

Eco-friendly approach for nanoparticles synthesis and mechanism behind antibacterial activity of silver and anticancer activity of gold nanoparticles

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Abstract This review covers general information about the eco-friendly process for the synthesis of silver nanoparticles (AgNP) and gold nanoparticles (AuNP) and focuses on mechanism of the antibacterial activity of AgNPs and the anticancer activity of AuNPs. Biomolecules in the plant extract are involved in reduction of metal ions to nanoparticle in a one-step and eco-friendly synthesis process. Natural plant extracts contain wide range of metabolites including carbohydrates, alkaloids, terpenoids, phenolic compounds, and enzymes. A variety of plant species and plant parts have been successfully extracted and utilized for AgNP and AuNP syntheses. Green-synthesized nanoparticles eliminate the need for a stabilizing and capping agent and show shape and size-dependent biological activities. Here, we describe some of the plant extracts involved in nanoparticle synthesis, characterization methods, and biological applications. Nanoparticles are important in the field of pharmaceuticals for their strong antibacterial and anticancer activity. Considering the importance and uniqueness of this concept, the synthesis, characterization, and application of AgNPs and AuNPs are discussed in this review.

Keywords Antibacterial activity · Anticancer activity · Green synthesis · Gold nanoparticles · Plant extract · Silver nanoparticle

Introduction

Nanotechnology is emerging as a cutting-edge technology involving many academic disciplines like chemistry, biology, material science, physics, and medicines. The word “nano” is derived from the Greek word nanos, which means “dwarf.” The size of a nanoparticle is one billionth of a meter (10^{-9} m), and materials with nano dimensions (1–100 nm) have significant activity compared to the same material in bulk form (Perez et al. 2005). The remarkable differences in structural and physical properties of atom and molecule of element are due to physicochemical characteristic and surface-to-volume ratio (Mody et al. 2010). Nanoparticles at the nanoscale system are attracting the attention of researchers because nanoparticles can be produced in different size and shapes and can be utilized for advanced biotechnology (Goodsell 2004). Because of their unique optoelectronic and physicochemical properties, nanoparticles are specifically of interest for a range of applications including chemical sensors, catalysts, electronic components, pharmaceutical products, medical diagnostic imaging (Choi et al. 2015; Coccia et al. 2012; Cavanagh et al. 2010), and antimicrobial activity (Nam et al. 2015; Beyth et al. 2015). For example, gold, silver, and platinum have been applied in cosmetics to pharmaceutical product; some known metallic nanoparticles including selenium (Kheradmand et al. 2014), copper (Ramya Devi et al. 2012), iron, and zinc oxide (Sirelkhatim et al. 2015) have also been used in antimicrobial application, cosmetics preparation, and medical treatment.

At present, the synthesis of nanoparticles using biological agents is becoming more attractive as an alternative to traditional methods. Biosynthesis of nanoparticles involves using an eco-friendly, green-chemistry-based approach that makes use of bacteria, fungi, actinomycetes, yeast, and algae (Li et al. 2011; Mukherjee et al. 2001), and plants (Mittal et al. 2013;

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Iravani 2011). Plant extract-based synthesis of nanoparticles offers an eco-friendly, clean, nontoxic, and inexpensive method producing nanoparticles with different shape, size, and morphology (Mohanpuria et al. 2008). The biosynthesis route can produce better sizes and shapes of metal nanoparticles compared to some physicochemical methods utilized in production (Raveendran et al. 2003). Plant extract-based nanoparticle synthesis is advantageous than using microorganisms, since it does not require multistep or complex procedures such as microorganism isolation, identification, growth optimization, culture preparation, and maintenance. Furthermore, plant-based synthesis is inexpensive, faster than using microorganisms, and easy to scale up for large-scale nanoparticle production (Li et al. 2011; Mittal et al. 2013; Iravani 2011). This review is concerned with the green synthesis of silver nanoparticles (AgNPs) and gold nanoparticles (AuNPs) using different plant extracts, giving a brief overview of the characterization methods and focusing on biological applications including antibacterial and anticancer activity against cancer cells and the possible mechanism of action.

Characterization of nanoparticles

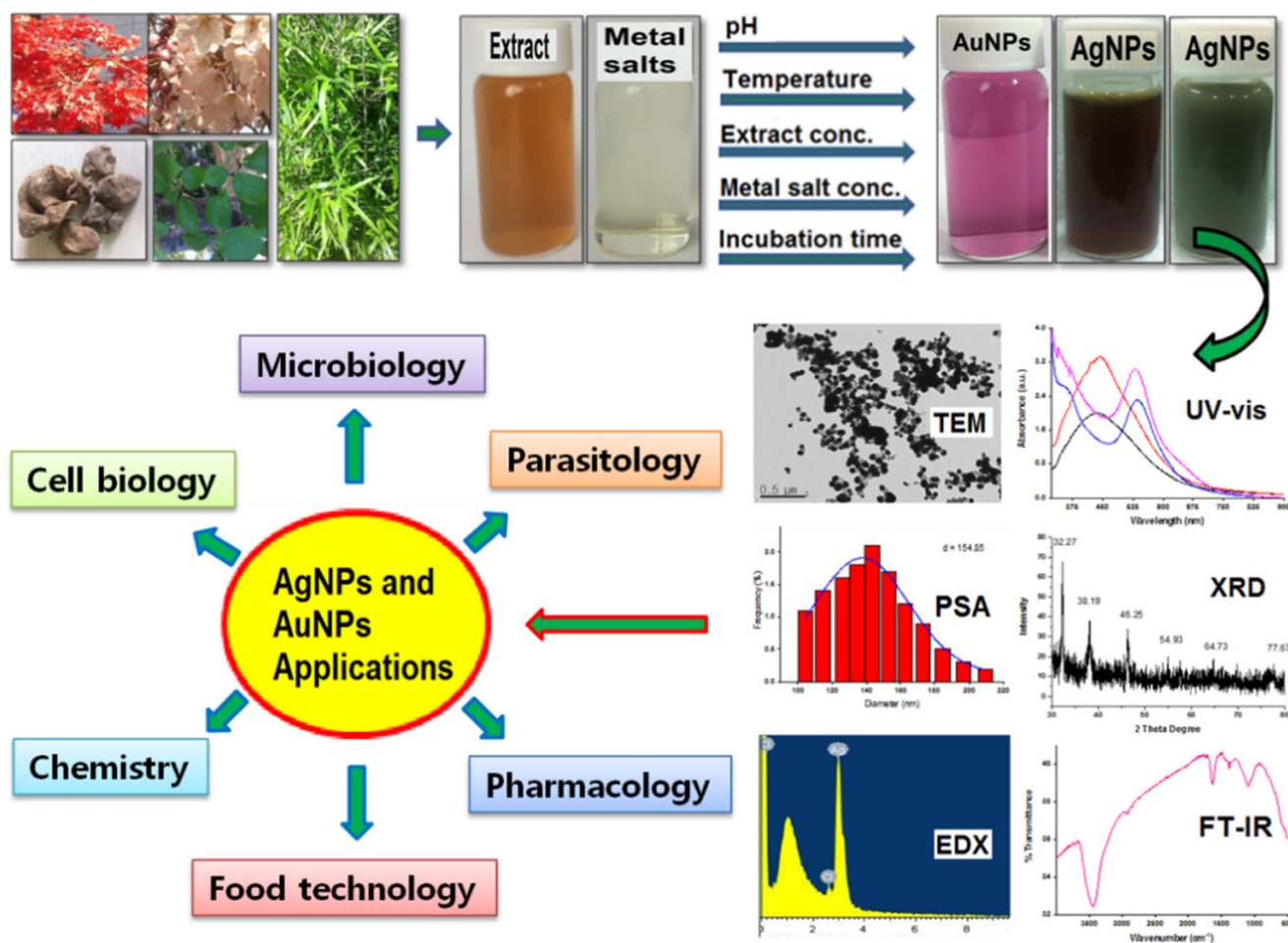
To date, there are many techniques employed for characterization of nanoparticles. However, the color change of reaction solution is the first qualitative indication for nanoparticle synthesis. The characteristic color change is from yellow to brown for AgNPs (Poinern 2014; Kuppusamy et al. 2015a, b) and yellow to deep red or purple for AuNPs (Lee et al. 2016; Malik et al. 2014) due to surface plasmon vibration. Nanoparticles are generally characterized based on their size, shape, and morphology; this is important for their applications. Usually, techniques applied in nanoparticle characterizations are ultraviolet-visible (UV-vis) spectrophotometer, dynamic light scattering (DLS), scanning electron microscopy (SEM), transmission electron microscopy (TEM), energy dispersive x-ray spectroscopy (EDX), X-ray powder diffraction (XRD), Fourier transform infrared spectroscopy (FT-IR), atomic force microscopy (AFM), particle size analysis (PSA), and Zeta potential measurement.

UV-vis spectrophotometer covers the UV range (190 to 380) and visible range (380 to 800); it is routinely applied for checking nanoparticle formation. Light wavelengths in between 300 and 800 are normally applied for nanoparticle (size range 2 to 100 nm) characterization (Poinern 2014). Spectrophotometric characteristic wavelength absorption peaks between 400–450 (Kuppusamy et al. 2015a, b; Banerjee et al. 2014) and 500–550 (MubarakAli et al. 2011; Singh and Srivastava 2015) are used for AgNPs and AuNPs, respectively. DLS is used to quantify surface charges and size distribution of nanoparticles suspended in a liquid (Tomaszewska et al. 2013; Patra et al. 2015). Microscopic

based techniques include SEM, TEM, and AFM and are extensively applied to visualize size, shape, and morphology of synthesized nanoparticles. Particularly, TEM analysis is for size and shape distribution (Klekotko et al. 2015; Ahmed and Ikram 2015), while SEM analysis is used to determine shape and morphology of nanoparticles (Kuppusamy et al. 2015a, b; Dhamecha et al. 2016). The elemental composition of silver and gold can be determined by EDX (Dubey et al. 2010a), while XRD is utilized for diffraction pattern identification and crystal structure of the nanoparticles (Nakkala et al. 2016; Verma et al. 2016). The Bragg's reflection of AgNPs by employing XRD is important for characterization and crystallographic planes specific for AgNPs are (111), (200), (220), (311), (331), and (222). Particle size analysis can be performed by TEM analysis and DLS (Tomaszewska et al. 2013; Patra et al. 2015; Hoshyar et al. 2016; Dubey et al. 2010b). FT-IR is used to investigate the functional groups of the plant extract involved in the salt reduction (silver nitrate and chloroauric acid) into its nanocrystalline form (Pugazhendhi et al. 2016; Geetha et al. 2013; Philip and Unni 2011). The zeta potential analysis provides information about the surface charges of nanoparticles, which affects their colloidal stability (Dubey et al. 2010a; Dubey et al. 2010b). Schematic presentation of nanoparticle synthesis using plant extracts with its characterization and applications in different fields is presented in Scheme 1.

Mechanism of nanoparticle synthesis

The plant extract acts as a reducing as well as capping or stabilizing agent in the green synthesis of AgNPs and AuNPs (Prabu and Johnson 2015; Ahmed et al. 2014; Nakkala et al. 2015; Mohammadi et al. 2016; Kumar et al. 2015). Chemically synthesized AgNPs need three things, namely silver nitrate, a reducing agent such as sodium borohydride, and a capping or stabilizing agent like polyvinyl alcohol for controlling the size of nanoparticles and preventing aggregation (Chung et al. 2016; Iravani et al. 2014). The nanoparticle synthesis process begins by mixing of plant extract with metal salt solution. Biochemical reduction of metal salt results in the change in reaction color (Mohapatra et al. 2015; Islam et al. 2015; Sathishkumar et al. 2016; Dwivedi and Gopal 2010). Different plants and plant parts are involved in nanoparticles synthesis. Plant extracts contain phytochemicals (Alkaloids, flavonoids, proteins, polysaccharides, cellulose, and phenolic compounds) and secondary metabolites, which are utilized for nanoparticle synthesis (Kuppusamy et al. 2015a, b; Mata et al. 2016; Patil et al. 2012). The variation in composition and concentration of reducing agents in plant extracts is responsible for different size, shape, and morphological nanoparticle synthesis (Lee et al. 2016; Dhand et al. 2016). The oxidation of hydroxyl group of polysaccharides to



Scheme 1 Schematic presentation of nanoparticle synthesis, characterization, and applications

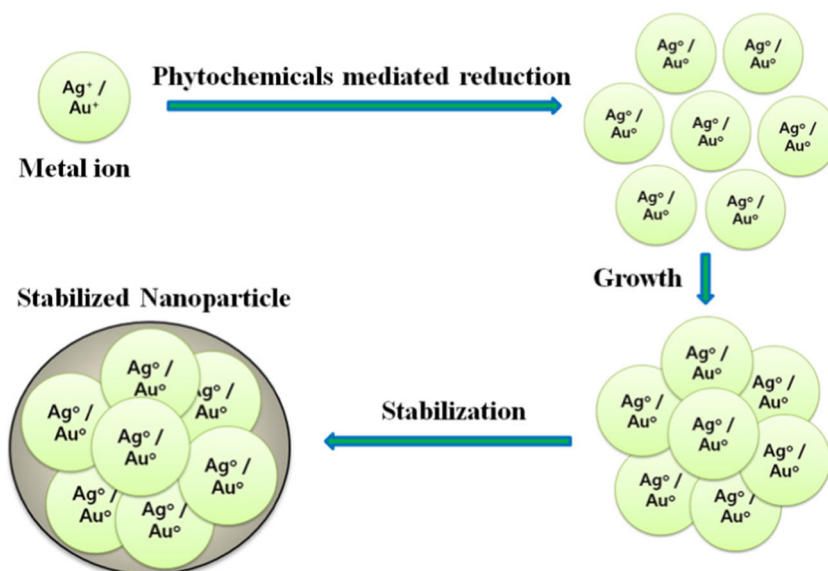
carboxyl group is most likely involved in the reduction of metal ions to produce metallic nanoparticles (Park et al. 2011). The initial activation of metal ions from their mono or divalent oxidation state to zero valent state and nucleation of the reduced metals atoms take place (Malik et al. 2014). A nanoparticle aggregates to form a variety of shapes such as sphere, spherical, cube, rods, hexagonal, triangle, pentagons, and wires (Akhtar et al. 2013; Naraginti et al. 2016). Researchers have reported significant influence of nanoparticle synthesis along with changes in size, shape, and morphology by changing synthesis parameters including pH, temperature, metal salt concentration, plant extract concentration, and reaction time (Mittal et al. 2013; Shah et al. 2015; Ghaffari-Moghaddam and Hadi-Dabanlou 2014). Figure 1 represents the mechanism for synthesis of nanoparticles in the presence of phytochemicals as a reducing agent. The nanoparticles are made up from smaller entities, like joining of atoms, molecules, and smaller particles (Mukherjee et al. 2001; Mittal et al. 2013); the nanostructure building blocks form first and assemble to form the final particle (Mittal et al. 2013; Shah et al. 2015). The synthesized nanoparticles were capped and stabilized by phytochemical components

(Dhamecha et al. 2016; Mohammadi et al. 2016; Mata et al. 2016; Shah et al. 2015).

Green synthesis of nanoparticles

The plant extract has been considered a green route and reliable method for the synthesis of silver and gold nanoparticles owing to its eco-friendly nature (Malik et al. 2014; Chung et al. 2016; Shah et al. 2015). In green synthesis of nanoparticles, a mixture of metal salt and plant extract is allowed to react at room temperature (Sadeghi and Gholamhoseinpoor 2015; Choudhary et al. 2016; Shankar et al. 2014). The reaction is completed within a few minutes to a few hours (Shankar et al. 2014; Devi et al. 2015). The AgNPs and AuNPs are successively synthesized in this way (Basavegowda and Lee 2013; Amooaghaie et al. 2015; Dong et al. 2014). Scheme 1 shows the flow of the procedure applied in nanoparticle synthesis, characterization, and application. Table 1 summarizes the list of plants reported for the synthesis of silver and gold nanoparticle. Leaves are most commonly used for nanoparticle synthesis; other plant parts

Fig. 1 Mechanism for synthesis of nanoparticles in the presence of phytochemicals as a reducing agent



like seed, bark, fruit, gum, aerial part, root, tuber, flower, and rhizome are utilized as well.

Use of plant extract in silver nanoparticle synthesis

In producing AgNPs using plant extract, the water extract of *Vigna radiata* seeds was reported to reduce silver ions to AgNPs at room temperature and spherical particles with sizes ranging from 5 to 30 nm were obtained (Choudhary et al. 2016). Sudhakar et al. have been able to synthesize spherical shaped AgNPs with different sizes using rhizome extract of *Acorus calamus* (Sudhakar et al. 2015).

Anisomeles indica leaf extract was reported to reduce silver ions to spherical AgNPs (50–100 nm) within 10 min at room temperature (Govindarajan et al. 2016), and extract of *Azadirachta indica* leaves was reported for synthesis of AgNPs with an average size of 34 nm (spherical shaped) (Ahmed et al. 2016). Krithiga et al. used leaf extract of *Clitoria ternatea* and *Solanum nigrum* for AgNP synthesis, spherical nanoparticles with sizes 20 and 28 nm, respectively, were recorded. It was found that as the reaction temperature increases, there is an increase in nanoparticle synthesis rate; also, different pH studies suggested that the formation of nanoparticles was suppressed in acidic conditions and enhanced in basic conditions (Krithiga et al. 2015). Kalaiyarasu et al. and Kharat et al. used leaf extract of *Digitaria radicola* and *Elephantopus scaber*, respectively, to produce spherical nanoparticles (Kalaiyarasu et al. 2016; Kharat and Mendhulkar 2016). *Lantana camara* leaf extract was applied to synthesize AgNPs; 20–34 nm homogenous spherical nanoparticles were obtained at room temperature in 10 min. This study concludes that as the plant extract concentration increases, there is a decrease in AgNP size (Ajitha et al. 2015). Spherical AgNPs with small size (5–15 nm) were

reported using leaf extract of *Salvinia molesta* (Verma et al. 2016). Leaf extract of *Terminalia arjuna* was used for a cost-effective bioreduction of silver ions to AgNPs without the need for a catalyst or temperature control (Ahmed and Ikram 2015). An aqueous extract of *Tinospora cordifolia* leaves was reported for production of silver nanoparticles (30 nm) (Selvam et al. 2016).

Saini et al. reported that the funicles of *Acacia auriculiformis* which were extracted using methanol and used for AgNPs synthesis showed that the desired shape and size of the nanoparticles were achieved by the optimization process which included the following parameters: frequency of ultrasound, temperature, plant extract, and metal solutions (Saini et al. 2016). While Velusamy et al. reported for the first time that the autoclave assisted aqueous extract of gum from *Azadirachta indica* was responsible for the formation of spherical AgNPs (12.09–29.65 nm) (Velusamy et al. 2015). It was also reported that polymorphic AgNPs, synthesized by using water-ethanol extracts from seeds of *Coffea arabica* with the size ranging from 20 to 30 nm, showed that the surface plasmon resonance (SPR) changed along with the change in the metal ions concentration (Dhand et al. 2016). Likewise, SPR changing with different concentrations of fruits extract from *Emblca officinalis* was observed by Ramesh et al.; they found that the SPR increased due to the increase in synthesized nanoparticle concentration (Ramesh et al. 2015).

A polydispersed spherical silver nanoparticle with an average size of 12 ± 6 nm was synthesized by using the latex of *Carica papaya* and latex containing NH_2 or OH groups of alcohol/phenol and aromatic C=C group of proteins which were found responsible for the reduction of silver nitrate into AgNPs by FT-IR (Chandrasekaran et al. 2016). Likewise, FT-IR studies of Rajkuberan et al. reported the involvement of N-

Table 1 Extracts from different plants and their parts in AgNP and AuNP eco-friendly synthesis with their shape, size, and applications

Plant name	Reducing agent	NPs	Size (nm)	Shape	Applications	Reference
<i>Acacia auriculiformis</i>	Methanol extract of funicles	Ag	15	Spherical and slightly irregular	Antifilaria activity against <i>Setaria cervi</i>	Saini et al. (2016)
<i>Anisomeles indica</i>	Water extract of leaves	Ag	50–100	Spherical	Mosquitocidal activity against <i>Anopheles subpictus</i> , <i>Aedes albopictus</i> , and <i>Culex tritaeniorhynchus</i> .	Govindarajan et al. (2016)
<i>Azadirachta indica</i>	Water extract of gum	Ag	12.09–29.65	Spherical	Antibacterial activity against <i>Salmonella enteritidis</i> and <i>Bacillus cereus</i> .	Velusamy et al. (2015)
<i>Azadirachta indica</i>	Water extract of leaves	Ag	34	Spherical	Antimicrobial activity against <i>Staphylococcus aureus</i> and <i>Escherichia coli</i> .	Ahmed et al. (2016)
<i>Bergenia ciliata</i>	Methanol extract of plant	Ag	35	Spherical	Antioxidant; antibacterial activity against <i>Micrococcus luteus</i> , <i>S. aureus</i> , <i>Bordetella bronchiseptica</i> , and <i>Enterobacter aerogenes</i> ; antifungal activity against <i>Aspergillus fumigatus</i> , <i>Fusarium solani</i> , <i>Aspergillus niger</i> , and <i>Aspergillus flavus</i> ; and cytotoxic activity against brine shrimp (<i>Artemia salina</i>); LD ₅₀ –33.92 µg/ml).	Phull et al. (2016)
<i>Clitoria ternatea</i>	Water extract of leaves	Ag	20	Spherical	Antibacterial activity against <i>Bacillus subtilis</i> , <i>S. aureus</i> , <i>Streptococcus pyogenes</i> , <i>E. coli</i> , <i>Pseudomonas aeruginosa</i> , and <i>Klebsiella aerogenes</i> .	Krithiga et al. (2015)
<i>Solanum nigrum</i>	Water extract of leaves	Ag	28	Spherical	Antibacterial activity against <i>S. aureus</i> and <i>E. coli</i>	Dhand et al. (2016)
<i>Coffea arabica</i>	Water: Ethanol extract of seeds	Ag	20–30	Polymorphic	Antioxidant, wound healing effect on albino Wistar rats.	Naraginti et al. (2016)
<i>Coleus forskohlii</i>	Water extract of root	Ag	5–15	Spherical		
		Au	5–18	Nanorods, prisms, and triangular		
<i>Curculigo orchioides</i>	Water extract of rhizome	Ag	15–18	Spherical	Antibacterial activity against <i>B. subtilis</i> , <i>E. coli</i> , <i>Klebsiella pneumoniae</i> , <i>P. vulgaris</i> , <i>P. aeruginosa</i> , <i>Salmonella typhimurium</i> , <i>S. aureus</i> , <i>Klebsiella oxytoca</i> , and <i>Morganella morganii</i> ; larvicidal activity against <i>A. subpictus</i> and <i>Culex quinquefasciatus</i> ; anticancer activity against MDA-MB-231, Vero cells (IC ₅₀ 18.86 and 42.43 µg/ml, respectively).	Kalayavizhi et al. (2016)
<i>Digitaria radicata</i>	Methanol extract of leaves	Ag	90	Spherical	Antioxidant, antibacterial activity against <i>S. aureus</i> and <i>E. coli</i> .	Kalayarasu et al. (2016)
<i>Dioscorea alata</i>	Water extract of tuber	Ag	10–20	Spherical, elongated	Antibacterial activity against <i>Staphylococcus auricularis</i> and <i>E. coli</i> .	Pugazhendhi et al. (2016)
<i>Diospyros paniculata</i>	Methanol extract of root	Ag	14–28	Spherical	Antimicrobial activity against (bacteria) <i>B. subtilis</i> , <i>S. aureus</i> , <i>S. pyogenes</i> , <i>K. pneumoniae</i> , <i>P. vulgaris</i> , <i>E. coli</i> , and <i>P. aeruginosa</i> as well as (fungi) <i>Penicillium notatum</i> , <i>A. flavus</i> , <i>A. niger</i> , <i>Candida albicans</i> , <i>Saccharomyces cerevisiae</i> .	Rao et al. (2016)
<i>Elephantopus scaber</i>	Water extract of leaves	Ag	11–100	Spherical	Antioxidant	Kharat and Mendhulkar (2016)
<i>Emblica officinalis</i>	Water extract of fruit	Ag	10–70	Spherical	Antibacterial activity against <i>S. aureus</i> , <i>B. subtilis</i> , <i>E. coli</i> , and <i>K. pneumoniae</i> .	Ramesh et al. (2015)

Table 1 (continued)

Green synthesis of silver (Ag) nanoparticles using plant extract						
Plant name	Reducing agent	NPs	Size (nm)	Shape	Applications	Reference
<i>Euphorbia antiquorum</i> L.	Latex extract	Ag	10–50	Spherical	Antimicrobial activity against <i>E. coli</i> , <i>K. pneumoniae</i> , <i>Proteus mirabilis</i> , <i>Vibrio cholerae</i> , <i>Enterococcus faecalis</i> ; larvicidal activity against <i>Aedes aegypti</i> and <i>C. quinquefasciatus</i> (LC ₅₀ 2.4 and 2.8 ppm, respectively); anticancer effect on HeLa cells (IC ₅₀ —28 µg/ml).	Rajkuberan et al. (2016)
<i>Ficus benghalensis</i>	Water extract of bark	Ag	40	Spherical	Antimicrobial activity against <i>B. subtilis</i> , <i>E. coli</i> , <i>P. aeruginosa</i> , and <i>V. cholerae</i> ; anticancer effect on MG-63 (IC ₅₀ 81.8 ± 2.6, 75.5 ± 2.4 µg/ml for <i>F. benghalensis</i> and <i>Azadirachta indica</i> mediated AgNPs, respectively).	Nayak et al. (2016)
<i>Azadirachta indica</i>	Water extract of bark	Ag	50	Spherical		
<i>Lantana camara</i>	Water extract of leaves	Ag	20–34	Spherical	Antibacterial activity against <i>E. coli</i> , <i>Staphylococcus</i> spp., <i>Pseudomonas</i> spp., and <i>Bacillus</i> spp.	Ajitha et al. (2015)
<i>Rhus chinensis</i>	Water extract of galls	Ag	150	Triangular and hexagonal	Antibacterial activity against <i>E. coli</i> , <i>P. aeruginosa</i> , <i>S. aureus</i> , and <i>Staphylococcus saprophyticus</i> .	Patil et al. (2016b)
<i>Salvinia molesta</i>	Water extract of leaves	Ag	5–15	Spherical	Antibacterial activity against <i>S. aureus</i> and <i>E. coli</i> .	Verma et al. (2016)
<i>Terminalia arjuna</i>	Water extract of leaf	Ag	3–50	Spherical and irregular	Antimicrobial activity against <i>S. aureus</i> and <i>E. coli</i> .	Ahmed and Ikram (2015)
<i>Terminalia cuneata</i>	Water extract of bark	Ag	25–50	Distorted spherical	Catalytic activity	Edison et al. (2016)
<i>Timospora cordifolia</i>	Water extract of leaves	Ag	30	Spherical	Antioxidant; antibacterial activity against <i>Klebsiella</i> spp., <i>Staphylococcus</i> spp., and <i>Bacillus</i> spp.	Selvam et al. (2016)
<i>Vigna radiata</i>	Water extract of seeds	Ag	5–30	Spherical	Antibacterial activity against <i>S. aureus</i> and <i>E. coli</i> .	Choudhary et al. (2016)
Green synthesis of gold (Au) nanoparticles using plant extract						
<i>Abutilon indicum</i>	Water extract of leaf	Au	1–20	Spherical	Antioxidant; anticancer activity on HT-29 cells (IC ₅₀ 210 and 180 µg/ml for 24 and 48 h, respectively).	Mata et al. (2016)
<i>Butea monosperma</i>	Water extract of leaf	Au	10–100	Spherical, rods, triangular, and hexagonal	Biocompatibility on normal cell lines (HUVAC, ECV-304) and anticancer effect on B16F10, MCF-7, and A549.	Patra et al. (2015)
<i>Carica papaya</i>	Water extract of Leaves	Au	15–28	Spherical and triangular	Antibacterial activity against <i>S. aureus</i> , <i>B. subtilis</i> , <i>E. coli</i> , and <i>P. vulgaris</i> ; anticancer effect on MCF7 and HepG2.	Muthukumar et al. (2016)
<i>Catharanthus roseus</i>	Water extract of Leaves	Au	15–28	Spherical and triangular		
<i>Cinnamomum zeylanicum</i>	water extract of Bark	Au	46.48	Spherical	Mosquitocidal activity against <i>Anopheles stephensi</i> and <i>C. quinquefasciatus</i> .	Soni and Prakash (2014)
<i>Citrus maxima</i>	Fruit juice	Au	25.7 ± 10	Rod and spherical	Catalytic activity	Yu et al. (2016)
<i>Couroupita guianensis</i> , <i>Aubl.</i>	Water extract of fruits	Au	25 ± 6	Spherical, triangular, and hexagonal	Antioxidant	Sathishkumar et al. (2016)
<i>Genipa americana</i>	Water extract of fruit	Au	15–40	Spherical and triangular	Antioxidant; cytotoxic activity against A549 and HeLa.	Kumar et al. (2016)
<i>Gynemna sylvestre</i>	Water extract of leaves	Au	72.8	Spherical	Anticancer effect on HT29 cells.	Arunachalam et al. (2014)
<i>Hibiscus sabdariffa</i>	Water extract of leaves	Au	10–60	Spherical	Anticancer effect on U87 cells.	Mishra et al. (2016)
<i>Hygrophila spinosa</i>	Water extract of leaf	Au	50–80	Triangular and spherical		Koperuncholan (2015)

Table 1 (continued)

Plant name	Reducing agent	NPs	Size (nm)	Shape	Applications	Reference
<i>Mentha piperita</i>	Water extract of leaves	Au	10–300	Spherical, triangular, and hexagonal	Antimicrobial activity against (bacteria) <i>Aeromonas liquefaciens</i> , <i>E. faecalis</i> , <i>K. pneumoniae</i> , <i>M. luteus</i> , <i>S. typhimurium</i> , and <i>V. cholerae</i> and (fungi) <i>C. albicans</i> , <i>Cryptococcus</i> spp., <i>Microsporium canis</i> , <i>Trichophyton rubrum</i> ; anticancer effect on HeLa cells (IC ₅₀ 2.5 and 4.5 µg/ml for 24 and 48 h, respectively). Cytotoxicity on Hek293.	Kleko et al. (2015)
<i>Moringa oleifera</i>	Water extract of leaves	Au	10–20	Spherical and polyhedral	Anticancer effect on A549 and SNO cells (IC ₅₀ 98.46 and 92.01 µg/ml, respectively).	Tiloke et al. (2016)
<i>Nepenthes khasiana</i>	Water extract of leaves	Au	125–180	Spherical and anisotropic	Biocompatibility on L929 cell lines.	Dhamecha et al. (2016)
<i>Nerium oleander</i>	Water extract of leaves	Au	2–10	Spherical	Antioxidant	Tahir et al. (2015)
<i>Ocimum sanctum</i>	Hexane extract of leaves	Au	1–50	Spherical, rods, and triangular	–	Lee et al. (2016)
	<i>n</i> -Butanol extract of leaves	Au	10–300	Irregular shape		
	Water extract of leaves	Au	50–300	Anisotropic		
	Chloroform extract of leaves	Au	> 200	Disk		
<i>Parkia roxburghii</i>	Water extract of leaves	Au	5–25	Spherical	Photocatalysis; antibacterial activity against <i>S. aureus</i> and <i>E. coli</i> .	Paul et al. (2016)
<i>Piper longum</i>	Water extract of fruits	Ag	5–25	Quasi-spherical	Antioxidant, catalytic.	Nakkala et al. (2016)
<i>Podophyllum hexandrum</i> L.	Water extract of leaves	Au	20–200	Spherical	Anticancer effect on HeLa (IC ₅₀ 20 µg/ml).	Jayaraj et al. 2014
<i>Rhus chinensis</i>	Water extract of galls	Au	5–35	Spherical		
<i>Stachys lavandulifolia</i> Vahl	Water extract of aerial part	Au	20–40	Oval and spherical	Cytotoxicity against MKN-28, Hep3B, and MG-63 cancer cells	Patil et al. (2016a)
<i>Tribulus terrestris</i>	Water extract of fruits	Au	34–80	Spherical, triangular, and hexagonal	–	Khademi-Azandehi and Moghaddam (2015)
	Water extract of fruits	Au	7 and 55	Anisotropic	Anti- <i>Helicobacter pylori</i> , cytotoxicity on AGS cells, and catalytic activity.	Gopinath et al. (2016)

H group, OH group, and C-C group of latex from *Euphorbia antiqorum* L. in the reduction of silver ions into nanoparticles (Rajkuberan et al. 2016). Same observations regarding the involvement of N-H group, OH group, and C-C group were reported for the AgNPs synthesis using mango peels extract along with the parameters pH, temperature and incubation time of reaction, concentration of extract, and silver nitrate being responsible for controlling nanoparticle size (Yang and Li 2013). Yang et al. concluded that as the pH (acidic to basic) and temperature increase (low to high), the reaction results in the shifting of wavelength towards shorter wavelength due to the formation of small nanoparticle nuclei which prevent secondary reduction. On the other hand, increasing the concentration of plant extract and silver nitrate resulting in the formation of bigger nanoparticles was observed in AgNPs synthesis by using mango peel extract and *Acacia* lignin extract. (Yang and Li 2013; Aadil et al. 2016).

Use of plant extract in gold nanoparticles synthesis

In producing gold nanoparticles using plant extract, water extract from leaves of *Abutilon indicum* was reported to reduce chloroauric acid to AuNPs. Mata et al. concluded that polyphenols were involved in the stabilization of AuNPs and found that the synthesized AuNPs are spherical with a size of 1–20 nm (Mata et al. 2016). *Butea monosperma* leaf extract-mediated AuNPs (10–100 nm) with triangular and hexagonal shapes were investigated and their element composition was detected by EDX (Patra et al. 2015). While a similar observation for the element composition by EDX was reported by Muthukumar et al. (2016), *C. papaya* leaf extract containing phytochemicals reduced chloroauric acid into AuNPs of spherical and triangular shapes with 15–28 nm size. Biosynthesis of AgNPs and AuNPs using *Cinnamomum zeylanicum* bark extract has been investigated and applied in mosquito control by Soni and Prakash (2014).

Citrus maxima fruit extract synthesized AuNPs with rod and spherical shapes, and an average size of 25.7 ± 10 nm was reported to be used for catalytic activity (Yu et al. 2016). The fruit extracts from *Couroupita guianensis*, Aubl. and *Genipa americana* were investigated for AuNPs synthesis; nanoparticles were polymorphic as detected by TEM analysis and had FCC crystalline structure as analyzed by XRD (Sathishkumar et al. 2016; Kumar et al. 2016). Green synthesis of polydispersed spherical AuNPs from different plants has been reported, among these are nanoparticles synthesized from *Nerium oleander* and *Podophyllum hexandrum* L. leaf extracts which are very small in size (2–10 and 5–35 nm, respectively) (Tahir et al. 2015; Jayaraj et al. 2014) and large-sized AuNPs (20–200 nm) synthesized by using *Piper longum* fruit extract (Nakkala et al. 2016). A similar synthesis of large nanoparticles by using leaves of *Mentha piperita* was reported which also explained the involvement of the plant

extract in the reduction of chloroauric acid into AuNPs with different shapes (spherical, triangular, and hexagonal) and sizes (10–300 nm) by Klekotko et al. (2015). Arunachalam et al. utilized water extract from leaves of *Gymnema sylvestris* for AuNPs formation and found that the resulting nanoparticles were spherical with an average size of 72.8 nm and SPR at 540 nm (Arunachalam et al. 2014). *Hibiscus sabdariffa* leaf extract-mediated AuNPs were reported to be spherical and small sized at around 10–60 nm (Mishra et al. 2016), after the following different parameters were utilized for optimized production: the acidic pH values 4.0–6.0, temperature at 100 °C, 0.45–1.0 mM metal salt with a reductant dilution factor of 200–300 and a reaction time of about 6 min, known to be suitable for maximum yield.

Green-synthesized nanoparticles vary in shapes and sizes due to different phytochemical compositions. A study on AuNPs biosynthesis by employing sequential fractional extract from leaves of *Ocimum sanctum* was reported by Lee et al. (2016). Different polarity solvents (hexane, chloroform, *n*-butanol and water) were utilized for the sequential fraction extraction from *O. sanctum* leaves, and it was concluded that differing solvent fractions (extract) are responsible for the synthesis of morphologically different AuNPs; chloroform extract produced a circular disk (>200 nm) shape with rough edges, hexane extract-produced spherical AuNPs 100 nm, water extract anisotropic nanoparticles, and *n*-butanol resulted in the aggregation of AuNPs (Lee et al. 2016).

The green synthesized AuNPs are polymorphic in nature due to the active involvement and interaction of different reducing groups present in plant extracts. The unique results from different studies shows that hydroxyl group (–OH) is the functional group most involved in AuNPs formation. The AuNPs formed from the leaf extract of *Moringa oleifera* and the extract itself were used in the FT-IR analysis for the investigation of functional group detection (Koperuncholan 2015), and it was concluded that the OH group is responsible for the reduction process. Likewise, Khademi-Azandehi et al. reported the involvement of OH and C=C groups from *Stachys lavandulifolia* Vahl leaf extract in nanoparticle formation. Similar observations regarding involvement of OH and C=O groups were reported in the AuNPs synthesis by using fruit extract from *Tribulus terrestris* (Gopinath et al. 2016).

Mechanisms of action

Green synthesis of AgNPs and AuNPs by using plant extracts gained significant interest over the years due to the remarkable antibacterial and anticancer properties of these nanoparticles. The excellent antibacterial activity of silver and anticancer activity of gold nanoparticles are mainly attributed to their high surface area-to-volume ratio which enables greater presence of atoms on the surface and, in turn, greater contact with

the environment. In addition to this, the nanosizes of these particles make penetration through cell membrane easier, interacting with intracellular materials and finally resulting in cell destruction in the process of multiplication. In the next section, we describe the possible mechanisms for the antibacterial activity of AgNPs and the anticancer activity of AuNPs.

Antibacterial approach of silver nanoparticles

AgNPs have been used in a wide range of antibacterial applications described in various studies, such as wound treatment (Nam et al. 2015), against clinical isolates (Kappusamy et al. 2015), nosocomial pathogens (Krithiga et al. 2015), food pathogens (Patil et al. 2016b), and many more; yet their mode of action has not been clearly explained. AgNPs obtained from various methods have been used in *in vitro* diagnosis of bactericidal activity against different bacterial species listed in Table 1. The antibacterial activity of AgNPs depends on the concentration of nanoparticles exposed to bacteria and the type of bacteria (Patil et al. 2016b). The bactericidal activity of AgNPs on gram-positive and gram-negative bacteria is different, but the superiority of one can be established over the other. There are mutually opposed reports about bactericidal activity against gram-positive and gram-negative bacteria. According to some research reports, gram-negative bacteria are found to be more sensitive to AgNPs compared with gram-positive bacteria (Kappusamy et al. 2015; Verma et al. 2016; Choudhary et al. 2016; Ramesh et al. 2015; Chandrasekaran et al. 2016; Kim et al. 2007); likewise, other researchers reported results in reverse (Kayalvizhi et al. 2016; Paul et al. 2016; Banerjee et al. 2014; Patil et al. 2016b). The varying results are due to different structural and molecular compositions of bacterial species (Kim et al. 2007) and also affected by the final inoculated concentration of bacteria, along with the concentration, size, and shape of AgNPs (Rai et al. 2012; Pal et al. 2007). The possible mechanisms of antibacterial activity of AgNPs are diagrammatically represented in Fig. 2.

The mechanism behind the antibacterial activity of AgNPs is quite complex and not well studied. Its mechanism is only explained provisionally. The smaller nanoparticles have more antibacterial activity due to them providing more surface exposure to the bacterial membrane (Rai et al. 2012; Pal et al. 2007). The positive charge of Ag^+ interacts with the negative charge on the cell wall of bacteria which leads to changes in cell wall morphology and increase in the cell permeability or leakage of the cell which consequently results in cell death (Patil et al. 2012; Dibrov et al. 2002). AgNPs have more affinity to interact with phosphorous and, sulfur-containing biomolecules present in extracellular (membrane protein), and intracellular components (DNA bases, protein); these biomolecules affect cell division, respiration, and ultimately, the survival of the cell. Other investigations have reported that the

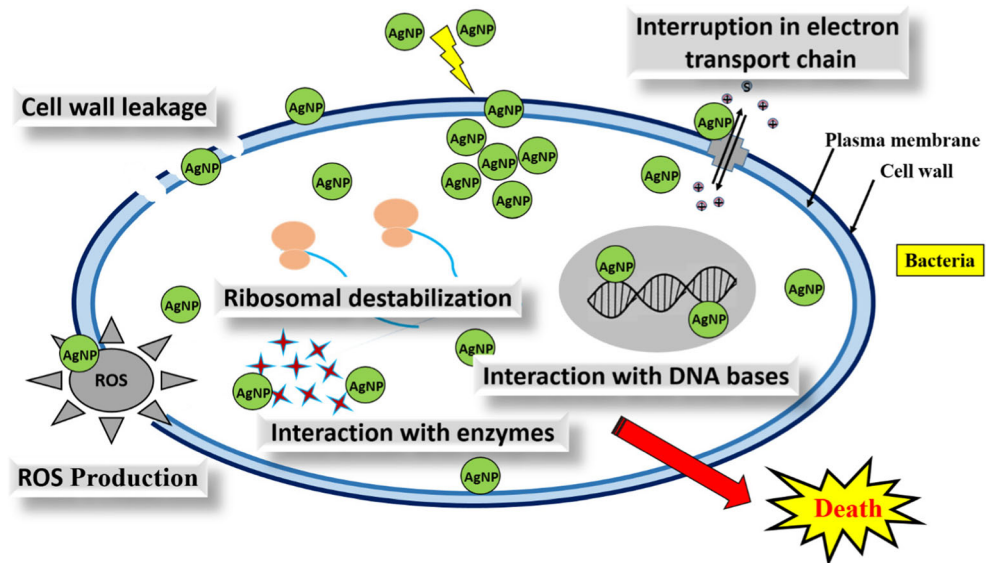
Ag^+ , which has an affinity for nitrogen and sulfur, can inhibit and disrupt protein structures by binding to thiol and amino groups (Choi et al. 2008). The interaction of nanoparticles with thiol group may be responsible for the induction of reactive oxygen species (ROS), which leads to the inhibition of respiratory enzymes and, consequently, death (Holt and Brad 2005; Ninganagouda et al. 2014). Silver ions act as an antibacterial by interacting with the peptidoglycan cell wall and plasma membrane (Radzig et al. 2013) and also prevent bacterial DNA replication by interfering with sulfhydryl groups in protein (Seth et al. 2011).

Anticancer approach of gold nanoparticles

Over the past decade, the investigation of inorganic nanoparticles for biomedical application becomes a fast growing area of research with great fascination. Distinct physicochemical properties of AuNPs make them ideal for biomedical applications. Green synthesis of AuNPs utilizes medicinally important plant extracts, which may remain on the surface of nanoparticles and, in such condition, AuNPs act as carriers. The cytotoxic activity of green synthesized AuNPs against different cancer cells is listed in Table 1. The use of AuNPs also minimizes the risk of side effects and limits the damage to normal (noncancerous) cells (Tiloke et al. 2016). AuNPs are a novel agent in cancer therapy and show aggregation (Cui et al. 2012) and size-dependent cytotoxic activity against different cancer cells (Pan et al. 2007; Cui et al. 2012) which also depends on the dose of nanoparticles (Raghunandan et al. 2011; Patil et al. 2016a). AuNPs are receiving significant attention from researchers because of their biocompatibility and unique property to conjugate with proteins (Fang et al. 2010). The possible mechanisms of anticancer activity for AuNPs are diagrammatically represented in Fig. 3.

The mechanism behind the anticancer activity of AuNP is quite complicated and not well understood. AuNPs are considered as a carrier for phytochemicals and may act as an anticancer agent. The mechanism behind its activity is only provisionally described. The interaction between AuNPs and cells differs in numerous ways; many researchers have reported the cellular internalization of AuNPs (Gong et al. 2015; Albanese et al. 2012). The surface properties of AuNPs are most important factor in internalization by cells. AuNPs carry positive charges, while cancer/normal cell membranes contain negatively charged materials like lipids (especially phosphate groups); having opposite charges is responsible for AuNPs uptake and internalization (Gong et al. 2015; Albanese et al. 2012). Another way for the entry of gold nanoparticles into cells is endocytosis, as shown by a study wherein tiny AuNPs were endocytosed and showed aggregation inside HeLa cells (Cui et al. 2012). The AuNPs showed cytotoxic activity via ROS production

Fig. 2 Mechanisms of antibacterial activity for AgNPs are diagrammatically representation



(Parida et al. 2014), by DNA damage (Patil et al. 2016a; Jayaraj et al. 2014; Mishra et al. 2016), and activation caspase cascade of apoptosis and mitochondrial dysfunctioning (Tiloke et al. 2016; Jayaraj et al. 2014).

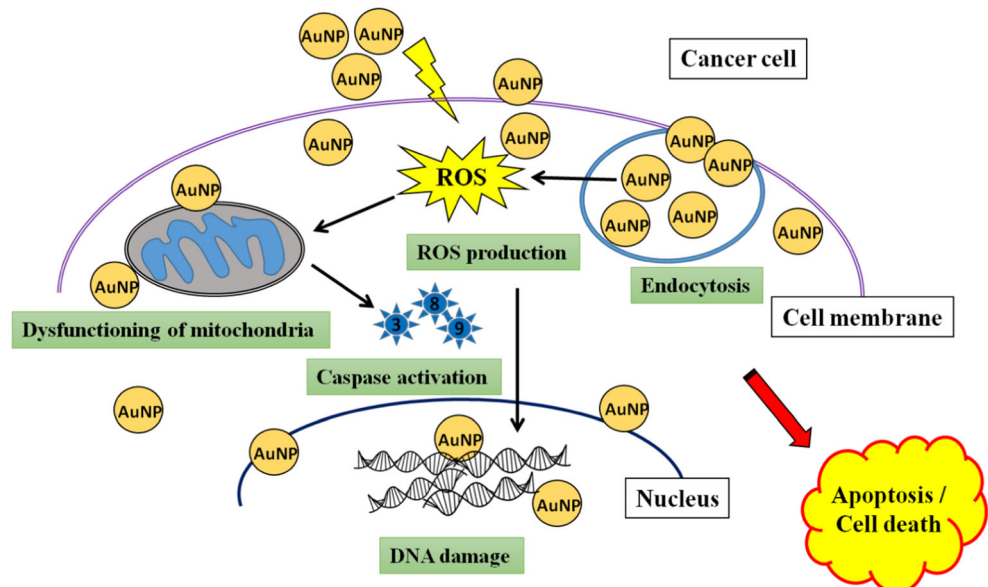
Syzygium aromaticum extract-mediated AuNPs were reported to be responsible for ROS production, dysfunctioning of mitochondria, and caspase-dependent apoptosis (Parida et al. 2014). *Rhus chinensis*-mediated AuNPs were applied on different cancer cells and showed cytotoxic activity by DNA damage (Patil et al. 2016a); similar results were observed on HeLa cells by using biologically synthesized (*P. hexandrum* L) AuNPs. Cell cycle arrest in G2/M phase and apoptosis by activation of caspase cascade including caspases 3, 8, and 9, were also

found (Jayaraj et al. 2014). Caspase-mediated apoptosis was also found in *M. oleifera*-mediated AuNPs in A549 cells by an increase in the activities of caspase-9 and caspase-3/7 and a significant decrease in the level of ATP, along with a significant increase of the protein concentration of p53 and Bax (Tiloke et al. 2016).

Conclusion and future prospects

The plant extract used for silver and gold nanoparticle syntheses is inexpensive, eco-friendly, simple, and easy to scale up. Nanoparticles synthesized by this method are more suitable for medicinal applications due to the

Fig. 3 Mechanisms of anticancer activity for AuNPs are diagrammatically representation



toxic chemical-free nature of synthesized nanoparticles, and simultaneously, the size, shape, and morphology of nanoparticles can be controlled by plant extract-based synthesis. The techniques utilized in nanoparticle characterization are not limited to those mentioned in this review. The most beneficial thing in green synthesis is the lack of necessity of additional agents for synthesis and stabilization of nanoparticles like reducing agent, capping agents, and stabilizing agents.

The major drawback of eco-friendly method by using plant extract in order to synthesize nanoparticles for commercially viable products is the anisotropic particle formation; plant extract contains more than one reducing agent which produces different size and shape of nanoparticles. The specific compound from plant extract involved in the synthesis of nanoparticle remains unclear. There is a need to find the exact component of the plant extract which is responsible for the reduction and stabilization of nanoparticles. The optimization studies reported controlling the high yield, shape, and size of nanoparticles, but there is a need to study about the stability of green synthesized nanoparticles. The significant different results were found during literature survey; the geographical different area, or environment conditions, and seasons were responsible for varying phytochemical compositions. This is the one major restriction to utilize plant extract for green synthesis technique, but this restriction may overcome by identify and investigating the biomolecules involve in reduction process. The antibacterial activity of silver nanoparticles is quite provisionally reported; there still exists a space to work on the molecular level, and it will be helpful to clear the picture for the mechanism of action of AgNPs. The anticancer or cytotoxic activity of AuNPs is quite clearly studied, but more studies are still required for finding its exact mechanism of action.

Research on eco-friendly synthesis of AgNPs and AuNPs is still ongoing, and many more applications may be studied like antimicrobial, antimosquito, antioxidant, catalytic, photocatalytic, and wound healing activities, which only add to the importance of nanoparticles. With the recent progress and ongoing works improving nanoparticle synthesis and exploring their applications, it is possible that nanoparticles will be the future alternatives to biomedical applications, pharmaceutical, therapeutics, and health cares.

Compliance with ethical standards This article does not contain any study with human participants or animals performed by any of the authors. All authors were involved in writing and have approved the submission of this manuscript.

Conflict of interest The authors declare that they have no conflict of interest.

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