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Phytosynthesis of silver nanoparticles by *Cissus quadrangularis*: influence of physicochemical factors

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Abstract

Phytosynthesis process of silver nanoparticles using plant extract is simple, cost-effective, and ecofriendly. In this present investigation, we report the green nanoparticles prepared by using stem extract of *Cissus quadrangularis* and assess the physical and chemical factors such as time duration, metal ion concentration, pH, and temperature that play vital role in the nanoparticles synthesis. The maximum synthesis of silver nanoparticles was attained within 1 h, at pH 8 and 1 mM $AgNO_3$ concentration, and 70°C. The nanoparticles obtained are characterized by UV–vis spectroscopy. Silver nanoparticles that were synthesized under these conditions show crystalline nature confirmed by X-ray diffraction and show mostly spherical and some rod and triangle shapes with sizes ranging from 37 to 44 nm, which were characterized by scanning electron microscopy. Fourier transform infrared spectroscopy shows that the functional groups are carboxyl, amine, and phenolic compounds of stem extract which are involved in the reduction of silver ions. Thus, synthesized silver nanoparticles show more antibacterial activity against *Klebsiella planticola* and *Bacillus subtilis*, which was analyzed by disc diffusion method.

Keywords: Stem extract, *Cissus quadrangularis*, XRD, SEM, Silver nanoparticles, Antibacterial activity

Background

Nanotechnology is a multidisciplinary research field that emerges from physical, chemical, and engineering sciences with novel techniques and produces material at nanoscale level [1]. Nanomaterials are the essential building elements of nanotechnology. Nanoparticles are very important and have a distinct property, that is, they exhibit larger surface area to volume ratio [2]. This increase in surface area to volume ratio leads to the change of the properties of nanoparticles better than the bulk particles [3]. Due to their great availability of surface area, they are involved in a many applications such as catalysis [4], drug delivery [5], biosensing [6,7], and optics [8]. The noble metals such as Au, Pt, Pd, and Ag are synthesized by various methods such as physical, chemical, and biological methods. Physically and chemically mediated syntheses necessitate high pressure and temperature, high cost, and toxicity [9]. The involvement of biological entities in the nanoparticle growth is gaining tremendous advantages such as production of anisotropic nanoparticles with size and shape control and intensive energy. Biological methods of nanoparticle synthesis by using microorganisms [10] such as bacteria [11], fungus [12] and algae [13], enzyme [14], and plants both intracellularly and extracellularly show time-consuming and ecofriendly process. Microorganism-assisted nanoparticles synthesis is a time-consuming process which takes 24 to 120 h [15,16]. But green-mediated synthesis of nanoparticles by using plants has more advantages over the micro-mediated synthesis process because it mainly eliminates the maintenance of cell cultures. The utilization of plant materials for biosynthesis of nanoparticles is called green synthesis [17]. Plant-mediated synthesis has been suggested as possibly eco-friendly, cost-effective, and less time-consuming process and do not involve any toxic chemicals [18-20]. The advantage of using plants for the synthesis of nanoparticles is that they are easily available, safe to

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handle, and possess a broad variability of metabolites that may aid in reduction. The time required for 90% of the reduction of silver ions was about 2 or 4 h [21]. The motivation for choosing the plant was its medicinal properties, and capping or reducing agents are available in this plant. The size, shape, stability, and the synthesis of nanoparticles were dependent on the various factors such as capping agents, pH, templates, and temperature. Gericke and Pinches reported that the intracellular particles formation and size of the particles could be controlled by altering the key factors such as pH, temperature, substrate concentration, and exposure time to the substrate [22].

Among the metal nanoparticles, silver has been consumed largely due to their antimicrobial and pharmaceutical applications [23,24]. Silver has long been recognized as having inhibitory effect on microbes present in medical and industrial process [25,26]. The most important application of silver and silver nanoparticles is in medical industry such as topical ointments to prevent infection against burn and open wounds [27]. Medical devices and implants were prepared with silver-impregnated polymers [28] and antibiotics [3,29]. The antimicrobial activity of silver nanoparticles mainly depends on the size property. The small-sized nanoparticles have large surface area to improve the antimicrobial activity in addition to improving chemical stability [30].

Cissus quadrangularis (Linn) has been used by common man in India for the promotion of fracture healing. It is also known as *Vitis quadrangularis* Wall and belongs to family Vitaceae. It is a common perennial climber and is distributed throughout India, particularly in tropical regions [31]. This plant was used in Ayurveda as an alternative anthelmintic, dyspeptic, digestive, tonic, analgesic in eye and ear diseases, and in the treatment of irregular menstruation and asthma. The whole plant is used in oral rehydration; while the leaf, stem, and root extracts of this plant are important in the management of various ailments. In addition, this plant is used in the management of obesity and metabolic disorders [32]. This plant has antioxidant property and free radical-scavenging activity [33].

In this work we report the synthesis of silver nanoparticles by the stem extract of *C. quadrangularis* as a reducing agent and find the effective factors for its synthesis process by varying the pH, temperature, metal ion concentration, and time duration. UV spectrophotometer was used to characterize the synthesized silver nanoparticles.

Results and discussion

Visible observation of nanoparticles synthesis

The formation of silver nanoparticles was preliminarily confirmed by the change of color of the solution (Figure 1). The color change from watery yellow to brown color of the aqueous solution was due to the excitation of the surface plasmon resonance [34] and SPR band which both play an important role in the confirmation of silver nanoparticles formation [35]. It also indicates that reduction of silver ion in the aqueous solution by the stem extract of *C. quadrangularis*.

UV–vis spectrum analysis of influencing the factor in synthesis process

Figure 2 shows UV spectra exposed from the reaction of reduction of silver ions which have polydispersed nanoparticles with broadening peak in the absorbance band at the wavelength of 450 nm. Nanoparticles formation was dependent on the different physical and chemical factors such as metal ion concentration, incubation time, pH, and temperature. These optimized factors affect the silver nanoparticles formation. This study identifies which factors should affect the nanoparticle synthesis.

The first factor is concentration of silver nitrate; the nanoparticles formation was increased while increasing the concentration of silver nitrate (Figure 3). At 1-mM concentration, the band at 410 nm shifts into 450 nm while increasing the silver nitrate concentration. Similar results are obtained using the seed extract of *Jatropha curcas* with different concentrations of silver nitrate [36]. It is because of the bioavailability of functional groups in the 10 ml of stem extract of *C. quadrangularis* is involved in the reduction of silver ion.

Figure 4 shows the factor of time-dependent formation of silver nanoparticles. As the time duration increased, the nanoparticle synthesis also increased. Nanoparticle formation was initiated within 5 min. The completion of nanoparticle synthesis was achieved after 1 h as identified in Figure 4. After 1 h the precipitation of nanoparticles occurred due to the instability of the silver

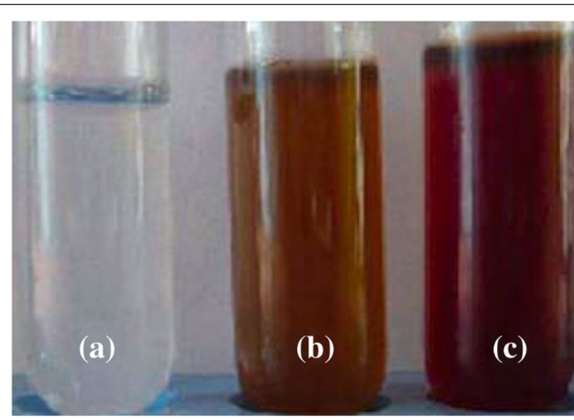

Figure 1 Color change indicates the formation of silver nanoparticles. (**a**) Silver nitrate solution, (**b**) after the 10 min reduction of silver nitrate, and (**c**) 1 h reaction time.

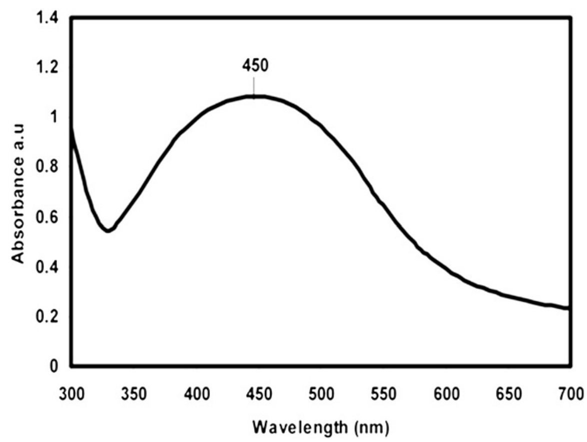

Figure 2 UV spectroscopy of aqueous solution of 1 mM $AgNO_3$ with stem extract. Spectroscopy recorded from the aqueous solution of 1 mM $AgNO_3$ with 10 ml of stem extract at different wavelengths.

nanoparticles. Agglomeration of nanoparticles shows the larger size of nanoparticles. So the optimum time duration for the formation of nanoparticles was 1 h. Earlier studies show it was a long-duration process when using some sources such as fungi which took 72 h in duration for synthesizing the silver nanoparticles [37], but in this study, it is a less time-consuming process. Some of the plants sources also took much longer time duration for silver ion reduction process.

The pH is one of the most important factor for nanoparticle formation. The shape and size of the nanoparticles are dependent on the pH of solution. In this study, the acidic pH 4 shows the peak at 420 nm. The absorbance band was increased while increasing the pH up to 8 due to the excitation of surface plasmon resonance. At pH 8 the absorbance peak was at 450 nm. The acidic pH suppresses the nanoparticle formation. At low pH, agglomeration took place because of the over nucleation and formation of larger nanoparticles. At high pH, a large number of nanoparticles with the small surface area are present due to the bioavailability of functional groups in the stem extract. Nanoparticle formation was increased with increasing pH. In the alkaline conditions, more precipitation or agglomeration occurred due to the instability of silver nanoparticles or due to lack of stabilizing agent. So the alkaline pH 8 was favorable for the nanoparticle formation (Figure 5).

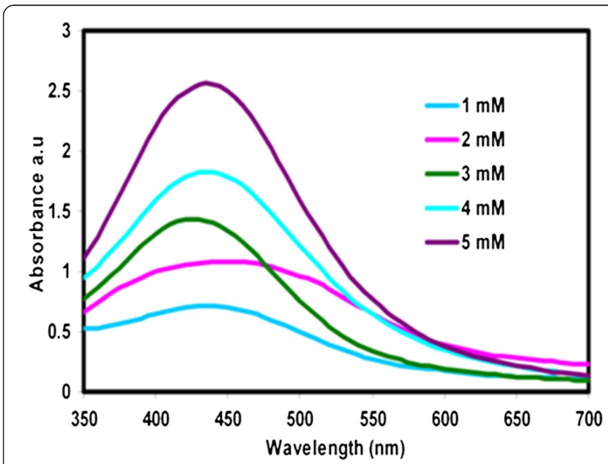

Figure 3 UV spectroscopy recorded from the aqueous solution of $AgNO_3$ at different concentrations at different wavelengths.

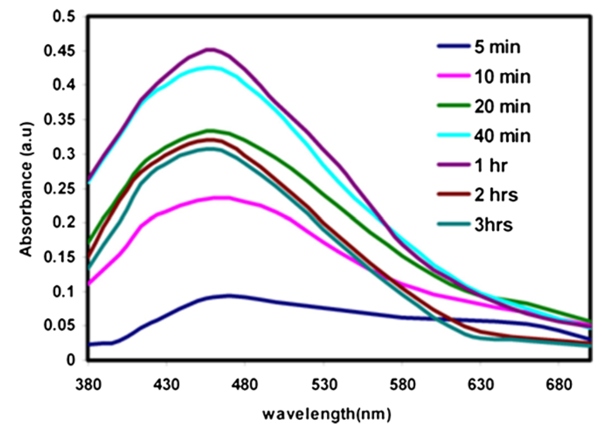

Figure 4 UV spectroscopy recorded from aqueous solution of $AgNO_3$ as a function time at different wavelengths.

Another one important factor is temperature. In this study nanoparticle synthesis was increased. While increasing the temperature (Figure 6), the absorption peak shifts from 460 nm in the temperature of 30°C; while by increasing the temperature up to 70°C, the band shifts to 440 nm, which is due to the localization of the surface

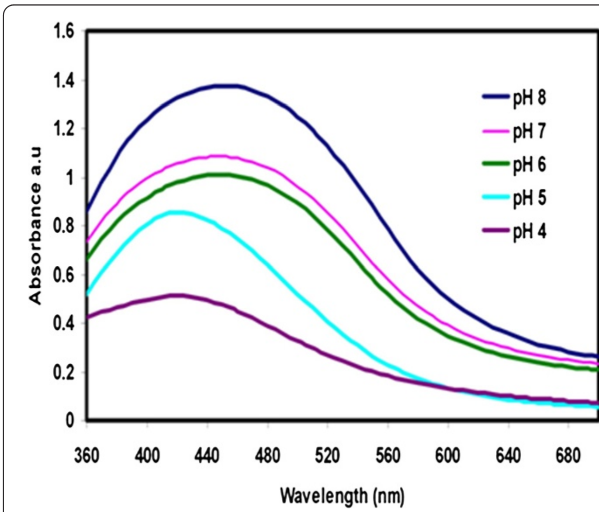

Figure 5 UV spectroscopic analysis shows effect of pH for nanoparticles formation.

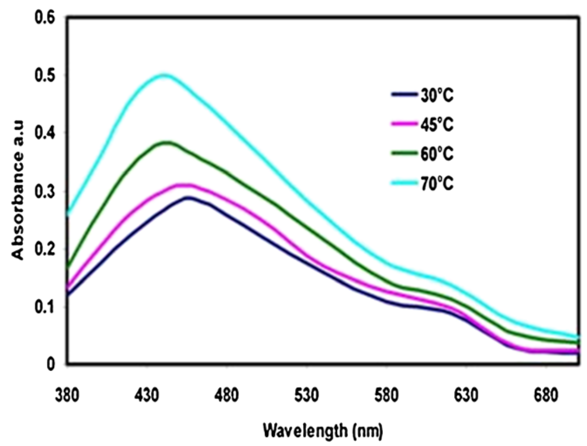

Figure 6 UV spectrum of silver nanoparticles after 1 h of reaction time at different temperatures.

plasmon resonance of the silver nanoparticles [38]. In higher temperature, the reduction occurred very hastily.

Characterization studies

X-ray diffraction

X-ray diffraction (XRD) pattern obtained for the silver nanoparticles was shown in Figure 7. XRD shows that the crystalline structure of silver is face centered cubic. In XRD, silver has similar diffraction profile with intense peaks at 2θ of 38.13°, 44.1°, 64.35°, and 77.88° corresponding to the planes (111), (200), (220), and (311), respectively. This indicates that the synthesized silver nanoparticles by using stem extract of *C. quadrangularis* had crystalline nature. Similar results were obtained by [39] using the *Bacillus cereus* microorganism.

SEM and EDX analysis

Scanning electron microscopy (SEM) image shows (Figure 8) the morphological character of silver nanoparticles synthesized by using extract of *C. quadrangularis.*

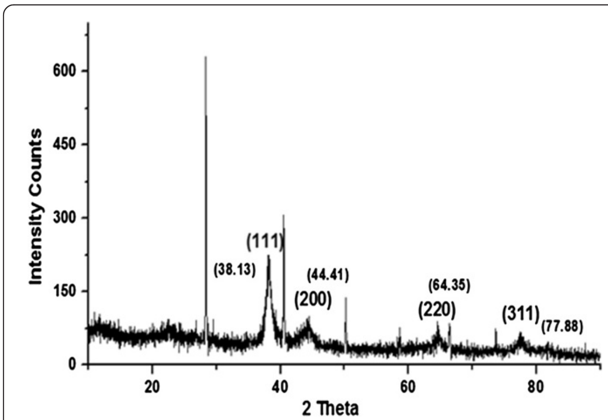

Figure 7 XRD analysis of synthesized silver nanoparticles.

This image shows that the sizes at around 37 to 43 nm with many shapes which are triangle, rod, and spherical are clearly observed. This SEM image also showed the aggregation of the silver nanoparticles.

Energy dispersive X-ray spectroscopy (EDX) shows the chemical analysis of synthesized silver nanoparticles using the *C. quadrangularis* stem extract (Figure 9). The strong signal was obtained at the energy of 3 keV for silver and also some of the weak signals from Cl, K, O, Ca, Mg, and S. The major emission energy at 3 keV indicates that the silver has been correctly identified.

FTIR analysis

FTIR analysis reveals the functional groups of the silver nanoparticles synthesized using stem extract of *C. quadrangularis* as shown in Figure 10. The band at 3,324 cm^{-1} shows O-H stretching vibrations of hydroxyl groups [40], H-bonded alcohols, phenols, or N-H stretching of 1° and 2° amines and amides. The band at 3,195 cm^{-1}appears due to the O-H stretching vibrations of carboxylic acids. A weak band at 2,966 cm^{-1} corresponds to C-H stretch alkanes and O-H stretch carboxylic acids [35]. The peak at 2,856 cm^{-1} shows C-H stretching for alkanes; peaks at 2,297 and 1668 cm^{-1} contribute to C=O stretch α, β-unsaturated aldehydes, and ketones; the peak at 1,596 cm^{-1} shows -C-C- skeletal vibrations [41]. The band at 1,400 cm^{-1} assigned to O-H bend indicates carboxylate [40]; the band 1,193 cm^{-1} shows C-O stretch alcohols and carboxylic acids. The peak at 1,115 cm^{-1} indicates presence of C-N stretch aliphatic amines; the peak at 605 $cm^{-1}$was assigned for C-Cl stretching vibrations of alkyl halides. The functional biomolecules are carboxylic and amine groups involved in the reduction of silver ions, as confirmed by FTIR spectrum.

Assay of antibacterial activity of synthesized silver nanoparticles

Antibacterial activity of synthesized silver nanoparticles against *Bacillus subtilis* and *K. planticola* was assayed by disc diffusion method (Figure 11). The concentration of silver nanoparticles varied from 10 to 50 μl. The zone of inhibition increased while increasing the concentration of silver nanoparticles. The zone of inhibition in diameter was tabulated (Table 1). Silver nanoparticles are very toxic in gram-negative bacteria than the gram-positive bacteria due to the difference in the cell wall of bacteria.

The mechanism of its antibacterial activity was not understood clearly. Some of the researchers explained the possible mechanism of antibacterial activity of silver nanoparticles. Silver nanoparticles attach with the cell wall of bacteria by electrostatic attraction and disrupt the cell permeability and respiration due to the generation of the reactive oxygen species. Silver nanoparticles

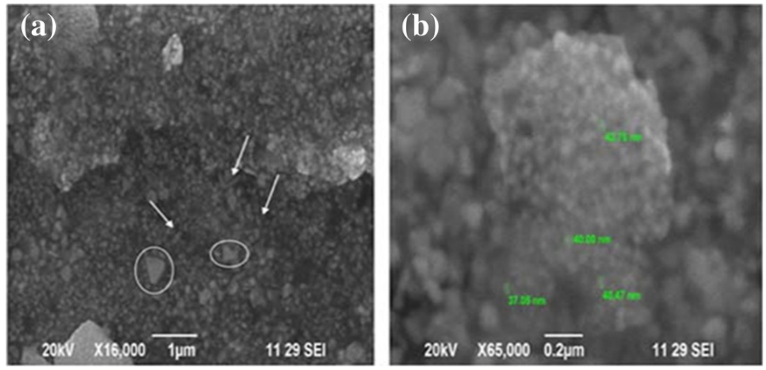

Figure 8 SEM analysis of silver nanoparticles at different magnifications (a) X16000 (b) X65000.

bind with thiol groups of DNA and RNA and affect the protein synthesis of the bacteria [42,43]. Silver nanoparticles form the pits on the cell surface and induce the proton leakage resulting in cell death [44]. The antimicrobial activity of silver nanoparticles depends on the size of the particles. Smaller particles having the larger surface area available for interaction will give more bactericidal effect than the larger particles and since they easily penetrate into the cell [45,46].

Conclusions

This present investigation shows the bioreduction of silver nanoparticles using the medicinal valuable stem extract of *C. quadrangularis* by optimizing the factors for rapid and stabilized nanoparticles synthesis. Nanoparticle synthesis process was controlled by incubation time, metal ion concentration, pH, and temperature. High pH and temperature influence the rapid synthesis of silver nanoparticles. Thus, synthesized silver nanoparticles are mostly spherical as confirmed by SEM. Mainly carboxylic and amine groups may responsible for the reduction process as revealed by FTIR. This green method is simple, rapid, eco-friendly and reliable, and it may have a potential use in the biomedical applications due to its high antibacterial activity.

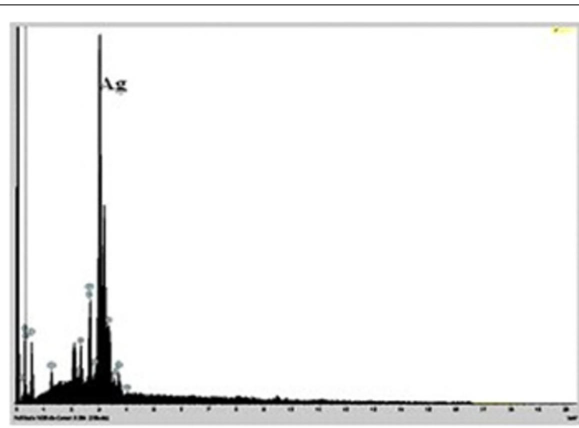

Figure 9 EDAX analysis of silver nanoparticles.

Methods

Preparation of reducing agent

All the chemicals and reagents are purchased from Himedia, Mumbai, India. The stem of *C. quadrangularis* was used as a reducing agent for the preparation of silver nanoparticles, collected from Western Ghats of Tamil Nadu. A 20 g of stem was thoroughly washed with Tween 20 and double-distilled water. The cleaned stem was cut into fine pieces which were boiled in 100 ml of double-distilled water for 5 min and filtered through Whatman no.1 filter paper. Filtered extract was stored at 4°C for 1 week for further experiment use.

Synthesis of silver nanoparticles

A 10 ml of stem extract was added into the 90 ml of aqueous solution of 1 mM of silver nitrate and kept at room temperature. The color change of the solution indicates the formation of silver nanoparticles. The reduced silver nanoparticles were monitored in UV–vis spectrophotometer.

Study of the effect of physicochemical factors in nanoparticles synthesis

To study the effect of silver ion concentration in nanoparticle synthesis, 10 ml of stem extract was added into different concentrations of silver ions (1, 2, 3, 4, and 5 mM). To study the effect of time duration on nanoparticle formation, the reaction solution was incubated at specific time intervals 0, 10, 20, and 40 and 1, 2, 3, 4, and 24 h. To study the effect of pH, experiments were carried out by varying the pH (4, 5, 6, 7, and 8) of the stem extract. The effect of temperature was studied by keeping the reaction solution at different temperatures (30°C, 45°C, 60°C, and 70°C). The influence of these

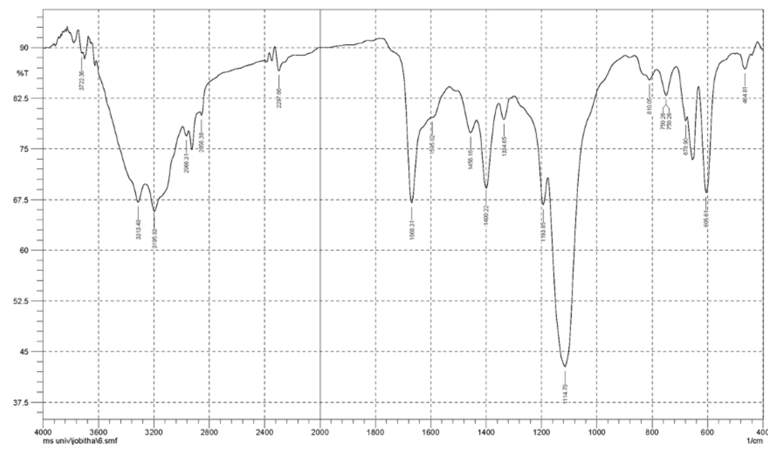

Figure 10 FTIR spectrum of synthesized silver nanoparticles.

physicochemical factors in silver nanoparticles formation was confirmed by UV–vis spectrophotometer at different wavelengths.

Characterization study of synthesized silver nanoparticles

The reduction process of the solution was monitored on a PerkinElmer double-beam UV spectrophotometer (PerkinElmer Inc., Waltham, MA, USA). The reaction of the aliquot solution analyzed at different reaction times in the wavelength ranges between 300 and 700 nm. The crystalline nature of the silver nanoparticles was characterized by X-ray diffraction. The reaction mixture was purified by repeated centrifugation at speed of 10,000 rpm for 10 min, and the pellets were dried at room temperature. The dried powder of silver nanoparticles was characterized by XRD (Philips PW 1830; Royal Philips Electronics, Amsterdam, Netherlands). The morphology and size of the silver nanoparticles were determined by scanning electron microscope (Philip model CM 200). The elemental analysis of silver was carried out by energy disperse analysis X-ray (EDAX). Fourier transform infrared spectroscopy (FTIR) measurements were carried out for dried biomass of leaf extract treated with silver nanoparticles to find out the compound responsible for the synthesis of silver nanoparticles. The FTIR was obtained on a Shimadzu instrument (Shimadzu Co. Ltd., Beijing, China) with the sample as KBR pellet.

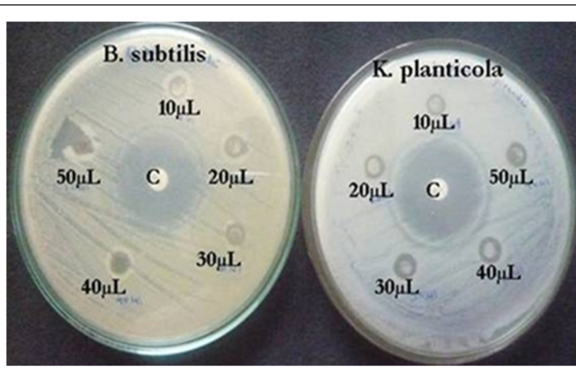

Figure 11 Antibacterial activity of silver nanoparticles synthesized by stem extract of *Cissus quadrangularis*.

Assay of antibacterial activity

Antimicrobial activities of synthesized silver nanoparticles against the two bacterial cultures of *Bacillus subtilis* (3053) and *K. planticola* (2727) (MTCC, Chandigarh, India) were assayed by disk diffusion method. These two bacteria were grown in LB broth for 24 h. Approximately 20 ml of molten and cooled Muller Hinton agar was poured into the Petri dishes. The two tested organisms are swapped over the agar medium, and the silver nanoparticle-containing disks were kept over the medium using sterile forceps. The silver nanoparticle disks were prepared at different concentrations (10, 20, 30, 40, and 50 µL) and incubated at 35°C for 24 h.

Table 1 Antibacterial activity of silver nanoparticles at various concentrations against *B. subtilis* and *K. planticola*

Concentration of silver nanoparticles (µL)	Zone of inhibition (diameter in mm)	
	Bacillus subtilis	*Klebsiella planticola*
10	8	9
20	9	10
30	9	9
40	11	12
50	11	13

Competing interests
The authors declare that they have no competing interests.

Authors' contributions
MV and GG carried out the nanoparticles synthesis. KP carried out the manuscript preparation. SR and CM carried out the antimicrobial activity. All authors read and approved the final manuscript.

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MV completed B.Sc. in Microbiology and M.Sc. Environmental Biotechnology in Sri Paramakalyani Centre for Environmental Sciences, Manonmaniam Sundaranar University, Alwarkurichi, Tirunelveli. She obtained her Ph.D. degree under the guidance of GA in the field of nanotechnology. Her research interests include ecofriendly synthesis of nanoparticles and its application in the environment. GG completed M.Sc. in Biotechnology in Periyar University and obtained her Ph.D. degree in Nanotechnology under the guidance of GA. Her research interests include green-meditated synthesis of nanoparticles and its agricultural applications for controlling plant diseases. KP received his B.Sc. (2005) and M.Sc. in Biotechnology (2007) from Madurai Kamaraj University and Periyar University, India, respectively. He is currently working to obtain his Ph.D. degree at Manonmaniam Sundaranar University in India. He is interested in the green and immobilized microbe-mediated synthesis process of nanoparticles and nanocomposites and their biomedical and textile industry applications. SR completed M.Sc. Biotechnology in Periyar University and obtained his Ph.D. degree in Nanotechnology under the guidance of GA. He published seven research articles in nanoparticles synthesis using algae. He is interested in the metallic nanoparticles synthesis using algae and algae-derived compounds and their potential applications in the biomedical field. CM obtained her M.Sc. in Environmental Biotechnology in Sri Paramakalyani Centre for Environmental Sciences, Manonmaniam Sundaranar University, Alwarkurichi, Tirunelveli. She achieved her Ph.D. degree under the guidance of GA in the field of nanotechnology. Her research interests include synthesis of semiconductor nanomaterials and its biomedical applications. GA received his M.Sc. in Applied Chemistry (1992) and Ph.D. in Environmental Biotechnology (1997) from Anna University, India. He had ten years (1999 to 2008) post-doctoral experiences from the National Taiwan University in Taiwan, National Institute of Advanced Industrial Science and Technology in Japan, and National Central University in Taiwan. He has received many research awards from Indian and other country governments. He is an associate editor in five international journals. At present, he is an associate professor of the Environmental Biotechnology and the leader of Environmental Nanobiotechnology Division at Sri Paramakalyani Centre for Environmental Sciences, Manonmaniam Sundaranar University, India. His main research interest is on biosynthesis of nanoparticles and nanomaterials, nanobiocatalyst, and environmental chemistry.

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References
1. Schmid, G: Nanoparticles from theory to applications. Wiley-VCH, Weinheim (2004)
2. Annamalai, A, Babu, ST, Jose, NJ, Sudha, D, Lyza, CV: Biosynthesis and characterization of silver and gold nanoparticles using aqueous leaf extraction of *Phyllanthusamarus* Schum & Thonn. World Appl Sci J **13**, 1833–1840 (2011)
3. Raimondi, F, Scherer, GG, Kotz, R, Wokaun, A: Nanoparticles in energy technology: examples from electochemistry and catalysis. Angew Chem Int Ed **44**, 2190–2209 (2005)
4. Tsang, C, Caps, V, Paraskevas, I, Chadwick, D, Thompsett, D: Magnetically separable, carbon-supported nanocrystals for the manufacture of fine chemical. Angew Chem **116**, 5763–5767 (2004)
5. Baptista, PV: Cancer nanotechnology - prospects for cancer diagnostics and therapy. Curr Canc Ther Rev **5**(2), 80–88 (2009)
6. Huang, CC, Yang, Z, Lee, KH, Chang, HT: Synthesis of highly fluorescent gold nanoparticles for sensing mercury (II). Angew Chem Int Ed **46**, 6824–6828 (2007)
7. Jain, PK, Lee, KS, El-Sayed, IH, El-Sayed, MA: Calculated absorption and scattering properties of gold nanoparticles of different size, shape, and composition: applications in biological imaging and biomedicine. J Phys Chem B **110**, 7238–7248 (2006)
8. Murphy, CJ, Sau, TK, Gole, AM, Orendorff, CJ, Gao, J, Gou, L, Hunyadi, SE, Li, T: Anisotropic metal nanoparticles: synthesis, assembly, and optical applications. J Phys Chem B **109**, 13857–13870 (2005)
9. Sulochana, S, Krishnamoorthy, P, Sivaranjani, K: Synthesis of silver nanoparticles using leaf extract of *Andrographis paniculata*. J Pharmacol Toxicol **7**(5), 251–258 (2012)
10. Schultz, S, Smith, DR, Mock, JJ, Schultz, DA: Single-target molecule detection with nonbleaching multicolor optical immunolabels. Proc Natl Acad Sci **97**, 996–1001 (2000)
11. Shahverdi, AR, Fakhimi, A, Shahverdi, HR, Minaian, SA: Synthesis and effect of silver nanoparticles on the antibacterial activity of different antibiotics against *Staphylococcus aureus* and *Escherichia coli*. Nanomed Nanotechnol Biol Med **3**, 168–171 (2007)
12. Shaligram, NS, Bule, M, Bhambure, R, Singhal, RS, Singh, SK, Szakacs, G, Pandey, A: Biosynthesis of silver nanoparticles using aqueous extract from the compactin producing fungal strain. Proc Biochem **44**, 939–943 (2009)
13. Singaravelu, G, Arockiamary, JS, Ganesh Kumar, V, Govindaraju, K: A novel extracellular synthesis of monodisperse gold nanoparticles using marine alga, *Sargassum wightii* Greville. Colloid Surf B: Biointer **57**, 97–101 (2007)
14. Nair, B, Pradeep, T: Coalescence of nanoclusters and formation of submicron crystallites assisted by *Lactobacillus* strains. Cryst Growth Des **2**, 293–298 (2002)
15. Ahmad, A, Mukherjee, P, Senapati, S, Mandal, D, Khan, DI, Kumar, R, Sastry, M: Extracellular biosynthesis of silver nanoparticles using the fungus *Fusarium oxysporum*. Colloids Surf B Biointerfaces **28**, 313–318 (2003)
16. Ahmad, A, Senapati, S, Khan, MI, Kumar, R, Ramani, R, Srinivas, V, Sastry, M: Intracellular synthesis of gold nanoparticles by a novel alkalotolerant actinomycete *Rhodococcus* species. Nanotechnology **14**, 824–825 (2003)
17. Sathishkumar, FM, Sneha, K, Won, SW, Cho, CW, Kim, S, Yun, YS: *Cinnamon zeylanicum* bark extract and powder mediated green synthesis of nano-crystalline silver particles and its bactericidal activity. Colloid Surf B: Biointerf **73**, 332–338 (2009)
18. Vigneshwaranm, N, Ashtaputre, NM, Varadarajan, PV, Nachane, RP, Paralikar, KM, Balasubramanya, RH: Biological synthesis of silver nanoparticles using the fungus *Aspergillus flavus*. Mater Lett **61**, 1413–1418 (2007)
19. Mohanpuria, P, Rana, NK, Yadav, SK: Biosynthesis of nanoparticles: technological concepts and future applications. J Nanopart Res **10**, 507–517 (2008)
20. Shankar, SS, Rai, A, Ahmad, A, Sastry, MJ: Rapid synthesis of Au, Ag, and bimetallic Au core-Ag shell nanoparticles using Neem (*Azadirachta indica*) leaf broth. J Colloid Interface Sci **275**, 496–502 (2004)
21. Chandran, SP, Chaudhary, M, Pasricha, R, Ahmad, A, Sastry, M: Synthesis of gold nanotriangles and silver nanoparticles using *Aloe vera* plant extract. Biotechnol Prog **22**, 577–583 (2006)
22. Gericke, M, Pinches, A: Microbial production of gold nanoparticles. Gold Bull **39**, 22–28 (2006)
23. Kim, YK, Lee, YS, Jeong, DH, Cho, MH, et al.: Antimicrobial effect of silver nanoparticles. Nanomed **3**, 95–101 (2007)
24. Choi, O, Deng, KK, Kim Jr, N, Ross, L, Rao, YS, Hu, Z: The inhibitory effects of silver nanoparticles, silver ions, and silver chloride colloids on microbial growth. Water Res **42**, 3066–3074 (2008)
25. Jose, RM, Jose, LEC: The bactericidal effect of silver nanoparticles. Alejandra Nanotech **16**, 2346–2353 (2005)
26. Lok, C, Ho, R, Chen, Q, He, W, Yu, H, Sun, PK, Tam, J, Chiu, C, Che, CM: Silver-nanoparticles: partial oxidation and antibacterial activities. J Biol Inorg Chem **12**, 527–534 (2007)
27. Ip, M, Lui, SL, Poon, VKM, Lung, I, Burd, A: Antimicrobial activities of silver dressings: an *in vitro* comparison. J Med Microbiol **55**, 59–63 (2006)
28. Silver, S: Bacterial silver resistance: molecular biology and use and misuses of silver compounds. FEMS Microbiol Rev **27**, 341–353 (2003)
29. Gajbhiye, M, Kesharwani, J, Ingle, A, Gade, A, Rai, M: Fungus-mediated synthesis of silver nanoparticles and their activity against pathogenic fungi in combination with fluconazole. Nanomed: Nanotechnol Biol Med **5**, 382–386 (2009)

30. Toshikazu, T: Antimicrobial agent composed of silica-gel with silver complex. Inorg Mat **6**, 505–511 (1999)
31. Mishra, G, Srivastava, S, Nagori, BP: Pharmacological and therapeutic activity of *Cissus quadrangularis*: an overview. Inter J Pharm Tech Res **2**, 1298–1310 (2010)
32. Mallika, J, Shyamala, CSD: *In vitro* and *in vivo* evaluation of free radical scavenging potential of *Cissus quadrangularis*. Afri J of Biomed Res **8**, 95–99 (2005)
33. Oben, J, Kuate, D, Agbor, G, Momo, C, Talla, X: The use of a *Cissus quadrangularis* formulation in the management of weight loss and metabolic syndrome. Lipids Health Dis **5**, 24 (2006)
34. Krishnaraj, C, Jagan, EG, Rajasekar, S, Selvakumar, P, Kalaichelvan, PT, Mohan, N: Synthesis of silver nanoparticles using *Acalypha indica* leaf extracts and its antibacterial activity against water borne pathogens. Colloids Surf B Biointerfaces **76**, 50–56 (2010)
35. Rajeshkumar, S, Kannan, C, Annadurai, G: Synthesis and characterization of antimicrobial silver nanoparticles using marine brown seaweed *Padina tetrastromatica*. Drug Invention Today **4**, 511–513 (2012)
36. Bar, H, Bhui, DK, Sahoo, GP, Sarkar, P, De, SP, Misra, A: Green synthesis of silver nanoparticles using latex of *Jatropha curcas*. Colloid Surf A **339**, 134–139 (2009)
37. Banu, A, Rathod, V, Ranganath, E: Silver nanoparticle production by *Rhizopus stolonifer* and its antibacterial activity against extended spectrum β-lactamase producing (ESBL) strains of *Enterobacteriaceae*. Mater Res Bull **46**, 1417–1423 (2011)
38. Fayaz, AM, Balaji, K, Kalaichelvan, PT, Venkatesan, R: Fungal based synthesis of silver nanoparticles - an effect of temperature on the size of particles. Colloid Surf B **74**, 123–126 (2009)
39. Ganesh Babu, MM, Gunasekaran, P: Production and structural characterization of crystalline silver nanoparticles from *Bacillus cereus* isolate. Colloid Surf B: Biointerf **74**, 191–195 (2009)
40. Rastogi, L, Arunachalam, J: Sunlight based irradiation strategy for rapid green synthesis of highly stable silver nanoparticles using aqueous garlic (*Allium sativum*) extract and their antibacterial potential. Mater Chem Phy **129**, 558–563 (2011)
41. Bansal, V, Rautray, D, Ahmad, A, Sastry, M: Biosynthesis of zirconia nanoparticles using the fungus *Fusarium oxysporum*. J Mater Chem **14**, 3303–3305 (2004)
42. Kulthong, K, Srisung, S, Boonpavanitchaku, K, Kangwansupamonkon, W, Maniratanachote, R: Determination of silver nanoparticle release from antibacterial fabrics into artificial sweat. Particle Fibre Toxicol **7**, 1–8 (2010)
43. Feng, QL, Wu, J, Chen, GQ, Cui, FZ, Kim, TN, Kim, JO: A mechanistic study of the antibacterial effect of silver ions on *Escherichia coli* and *Staphylococcus aureus*. J Biomed Mater Res **52**, 662–668 (2000)
44. Dibrov, P, Dzioba, J, Gosink, KK, Hase, CC: Chemiosmotic mechanism of antimicrobial activity of Ag(+) in *Vibrio cholerae*. Antimicrob Agents Chemother **46**, 2668–2670 (2002)
45. Panacek, A, Kvytek, L, Prucek, R, Kolar, M, Vecerova, R, Pizurova, N, Virender, KS, Nevecna, T, Zboril, R: Silver colloid nanoparticles: synthesis, characterization, and their antibacterial activity. J Phys Chem B **110**, 16248–16253 (2006)
46. Shrivastava, S, Bera, T, Roy, A, Singh, G, Ramachandran, P, Das, D: Characterization of enhanced antibacterial effect of novel silver nanoparticle. Nanotechnology **18**, 1–9 (2007)

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