Journal of Photochemistry & Photobiology, B: Biology 153 (2015) 261–266

Contents lists available at ScienceDirect

Journal of Photochemistry & Photobiology, B: Biology

journal homepage: www.elsevier.com/locate/jpb

Enhanced visible light photocatalytic inactivation of *Escherichia coli* using silver nanoparticles as photocatalyst

Kamran Tahir [a,b], Sadia Nazir [b], Baoshan Li [a,*], Arif Ullah Khan [a], Zia Ul Haq Khan [a], Aftab Ahmad [a], Qudrat Ullah Khan [a], Yunchen Zhao [a]

[a] State Key Laboratory of Chemical Resource Engineering, Beijing University of Chemical Technology, Beijing 100029, PR China
[b] Institute of Chemical Sciences, Gomal University D.I.Khan, KP, Pakistan

ARTICLE INFO

Article history:
Received 23 June 2015
Received in revised form 11 September 2015
Accepted 14 September 2015
Available online 16 September 2015

Keywords:
Cirsium arvense
Green synthesis
Nanoparticles
Antibacterial activity
Escherichia coli

ABSTRACT

The silver nanoparticles (AgNPs) were green synthesized using *Cirsium arvense* plant extract as a reducing and stabilizing agent, with superior photo inactivation activity against *Escherichia coli* (*E. coli*). The synthesized AgNPs had crystalline structure and were characterized by UV–vis spectroscopy, XRD, HRTEM, SEM, EDX and FT-IR. The formation of nanoparticles was observed at different pH and different plant extract concentrations and it was found that at higher pH (pH > 6) and at lower concentration (10 mL), the reducing and stabilizing efficiency of plant extract was increased. The synthesized AgNPs had small size (<15 nm) and spherical shape. The AgNPs were evaluated for antibacterial activity against *E. coli*. Before transferring it to antibacterial activity, it was placed under visible light for 120 min. The same experiment was performed in dark as a control medium. The photo irradiated AgNPs were observed to be more effective against *E. coli*. The results showed, that the diameter of zone of inhibition of visible light irradiated AgNPs against *E. coli* was 23 (±0.5) mm and in dark was 11 (±0.4) mm.

1. Introduction

Natural sources i.e. plants have the strong ability to reduce metal ions into metal nanoparticles [1,2]. AgNPs are multi-applicative and widely used in catalysis, electrical conductance [3,4] and antimicrobial activity [5–7]. The small size, spherical shape and dispersion of metal nanoparticles play an important role in controlling the physical and chemical properties. The catalytic and biological activities of Ag nanoparticles are greatly depending upon specific surface area of Ag nanoparticles. Various methods have been used to prepare nanoparticles such as laser ablation [8], UV irradiation reduction [9], microwave processing [10], electrolysis method [11], thermal decomposition [12], ion implantation [13] and chemical reduction [14,15]. However chemical reduction is the simplest process and has the capability of synthesizing highly dispersed, uniform and small size nanoparticles [16]. But most of the chemicals used in chemical reduction method are toxic, making nanoparticles unsuitable for biological purposes [16–18]. Recently the most important nanoparticles i.e. silver [19], gold [20] and platinum [21] were prepared from noble metals by green method. These nanoparticles were extensively used in soaps, shampoos, detergents, toothpaste and cosmetic products as well as pharmaceutical applications. Green synthesis is an eco-friendly, easily accessible and important method to obtain nanoparticles with unique structure, small size and high dispersion. Green synthetic method is a convenient way to synthesize nanoparticles as compared to other conventional methods i.e. chemical and physical methods. Various microorganisms including fungi [22–24], bacteria [25,26], and plant leaves [27] have been used for biologically synthesized Ag and Au nanoparticles. Among all metallic nanoparticles, silver has got a specific position due to its high activity, low cost, non toxic, and high electrical properties which make AgNPs an important agent in various fields like antimicrobial, catalysis and sensor.

In this work, we prepared photoactive AgNPs by green way using *Cirsium arvense* plant extract as reducing and stabilizing source. The AgNPs were evaluated for photo inhibition of *Escherichia coli* under visible light irradiations. The same experiment was also performed in dark as a control. The results showed that irradiated AgNPs have higher antibacterial activity than in dark. To the best of our knowledge it is the first report about photo inactivation of *E. coli* by green synthesized AgNPs.

2. Materials and Method

A 100 g of *C. arvense* plant was collected from District Bannu, Pakistan and washed with distilled water to remove impurities. The plant was shade dried, and grinded it to fine powder. Then 10 g of it was mixed with 100 mL of distilled water and stirred at 80 °C for 3 h and then filtered to get the extract.

For AgNPs synthesis, 10 mL of plant extract was added to 70 mL of 3×10^{-2} M aqueous solution of silver nitrate in 150 mL beaker. As a

* Corresponding author.
E-mail address: bsli@mail.buct.edu.cn (B. Li).

http://dx.doi.org/10.1016/j.jphotobiol.2015.09.015
1011-1344/

visual examination, a color change occurs from yellow to brown within 5 min. The AgNPs suspension thus obtained was purified with the help of repeated centrifugation at 10,000 rpm for 15 min and then the AgNPs were freeze dried.

2.1. Microorganisms

Bacterial species, *E. coli* was used in antimicrobial assay. Strains were obtained from State Key Laboratory of Chemical Resource Engineering, Beijing University of Chemical Technology, Beijing 100029, Laboratory No. 1102, PR China, where these were identified and characterized. These strains were maintained on agar slants at 4 °C for antimicrobial test. Microorganisms were incubated overnight at 37 °C in Mueller–Hinton Broth (Oxoid) at pH 7.2.

2.2. Screening for Antibacterial Activity by Agar Well Diffusion Method

For determination of antibacterial activity of AgNPs, the agar well diffusion method was carried out [28]. All bacterial strains were grown in nutrient broth at 37 °C for 24 h incubated till turbidity became equal to McFarland 0.5 turbidity standard. Using a sterile swab, the inocula of the respective bacteria were streaked on to the Muller Hinton agar (Oxoid) plates in order to make sure a uniform thick lawn of growth following incubation. Using sterile cork borer, wells of 6 mm in diameter were formed on to nutrient agar plates. On the other hand 1 mg of AgNPs were kept in 1 mL water under the visible light for 120 min and then wells were filled with 50 μL of the irradiated AgNPs and the plates were then kept to stay for 2 h at 25 °C. The same experiment was performed in dark as a control medium. After that the plates were incubated at 37 °C for 24 h and the resultant diameters of zones of inhibition were measured carefully.

2.3. Determination of Minimum Inhibitory Concentration (MIC)

In order to determine MIC of the AgNPs, serial dilution protocol was used. With 1 mL of different concentrations of irradiated AgNPs in sterilized test tubes was mixed with 1 mL of *E. coli* solution having turbidity of 0.5 McFarland turbidity standards. After mixing, these test tubes were placed in an incubator at 37 °C for 24 h. A test tube having only growth media and bacteria was used as a control. 1 mg/mL to 0.125 mg/mL concentrations of AgNPs were used in these tests. The minimum concentration of the compound, which inhibited the growth of the respective organism, was considered as MIC. The assay was carried out in triplicate.

3. Result and Discussion

3.1. Characterization

The biogenic Ag nanoparticles were studied regularly by scanning the aliquot sample in the wavelength range of 200–800 nm and recorded the absorption maxima in Shimadzu UV-2400 spectrophotometer at a resolution of 1 nm. The wide angle X-ray diffraction (XRD) measurements were carried out on a Rigaku D/Max 2500 VBZ + /PC diffractometer using CuK_a radiation at a scanning rate of 20 min^{-1} with an operating voltage of 40 kV and a current of 200 mA. High-resolution transmission electron microscopy (HRTEM) on a JEM-3010 microscope with an accelerating voltage of 200 kV was used to examine the morphologies and size of the nanoparticles. Infrared (IR) spectrum of the extract was obtained using the KBr pellet technique on an ABB MB3000 spectrophotometer where it was scanned between 4000 and 500 cm^{-1} at a resolution of 4 cm^{-1} in transmittance mode.

3.1.1. UV–Vis Spectral Analysis

UV–vis spectroscopy is the most suitable method for characterization of Ag nanoparticles. The phyto-reduction of Ag ions into Ag

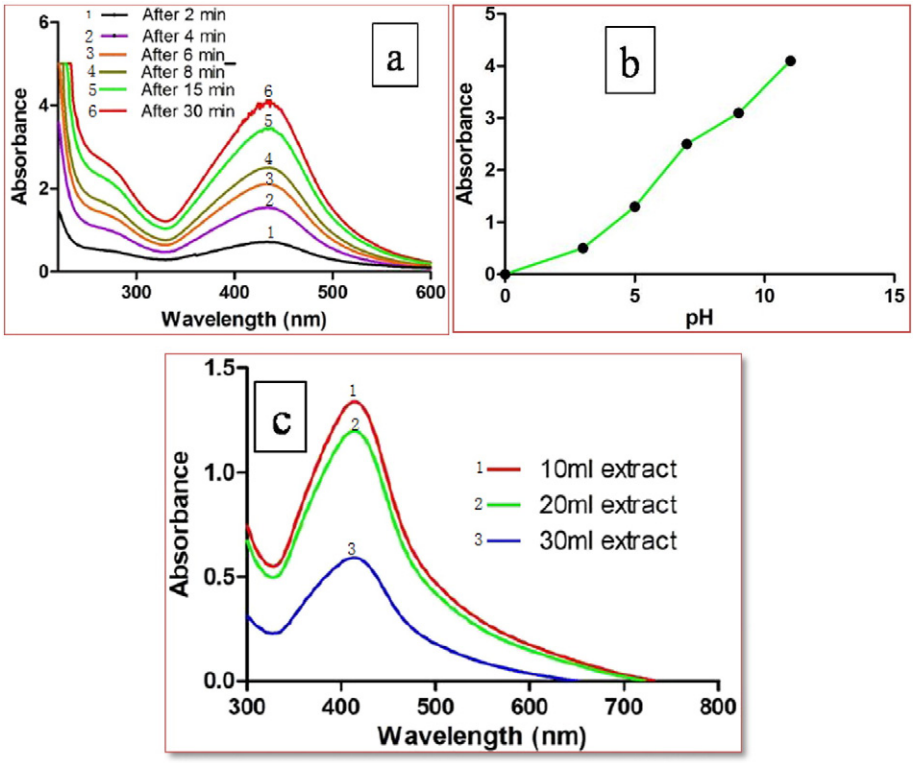

Fig. 1. Absorption spectra and plot of formation of AgNPs examined (a) at different time (b) at different pH and (c) at different extract concentration.