

Research Article

Detection of Phenols from Industrial Effluents Using *Streptomyces* Mediated Gold Nanoparticles

Bi Bi Zainab Mazhari and Dayanand Agsar

A-DBT Research Laboratory, Department of Microbiology, Gulbarga University, Gulbarga 585 106, India

Correspondence should be addressed to Bi Bi Zainab Mazhari; mbzainab@gmail.com

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Extracellular gold nanoparticles synthesized by *Streptomyces tuius* DBZ39 were explored for the detection of phenols in the effluent of fertilizer and distillery industries. An average size of 27–56 nm gold nanoparticles was produced and confirmed by UV-vis absorption spectrum, scanning electron microscopy, and energy dispersive X-ray analysis. In the present investigation visual detection of phenols in the effluent samples by gold nanoparticles is enhanced by sodium sulphate. The detection is achieved successfully within 2 min, with change in color of the effluent samples. Use of biologically originated gold nanoparticles along with salt for the detection of phenols from industrial effluents is a novel approach.

1. Introduction

Phenols of anthropogenic origin exist in the environment due to the activity of the chemical, petrol, tinctorial, or pharmaceutical industries. Phenols are among the most abundant organic impurities penetrating into the aquatic environment as a result of their use in a large number of processes, including petroleum and paper industry and synthesis of plastics and pharmaceuticals [1]. Phenol is most widely used in the organic compounds in existence and is a basic structural unit for a variety of synthetic organic compounds including agricultural chemicals and pesticides. It is naturally found in decaying dead organic matters like rotting vegetables and in coal. Phenol toxicity is related to two main processes, unspecified toxicity related with hydrophobicity of the individual compound and formation of free radicals [2]. Because most phenolic compounds exhibit a high degree of toxicity, they have been included in the list of high priority pollutants by the US Environmental Protection Agency (EPA) and several other countries [3]. The European Union has set the maximum total and individual phenol permitted concentrations in water used for human consumption at 0.5 and 0.1 mg/L, respectively [4]. To evaluate the risks which these compounds pose, a rapid and reliable process for their determination is therefore necessary. Nanoparticles with unique physical and chemical properties are increasingly

being used in different emergent areas. Each of these features allows the researchers to design a novel detection system for the detection of phenol constituents using gold nanoparticles which is significantly advantageous in terms of sensitivity [5], surface bioconjugation, remarkable plasmon resonance, optical properties, chemical stability, and nontoxicity [6]. The use of toxic chemicals in nanoparticles synthesis limits their application. There is a need for development of clean biocompatible, nonhazardous, and eco-friendly methods for the synthesis [7]. Novel and green method for the synthesis of gold nanoparticles was the use of microbial cell.

Among microorganisms, actinomycetes especially are very potential for the synthesis of several nanoparticles with much improved controlled size, shape, and composition of nanoparticles. Recently, the genus *Streptomyces* has been studied as potential producer of certain bioactive molecules with diverse, chemical structures and biological activities. The basic steps for gold nanoparticles biosynthesis include the microbial growth and the metal reduction process, which takes place by intra- or extracellular reduction [8]. In actinomycetes, reduction of metal ions occurs on the surface mycelia along with cytoplasmic membrane leading to the formation of nanoparticles [7, 8].

In the present analysis, change of color of industrial effluent samples treated with enhanced gold nanoparticles was used for the detection of phenols. Hence, the work

was proposed for the detection of phenols from different industrial effluents using *Streptomyces* mediated gold nanoparticles.

2. Materials and Methods

2.1. Biosynthesis of Extracellular Gold Nanoparticles. An isolate of *Streptomyces tuius* DBZ39 obtained from limestone quarry soil earlier [9] was employed for the synthesis of extracellular gold nanoparticles, as per the standard protocol prescribed by Ahmad et al. [8] and Shahverdi et al. [10]. A loopful of three-day-old culture of *Streptomyces tuius* DBZ39 was inoculated into starch casein broth containing starch, 1 g; casein, 0.003 g; KH_2PO_4 , 2.0 g; KNO_3 , 2.0 g; NaCl , 2.0 g; MgSO_4 , 0.002 g; FeSO_4 , 0.001 g; and CaCO_3 , 0.001 g and incubated at 40°C for 5 days on rotatory shaker (200 rpm). After incubation, the broth culture was centrifuged at 8000 rpm. The biomass obtained was suspended in aurum (AuCl_4) solution and kept for incubation at 37°C on shaker (200 rpm) for three days. The gold nanoparticles synthesized in the solution were visually confirmed by the development of deep purple color and UV-visible absorption spectrum in the range of 500–550 nm. The gold nanoparticles produced were characterized by scanning electron microscopy [11, 12] for their size, shape, and dispersion and EDAX for elemental confirmation of gold in the nanosolution as per the standard protocols.

2.2. Enhancement of Gold Nanoparticles. Effect of salts such as Na_2SO_4 , CuSO_4 , NaCl , and K_2HPO_4 on the enhanced detection of phenols was studied [13]. Enhancement of gold nanoparticles with salts increased the color intensity in the detection process. The UV-vis absorption spectra of the gold nanoparticles + standard phenol and gold nanoparticles along with salt + standard phenol were recorded at similar wavelength. The salt which enhances the detection of phenols with higher color intensity along with gold nanoparticles was used in the further study for the detection of phenols from the industrial effluent samples.

2.3. Phenol Detection. Fertilizers and distillery industrial effluents were collected and stored at room temperature overnight for the settling of precipitate. After settling the sample was filtered and used to detect the presence of phenols by gold nanoparticles enhanced with salt as per the protocol prescribed by Lisha et al. [13, 14]. 1.0 mL of salt solution (0.5%) was mixed with 0.5 mL (1 mM) of gold nanosolution. The effluent sample of fertilizers and distillery industries in the range of 0.5, 1.0, 1.5, and 2.0 mL was added to the above mixture and the color change was observed visually. A blank was prepared with salt and gold nanosolution.

3. Results and Discussion

3.1. Biosynthesis of Gold Nanoparticles. Actinomycetes are known to produce highly diverse bioactive compounds [15–18]. *Streptomyces* is a predominant genus among actinomycetes for the synthesis of variety of bioactive compounds.

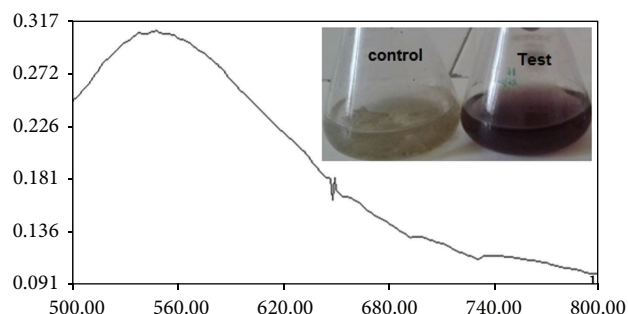


FIGURE 1: Visual observation and UV-vis absorbance spectra of gold nanoparticles synthesized by *Streptomyces tuius* DBZ39.

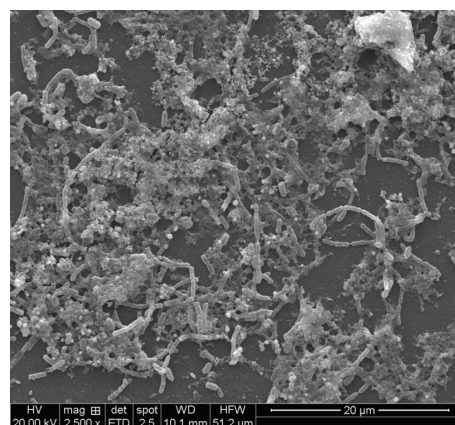


FIGURE 2: Scanning electron micrograph of gold nanoparticles synthesized by *Streptomyces tuius* DBZ39.

In the present investigation, extracellular gold nanoparticles were synthesized by *Streptomyces tuius* DBZ39. Figure 1 reveals the presence of gold nanoparticles in aurum solution. The development of deep purple color exhibits the presence of gold nanoparticles, when compared to the test solution, which is colorless. A higher absorption peak (Figure 1) in UV-visible range of 500–550 nm confirms the synthesis of extracellular gold nanoparticles by *Streptomyces tuius* DBZ39.

Gold nanoparticles synthesized by biological sources have been investigated extensively in recent years because of their potential applications. Gold nanoparticles exhibit intriguing properties compared to metallic gold which imply their tremendous biomedical applications [19–22]. Highly regulated composition including size, shape, and dispersion of gold nanoparticles would exhibit varied biological applications. Scanning electron microscopy (SEM), Transmission Electron Microscopy (TEM), EDAX, and X-ray diffraction (XRD) are the important analytical techniques generally employed to understand size, shape, and dispersion as characteristic properties of gold nanoparticles [23, 24]. In the present investigation, gold nanoparticles synthesized by *Streptomyces tuius* DBZ39 were characterized by SEM analysis. Scanning (Figure 2) revealed average size of 27–56 nm which is dispersed uniformly. The spectrum of EDAX (Figure 3) confirms the purity of gold nanoparticles.

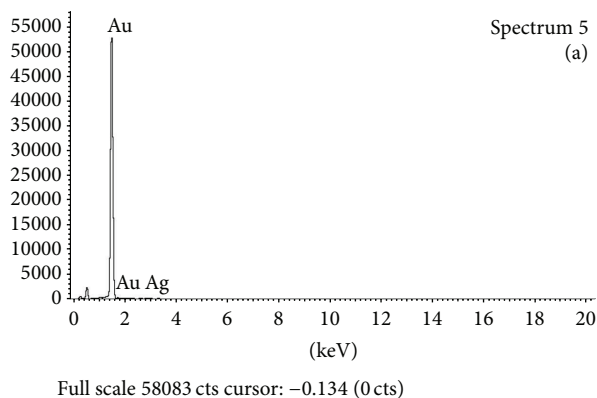


FIGURE 3: EDAX features of gold nanoparticles synthesized by *Streptomyces tuius* DBZ39.

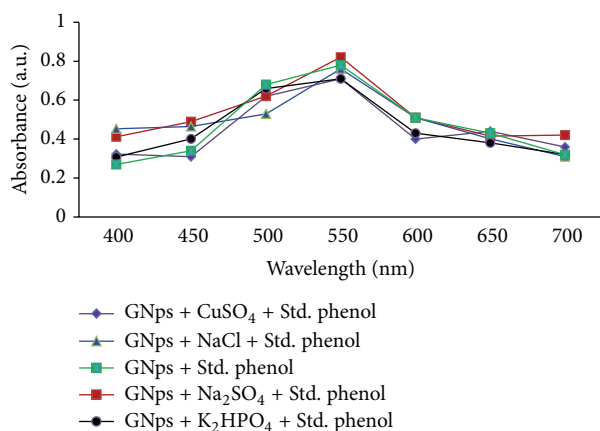


FIGURE 4: Effect of different salts on gold nanoparticles for enhanced detection of phenol.

3.2. Enhancement of Gold Nanoparticles. Gold nanoparticles synthesized were enhanced by salts for detecting the phenols. When salt was added to gold nanoparticles it enhanced the color intensity and time required for detecting the phenol by aggregation of gold nanoparticles. Salts like Na_2SO_4 , K_2HPO_4 , CuSO_4 , and NaCl were used for enhancement. UV-vis spectra of the gold nanoparticles before and after the addition of salts and standard phenol showed significant variation. Figure 4 shows the UV-vis spectra of gold nanoparticles + standard phenol (100 mg) and gold nanoparticles + salts + standard phenol mixture (100 mg). UV-vis spectrum of gold nanoparticles + standard phenol exhibits characteristic absorption at 550 nm. The gold nanoparticles + Na_2SO_4 + standard phenol mixture showed the highest absorption spectrum compared to gold nanoparticles + standard phenol mixture. This may be due to the aggregation of gold nanoparticles by the adsorption of cations on the surface [13] by sodium sulphate (Na_2SO_4). The response of other salts, that is, K_2HPO_4 , CuSO_4 , and NaCl , to color change is not as good as compared to Na_2SO_4 . By adding K_2HPO_4 , CuSO_4 , and NaCl , the time required for the color change is longer (5 min). Similarly, the addition of Na_2SO_4 detects the phenols with higher color intensity and

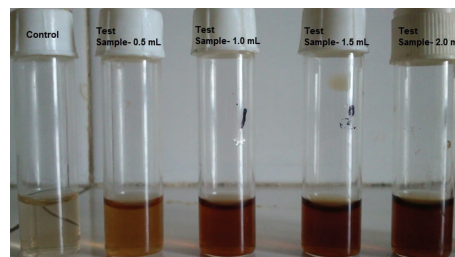


FIGURE 5: Visual observation for the detection of phenol in fertilizer industrial effluents.

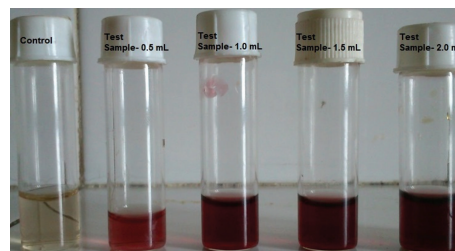


FIGURE 6: Visual observation for the detection of phenol in distillery industrial effluents.

less time (2 min). Thus sodium sulphate was used for further detection method with gold nanoparticles.

3.3. Phenol Detection. Industrial effluents collected from fertilizers and distillery industries were treated with sodium sulphate enhanced gold nanoparticles in order to accomplish greater detection of phenol by salt aggregation principle. The color intensities of the treated samples were quantified visually (Figures 5 and 6). The intensity of color generated during 2 min of the reaction period was proportional to the presence of phenol in samples. Intensity of color increased, as the concentration of effluent sample was increased, that is, 0.5 to 2.0 mL. Figures 5 and 6 reveal the color intensity pattern for effluent samples of fertilizer (Figure 5) and distillery (Figure 6) industries at different concentration. The results exhibited that detection of phenol by gold nanoparticles was quite good for qualitative detection of phenols from effluents samples of both fertilizer and distillery industry.

Industrial effluent invariably containing various phenol contents is known for causing water pollution and is a major problem in developing countries. Several basic and advanced physicochemical methods exist to monitor the presence of pesticides in polluted water samples [25]. Advanced techniques such as liquid and gas chromatography and mass photometry are highly sensitive, time consuming, and expensive and have greater limitations for the on-site or in-field analytical applications [26, 27]. Gold nanoparticles were used for extraction of polycyclic aromatic hydrocarbons in drinking water [28]. Liu and Mattiasson reviewed the use of nanoparticles for detection and remediation of environmental pollutants [29]. Nanoparticles are frequently employed to aid the detection of environmental pollutants as a preconcentration medium or analytical sensor [30].

These applications take advantage of the unique features of nanoparticles such as their large surface areas and their unique photochemical, electronic, or magnetic properties. The most important feature of the investigation was using *Streptomyces* gold nanoparticles for the detection of phenol without any electrodes underlying with electrochemical principles or so-called biosensors. According to literature no reports were available for the detection of phenol by biosynthesized gold nanoparticles using salt enhanced method, which is an important and significant observation of the present investigation.

4. Conclusions

In the present investigation, *Streptomyces tuius* DBZ39 was employed for the synthesis of gold nanoparticles, aiming at the detection of phenol present in different industrial effluents. A simple method based on enhancement of gold nanoparticles by salt for the rapid detection of phenol from industrial effluents was effectively confirmed, which is a new criterion. Visual detection of phenol within 2 min was an important and significant observation. The developed method is very simple and response is rapid and visually observable. This method can be effectively utilized for on-site detecting of phenol qualitatively.

Competing Interests

The authors declare that there is no conflict of interests.

Acknowledgments

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