Research Article

Biosynthesis of Silver and Gold Nanoparticles Using Aqueous Extract of *Codonopsis pilosula* Roots for Antibacterial and Catalytic Applications

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In this study, biogenic silver nanoparticles (AgNPs) and gold nanoparticles (AuNPs) were synthesized by a green approach using an aqueous extract from *Codonopsis pilosula* (CP) roots as a reducing and stabilizing agent. The formation of CP-AgNPs and CP-AuNPs was confirmed and optimized by UV-Vis spectroscopy. The CP-AgNPs and CP-AuNPs obtained under optimum conditions of metal ion concentration, reaction temperature, and reaction time were characterized by high-resolution transition electron microscopy (HR-TEM), selected area electron diffraction (SAED) analysis, field-emission scan electron microscopy (FE-SEM), powder X-ray diffraction (XRD) analysis, Fourier transform infrared (FTIR) spectroscopy, dispersive X-ray spectroscopy (EDX), and dynamic light scattering (DLS) method. It has been found that the biosynthesized CP-AgNPs and CP-AuNPs were formed in spherical shape with an average size of 10 ± 2.5 nm and 20 ± 3.2 nm, respectively. The biosynthesized metallic nanoparticles exhibited selective bacterial activity against three bacterial strains including two Gram-positive bacteria of *Bacillus subtilis* and *Staphylococcus aureus* and one Gram-negative bacteria of *Escherichia coli*. Meanwhile, there was no antibacterial activity detected toward Gram-negative *Salmonella enteritidis*. CP-AgNPs and CP-AuNPs also manifested an excellent catalytic performance in the reduction of 1,4-dinitrobenzene, 2-nitrophenol, 3-nitrophenol, and 4-nitrophenol.

1. Introduction

In recent years, there has been a growing interest in the synthesis of metal nanoparticles (MNPs) such as silver nanoparticles (AgNPs) and gold nanoparticles (AuNPs) due to their useful properties for applications in different areas of medicine, biology, catalysis, and antibacterial [1–4]. Along with the rapid development of nanotechnology, several promising approaches were utilized to synthesize AgNPs and AuNPs [5–7]. Owing to the simplicity and rapid operation, chemical reduction methods using commercial chemicals such as

hydrazine [8], sodium borohydride [9], ascorbic acid [10], and ethylene glycol [11] are widely applied to convert gold and silver ions into metals at nanoscale. However, these methods have a major drawback related to the toxicity originated from the excess amount of the used chemicals that can affect the quality of nanoproducts. Recently, green approaches have been preferred to use. Among them, the way of using extracts of plants as reducing agents is quite popular, especially in developing countries, with several advantages such as high efficiency and potential for practical applications [12–14]. As reported in literatures [15–18],

different parts of plants such as leaves, stems, roots, tubers, and flowers that contain a high amount of bioactive molecules with water-soluble polyol components responsible in reduction and stabilization of biogenic AgNPs and AuNPs were deployed already for the synthesis.

The biogenic MNPs are well known as effective catalysts for the complete degradation of toxic effluents and hydrogenation of derivatives based on nitroaromatic compounds. Among them, 4-nitrophenol (4-NP), 3-nitrophenol (3-NP), 2-nitrophenol (2-NP), and 1,4-dinitrobenzene (1,4-DNB) are dangerous pollutants contained in industrial wastewater that is discharged mainly from petrochemical refining, pesticides, fertilizer production, and dyes-related manufacturing activities [19]. The catalytic reduction of the mentioned above nitroaromatic compounds using AgNPs and AuNPs in the role of catalysts has been widely studied [20]. Furthermore, it is determined that the biosynthesized AgNPs and AuNPs at nanosize ranged from about 6 up to 100 nm also demonstrated a strong antibacterial activity against various microorganisms as *Escherichia coli* (*E. coli*), *Staphylococcus aureus* (*S. aureus*), *Bacillus subtilis* (*B. subtilis*), *Salmonella typhimurium* (*S. typhimurium*) [21, 22], *Streptococcus pyogenes* [23], *Pantoea agglomerans*, *Staphylococcus* sp., *Klebsiella* sp., and *Rahnella* sp. bacteria [24].

Codonopsis pilosula (Franch.) Nannf. (CP) belonging to Campanulaceae family is a perennial herb grows naturally in mountains of Vietnam, China, and India with bellflower and wire stem. Commonly, the extracts of CP roots were used as a traditional medicine for health promotion, prevention, and treatment of many diseases [25]. The main chemical constituents of CP roots include phytosteroids, sesquiterpenes, triterpenes, alkaloids, alkyl alcohol glycosides, phenylpropanoid glycosides, polyacetylene glycosides, and polysaccharides that could be an excellent source for synthesizing AgNPs and AuNPs [26, 27].

In this study, the aqueous extract of CP roots was used as a reducing and stabilizing agent simultaneously for the biosynthesis of AgNPs and AuNPs. The biosynthesized MNPs were studied for antimicrobial activity toward four bacterial strains including two Gram-positive bacteria (*B. subtilis* and *S. aureus*) and two Gram-negative bacteria (*Salmonella Enteritidis* (*S. Enteritidis*) and *E. Coli*), and for catalytic degradation of 1,4-DNB, 2-NP, 3-NP, and 4NP in aqueous medium.

2. Materials and Methods

2.1. Materials and Chemicals. All chemicals used were of analytical grade and utilized without further purification. Silver nitrate (AgNO_3) and hydrogen tetrachloroaurate(III) hydrate ($\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$) were purchased from Acros (Belgium). Sodium tetrahydridoborate (NaBH_4), 1,4-dinitrobenzene ($\text{C}_6\text{H}_4\text{N}_2\text{O}_4$), 2-nitrophenol ($\text{C}_6\text{H}_5\text{NO}_3$), 3-nitrophenol ($\text{C}_6\text{H}_5\text{NO}_3$), and 4-nitrophenol ($\text{C}_6\text{H}_5\text{NO}_3$) were supplied by Merck (India). CP roots were collected from mountainous province Gialai, Vietnam. Four bacterial strains including two Gram-positive bacteria (*B. subtilis* and *S. aureus*) and two Gram-negative bacteria (*S. Enteritidis* and *E. Coli*) were provided by the Department of Biotechnology, Institute of

Food and Biotechnology, Industrial University of Ho Chi Minh City, Vietnam.

2.2. Preparation of CP Aqueous Extract. The dried CP roots were finely ground up powder using an electronic blender. The CP root powder (5 g) was boiled in distilled water (300 mL) with reflux for 1 h. The obtained mixture was filtered with Whatman filter paper No.1 to remove the solid, and the extract was stored in a refrigerator at 4–10°C for further experiments.

2.3. Synthesis of Biogenic AuNPs and AgNPs. Synthesis of biogenic AuNPs and AgNPs was performed with HAuCl_4 , AgNO_3 solutions, and aqueous CP extract. Briefly, 10 mL of CP extract was mixed with 10 mL of metallic ion solutions under vigorous stirring in the dark. The change in the solution color after reactions complete acts as a visual sign for the success of the synthesis process. Factors that affect the synthesis process, such as concentration (0.5–2 mmol/L), temperature (60–100°C), and time (15–180 min) were also investigated to determine the optimal conditions using UV-Vis measurements on an Evolution 300 UV-Vis spectrophotometer with characterized maximum absorption peaks of AgNPs and AuNPs at around 420 and 540 nm, respectively. The obtained MNPs under optimal conditions were centrifuged, dried, and then used to study their physico-chemical characteristics and antimicrobial and catalytic activities. The procedure for the biosynthesis of CP-AgNPs and CP-AuNPs with their applications can be illustrated in Figure 1.

2.4. Characterization of Biogenic AgNPs and AuNPs Nanoparticles. Fourier transform infrared spectroscopy (FTIR) in the range of 4000–500 cm^{-1} on a Bruker Tensor 27 (Germany) was applied to detect covalent bonds of possible functional groups presented in the dried CP extract and powdered MNPs. Powder X-ray diffraction (XRD) analysis on a Shimadzu 6100 X-ray diffractometer (Japan) operating at the voltage of 40 kV, the current of 30 mA with $\text{CuK}\alpha$ radiation at the wavelength of 1.5406 nm, scanning speed of 0.05°/s and step size of 0.02° in the range 2θ from 10° to 80° was used to determine the crystalline structure and composition of MNPs. The morphology of the MNPs in colloidal solution was determined by transmission electron microscope (TEM) and high-resolution transmission electron microscope (HR-TEM) on a JEOL JEM-2100 (Japan) at an accelerated voltage of 120 and 200 kV, respectively. The selected area electron diffraction pattern (SAED) of the nanoparticles was also recorded. Morphology of biosynthesized MNPs in the aggregation form after centrifugation was also examined by field-emission scanning electron microscopy (FE-SEM) on a Hitachi S-4800 HI-9057-0006 (Japan) at an accelerating voltage of 10 kV. A Horiba EMAX Energy EX-400 analyzer (Japan) was used to perform energy dispersive X-ray spectroscopy (EDX) for the determination of the chemical elemental composition of the powder nanoparticles. Finally, the dynamic diameter of MNPs in colloidal solution was examined by a Horiba SZ-100 (Japan) using dynamic light scattering (DLS) technology.

FIGURE 1: Schematic illustration for biosynthesis of CP-AgNPs and CP-AuNPs with their applications.

2.5. Antibacterial Activity. Antibacterial performance of the CP-AgNPs and CP-AuNPs in the form of optimized stable colloidal solution was studied by the agar disk diffusion method. The standard antibiotic ampicillin ($0.1 \mu\text{g/mL}$) was used as a positive control, while the aqueous CP-extract was used as a negative control. The used concentrations of MNPs were of 80, 40, 20, 10, 5 ppm for CP-AgNPs, and 120, 60, 30, 15, 7.5 ppm for CP-AuNPs. In the study, four bacterial strains including two Gram-positive bacteria (*B. subtilis* and *S. aureus*) and two Gram-negative bacteria (*S. Enteritidis* and *E. Coli*) were applied to evaluate the antibacterial activity of the biosynthesized CP-AgNPs and CP-AuNPs. Briefly, aliquots ($50 \mu\text{L}$) of CP-AgNPs and CP-AuNPs suspension were put into 6 mm-diameter paper disks on Petri plates with the bacterial culture of brain-heart infusion ($100 \mu\text{L}$, 10^6CFU/mL) by Mueller Hinton agar. The plates were kept at 37°C for 24 h, and the antibacterial activity was determined via the inhibition zone diameter of tested bacteria.

2.6. Catalytic Activity of CP-AgNPs and CP-AuNPs. It has been reported that AgNPs and AuNPs exhibit strong catalytic activity for the complete hydrogenation of toxic nitrophenolic compounds to unarmful substances of respective aminophenols [28, 29]. The catalytic performance of CP-AgNPs and CP-AuNPs was investigated via reduction reaction of contaminated organic substances (1,4-DNB, 2-NP, 3-NP, and 4-NP) using NaBH_4 solution as reducing agent in a cuvette at room temperature. The pollutants (2.5 mL of 0.1 mmol/L) was mixed with the excess amount of NaBH_4 (0.5 mL of 0.1 mol/L) and CP-AgNPs and CP-AuNPs (3 mg) added then. After the reaction completed, MNPs were recovered by centrifugation, washed thoroughly with ethanol for reuse. The catalytic performance and kinetics were evaluated using UV-Vis spectroscopy at the wavelength of 380, 410, 390, and 400 nm for 2-NP, 3-NP, 4-NP, and 1,4-DNB, respectively. In the context, the amount of NaBH_4 was used far beyond the concentration of pollutants, so the concentra-

tion of NaBH_4 could be considered as a constant during the reaction. In this regard, the reaction could be pseudo-first-order one which the kinetics described by the linear equation $\ln (At) = -kt + \ln (Ao)$ [30], where k is the rate constant, t is the reaction time, At and Ao are the concentrations of pollutants at the time t and initial concentration, respectively. The rate constant k is determined from the slope of the straight line using linear regression of $\ln (At)$ over the reaction time t . All catalytic and antibacterial experiments were performed in triplicate to confirm the reproducibility of the results and data are presented as mean and standard deviation.

3. Results and Discussions

3.1. Optimization of CP-AgNPs and CP-AuNPs Synthesis. The optimization procedure is extremely necessary to ensure the stability of any preparation process in general, as well as the quality of MNPs obtained under optimum conditions using aqueous extract of plants as reducing and capping agents in particular [31]. In this study, the significant synthesis conditions including the concentration of metal ions, reaction temperature, and reaction time were optimized through UV-Vis measurements based on the surface plasmon resonance phenomenon in MNPs [32].

The optimum concentrations of metal ions were investigated by adjusting the respective concentrations of the HAuCl_4 and AgNO_3 solutions in the range of $0.5\text{--}1.5 \text{ mmol/L}$ for Au^{3+} and $0.5\text{--}2 \text{ mmol/L}$ for Ag^+ , while the reaction temperature and reaction time were kept constant at 80°C and 60 min, respectively (Figures 2(a) and 2(a')). The results show that the concentration of metal ions strongly affects the formation of MNPs, avoiding the use of the excess amount of expensive precious metals. In fact, in small concentration range, the increase in metal ion concentration led to the higher UV-Vis absorbance. In the high concentration range, MNPs have been partially coagulated, resulting in a decrease in its UV-Vis absorbance. It has been found that the

FIGURE 2: Continued.

FIGURE 2: UV-Vis spectra of CP-AuNPs (a, b, c) and CP-AgNPs (a', b', c') suspensions: effects of concentration (a, a'), reaction time (b, b'), and reaction temperature (c, c').

appropriate concentrations of HAuCl_4 and AgNO_3 necessary to form stable CP-AuNPs and CP-AgNPs with maximum yield are 1.25 mmol/L and 1.5 mmol/L, respectively.

To find the suitable time for the formation of MNPs, the synthesis mixture (2.5 mL) was taken out to perform UV-Vis measurement for every 15 min toward CP-AuNPs and every 20 min toward CP-AgNPs while fixing the two other parameters (80°C, 1.25 mmol/L of Au^{3+} , and 1.5 mmol/L of Ag^+). As seen in Figures 2(b) and 2(b'), the reaction time plays an important role in the formation of CP-AgNPs but has less influence on the CP-AuNPs during the survey period due to the superiority in reducing the ability of Au^{3+} ($E_{\text{Au}^{3+}/\text{Au}}^0 = +1.5 \text{ V}$) as compared to Ag^+ ions ($E_{\text{Ag}^+/\text{Ag}}^0 = +0.799 \text{ V}$) [33]. For CP-AuNPs, in the first stage (0–45 min), the longer the reaction

time, the higher the maximum UV-Vis absorption was observed. In the case of reaction time longer than 45 min, the decrease in UV-Vis absorbance and the slight shift of maximum wavelength toward larger values indicated the formation of larger size CP-AuNPs. Therefore, 45 min was chosen as the optimum reaction time for the synthesis of CP-AuNPs. For CP-AgNPs, with the reaction time of 40 to 120 min, the maximum UV-Vis absorbance slightly increased. However, it reached stable values after 120 min. Therefore, it can be concluded that the reaction time of 120 min is the best choice to perform the synthesis of CP-AgNPs.

Finally, the optimum reaction temperature was investigated in the range of 60–100°C, while the concentration of metal ions and reaction time were kept constant (1.25 mmol/L and 45 min for CP-AuNPs; 1.5 mmol/L and

FIGURE 3: (a) XRD patterns and (b) FTIR spectra of CP-AgNPs and CP-AuNPs.

120 min for CP-AgNPs). The obtained results shown in Figures 2(c) and 2(c') indicate that the reaction temperature significantly affected the formation of both CP-AuNPs and CP-AgNPs. The increased temperature in the range of 60–80°C for Au^{3+} and Ag^{+60} –90°C provided more energy to the metal ions, leading to its faster conversion into nanoparticles.

At higher temperatures, the ions could move faster, and the number of effective collisions might increase rapidly, resulting in partial coagulation of newly formed nanoparticles with the larger size, causing a decrease in optical density. Therefore, the optimal temperatures of 90°C and 80°C were chosen for CP-AuNPs CP-AgNPs, respectively.

FIGURE 4: SEM images (a,b) and EDX spectra (c, d) for CP-AuNPs (a, c) and CP-AgNPs (b, d).

3.2. Characterizations of CP-AgNPs and CP-AuNPs. The features of crystalline structures of CP-AgNPs and CP-AuNPs were determined by XRD patterns as presented in Figure 3(a). The XRD pattern of CP-AgNPs shows characteristic peaks at 2θ angles of 38.12° (111), 44.27° (200), 64.42° (220), and 77.47° (311) that are typical for the face-centered cubic structure of Ag (ICDD PDF card number 00-004-0783) [3] [34]. In addition, the crystalline AgCl was observed with featured diffraction peaks at 2θ angle of 27.80° , 32.31° , 46.24° , 54.80° , 57.44° , and 67.30° due to reaction of Ag^+ ion with chloride ion presented in CP extract. The presence of Cl^- in aqueous plant extract was reported in similar studies [35] [36]. Besides, the detected peaks typical for biosynthesized CP-AuNPs at 38.2° (111), 44.4° (200), 64.57° (220), and 77.54° (311) corresponding to the face-centered cubic structure of Au with the ICDD PDF card number 00-004-0784 [35] [36]. In particular, the highest diffraction peaks of CP-AuNPs and CP-AgNPs at 2θ angle about 38° indicated that the crystals have a preferred growth direction in Miller indices planes (111). Thus, when determining the full width

at half maximum of peaks on (111) plane, the average crystal size of CP-AgNPs and CP-AuNPs can be calculated, according to Debye-Scherrer equation $d = 0.9\lambda/\beta\cos\theta$, where d (nm) is the average crystal size, β (radian) is the full width at half maximum, λ (0.1540 nm) is the wavelength of used $\text{CuK}\alpha$ X-ray radiation and " θ " (degree) is the Bragg diffraction angle. Accordingly, the average crystal size of CP-AgNPs and CP-AuNPs is 16 ± 0.8 nm and 19.8 ± 1.2 nm, respectively.

The FTIR spectra of CP-AgNPs, CP-AuNPs, and dried CP extract as presented in Figure 3(b) show the appearance of major bands at 3362, 2928, 1701, 1596, 1396, 1026, and 1208 cm^{-1} . The adsorption peaks of CP-AgNPs and CP-AuNPs have a minor difference in position compared to those of dried CP extract due to the presence of metals and the change in chemical functional groups of extract after the reaction. The broad band at 3362 cm^{-1} is assigned to the O-H stretching vibration of phytosteroids, alkaloids, alkyl alcohol glycosides, phenylpropanoid glycosides, polyacetylene glycosides, and polysaccharides presented in the CP-extract

FIGURE 5: Continued.

FIGURE 5: TEM images (a and a'), HR-TEM image (b and b'), SEAD (inset of b and b'), and DLS (c and c') of CP-AuNPs (a, b, and c) and CP-AgNPs (a', b, and c').

[37, 38]. Therein, the water-soluble polyol compounds are mainly responsible for the reduction and stabilization of bio-synthesized CP-AgNPs and CP-AuNPs [16] [17]. In addition, the absorption bands at 2928 , 1396 , and 1026 cm^{-1} characterized for $-\text{CH}$, $-\text{NH}$, and $-\text{CN}$ groups, respectively, were also observed in the FTIR spectra of all samples [39]. The sharp peaks at 1596 and 1701 cm^{-1} are assigned to $\text{C}=\text{C}$ and $\text{C}=\text{O}$ groups in aromatic compounds [23]. Consequently, the FTIR spectra indicate that the organic constituents of the CP extract acted as an effective reducing agent and stabilizer for CP-AgNPs and CP-AuNPs nanoparticles.

The surface morphology and element composition of biosynthesized MNPs after undergoing coagulation were

expressed by SEM images and EDX analysis, respectively (Figure 4). In this work, SEM microscopy was applied for dried solid CP-AgNPs and CP-AuNPs samples. For this purpose, colloidal stable CP-AgNPs and CP-AuNPs solutions were centrifuged to separate the solids. As presented in SEM images of CP-AuNPs (Figure 4(a)) and CP-AgNPs (Figure 4(b)), both obtained MNPs are spherical in shape and fairly uniform in size. In powdered form, that may be a part of nanoparticles was agglomerated because after the centrifugation at high speed, the organic layer acting as a capping agent could be separated from the metal cores, leading to partial agglomeration. In the EDX spectrum for CP-AuNPs (Figure 4(c)), the strong peaks appeared at 1.65, 2.2,

FIGURE 6: Antibacterial effect of CP-AgNPs at various concentrations.

2.4, 8.5, and 9.75 keV indicated the existence of Au element. In addition, the characteristic signals for elements of C (0.4 keV), O (0.55 keV), and Cl (0.2 keV) were also observed, confirming the results of previous work about the presence of nutritive constituents in the CP extract [40]. The high content of silver element was certified by characteristic signals at 2.65, 2.85, and 2.95 keV (Figure 4(d)). It should be noted that the average content of gold (29.78 w%) is much lower than that of the silver element (65.57 w%); meanwhile, the average total content of carbon and oxygen in CP-AuNPs (66 w%) is superior to that of CP-AgNPs (21.01 w%). This can be understood because CP-AgNPs contained more capping agents of organic molecules in CP-extract than CP-AuNPs.

The particle size, shape, and distribution of MNPs were evaluated by TEM, SAED, and DLS measurements, and the obtained results are presented in Figure 5. It can be seen from Figures 5(a) and 5(a'), the CP-AgNPs and CP-AuNPs crystals were mostly formed in uniform spheres dispersed well in colloidal solution with the respective average sizes of about 10 ± 2.5 nm and 20 ± 3.2 nm determined by Debye-Scherrer equation. The crystalline nature of CP-AuNPs and CP-AgNPs can be visually observed by HR-TEM and SEAD images (Figures 5(b) and 5(b')). The lattice fringe of CP-AuNPs and CP-AgNPs corresponding to the (111) plane had d-spacing of 0.24 and 0.22 nm, respectively. The bright circular rings in SAED images related to (111), (200), (220), and (311) lattice planes also indicated the existence of MNPs in cubic crystal form. Furthermore, the particle size distribution diagrams of CP-AuNPs and CP-AgNPs (Figures 5(c) and 5(c')) indicated dynamic diameters of 89 ± 4.5 nm and

112 ± 5.8 nm, respectively. The difference in particle size between TEM and DLS measurements confirmed that CP-AuNPs and CP-AgNPs in colloidal solutions were covered with a thick layer of organic molecules acted as a stabilizing agent. In fact, both CP-AgNPs and CP-AuNPs samples in colloidal form were stable for more than 3 weeks under normal room conditions at 25°C. The thickness of this organic matter layer covering CP-AgNPs sample is larger than that of CP-AuNPs sample. This is confirmed by the difference in EDX results of total content of carbon and oxygen presented in the two MNPs.

3.3. Antibacterial Assay. Antibacterial activity of both bio-synthesized MNPs in the form of colloidal solutions was tested against four bacterial strains: two Gram-positive (*B. subtilis* and *S. aureus*) and two Gram-negative (*S. Enteritidis* and *E. Coli*) with various MNPs concentrations. The antibacterial effects of CP-AuNPs and CP-AgNPs at various concentrations are illustrated in Figures 6 and 7, respectively. It has been found that CP-AuNPs did not exhibit bioactivity against any bacterial strain even at the highest concentration (120 ppm), while the colloidal sample of CP-AgNPs possessed a good antibacterial activity against three tested bacterial strains. The observed nonantibacterial activity of CP-AuNPs might be due to its minimum concentration required for antibacterial performance is still higher than the optimum. In addition, the surface area, size, and shape of MNPs can also affect bacterial inhibition [22, 41, 42]. For CP-AgNPs, a high antibacterial activity against three bacteria strains including *B. subtilis*, *S. aureus*, and *E. coli* was observed; however, it did not inhibit *S. enteritidis* at tested

FIGURE 7: Antibacterial effect of CP-AgNPs at various concentrations.

TABLE 1: Comparative antibacterial activity of AgNPs.

Bacterial strains	Plant part	Size (nm)	Concentration of AgNPs (ppm)	Zone of inhibition (mm)	References
<i>S. aureus</i>	<i>Capparis spinosa</i> L. leaf	5-30	100	14.1	[43]
	<i>Annona squamosa</i>	7-8	25	19.2	[3]
	<i>Aspergillus fumigatus</i>	94	7	27.0	[44]
	<i>Parkia speciosa</i>	35	100	10.0	[45]
	Corn-cob	11	8	15.0	[30]
	<i>Albizia procera</i>	6.18	100	18.5	[22]
	<i>Codonopsis pilosula</i>	10	80	17.0 ± 1.2	This work
<i>B. cereus</i>	<i>Capparis spinosa</i> L. leaf	5-30	100	13.1	[43]
	<i>Annona squamosa</i>	7-8	25	17.8	[3]
	<i>Parkia speciosa</i>	35	100	5.0	[45]
	Corn-cob	11	8	16.0	[30]
	<i>Codonopsis pilosula</i>	10	80	12.0 ± 0.85	This work
<i>E. coli</i>	<i>Aspergillus fumigatus</i>	94	7	25.0	[44]
	<i>Capparis spinosa</i> L. leaf	5-30	100	16.0	[43]
	<i>Annona squamosa</i>	7-8	25	12.0	[3]
	<i>Parkia speciosa</i>	35	100	9.0	[3]
	<i>Albizia procera</i>	6.18	100	13.5	[22]
	<i>Holoptelea integrifolia</i>	32-38	400	10.0	[41]
	<i>Codonopsis pilosula</i>	10	80	7.0 ± 0.42	This work
<i>S. enteritidis</i>	<i>Capparis spinosa</i> L. leaf	5-30	100	15.1	[43]
	Corn-cob	11	8	11.0	[30]
	<i>Holoptelea integrifolia</i>	32-38	400	13.0	[41]
	<i>Codonopsis pilosula</i>	10	80	0.0	This work

FIGURE 8: UV-Vis spectra (a, b) and first-order kinetics (c, d) for degradation of 1,4-DNB by NaBH₄ in the presence of CP-AuNPs (a, c) and CP-AgNPs (b, d).

concentrations. Moreover, the antibacterial activity increased with an increase in the concentration of the biosynthesized CP-AgNPs. The comparison of antibacterial activity for the CP-AgNPs with those reported in previous works are listed in Table 1. It is evident that the antibacterial activity of CP-AgNPs depends not only on AgNPs size but also strongly on the stabilizing agents.

3.4. Catalytic Performance for Reduction of Nitrophenols. Resistant substituted phenols, especially nitrophenols, are widely used in chemical industries for many applications. However, they can cause a serious threat to the aquatic animals even at low concentrations due to high toxicity and difficulty in degradation [46]. In this work, the catalytic performance of CP-AgNPs and CP-AuNPs was evaluated by degradation of 2-NP, 3-NP, 4-NP, and 1,4-DNB using

NaBH₄ as reductant. It is well known that the reduction of those organic substances by NaBH₄ without a catalyst is a thermodynamically favorable reaction but kinetically unfavorable due to the kinetic barrier between the BH₄⁻ and nitrophenolate ions [14]. This barrier can be quickly overcome by using AgNPs and AuNPs via an electron transfer mechanism [28]. The results for the reduction of 1,4-DNB, 2-NP, 3-NP, and 4-NP by NaBH₄ in the presence of the biosynthesized CP-AgNPs and CP-AuNPs are shown in Figures 8–11, respectively.

3.4.1. Catalytic Activity of CP-AgNPs and CP-AuNPs for Reduction of 1,4-DNB. After adding NaBH₄ to the yellowish 1,4-DNB solution, the color of the solution turns into dark red, and the maximum absorbance peak at 380 nm appeared regardless of that the decomposition reaction has not started

FIGURE 9: UV-Vis spectra (a, b) and first-order kinetics (c, d) for degradation of 2-NP by NaBH₄ in the presence of CP-AuNPs (a, c) and CP-AgNPs (b, d).

yet. As soon as CP-AgNPs and CP-AuNPs were added, the color of the solution gradually disappears. Figure 8 shows the UV-Vis spectra and first-order kinetics for the degradation of 1,4-DNB by NaBH₄ in the presence of CP-AuNPs and CP-AgNPs. A gradual decrease in UV-Vis maximum absorbance at the wavelength of 380 nm and a simultaneous increase in peak at 300 nm demonstrated the occurrence of 1,4-DNB decomposition process with the formation of 1,4-diaminobenzene. The results indicated that the 1,4-diaminobenzene decomposition in the presence of CP-AgNPs and CP-AuNPs was completed within 15 minutes as evidenced by near-zero absorbance at 380 nm (Figures 8(a) and 8(b)). The linear relationship can be observed from the equation $\ln(A_t)$ over the reaction time (Figures 8(b) and 8(d)), wherein the first-order reaction

rate constants k were determined as $(2.58 \pm 0.15) \times 10^{-3} \text{ sec}^{-1}$ for CP-AuNPs and $(2.34 \pm 0.18) \times 10^{-3} \text{ sec}^{-1}$ for CP-AgNPs.

3.4.2. Catalytic Activity of CP-AgNPs and CP-AuNPs for Reduction of 2-NP. Quite similarly as above, the color of 2-NP solution was changed from light to dark yellow after the addition of NaBH₄ due to the formation of 2-nitrophenolate ions in a slightly alkaline environment without reaction. UV-Vis spectroscopy (Figures 9(a) and 9(b)) shows a gradual decrease in absorbance at 410 nm and a simultaneous increase in a new peak at 290 nm, suggesting that 2-NP was reduced to 2-aminophenol. The reduction reaction was also completed within 15 min with the reaction rate constants $k = (2.54 \pm 0.13) \times 10^{-3} \text{ sec}^{-1}$ for

FIGURE 10: UV-Vis spectra (a, b) and first order kinetics (c, d) of 3-NP by NaBH₄ in the presence of CP-AuNPs (a, c) and CP-AgNPs (b, d).

CP-AuNPs and $k = (1.22 \pm 0.24) \times 10^{-3} \text{ sec}^{-1}$ for CP-AgNPs (Figures 9(c) and 9(d)).

3.4.3. Catalytic Activity of CP-AgNPs and CP-AuNPs for Reduction of 3-NP. For 3-NP, the color of the solution turned from pale yellow to yellow by the addition of NaBH₄ and simultaneously disappeared in the presence of CP-AgNPs and CP-AuNPs after 13 min. UV-Vis spectroscopy shows changes in UV-Vis maximum absorbance during the reaction (Figures 10(a) and 10(c)), wherein a gradual decrease in absorbance at 390 nm and a simultaneous increase in a new peak at 300 nm suggests that the decomposition of 3-NP occurred to form 3-aminophenol with the first-order reaction rate constants $k = (1.71 \pm 0.075) \times 10^{-3} \text{ sec}^{-1}$ for CP-AuNPs and $k = (4.09 \pm 0.16) \times 10^{-3} \text{ sec}^{-1}$ for CP-AgNPs (Figures 10(b) and 10(d)).

3.4.4. Catalytic Activity of CP-AgNPs and CP-AuNPs for Reduction of 4-NP. For the reduction of 4-NP by NaBH₄ using MNPs as a catalyst, UV-Vis spectroscopy showed a decrease in absorbance at 400 nm characterized for the dark yellow 4-nitrophenolate solution and the simultaneous increase in the peak at 300 nm related to transparent 4-aminophenol (Figures 11(a) and 11(b)). It has been found that the 4-NP reduction in the presence CP-AuNPs and CP-AgNPs completed within 14 minutes with reaction rate constant values of $(3.84 \pm 0.32) \times 10^{-3} \text{ sec}^{-1}$ and $(2.88 \pm 0.34) \times 10^{-3} \text{ sec}^{-1}$, respectively (Figures 11(c) and 11(d)). Thus, the catalytic ability of CP-AuNPs showed the best performance for the reduction of 4-NP, almost double that of 3-NP. Meanwhile, CP-AgNPs exhibited the best catalytic activity with 3-NP by 3.35 times greater than that of 2-NP. The CP-AuNPs revealed better catalytic performance than

FIGURE 11: UV-Vis spectra (a, b) and first-order kinetics (c, d) of 4-NP by NaBH₄ in the presence of CP-AuNPs (a, c) and CP-AgNPs (b, d).

CP-AgNPs for all nitrophenols, while the reduction time was only slightly different. The CP-AuNPs and CP-AgNPs biosynthesized by the extract of CP also demonstrated a good catalyst performance in comparison with those of AuNPs and AgNPs prepared by the extract from different plants (Table 2).

3.4.5. The Reusability of MNPs. The reusability of catalysts based on MNPs is especially necessary for practical applications by means of confirming their stability for long-term use and reducing the cost. In this work, the recyclable performance was tested for four reaction cycles toward the reduction of 4-NP as representative for nitrophenols.

After the initial use, the MNPs were recovered by centrifugation, washed carefully with ethanol, and applied for reuse according to the mentioned above procedure. In the first reuse for both CP-AuNPs and CP-AgNPs, the UV-Vis absorbance

peak at 400 nm corresponded to yellow 4-NP disappeared in about 20 minutes and a peak at 300 nm increased over time, indicating the formation of colorless 4-aminophenol. For the subsequent reuses, the same results were also observed. However, the reduction of 4-NP lasted longer, about 30 and 35 min, respectively. The recycling process confirmed a good recyclability of biosynthesized MNPs in the reduction of 4-NP after 4 successive cycles with the yield greater than 96% for CP-AuNPs and 95% for CP-AgNPs (Figure 12).

4. Conclusions

In this study, the aqueous extract from *Codonopsis pilosula* roots was successfully used as both reducing and stabilizing agents for the synthesis of AgNPs and AuNPs. The obtained biosynthesized MNPs were formed in a spherical

TABLE 2: Comparative catalytic performance of biosynthesized MNPs for reduction of polyphenols by NaBH_4 .

Pollutants	MNPs	Plant part	Average size (nm)	k (sec^{-1})	References
1,4-DNB	AuNPs	<i>Codonopsis pilosula</i> root	20	$(2.58 \pm 0.15) \times 10^{-3}$	This work
	AgNPs		10	$(2.34 \pm 0.18) \times 10^{-3}$	
2-NP	AuNPs	Seaweed <i>Lobophora variegata</i>	2-12	1.21×10^{-3}	[27]
	AuNPs	Corn-cob	35	3.00×10^{-3}	[30]
	AgNPs		11	2.10×10^{-3}	
	AuNPs	<i>Codonopsis pilosula</i> root	20	$(1.22 \pm 0.24) \times 10^{-3}$	This work
	AgNPs		10	$(2.54 \pm 0.13) \times 10^{-3}$	
	AuNPs	Seaweed <i>Lobophora variegata</i>	2-12	4.50×10^{-3}	[37]
3-NP	AuNPs	Corn-cob	35	8.00×10^{-3}	[30]
	AgNPs		11	2.80×10^{-3}	
	AuNPs	<i>Codonopsis pilosula</i> root	20	$(4.09 \pm 0.16) \times 10^{-3}$	This work
	AgNPs		10	$(1.71 \pm 0.075) \times 10^{-3}$	
4-NP	AuNPs	<i>Coffea arabica</i> seed	16-22	5.22×10^{-3}	[1]
	AuNPs	Burdock root	24.7	6.87×10^{-3}	[29]
	AgNPs		21.3	6.77×10^{-3}	
	AuNPs	<i>L. indica</i> leaf	14.5	1.30×10^{-3}	[35]
	AgNPs		13.5	2.10×10^{-3}	
	AuNPs	<i>Breynia rhamnoides</i>	25	9.10×10^{-3}	[23]
	AgNPs		64	4.00×10^{-3}	
	AuNPs	Corn-cob	35	5.80×10^{-3}	[30]
	AgNPs		11	5.00×10^{-3}	
	AuNPs	<i>Codonopsis pilosula</i> root	20	$(2.88 \pm 0.34) \times 10^{-3}$	This work
	AgNPs		10	$(3.84 \pm 0.32) \times 10^{-3}$

FIGURE 12: Reusability of CP-AuNPs (a) and CP-AgNPs (b).

shape with an average size of about 10–20 nm. The colloidal CP-AgNPs exhibited selective, strong antibacterial activity against three bacterial strains including two Gram-positive *B. subtilis* and *S. aureus*, and one Gram-negative *E. coli*, but there was no bacterial activity detected toward Gram-negative *S. enteritidis* at all tested concentrations. The biogenic MNPs also possessed a high catalytic activity in degradation of 1,4-DNB, 2-NP, 3-NP, and 4-NP. Therefore, the novel CP-AgNPs and CP-AuNPs synthesized by the aqueous extract of *Codonopsis pilosula* roots can be considered as perspective nanomaterials for large-scale biological and catalytic applications.

Data Availability

The data used to support the findings of this study are included within the article.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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