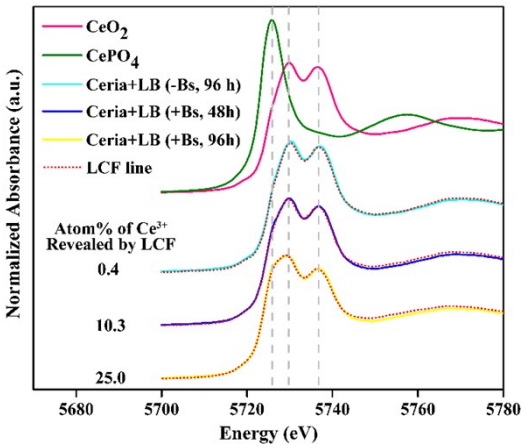

Fig. S3. Ce LIII-edge XANES spectra. XANES spectra of reference standards: bulk CeO_2 and $CePO_4$; XANES spectra of samples: 100 μg/mL rod-ceria incubated in the LB medium with or without *B. subtilis* for 48 or 96 hours; the LCF line and the fitting result.

MTT Assay with or without the Presence of Rod-ceria or Ce^{3+}. MTT assay was conducted immediately after the addition of 100 μg/mL of rod-ceria or Ce^{3+} into the *B. subtilis* suspension in LB medium. The *B. subtilis* suspension without rod-ceria or Ce^{3+} was used as a control for MTT assay.

Table S1. The viability of *B. subtilis* with or without the presence of rod-ceria or Ce^{3+}

Control	Rod-ceria	Ce^{3+}
100.0±8.3%	99.3±8.5%	96.7±6.6%

Cytotoxicity of Rod-ceria against *B. subtilis*. The viability of *B. subtilis* was determined by MTT

assay after a 6-h incubation with rod-ceria or Ce^{3+} ions in LB medium.

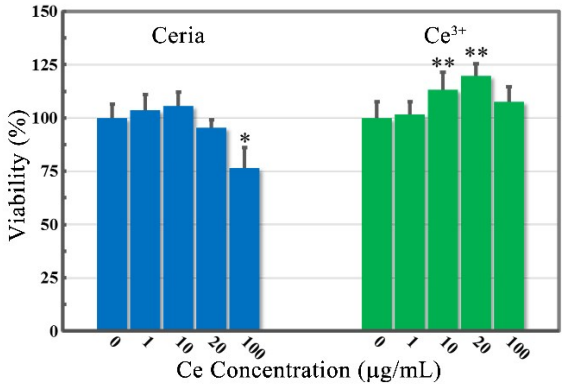

Fig. S4. The viability of *B. subtilis* after a 6-h incubation with rod-ceria or Ce^{3+} ions in LB medium. Data are expressed as mean ± SD (n=6). Statistical significance: *, $0.01 < p < 0.05$; **, $p < 0.01$ versus control.

Determination of Lipid Peroxidation. Lipid peroxidation assessment of the rod-ceria-treated *B. subtilis* was carried out with an assay kit (Nanjing Jiancheng Bioengineering Institute, Nanjing, China), according to the description by Ashutosh Kumar *et al.*[2] Malondialdehyde (MDA) and thiobarbituric acid form a pink colored complex, which can be quantitatively measured at λ max = 532 nm. The *B. subtilis* without rod-ceria were used as a negative control, and the *B. subtilis* treated with Fenton reagent were used as a positive control. The result showed that the incubation of *B. subtilis* with rod-ceria in the LB medium for 6 h did not cause any lipid peroxidation in *B. subtilis*.

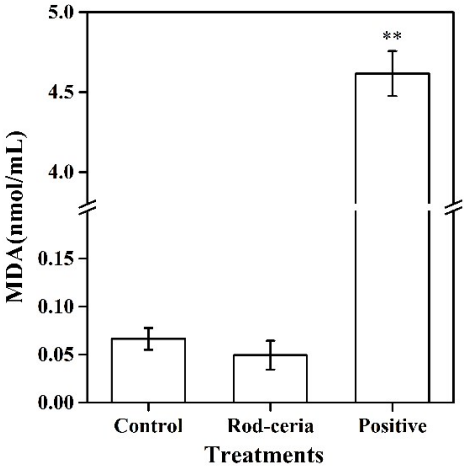

Fig. S5. The lipid peroxidation levels of *B. subtilis* treated with rod-ceria (100 mg/L) and Fenton reagent (Positive control) in LB medium for 6 h. Data were expressed as mean ± SD (n=6). Statistical significance: **, $p < 0.01$ versus control.

Notes and references

1. Mai, H.; Sun, L.; Zhang, Y.; Si, R.; Feng, W.; Zhang, H.; Liu, H.; Yan, C. Shape-selective synthesis and oxygen storage behavior of ceria nanopolyhedra, nanorods, and nanocubes. *The Journal of Physical Chemistry B* **2005**, *109* (51), 24380-24385.

2. Kumar, A.; Pandey, A. K.; Singh, S. S.; Shanker, R.; Dhawan, A. Engineered zno and tio 2 nanoparticles induce oxidative stress and DNA damage leading to reduced viability of escherichia coli. *Free Radic. Biol. Med.* **2011**, *51* (10), 1872-1881.