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Harnessing Intracellular Biochemical Pathways for In Vitro Synthesis of Designer Tellurium Nanorods

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Abstract

Synthesizing nanomaterials of desired properties is a big challenge, which requires extremely harsh conditions and/or use of toxic materials. More recently developed in vivo methods have brought a different set of problems such as separation and purification of nanomaterials made in vivo. Here, a novel approach that harnesses cellular pathways for in vitro synthesis of high-quality

Supporting Information

Supporting Information is available from the Wiley Online Library or from the author.

tellurium nanorods with tunable lengths and optical properties is reported. It is first demonstrated that in vivo biochemical pathways could be used to synthesize Te nanorods via the intracellular reduction of TeO_3^{2-} in living *Staphylococcus aureus* cells. The pathways to set up a quasi-biological system for Te precursor formation are then utilized, which could further synthesize Te nanorods in vitro. This allows to successfully synthesize in vitro, under routine laboratory conditions, Te nanorods with uniform and tunable lengths, ranging from about 10 to 200 nm, and controllable optical properties with high molar extinction coefficients. The approach here should open new avenues for controllable, facile, and efficient synthesis of designer nanomaterials for diverse industrial and biomedical applications.

1. Introduction

Nanomaterials, due to their diverse sizes, shapes, compositions, and versatile optical, electronic, and magnetic properties, have attracted tremendous research interests in the last decade or two, leading to a wide range of applications in different areas such as tracking,^[1] biodetection,^[2] and optoelectronic and photovoltaic devices.^[3,4] The success of such applications is critically dependent on the ability to synthesize nanomaterials with desired structure and properties. However, it has been a significant challenge to design and fabricate such nanomaterials. In vitro fabrication methods often involve strong reducing agents, harsh reaction conditions, and extremely rapid process, which are exquisitely difficult to control. This has made it attractive to manipulate biochemical reactions in living cells to synthesize nanomaterials.^[5,6] Live cells have been exploited to synthesize nanomaterials with exquisitely controlled size, shape, composition, and crystal structure of the desired inorganic nanomaterials due to the high degree of organization and sophisticated molecular controls in these biological systems.^[7,8] As the intrinsic properties of nanomaterials are mainly determined by their composition, size, shape, crystallinity, and structure, one can potentially fine-tune the properties of nanomaterials by controlling any of these parameters.^[9] Thus, using biochemical reactions in living cells to synthesize materials offers a very attractive way to obtain the designer nanomaterials under easily feasible conditions.

Toward this goal, we have previously developed a novel “space–time coupling” strategy that allows the synthesis of inorganic nanomaterials in a highly tunable way in live cells.^[5,6,10,11] The key feature is to temporally and spatially couple intracellular unrelated biochemical reactions or metabolic pathways that do not encounter each other under normal condition to enable the synthesis of nanomaterials with desired properties through elaborate design and facile chemical handling. For example, by coupling of intracellular selenium reduction and Cd(II) detoxification, fluorescent CdSe quantum dots (QDs) with tunable fluorescence spectra can be synthesized in live yeast cells just by “culturing” the live cells with chemical compounds.^[5,10,11] More recently, we have successfully transformed *Staphylococcus aureus* (*S. aureus*) to highly fluorescent cellular beacons via this space–time coupling strategy.^[6] Specifically, exogenous high-valenced inorganic selenium compounds are uptaken by *S. aureus* cells to produce intracellular low-valenced organoselenium compounds via intracellular selenium metabolic pathways. These compounds then react with cadmium complexes formed from detoxification of exogenous Cd(II) and endogenous biomolecules containing mercapto groups to yield fluorescent $\text{CdS}_{0.5}\text{Se}_{0.5}$ QDs in the cytoplasm. The final

outcome is the transformation of whole cell into an ultra bright bulb due to intracellular QDs' fluorescence, which can be directly used as cellular beacons with outstanding photostability, high luminance, near perfect uniformity, good monodispersity, and repeatability.^[6]

Despite such progresses, it is difficult to utilize such live cell synthesis strategy for large-scale synthesis of nanomaterials and/or their diverse applications, which often require highly purified nanomaterials. This is mainly due to the lack of knowledge on the biosynthetic mechanisms underlying the intracellular nanomaterial formation.^[12] To overcome this bottleneck, we hypothesize the use of the biochemical principles involved in intracellular nanomaterial synthesis to direct in vitro synthesis of designer nanomaterials. Here, we have tested this idea by utilizing an intracellular process for precursor formation to successfully synthesize high-quality Te (tellurium) nanorods with adjustable lengths in vitro in a simple and efficient process without using any combustible, explosive, and toxic organic reagents (Figure 1).

2. Results and Discussion

We chose the synthesis of Tenanorods to investigate the possibility of using intracellular pathways for in vitro synthesis of nanomaterials. We first determined whether Te (tellurium) nanorods could be synthesized in vivo. We incubated *S. aureus* cells with Na₂TeO₃ solution for 5 h, which led to a gradual, Na₂TeO₃-dependent color change of the cell culture from milky yellow to grayish black (Figure S1, Supporting Information), suggesting the formation of Te-containing nanomaterials. Atomic composition analysis by using inductively coupled plasma atomic emission spectroscopy (ICP-AES) showed the total content of tellurium was 15.2 mg per gram wet weight of the cells cultured in the presence of Na₂TeO₃ in the culture medium, indicating that exogenous Na₂TeO₃ was taken up by the cells. Control cells cultured in the absence of Na₂TeO₃ had no color change in the cell culture or tellurium in the cells, illustrating exogenous Na₂TeO₃ was indispensable for the intracellular Te accumulation.

To characterize the intracellular Te-containing materials, the cell samples were analyzed by using transmission electron microscopy (TEM) images. Cells in the stationary phase appeared to accumulate Te particles within the cytoplasm of cells cultured in the presence of Na₂TeO₃ (Figure 2a) but not in its absence (Figure 2b). The particles appeared rodlike, with at least one nanorod per cell based on the analysis of the TEM images of ~300 individual cells, suggesting that the reduction of Te(IV) to Te⁰ took place in the cells, which was consistent with the above ICP-AES analysis. Further analysis with in situ high-resolution transmission electron microscopy (HRTEM) revealed that the individual rod was an apparent single crystalline particle with the (101) lattice planes (~0.32 nm) (Figure 2c). Furthermore, energy-dispersive X-ray (EDX) spectroscopy showed that Te was the predominant component of the nanorods, although detectable S element, likely from the endogenous intracellular biomolecules containing sulfur, was also found (Figure 2d). Finally, powder X-ray diffraction (XRD) analysis indicated that the positions and relative intensities of all diffraction peaks of Te nanorods matched well with the hexagonal Te (JCPDS Card No. 01-0727) (Figure S2, Supporting Information).

We have shown previously that intracellular reduction of selenite (Na_2SeO_3) by glutathione reductase (GR) leads to the formation of low-valenced organoselenium in the *S. aureus* cells. [6] Since Se and Te are in the same family within the periodic table, we reasoned that Te in Na_2TeO_3 might also be reduced to low-valenced tellurium by the intracellular glutathione reductase in the cells under our experimental conditions. Thus, we hypothesized that TeO_3^{2-} could be reduced by the glutathione (GSH), reduced nicotinamide dinucleotide phosphate (NADPH), and GR to GSTeH as shown in the chemical equations (1) and (2). It is known that glutathioselenol (GSSeH) can spontaneously decompose into elemental selenium ($\text{Se}^{(0)}$). [13] We hypothesized that the resulting GSTeH could also decompose into elemental tellurium ($\text{Te}^{(0)}$), as described in chemical equation (3), leading to the formation of Te nanorods

To test the above hypotheses about the intracellular biochemical pathways for the in vivo synthesis of Te nanorods, we carried out in vitro reactions to mimic the intracellular processes shown in the reactions (1)–(3) above. To biomimic the reduction of TeO_3^{2-} to GSTeSG with GSH in vitro, we mixed 40 μL of 0.05 M Na_2TeO_3 , 2.1 mL of 0.1 M Britton–Robison (BR) buffer ($\text{pH} = 7.1$), and 80 μL of 0.1 M GSH and incubated the mixture for 30 s at room temperature. High-performance liquid chromatography coupled with mass spectrometry (HPLC-MS) was performed to determine products in the reaction mixture. Mass spectra analyses of the three HPLC peaks showed that they were GSH, GSSG, and GSTeSG, respectively (Figure S3b–d, Supporting Information), as predicted based on reaction (1) above. Thus, Na_2TeO_3 can indeed be reduced by GSH in vitro, mimicking the biological process in the cells.

GSTeSG reacting with NADPH was the key step of the reduction process. We used HPLC-MS to detect the formation of the hypothesized Te precursor (GSTeH) from the in vitro reaction. 40 μL of 0.05 M Na_2TeO_3 , 2.1 mL of 0.1 M Britton–Robison buffer ($\text{pH} 7.1$), and 80 μL of 0.1 M GSH was mixed and incubated for 30 s at room temperature, next 10 μL of the resulting mixture was mixed with 20 μL of 0.1 M NADPH and 8 μL of GR for 30 s at room temperature. HPLC-MS analysis of the reaction mixture confirmed the production of GSTeH (Figure S4, Supporting Information), indicated both of reactions (1) and (2) above could be biomimicked in vitro. GSTeH is a key intermediate for Te nanorod formation as shown in decomposition reaction (3) above. Therefore, to biomimic the entire in vivo pathways leading to the formation of Te nanorods in the cells, we created a quasi-biological

system containing electrolytes, reduced GSH, reduced NADPH, and GR to allow reactions (1) and (2) to take place. At the end of the reaction, the mixture was injected into 0.01 M NaOH solution, leading to a gradual change of the color of the solution to brown (Figure 3a, inset), suggesting the formation of Te nanorods.

The UV–visible spectrum of the brown solution revealed two distinct interband transitions of Te nanorods (Figure S5a, Supporting Information). Based on the findings from the Hartree–Fock–Slater study,^[14] peak I at 4.66 eV originated from the transition from valence band (p-bonding VB2) to the conduction band (p-antibonding CB1), which typically appears in the range of 3–6 eV, whereas peak II at 2.99 eV was due to the transitions from the p-lone pair valence band (VB3) to the p-antibonding conduction band (CB1), which typically appears in the range of 0–3 eV. The molar extinction coefficient of Te nanorod was found to be $0.806 \times 10^9 \text{ M}^{-1} \text{ cm}^{-1}$ (see the Experimental Section).

TEM analyses of the Te nanorods formed in our in vitro system showed that the nanorods were uniform both in size and shape (Figure 3a). XRD was carried out to determine the phase and purity of the Te nanorods (Figure 4d, the upper curve labeled with 0.01 M). Intensities and positions of the diffraction peaks (upper curve) matched those of the hexagonal Te (Figure 4d bottom curve: the XRD pattern from the standard Te powder, JCPDS No. 01–0727). The absence of peaks corresponding to any other phases indicated the high purity of the products.

The HRTEM image (Figure 4a) revealed the Te nanorods as a single crystalline structure with the lattice spacing of $\approx 0.32 \text{ nm}$, which is the same as the Te nanorods synthesized in vivo (Figure 2c) and agrees well with the known distance between adjacent facets (101) of hexagonal Te. To determine the composition of nanorods, we then used energy-dispersive X-ray spectroscopy. The results showed that the predominant component is Te (Figure S6a, Supporting Information). A small amount of S was also present, likely originated from the GSH that passivated the nanorod surfaces. X-ray photoelectron spectroscopy (XPS) further confirmed that the nanomaterials were made of Te (Figure S6b, Supporting Information). These results demonstrated that reactions (1)–(3) involved in the synthesis of intracellular Te nanorods could be mimicked to obtain Te precursors for Te nanorod formation in vitro.

As we all know, Te^0 produced in reaction (3) can readily disproportionate in an alkaline solution to generate Te^{2-} and TeO_3^{2-} ,^[15,16] as shown in chemical equation (4) below. We hypothesized that the size of the Te nanorods synthesized in vitro can be regulated by changing the concentration of NaOH, thus enabling the synthesis of the designer nanomaterials. Indeed, when we used different NaOH concentrations in our in vitro system, we found that the lengths but not the diameters of the Te nanomaterials formed changed inversely with the concentration of NaOH used (Figure 3). The Te nanocrystals formed under NaOH concentration were all rod-shaped and retained high uniformity within a narrow size distribution (Figures 3 and 4). Furthermore, within each type of tellurium nanorods, the lattice space of Te nanorods was kept at $\approx 0.32 \text{ nm}$ (Figure 4a–c), corresponding to the (101) facet of tellurium, and all were exclusively indexed as the hexagonal Te crystal structure (Figure 4d). Thus, by simply changing NaOH concentration from 0.01 to 0.05 or 0.08 M in a 10 min in vitro reaction, we could produce nanorods that

were essentially identical in diameter, at 6.8 ± 0.8 , 6.2 ± 1.8 , and 6.6 ± 2.5 nm, respectively, but with very different length, at 43.0 ± 8.0 , 22.0 ± 6.3 , and 16.4 ± 3.8 nm, respectively (Figure 3a–f). These results suggested that the Te nanorods grew along their longitudinal direction, while the lateral dimensions were maintained essentially constant under our in vitro conditions. This was likely because the lateral dimensions are mainly confined by the highly anisotropic crystal structure of the trigonal tellurium^[17,18]

To explain the inverse correlation between NaOH concentration and the length of the nanorods, we realized that according to chemical equation (4) above, Te^0 concentration can be affected by the NaOH concentration. With increasing NaOH concentration, more Te^0 can be dissolved in this disproportionation reaction, thus reducing Te monomer needed for the growth of nanocrystals, leading to shorter nanocrystals in the solution.^[19,20] To test this hypothesis, we carried out ICP-AES analyses of the reaction solutions at different concentrations. With the same amount of input materials but increasing NaOH concentration, we found that Te^0 in the solution decreased with increasing NaOH concentration (Table S1, Supporting Information). As Te^0 is required for Te nanorod formation, the reduction in Te monomer (Te^0) due to increased NaOH concentration might be responsible for the observed reduction in the lengths of the Te nanorods formed. Such a mechanism would also suggest that continued presence of NaOH solution would lead to the dissolution of the nanorods already formed due to continued disproportionation. Indeed, when we incubated Te nanorods formed in 0.01 M NaOH with 0.1 M NaOH solution for 40 min, the length of the nanorods shortened from 43.0 ± 8.0 to 27.4 ± 5.4 nm (Figure S7, Supporting Information).

While changing the concentration of NaOH allowed us to tune the length of the nanorods, the range was fairly limited, from around 16 nm at 0.08 M NaOH to 43 nm at 0.01 M NaOH, and there is only a limited range of NaOH concentrations that can be used. To increase the range of the length for the in vitro synthesized nanorods, we reasoned that lower concentration of starting Te^0 would reduce the nucleation of nanorod formation. On the other hand, having continuous presence of Te^0 should enable continued growth of the nanorods, thus lengthening the nanorods. According to chemical equation (2), the formation of GSTeH would be slower if the reaction were conducted in the absence of GR, this would in turn lead to the slower formation of Te^0 (reaction (3), leading to reduced Te^0 and thus less nucleation for nanorod formation. If the amounts of the other reactants/compounds and NaOH concentration were kept the same, then the lengths of Te nanorods formed would be longer since more Te monomer left would be supplied for growing in size. To test this possibility, we carried out the same in vitro synthesis as above but leaving out GR. Again, after injecting the Te precursors into 0.01 M NaOH, the solution became grayish black (Figure S8a, inset, Supporting Information), indicating the formation of Te nanorods. Analysis of the products indicated that there were two distinct interband transitions of Te nanorods appearing in the UV–visible spectrum of the grayish black solution (Figure S5b, Supporting Information), which was similar to the Te nanorods formed with GR added

except for some redshifts in the absorption peak positions, arising from the change of the size of Te nanorods.^[21] And the molar extinction coefficient of Te nanorod was calculated to be $1.542 \times 10^9 \text{ M}^{-1} \text{ cm}^{-1}$ (see the Experimental Section). Importantly, the Te nanorods were found to have a length of 175.6 nm (Figure S8b, Supporting Information), much longer than those formed in the presence of GR, consistent with our prediction above. The HRTEM image confirmed the crystallinity of these Te nanorods (Figure S9a, Supporting Information) and the fringe spacing (≈ 0.32 nm) agreed well with the (101) lattice plane of Te nanorods. Likewise, the XRD corroborated that the nanorods were phase pure Te, agreeing well with the standard Te powder on JCPDS No. 01-0727, with no other crystalline impurities observed (Figure S10, Supporting Information). EDX and XPS results also indicated that the nanorods were composed of tellurium element (Figure S11, Supporting Information), with a Te 3d binding energy of 572.9 and 575.5 eV.^[22] Thus, by including or omitting GR in the in vitro system, we could synthesize essentially identical Te nanorods with very different lengths.

We also tested the effect of different NaOH concentrations on the lengths of Te nanorods formed in the absence of GR (Figure S8, Supporting Information). Based on the measurement of ≈ 200 such Te nanorods under each condition, we found that the mean lengths were 60.7 and 35.9 nm for the nanorods formed under 0.05 and 0.08 M of NaOH, respectively (Figure S8d,f, Supporting Information). Again, the corresponding HRTEM results (Figure S9b,c, Supporting Information) demonstrated that the Te nanorod was structurally single crystalline with a periodic fringe spacing of 0.32 nm, which corresponds to the interplanar spacing between the (101) planes of the hexagonal lattice of Te, just like those obtained at 0.01 M NaOH. Thus, by utilizing intracellular pathways to in vitro applications and altering GR and NaOH concentrations, we have succeeded in synthesizing Te nanorods with the length ranging from about 10 nm to nearly 200 nm in a simple step in vitro system.

3. Conclusion

We have presented here the very first example of successfully harnessing intracellular biochemical pathways for in vitro synthesis of Te nanorods. Our strategy was to first identify an in vivo system for the synthesis of a nanomaterial of interest, then deduce the likely underlying intracellular pathways, and finally utilize the pathways for in vitro synthesis of the nanomaterial. We have validated this strategy by utilizing intracellular processes to synthesize Te nanorods under mild experimental conditions that can be easily manipulated to produce uniform Te nanorods of tunable lengths ranging from 10 to 200 nm. We envision that such a strategy should allow the discovery of other in vivo pathways for in vitro adaptations to synthesize other nanomaterials. It is likely that the same in vitro system that we have developed here can be used to synthesize other nanomaterials as well, such as using other elements in the same Te family of the periodic table. In addition, it may be possible to synthesize Te nanomaterials of different structures and properties by altering the in vitro chemical reaction conditions. Furthermore, the molar extinction coefficients were 1.54×10^9 and $0.806 \times 10^9 \text{ M}^{-1} \text{ cm}^{-1}$ for 549 and 410 nm peaks of Te nanorods, respectively, which are comparable to gold nanorods with the similar lengths.^[23] Thus, similar to gold nanorods, the Te nanorods may serve as a new kind of photothermal material that can be applied in the

tumor therapy. More important to the ever expanding applications of nanomaterials, the in vitro systems such as the one reported here can be easily scaled up and manipulated to produce and purify large amounts of high quality and homogenous nanomaterials under conditions that are user friendly and reliable, which has long been a major road block for broad applications of nanomaterials.

4. Experimental Section

Material:

Ultrapure water was used in all of the experiments (18.2 mΩ cm, Millipore, France). Sodium tellurite (Na₂TeO₃) and sodium chloride (NaCl, AR) were purchased from Sinopharm Chemical Reagent Co., Ltd. Yeast extract and tryptone were obtained from Bectone, Dickinson and Company, USA. Reduced GSH was purchased from Amresco. NADPH (reduced β-nicotinamide adenine dinucleotide 2'-phosphate tetrasodium salt) was purchased from Biomol. Yeast GR was purchased from Calbiochem. The Amicon centrifugal filter units were purchased from Millipore.

Intracellular Synthesis of Te Nanorod:

S. aureus (CCTCC AB91093) was obtained from the Chinese Center for Type Culture Collections of Wuhan University (Wuhan, P. R. China). The *S. aureus* cells were cultured for 12 h with LB broth (yeast extract 5 g L⁻¹, tryptone 10 g L⁻¹, and NaCl 5 g L⁻¹) at 37 °C to ensure that the cells were in the stationary phase. The cells were then incubated with Na₂TeO₃ (final concentration: 2.5 × 10⁻³ M) for 5 h to allow the synthesis of Te nanorods in the cells.

HPLC-MS Analysis of GSH, GSSG, and GSTeSG:

This was done with an HPLC system equipped with an Agilent 1100 quaternary gradient pump and a diode array detector for chromatographic separations. The HPLC system was interfaced with an LC-MSD-Trip-XCT mass spectrometer (Agilent). The components of the reduced product of Na₂TeO₃ were separated and analyzed by HPLC-MS. All separations were performed on a Kylin C18HC column (5 μm, 4.6 × 150 mm, Dalian Sipore Co., Ltd.), with a liquid phase flow rate of 0.8 mL min⁻¹ and column temperature of 25 °C. The liquid phase consisted of 0.1% formic acid in water and acetonitrile at a ratio of 100:1 (v/v). Absorption at 220 nm was used as the detection wavelength of the UV spectrometry detector. 40 μL of Na₂TeO₃ (0.05 M), 2.1 mL of Britton–Robison buffer (0.1 M, pH 7.1), and 80 μL of GSH (0.1 M) were incubated together for 30 s. 5 μL of the resulting mixture was injected into the HPLC-MS system for analysis. Electrospray ionization (ESI) instrument in the positive polarity was used for the analysis of GSH, GSSG, and GSTeSG with a capillary voltage of 3.5 kV, dry gas of 10.00 L min⁻¹, and nebulizer 40.00 psi.

HPLC-MS Analysis for GSTeH:

This was performed on an HPLC-ESI/MS system (Shimadzu LC/MS-2010EV, Tokyo, Japan). All separations were performed on a Tigerkin C18TDE column (5 μm, 2.1 × 150 mm, Dalian Sipore Co., Ltd), with a liquid phase flow rate of 0.2 mL min⁻¹ and column temperature of 30 °C. The liquid phase consisted of 0.1% formic acid in water and

acetonitrile at a ratio of 100:1 (v/v). Absorption at 220 nm was used as the detection wavelength of UV spectrometry detector. 40 μ L of 0.05 M Na_2TeO_3 , 2.1 mL of 0.1 M pH 7.1 BR buffer, and 80 μ L of 0.1 M GSH were mixed and incubated for 30 s. 10 μ L of the resulting mixture was mixed with 20 μ L of 0.1 M NADPH and 8 μ L of GR and were injected into HPLC-ESI/MS system. Electrospray ionization instrument in the positive polarity was used for the analysis of GSTeH with capillary voltage of 4.5 kV, dry gas of 1.5 L min^{-1} , and nebulizer 0.02 MPa.

In Vitro Synthesis of Te Nanorod:

11.2 μ moles of GSH, 2.8 μ moles of Na_2TeO_3 , 2.8 μ moles of NADPH, and 8 μ L of 5 mg mL^{-1} (protein concentration) GR were dissolved in 3 mL of BR buffer (pH 7.1) and incubated for 1 min at room temperature to obtain fresh Te precursors. The Te precursors were quickly injected into a flask containing 11 mL of NaOH (0.01 M), and the mixture was stirred at 80 $^{\circ}\text{C}$ for 10 min under Ar atmosphere, during which the color of the solution changed from colorless to brownish, indicating the formation of Te nanorods. The product was purified with an Amicon Centrifugal Filter Unit (MWCO 10 kDa) to remove free substances. The Te nanorods with different sizes were obtained by using a constant amount of Te precursors while adjusting the concentrations of NaOH.

Characterization of Te Nanorods Synthesized In Vivo:

In situ high-resolution field-emission transmission electron microscopy of cells was done by using JEM-2010 FEE microscopy with an acceleration voltage of 200 kV. The cells were harvested by centrifuging at 4000 rpm for 5 min, washed three times with ultra-pure water (18.2 $\text{M}\Omega$ cm, Millipore, Molsheim, France), and re-dispersed in ultra-pure water. The HRTEM samples were prepared by dispersing the cells onto copper grid coated with ultrathin carbon film, and dried at room temperature. In situ EDX analysis of the cells was performed by using JEM-2010 FEE microscope equipped with EDX spectrometry (EDAX Inc.). The X-ray powder diffraction pattern was obtained with a Bruker AXS D8 Advance X-ray diffractometer. Te nanorods isolated from cells were dropped onto the slide and allowed to dry to a uniform thin film for X-ray powder diffraction.

Characterization of Te Nanorods Synthesized In Vitro:

For TEM and HRTEM analyses, 6 μ L of sample onto ultrathin-carbon-coated copper grids and then air-dried. TEM and HRTEM were performed on a JEOL JEM-2100F high-resolution field-emission transmission electron microscope. The X-ray powder diffraction pattern was obtained with Bruker AXS D8 advance X-ray diffractometer. Absorption spectra were recorded with a Shimadzu UV-2550 UV-vis spectrometer. XPS spectra were measured with a VG Multilab 2000 X-ray photoelectron spectrometer. The elementary analysis of Te nanorods was done with ICP-AES (IRIS Intrepid II XSP, Thermo, Electron Corporation). For this purpose, the purified nanocrystal samples were dissolved in 65% nitric acid. The solution was diluted with water to a final nitric acid concentration of about 2%. The concentration of Te in the solution was calculated from the ICP-AES measurements.

Determination of the Molar Extinction Coefficient:

This was done as reported.^[24] Briefly, the molar concentration of elemental Te (C_{Te}) in Te nanorods was determined from ICP-AES data. The total weight of Te nanorods in the solution was calculated according to the following equation

$$(C_{\text{Te}} \cdot MW_{\text{Te}}) \cdot V \quad (5)$$

where V is the volume of the solution and MW_{Te} is the molecular weight of Te (127.60 g mol⁻¹).

Assuming that the nanorod is a regular hexahedron of lantern (based on the HRTEM and XRD results), the weight of each nanorod could be calculated according to the following equation

$$\rho(\text{Te}) \cdot (3/2) \sqrt{3} \cdot (d/2)^2 L \quad (6)$$

where $\rho(\text{Te})$ is the density of the Te crystal (6.24×10^3 g dm⁻³), d is the average diameter, and L is the height of the nanorod as determined from the HRTEM data.

The molar concentration (c) of Te nanorod is calculated according to the following equation

$$c = C_{\text{Te}} \cdot (MW_{\text{Te}})] \cdot [\rho(\text{Te}) \cdot (3/2) \sqrt{3} \cdot (d/2)^2 L N_A]^{-1} (\text{mol/L}) \quad (7)$$

where N_A is Avogadro's number.

Finally, the molar extinction coefficient (ϵ) is determined according to the following equation: $\epsilon = \text{Abs} \cdot (bc)^{-1}$, where Abs is the absorption value of spectrum peak, b is optical path difference, and c is the molar concentration of Te nanorod.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Figure 1. Schematic diagram for utilizing *in vivo* pathways for efficient *in vitro* synthesis of designer Te nanorods. *S. aureus* cells are used to demonstrate that intracellular reduction of TeO_3^{2-} lead to the synthesis of Te nanorods. The biochemical pathways for the *in vivo* synthesis of Te nanorods are inferred and used to set up a quasi-biological system for Te nanorod synthesis under mild conditions *in vitro*. This involves first the synthesis of the key intermediate, the Te precursor, which can be identified by HPLC-MS, the intermediate is then used in a simple chemical reaction *in vitro* to form Te nanorods with uniform and tunable lengths ranging from about 10 to 200 nm.

Figure 2.

TEM images of *S. aureus* cells reveal the formation of Te nanorods in the presence of Na_2TeO_3 . *S. aureus* cells were incubated at 37 °C for 5 h in the a) presence or b) absence of Na_2TeO_3 . The cells were then analyzed directly by TEM. c) In situ HRTEM image of intracellular Te nanorods revealed that the individual rod was an apparent single crystalline particle with the (101) lattice planes displayed (≈ 0.32 nm). d) In situ energy-dispersive X-ray spectroscopy analysis of intracellularly synthesized Te nanorods showed that Te was the predominant component of the nanorods, although detectable S element, likely from the endogenous intracellular biomolecules containing sulfur, was also present.

Figure 3.

TEM analyses show that in vitro synthesized Te nanorods under different conditions have uniform but different size distributions. TEM images of Te nanorods formed in the presence of a) 0.01, c) 0.05, and e) 0.08 M NaOH, respectively. The inset in each panel shows a photo, with a distinct color, of the vial containing Te nanorods in the respective NaOH solution shortly after injecting the Te precursor. b,d,f) The respective histograms for the width and length distributions.

Figure 4. HRTEM and XRD analyses reveal that Te nanorods formed in vitro in different NaOH concentrations have the same interplanar spacing and crystal structure. HRTEM images of a single Te nanorod formed in the presence of a) 0.01, b) 0.05, and c) 0.08 M NaOH, respectively. Note the rods of different sizes but with the same (101) lattice planes (0.32 nm). d) XRD patterns of the nanorods formed under the different NaOH conditions indicating that all were made of Te. The vertical lines under the spectra represent the JCPDS pattern for Te.