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Green synthesis of colloidal silver nanoparticles through electrochemical method and their antibacterial activity

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ABSTRACT

This paper presents a facile, and ecofriendly method to prepare colloidal silver nanoparticles (AgNPs) at room temperature using bulk silver bars, bi-distilled water, trisodium citrate, and a direct-current voltage source. AgNP formation was confirmed and characterized by ultraviolet–visible spectroscopy, transmission electron microscopy and X-ray diffraction. Their antibacterial activity was examined by disc diffusion and minimum inhibitory concentration (MIC) techniques against three bacterial strains, including the Gram-positive bacteria *Staphylococcus aureus* and the Gram-negative bacteria *Escherichia coli* and *Pseudomonas aeruginosa*. Results showed that the size of AgNPs formed was about 19.7 ± 4.3 nm with nearly spherical shape and high purity. The antibacterial activity of as-prepared AgNPs was determined through zones of inhibition against the growth of three bacterial strains on agar. MIC analysis showed that *S. aureus* had resistance to AgNPs that was about 2–3 times higher than those of *E. coli* and *P. aeruginosa*, and that resistance ability depended on the bacterial concentrations inoculated. This work revealed the effective method of synthesizing a large quantity of colloidal AgNPs with high antibacterial effectiveness and potential application in different fields.

1. Introduction

Silver nanoparticles (AgNPs) are well known by their unique optical, electrical, and biological properties, especially high antibacterial activity [1,2]. In recent years, AgNPs have attracted intensive interest for different applications, such as in wound dressings [3], catheters [4], disinfectant sprays [5], food containers [6], paints [7], and textile products [8]. Several methods have been introduced for the synthesis of AgNPs, such as physical [9–11], chemical [12,13], and biological approaches [14,15]. Each of them has different advantages and disadvantages in terms of cost, time consumption, stability, size distribution, and application purpose [16].

The physical approach can produce large quantities of AgNPs from bulk silver in a single process [9–11], but the cost of the required equipment should be considered. Recently, Slepička et al. [17] described an inexpensive method for the preparation of stable gold and silver nanoparticles by direct sputtering of metal targets into polyethylene glycol solution. In fact, the chemical reduction is the most common method employed to synthesize AgNPs because of the convenience and simple equipment [18,19]. Nonetheless, the eco-unfriendly and expensive reagents, as well as the pH control and purity of synthesized AgNPs, should be carefully considered. The biological approach is an environmentally friendly and low-cost technique [20]. This approach uses naturally reducing agents such as microorganism (bacteria or fungi) [21,22], polysaccharides [23], or plants extracts [24] to obtain AgNPs from silver salts. The main drawback of this method is the difficulty of obtaining a large quantity and uncontrollable size of AgNPs [16].

Several electrochemical approaches were also introduced as simple and cost-effective solution to prepare large quantities of colloidal AgNPs, but their antibacterial effectiveness have not been demonstrated [25,26]. Actually, the antibacterial activity of AgNPs is widely recognized, but their real antibacterial effectiveness is still unclear [16]. It depends on the synthesis [27], shape and size [28], surfactants, strains and concentration of bacteria, and testing methods [29].

In this paper, we describe a facile electrochemical method for green synthesis of colloidal AgNPs at room temperature using bulk silver, bi-distilled water, trisodium citrate, and an externally applied direct-current (DC) voltage source. This method could obtain a large quantity of colloidal AgNPs with high antibacterial

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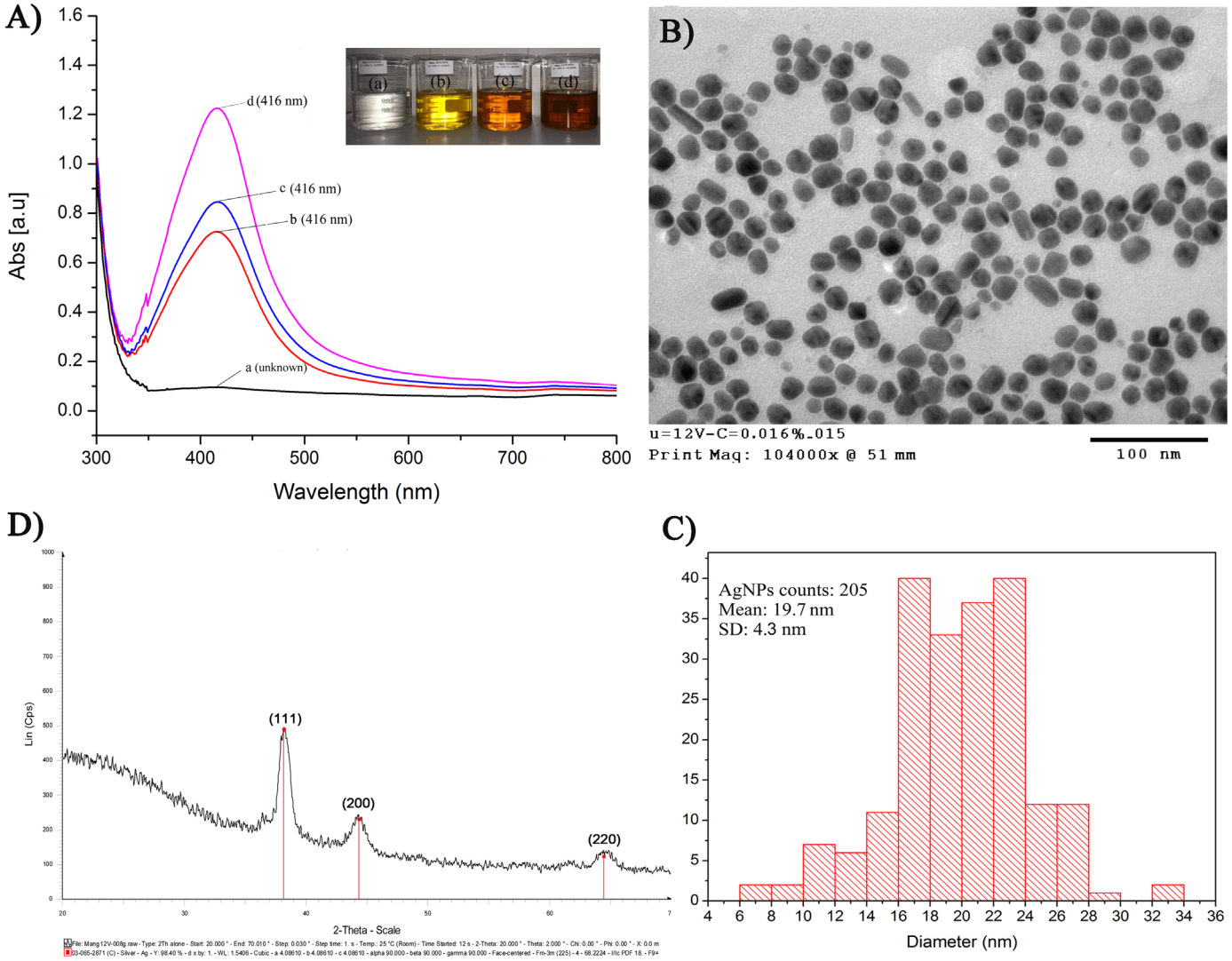

Fig. 1. (A) UV–vis spectra of AgNPs synthesized corresponding to different concentrations of trisodium citrate: 0.002% (a), 0.010% (b), 0.016% (c), and 0.024%; (B) TEM image of AgNPs synthesized at 0.016% of trisodium citrate; (C) Histogram for particle-size distribution (16–24 nm); and (D) X-ray diffraction pattern of as-synthesized AgNPs.

effectiveness in a single process, and as-synthesized AgNPs could be applied for different applications in biomedicine.

2. Experimental

2.1. Reagents

Two bulk silver bars (99.999% purity) with dimensions of 150 mm × 10 mm × 0.5 mm (L × W × T) used as electrodes, were purchased from a domestic jewelry company in Hanoi, Vietnam. Trisodium citrate ($Na_3C_6H_5O_7$) was purchased from Sigma–Aldrich. All other chemicals were analytical grade. A DC voltage source of 12 V was supplied to silver bars.

Three bacterial strains, namely, *Staphylococcus aureus* (ATCC 25922), *Escherichia coli* (ATCC 25923), and *Pseudomonas aeruginosa* (ATCC 27853) were provided by the Dept. of Bacteriology, National Institute of Hygiene and Epidemiology, Vietnam.

2.2. Preparation of colloidal AgNPs

Two silver bars were connected to the DC voltage source in parallel as electrodes in the 500 mL glass beaker placed on the magnetic stirrer. The distance between the two electrodes was 6.5 cm, and bi-distilled water was poured until 7 cm of silver electrodes. Then, 0.016% (wt%) of trisodium citrate was added, and magnetic stirring was performed for 15 min. A DC voltage of 12 V was then supplied to the silver bars for 2 h under magnetic stirring and at room temperature. Afterwards, the silver electrodes were slightly removed to avoid the dispersion of residue produced during synthesis; the stirring process was still remained for 30 min. The beaker containing the solution was kept for 24 h in darkness to complete AgNP formation. The AgNPs solution was decanted and slightly centrifuged at 2000 rpm for 10 min to remove the sediments before use and measurements.

2.3. Characterizations of AgNPs

Ultraviolet–visible (UV–vis) spectrometry (HP 8453 spectrophotometer) was performed within the wavelength range of 300–800 nm to confirm AgNP formation. AgNP morphology and size were observed by transmission electron microscopy (TEM; JEM 1010, JEOL) at 80 kV. The particle size distributions of AgNPs were calculated from TEM images using ImageJ 1.46 program.

The crystal structure and purity of as-synthesized AgNPs were investigated by X-ray diffractometer (D8-Advance, Bruker).