

Study the Antibacterial Effect of Bismuth Oxide and Tellurium Nanoparticles

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Abstract

Noble metals nanoparticles (NPs) were synthesized directly by pulsed laser ablation (Nd : YAG, $\lambda = 1064$ nm) of bismuth and tellurium plates immersed in pure water. Concentrations of the NPs were determined by Atomic Absorption Spectroscopy (AAS) measurement. Atomic Force Microscope (AFM) and Transmission Electron Microscope (TEM) analysis were used to characterize the size and size distributions of the metals NPs. The present study was conducted to synthesize bismuth and tellurium nanoparticles by laser ablation of bulk target immersed in liquid environment and study their antibacterial activity against some pathogenic bacteria. The results revealed that the concentration of NPs increase with increasing the pulses energy, while the average diameter, in general, decreased. Antibacterial properties of nanoparticles are attributed to their total surface area, as a larger surface to volume ratio of TeNPs provides more efficient means for enhanced antibacterial activity against pathogenic bacteria. *Pseudomonas earuginosa* was more sensitive to TeNPs than *Acinetobacter baumannii* and *Escherichia coli*. While, no effect of Bismuth Oxide NPs at different concentration on tested bacteria.

Keywords

Nanoparticles, Bismuth Oxide, Tellurium, Antibacterial Activity

Received: July 4, 2015 / Accepted: July 10, 2015 / Published online: July 24, 2015

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1. Introduction

Nanotechnology involves the characterization, fabrication and/or manipulation of structures, devices or materials that have at least one dimension (or contain components with at least one dimension) that is approximately 1-100 nm in length. When particle size is reduced below this threshold, the resulting material exhibits physical and chemical properties that are significantly different from the properties of macro scale materials composed of the same substance(1). It is applied to various fields such as physical, chemical, biological and engineering sciences where novel techniques are being developed to probe and manipulate single atoms and molecules. Among all NPs the metallic one have

applications in diverse areas such as electronics, cosmetics, coating, packaging and biotechnology (2). NPs can traverse through the vasculature and localize any target organ, this leads to novel therapeutic, imaging and biomedical application (3). NPs have an increased surface area and therefore have increased interaction with biological targets. Bismuth (Bi) is a metallic element of the VA group, together with nitrogen, phosphorus, antimony, and arsenic. Its oxidation numbers are +3 and +5. It is found in the same proportions as silver in the earth's crust, and it occupies the 73rd place in abundance (4). Theoretical studies predicted that nano-structured bismuth is potentially useful for optical, electrooptic device applications, enhanced thermoelectric feature and catalysts (5). Tellurium (Te) is a semiconductor and is frequently doped with copper, tin, gold or silver. It is

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used in metallurgy as a secondary vulcanizing agent for rubber, in color glass and ceramics, in alloying agent, small amount of Te are add to copper and stainless steel to make them easier to machine and mill. Possible future uses are Te-methionine as heavy-atom label for X-ray studies in proteins and enzymes, Te-agents in cancer drug development, Te-agents as (selective) antibiotics and TeNPs in medicine (6,7).

Alternative-novel, easy, fast and one-step (based on Pulsed Laser Ablation in Liquids medium denoted by PLAL) method for the preparation of pure and stable noble metal versatile NPs in a high ablation rate and size-selected manner with a high concentration, long period of stability, less aggregation, nontoxic and contamination(8). In recent decades, laser ablation of a solid target in a liquid environment has been widely used in preparation of nanomaterials and fabrication of nanostructures. Remarkably, there are many groups that pay attention to this issue in the world, and a large variety of nanomaterials such as metals, metallic alloys, semiconductors, polymers, etc, have been synthesized using laser ablation of solid in liquid. Therefore, laser ablation in liquids has been recognized to be an effective and general route to synthesize nanocrystals and fabricate nanostructures(9). Noble metal $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs are synthesized by using pulsed laser ablation (Q-switched, 1064 nm- Nd: YAG) of Bi and Te metal plates immersed in distilled deionized water (DDW) (10).

Synthesis of nanosized drug particles with tailored physical and chemical properties is of great interest in the development of new pharmaceutical products (11). Emergence of new resistant bacterial strains to current antibiotics has become a serious public health issue, which raised the need to develop new bactericidal materials. However, the phenomenon of enhanced biological activity and certain material changes resulting from nanoparticles is not yet understood fairly. Investigations have shown encouraging results about the activity of different drugs and antimicrobial formulation in the form of nanoparticles (12). The present study was conducted to synthesize bismuth and tellurium nanoparticles by laser ablation of bulk target immersed in liquid environment and study their antibacterial activity against some Gram negative bacteria.

2. Materials and Methods

2.1. Preparation and Characterization of $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs

$\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs were prepared by irradiating a metallic target plates with a thickness of 1mm placed on the bottom of quartz vessel containing DDW with a pulsed laser beam. The ablation was performed with the 1064 nm of Nd: YAG laser

(HUAFEI) operating at 10 Hz repetition rate, with a pulse width of 10 ns at energy set in (500, 600, 700) mJ/pulse for $\text{Bi}_2\text{O}_3\text{NPs}$ and (500, 600, 700) mJ/Pulse for TeNPs with a positive lens having a focal length of 9 cm. The spot size was about 1.5 mm in diameter and the liquid thickness was changed in the range from 2-14 mm to increase the shock wave and it's adjusted by using different dimension of cells. The number of laser shots applied for the metal target was 20 pulses. Atomic Force Microscope (model AA3000, Angstrom Advance Inc., USA) and Transmission Electron Microscope (CM10-PW6020, Philips, Germany) were used to examine the size and size distributions of the metals NPs (10, 13). The NPs concentration was also characterized by Atomic Absorption Spectroscopy (AA-680, shimadzu-Japan).

2.2. Antibacterial Activity of $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs

A stock solution (11.29 ppm) concentration of $\text{Bi}_2\text{O}_3\text{NPs}$ (700 mJ/Pulse) and (10.2 ppm) concentration of TeNPs (700 mJ/Pulse) and then the following concentrations (A=11.29, B=9, C=7, D=5, E=3, F=1) ppm and (G=10.2, H=7, I=5, J=3, K=1, L=0.5) ppm for each $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs, respectively, were prepared by serial dilution from the stock solution with DDW.

The agar well diffusion method was used to detect the effect of the concentrations of both $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs against *Pseudomonas earuginosa*, *Acinetobacter baumannii* and *E.coli* (obtained from Department of Biology, College of Science, Al-Mustansiriyah University). Plates were prepared by spreading approximately 10^5cfu/ml culture broth of each indicator bacterial isolates on nutrient agar surface. The agar plates were left for about 15 min before aseptically dispensing the 50 μl of $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs into the agar wells already bored in the agar plates. The plates were then incubated at 37°C for 18 - 24 h. Zones of inhibition were measured and recorded in millimeter diameter (14).

3. Results and Discussion

Nanotechnology is a new discipline with many applications in fields like biological sciences and medicine. Nanomaterials are applied as coating materials, as well as in treatment and diagnosis. The PLAL process is currently explored as a prospective top-down (dispersion method) strategy of metals NPs preparation (10). This research addresses on preparation of pure noble metals of $\text{Bi}_2\text{O}_3\text{NPs}$ and TeNPs and investigation their antibacterial activity.

The concentrations of both NPs were shown in Table 1 for each pulse energy in liquid, which measured by using atomic absorption spectrophotometer, as a function of standard concentration samples to make the calibration curve. Average

size, of NPs was studied by using AFM and TEM.

Table 1. The concentration (ppm) by AAS and the average diameter (nm) for Bi₂O₃NPs (1, 2, 3) and TeNPs (4, 5, 6).

Energy (mJ)	Average diameter (nm)	Concentration (ppm)	Energy (mJ)	Average diameter (nm)	Concentration (ppm)
1-Bi-700 mJ	75.87	11.29	4-Te-700 mJ	31.99	10.2
2-Bi-600 mJ	89.04	9.3	5-Te-600 mJ	44.76	8.6
3-Bi-500 mJ	76.58	2.0	6-Te-500 mJ	46.53	7.4

Table 2. showed the inhibition zone of Bi₂O₃NPs and TeNPs against *Pseudomonas aeruginosa*, *Acinetobacter baumannii* and *E.coli* as a function of concentrations of Bi₂O₃NPs (1=11.29, 2=9.3, 3=2.0) ppm (in Table 1) and the serial dilution (A=11.29, B=9, C=7, D=5, E=3, F=1) ppm, also for TeNPs (4=10.2, 5=8.6, 6=7.4) ppm (in Table 1) and the serial dilution (G=10.2, H=7, I=5, J=3, K=1, L=0.5) ppm.

Table 2. Antibacterial effect of Bi₂O₃NPs (1, 2, 3) and TeNPs (4, 5, 6) with different dilution against *Pseudomonas aeruginosa*, *Acinetobacter baumannii* and *E.coli*.

Nanoparticles	Inhibition zone diameter (mm) against <i>P. aeruginosa</i>	Inhibition zone diameter (mm) against <i>A. baumannii</i>	Inhibition zone diameter (mm) against <i>E. coli</i>
1	-	-	-
2	-	-	-
3	-	-	-
A	-	-	-
B	-	-	-
C	-	-	-
D	-	-	-
E	-	-	-
F	-	-	-
4	25	20	20
5	25	15	20
6	22	15	16
G	25	19	21
H	-	11	20
I	-	12	18
J	-	-	16
K	-	-	-
L	-	-	-

(-) no inhibition zone.

It can be seen that, the highest effect was achieved when the TeNPs concentration at 10.2 ppm, for each type of bacteria and the inhibition zone increased with increasing the concentration of TeNPs and the inhibition was decreased with dilution of TeNPs. *Pseudomonas aeruginosa* was more sensitive than *Acinetobacter baumannii* and *E. coli*. While, in studying the effect of different concentrations of Bi₂O₃NPs, we can be seen that there is no effect at different concentration against all types of bacteria. Bijan *et al.* (6) reported that Te NPs had good antibacterial activity against *S.aureus* and *S.typhi*.

Gram-negative bacterium exhibit a thin layer of peptidoglycan (about 2–3 nm) between the cytoplasmic membrane and the outer cell wall. Outer membrane of *E. coli* cells is predominantly constructed from tightly packed lipopolysaccharide (LPS) molecules, which provides an effective permeability barrier (15). The overall charge of bacterial cells at biological pH values is negative because of excess number of carboxylic groups, which upon dissociation makes the cell surface negative. The opposite charges of bacteria and nanoparticles are attributed to their adhesion and

bioactivity due to electrostatic forces. It is logical to state that binding of nanoparticles to the bacteria depends on the surface area available for interaction. Nanoparticles have larger surface area available for interactions, which enhances bactericidal effect than the large sized particles; hence they impart cytotoxicity to the microorganisms(16). The mechanism by which the nanoparticles are able to penetrate the bacteria is not understood completely, but studies suggest that when *E. coli* was treated with some nanoparticles, changes took place in its membrane morphology that produced a significant increase in its permeability affecting proper transport through the plasma membrane, leaving the bacterial cells incapable of properly regulating transport through the plasma membrane, resulting into cell death(17). Heavy metals are toxic and react with proteins, therefore they bind protein molecules(18), heavy metals strongly interacts with thiol groups of vital enzymes and inactivates them (19).

Metal depletion causes formation of irregular shaped pits in the outer membrane of bacteria which is caused by progressive release of LPS molecules and membrane proteins

believed that metal binds to functional groups of proteins, resulting in protein denaturation(20). Complete bacterial inhibition depends upon the concentrations of nanoparticles and on the number of bacterial cells, we conclude that TeNPs have an excellent biocidal and potential effect in reducing bacterial growth in practical applications.

The advantages of NPs are their high surface-to-volume ratios, their quantum confinement, and their nanoscale size, which allow more active site to interact with biological systems (4).

4. Conclusion

Antibacterial properties of nanoparticles are attributed to their total surface area, as a larger surface to volume ratio of nanoparticles provides more efficient means for enhanced antibacterial activity against pathogenic bacteria. TeNPs was reducing gram negative bacterial growth.

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