Synthesis of metallic nanoparticles using plant extracts

Amit Kumar Mittal a, Yusuf Chisti b, Uttam Chand Banerjee a,⁎

a Department of Pharmaceutical Technology (Biotechnology), National Institute of Pharmaceutical Education and Research, Sector-67, SAS Nagar, 160062 Punjab, India
b School of Engineering, Massey University, Private Bag 11 222, Palmerston North, New Zealand

ABSTRACT

Biomolecules present in plant extracts can be used to reduce metal ions to nanoparticles in a single-step green synthesis process. This biogenic reduction of metal ion to base metal is quite rapid, readily conducted at room temperature and pressure, and easily scaled up. Synthesis mediated by plant extracts is environmentally benign. The reducing agents involved include the various water soluble plant metabolites (e.g. alkaloids, phenolic compounds, terpenoids) and co-enzymes. Silver (Ag) and gold (Au) nanoparticles have been the particular focus of plant-based syntheses. Extracts of a diverse range of plant species have been successfully used in making nanoparticles. In addition to plant extracts, live plants can be used for the synthesis. Here we review the methods of making nanoparticles using plant extracts. Methods of particle characterization are reviewed and potential applications of the particles in medicine are discussed.

© 2013 Elsevier Inc. All rights reserved.

Biomolecules present in plant extracts can be used to reduce metal ions to nanoparticles in a single-step green synthesis process. This biogenic reduction of metal ion to base metal is quite rapid, readily conducted at room temperature and pressure, and easily scaled up. Synthesis mediated by plant extracts is environmentally benign. The reducing agents involved include the various water soluble plant metabolites (e.g. alkaloids, phenolic compounds, terpenoids) and co-enzymes. Silver (Ag) and gold (Au) nanoparticles have been the particular focus of plant-based syntheses. Extracts of a diverse range of plant species have been successfully used in making nanoparticles. In addition to plant extracts, live plants can be used for the synthesis. Here we review the methods of making nanoparticles using plant extracts. Methods of particle characterization are reviewed and potential applications of the particles in medicine are discussed.

© 2013 Elsevier Inc. All rights reserved.

Contents

1. Introduction .......................................................................................................................... 346
2. Characterization of nanoparticles ......................................................................................... 347
3. Synthesis of nanoparticles ..................................................................................................... 347
4. Use of plant extracts in nanoparticle synthesis ....................................................................... 347
5. Applications of nanoparticles ................................................................................................. 352
6. Concluding remarks ............................................................................................................... 353
Acknowledgment ................................................................................................................... 353
References .................................................................................................................................. 353

1. Introduction

This review is concerned with the synthesis of metallic nanoparticles using plant extracts. The methods used in characterizing the nanoparticles are outlined and the emerging applications of nanoparticles in clinical diagnostics and therapy (Azzazy et al., 2012; Chen et al., 2012; Doria et al., 2012; Fortina et al., 2007; Larguinho and Baptista, 2012; Sahoo et al., 2007; Salata, 2004; Seil and Webster, 2012; Wagner et al., 2006; Youns et al., 2011; Zhang et al., 2008) are discussed.

Although nanoparticles can be made using various physicochemical methods (Cao, 2004; Sepeur, 2008), their synthesis using nontoxic and environmentally benign biological methods is attractive specially if they are intended for invasive applications in medicine. Several routes have been developed for biological or biogenic synthesis of nanoparticles from salts of the corresponding metals (Bar et al., 2009; Dhillon et al., 2012; Duran and Seabra, 2012; Gan and Li, 2012; Gericke and Pinches, 2006; Korbekandi et al., 2009; Luangpipat et al., 2011; Mohanpura et al., 2008; Mukherjee et al., 2001; Parsons et al., 2007; Ray et al., 2011; Shankar et al., 2003, 2004; Singaravelu et al., 2007; Thakkar et al., 2010). Microorganisms, whole plants, plant tissue and fruits, plant extracts and marine algae (Luangpipat et al., 2011; Rajesh et al., 2012; Singaravelu et al., 2007) have been used to produce nanoparticles.

Biogenic synthesis is useful not only because of its reduced environmental impact (Anastas and Zimmerman, 2007; Dahl et al., 2007; Shankar et al., 2004) compared with some of the physicochemical production methods, but also because it can be used to produce large quantities of nanoparticles that are free of contamination and have a
well-defined size and morphology (Hutchison, 2008). Biosynthetic routes can actually provide nanoparticles of a better defined size and morphology than some of the physicochemical methods of production (Raveendran et al., 2003).

The ability of plant extracts to reduce metal ions has been known since the early 1900s, although the nature of the reducing agents involved was not well understood. In view of its simplicity, the use of live plants or whole plant extract and plant tissue for reducing metal salts to nanoparticles has attracted considerable attention within the last 30-years (Ankamwar, 2010; Armendariz et al., 2004; Beattie and Haverkamp, 2011; Gan and Li, 2012; Gardea-Torresdey et al., 2003; Gercke and Pinches, 2006; Haverkamp and Marshall, 2009; Iravani, 2011; Kandasamy et al., 2012; Kumar and Yadav, 2009; Marshall et al., 2007; Park et al., 2011; Parsons et al., 2007).

Compared with the use of plant extracts for making nanoparticles is simpler. Plant extract mediated synthesis is an increasing focus of attention (Ali et al., 2011; Ankamwar, 2010; Babu and Prabu, 2011; Banerjee, 2011; Bankar et al., 2010; Bar et al., 2009; Baskaralingam et al., 2012; Castro et al., 2011; Chandran et al., 2006; Daisy and Saipriya, 2012; Dubey et al., 2009, 2010a; Kaler et al., 2011; Kesharwani et al., 2009; Lee et al., 2011; Park et al., 2011; Parsons et al., 2007; Singh et al., 2010; Song et al., 2009). Processes for making nanoparticles using plant extracts are readily scalable and may be less expensive (Iravani, 2011) compared with the relatively expensive methods based on microbial processes (Dhillon et al., 2012; Li et al., 2011; Luangpipat et al., 2011; Sastry et al., 2003) and whole plants (Armendariz et al., 2004; Beattie and Haverkamp, 2011; Haverkamp and Marshall, 2009; Kumar and Yadav, 2009; Marshall et al., 2007).

Plant extracts may act both as reducing agents and stabilizing agents in the synthesis of nanoparticles (Kumar and Yadav, 2009). The source of the plant extract is known to influence the characteristics of the nanoparticles (Kumar and Yadav, 2009). This is because different extracts contain different concentrations and combinations of organic reducing agents (Mukunthan and Balaji, 2012). Typically, a plant extract-mediated bioreduction involves mixing the aqueous extract with an aqueous solution of the relevant metal salt. The reaction occurs at room temperature and is generally complete within a few minutes. In view of the number of different chemicals involved, the bioreduction process is relatively complex. Nanoparticles are already used in numerous applications (Virkutyte and Varma, 2011) including in vitro diagnostics, but their use in medicine is mostly on an experimental basis. Drugs bound to nanoparticles have been claimed to have advantages compared with the conventional forms of the drugs (Wagner et al., 2006). Nanoparticle bound drugs have an extended half-life in vivo, longer circulation times and can convey high concentrations of a potent drug to where it is needed (Sahoo et al., 2007). The size of the drug nanoparticle and its surface characteristics can be modified to achieve the desired delivery characteristics (Mohanraj and Chen, 2007). As the nanoparticle-bound drug is not able to circulate broadly, its side effects are reduced and a high localized concentration can be achieved where it is needed (Panyam and Labhasetwar, 2003). In view of the large surface area per unit mass of nanoparticles, the drug loading can be relatively high (Han et al., 2007). Nanoparticle-bound drugs are easily suspended in liquids and are able to penetrate deep in organs and tissues.

2. Characterization of nanoparticles

Nanoparticles are generally characterized by their size, shape, surface area, and dispersity (Jiang et al., 2009). A homogeneity of these properties is important in many applications. The common techniques of characterizing nanoparticles are as follows: UV–visible spectrophotometry, dynamic light scattering (DLS), scanning electron microscopy (SEM), transmission electron microscopy (TEM), Fourier transform infrared spectroscopy (FTIR), powder X-ray diffraction (XRD) and energy dispersive spectroscopy (EDS) (Feldheim and Foss, 2002; Sepeur, 2008; Shahverdi et al., 2011).

The UV–visible spectroscopy is a commonly used techniques (Pal et al., 2007). Light wavelengths in the 300–800 nm are generally used for characterizing various metal nanoparticles in the size range of 2 to 100 nm (Feldheim and Foss, 2002). Spectrophotometric absorption measurements in the wavelength ranges of 400–450 nm (Huang and Yang, 2004) and 500–550 nm (Shankar et al., 2004) are used in characterizing the silver and gold nanoparticles, respectively. The dynamic light scattering (DLS) is used to characterize the surface charge and the size distribution of the particles suspended in a liquid (Jiang et al., 2009).

Electron microscopy is another commonly used method of characterisation (Cao, 2004). Scanning electron microscopy and transmission electron microscopy are used for morphological characterization at the nanometer to micrometer scale (Schaffer et al., 2009). The transmission electron microscopy has a 1000-fold higher resolution compared with the scanning electron microscopy (Eppler et al., 2000). FTIR spectroscopy is useful for characterizing the surface chemistry (Chithrani et al., 2006). Organic functional groups (e.g. carbonyls, hydroxyls) attached to the surface of nanoparticles and the other surface chemical residues are detected using FTIR.

XRD is used for the phase identification and characterization of the crystal structure of the nanoparticles (Sun et al., 2000). X-rays penetrate into the nanomaterial and the resulting diffraction pattern is compared with standards to obtain structural information. Elemental composition of metal nanoparticles is commonly established using energy dispersive spectroscopy (EDS) (Strasser et al., 2010).

3. Synthesis of nanoparticles

The methods for making nanoparticles can generally involve either a “top down” approach or a “bottom up” approach (Sepeur, 2008). In top-down synthesis (Fig. 1), nanoparticles are produced by size reduction from a suitable starting material (Meyers et al., 2006). Size reduction is achieved by various physical and chemical treatments (Fig. 1). Top down production methods introduce imperfections in the structure of the product and this is a major limitation because the surface chemistry and the other physical properties of nanoparticles are highly dependent on the surface structure (Thakkar et al., 2010).

In bottom up synthesis, the nanoparticles are built from smaller entities, for example by joining atoms, molecules and smaller particles (Mukherjee et al., 2001). In bottom up synthesis, the nanostructured building blocks of the nanoparticles are formed first and then assembled to produce the final particle (Thakkar et al., 2010). The bottom up synthesis mostly relies on chemical and biological methods of production. The probable mechanism of nanoparticle synthesis by bottom up approach is shown in Fig. 2.

Of the biological methods of synthesis, the methods based on microorganisms have been widely reported (Dhillon et al., 2012; Gercke and Pinches, 2006; Kaler et al., 2011; Korbekandi et al., 2009; Li et al., 2011; Luangpipat et al., 2011; Mohanpura et al., 2008; Sanghi and Verma, 2010; Sastry et al., 2003). Microbial synthesis is of course readily scalable, environmentally benign and compatible with the use of the product for medical applications, but production of microorganisms is often more expensive than the production of plant extracts. Plant mediated nanoparticle synthesis using whole plant extract or by living plant were also reported in literature (Gardea-Torresdey et al., 2003; Park et al., 2011).

4. Use of plant extracts in nanoparticle synthesis

In producing nanoparticles using plant extracts, the extract is simply mixed with a solution of the metal salt at room temperature. The reaction is complete within minutes. Nanoparticles of silver, gold
and many other metals have been produced this way (Li et al., 2011). Fig. 3 shows picture of various plants used for the biosynthesis of nanoparticles. The nature of the plant extract, its concentration, the concentration of the metal salt, the pH, temperature and contact time are known to affect the rate of production of the nanoparticles, their quantity and other characteristics (Dwivedi and Gopal, 2010). Table 1 summarizes some of the reports pertaining to nanoparticle synthesis mediated by extracts of various plants.

Synthesis of silver nanoparticles using a leaf extract of *Polyalthia longifolia* was reported by Prasad and Elumalai (2011). An average particle size of about 58 nm was obtained. Silver and gold ions could be reduced to nanoparticles using a leaf extract of *Cinnamomum camphora* (Huang et al., 2007). The reduction was ascribed to the phenolics, terpenoids, polysaccharides and flavones compounds present in the extract. These nanoparticles were found to have a peak bactericidal activity at a concentration of 45 μg/mL (Huang et al., 2007). They were most active against the yeast *Candida albicans*. Saxena et al. (2011) used an aqueous extract of *Ficus benghalensis* leaves to produce silver nanoparticles that had an average diameter of 16 nm.

**Fig. 1.** Various approaches for making nanoparticles and cofactor dependent bioreduction.

**Fig. 2.** Mechanisms of nanoparticle synthesis (M⁺-metal ion).
In synthesis of silver nanoparticles using a geranium (Pelargonium graveolens) leaf extract, the particles formed rapidly and a stable size of 16–40 nm was achieved (Shankar et al., 2003). Safaepour et al. (2009) used geraniol (C_{10}H_{18}O), a natural monoterpene alcohol found in some plants, and silver nitrate to produce uniformly shaped silver nanoparticles in the size range of 1–10 nm. These nanoparticles were found to inhibit the growth of fibrosarcoma Wehi 164 cancer cell line in vitro by up to 60% at a concentration of 5 μg/mL (Safaepour et al., 2009). Kaviya et al. (2011a) reported the synthesis if silver nanoflakes using leaf extract of Crossandra infundibuliformis.

Reduction of silver ions to nanoparticles using extract of Desmodium trifolium was ascribed to the presence of H⁺ ions, NAD⁺ and ascorbic acid in the extract (Ahmad et al., 2010). Synthesis of highly stable silver nanoparticles (16–40 nm) using leaf extract of Datura metel has been reported (Kesharwani et al., 2009). The extract contained alkaloids, proteins, enzymes, amino acids, alcoholic compounds, and polysaccharides which were said to be responsible for the reduction of the silver ions to nanoparticles (Kesharwani et al., 2009). Quinol and chlorophyll pigments present in the extract also contributed to the reduction of silver ions and stabilization of the nanoparticles. Fig. 4 shows the probable chemical constituents present in the plant extract responsible for the bioreduction of metal ions, their growth and stabilization.

A tuber extract of Dioscorea bulbifera was used to produce gold and silver nanoparticles of various shapes (Ghosh et al., 2011, 2012). These nanoparticles in combination with antibiotics were found to have a synergistic antibacterial activity against test microorganisms, particularly against Pseudomonas aeruginosa, Escherichia coli and Acinetobacter baumanii (Ghosh et al., 2012). Use of antibodies in combination with silver nanoparticles has been reported also for effective control of otherwise antibiotic-resistant microorganisms.

Sukirtha et al. (2011) synthesized silver nanoparticles using a leaf extract of Melia azedarach and showed them to be active against the HeLa cervical cancer cell line. Eclipta prostrata leaf extract-mediated synthesis of silver nanoparticles has been reported (Jha et al., 2009). Silver nanoparticles made using leaf extract of E. prostrata were reported to have larvicidal activity against filariasis and malaria vectors (Rajakumar and Abdul Rahuman, 2011). Synthesis of silver nanoparticles using a methanolic extract of Eucalyptus hybrid (safeda) leaves has been reported (Dubey et al., 2009). Flavonoid and terpenoid compounds present in the extract were claimed to be responsible for the stabilization of nanoparticles.

Cinnamomum zeylanicum bark is rich in linalool, methyl chavicol and eugenol and these compounds were said to be responsible for the bioreduction of silver and palladium ions to corresponding nanoparticles in separate experiments (Sathishkumar et al., 2009a,b). The protein from C. zeylanicum bark helped to stabilize the synthesized silver nanoparticles (Sathishkumar et al., 2009b).

Jacob et al. (2011) reported the synthesis of silver nanoparticles using Piper longum leaf extracts. The particles had a uniform spherical shape and ranged in size from about 18 to 41 nm. These nanoparticles were found to have a significant cytotoxic effect on HeP-2 cancer cells. Panda et al. (2011) reported the synthesis of silver nanoparticles using a broth prepared from the aromatic spath of male inflorescence of screw pine (Pandanus odorifer Forssk). These silver nanoparticles were less cytotoxic in comparison with Ag⁺ ion, but were more genotoxic in an assay based on onion plant cells.

Banerjee (2011) used an extract of Syzygium cumini (jambul) seeds to produce silver nanoparticles. The seed extract had antioxidant properties in vitro. The nanoparticles formed using the extract were found to have higher antioxidant activity compared with the seed extract. This may have been due to a preferential adsorption of the antioxidant material from the extract onto the surface of the nanoparticles.

An extract of Ocimum sanctum leaves was reported to reduce silver ions to nanoparticles (4–30 nm) within 8 min (Mallikarjun et al., 2011). This high activity of the extract was ascribed to the relatively high levels of ascorbic acid contained in the extract. In other studies, silver nanoparticles produced using O. sanctum leaf extracts have been found to have a high antimicrobial activity against both Gram-negative (E. coli) and Gram-positive (Streptococcus aureus) microorganisms (Singhal et al., 2011).

Extract of banana (Musa paradisiaca) peels has been used to produce silver nanoparticles (Bankar et al., 2010). These nanoparticles displayed antifungal activity against the yeasts C. albicans and Candida lipolytica, and antibacterial activity against E. coli, Shigella sp., Klebsiella sp. and Enterobacter aerogenes. Ahmad et al. (2010) used extracts of the legume Desmodium triforum to synthesize silver nanoparticles.
in the size range of 5–20 nm. Ali et al. (2011) reported the synthesis of silver and gold nanoparticles using extracts of Mentha piperita (peppermint) plant. These nanoparticles had antibacterial activity against clinically isolated human pathogens such as E. coli and S. aureus.

Prathna et al. (2011a) reported the synthesis of silver nanoparticles using an extract of Azadirachta indica leaves and a solution of silver nitrate. Increasing the reaction time from 30 min to 4 h resulted in a progressive increase in the particle size from around 10 nm to around 35 nm (Prathna et al., 2011a). Prathna et al. (2011b) synthesized 50 nm sized silver spherical nanoparticles using juice of Citrus limon (lemon). The nanoparticles were produced by incubating the juice (20 g/L citric acid, 5 g/L ascorbic acid) for 4 h with $10^{-2}$ M silver nitrate solution such that the ratio of the juice to salt solution was 4:1 by volume. Citric acid present in the juice was the principal reducing agent.

Table 1

<table>
<thead>
<tr>
<th>Plant</th>
<th>Type of nanoparticle</th>
<th>Size and shape</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acalypha indica</td>
<td>Ag</td>
<td>20–30 nm; spherical</td>
<td>Krishnaraj et al. (2010)</td>
</tr>
<tr>
<td>Allium sativum (garlic clove)</td>
<td>Ag</td>
<td>4–22 nm; spherical</td>
<td>Ahamed et al. (2011)</td>
</tr>
<tr>
<td>Aloe vera</td>
<td>Au, Ag</td>
<td>50–350 nm; spherical, triangular</td>
<td>Chandran et al. (2006)</td>
</tr>
<tr>
<td>Aloe vera (Aloe barbadensis Miller)</td>
<td>Indium oxide</td>
<td>5–50 nm; spherical</td>
<td>Maersiri et al. (2008)</td>
</tr>
<tr>
<td>Anacardium occidentale</td>
<td>Au/Ag bimetallic</td>
<td>≤6 nm at 27 °C; 17 nm at 100 °C</td>
<td>Sheny et al. (2011)</td>
</tr>
<tr>
<td>Apiniextracted from henna (Lawsonia inermis) leaves</td>
<td>Au, Ag</td>
<td>7.5–65 nm; spherical, triangular, quasi-spherical</td>
<td>Kasthuri et al. (2009b)</td>
</tr>
<tr>
<td>Azadirachta indica (neem)</td>
<td>Ag/Au bimetallic</td>
<td>50–100 nm</td>
<td>Shankar et al. (2004)</td>
</tr>
<tr>
<td>Boswellia ovalifoliolata</td>
<td>Ag</td>
<td>30–40 nm</td>
<td>Ankanna et al. (2010)</td>
</tr>
<tr>
<td>Calotropis procera</td>
<td>Ag</td>
<td>150–1000 nm</td>
<td>Babu and Prabu (2011)</td>
</tr>
<tr>
<td>Camellia sinensis</td>
<td>Ag, Au</td>
<td>30–40 nm</td>
<td>Vijhish-Nestor et al. (2008)</td>
</tr>
<tr>
<td>Carica papaya</td>
<td>Ag</td>
<td>25–50 nm</td>
<td>Jain et al. (2009)</td>
</tr>
<tr>
<td>Catharanthus roseus</td>
<td>Ag</td>
<td>48–67 nm</td>
<td>Kanna et al. (2011); Ponrulselsvan et al. (2012)</td>
</tr>
<tr>
<td>Chenopodium album</td>
<td>Au, Ag</td>
<td>10–30 nm; quasi-spherical shape</td>
<td>Dwivedi and Gopal (2010)</td>
</tr>
<tr>
<td>Cinnamomum camphora</td>
<td>Ag, Au</td>
<td>55–80 nm</td>
<td>Huang et al. (2007)</td>
</tr>
<tr>
<td>Cinnamomum camphora</td>
<td>Au, Pd</td>
<td>3.2–20 nm; cubic hexagonal crystalline</td>
<td>Yang et al. (2010)</td>
</tr>
<tr>
<td>Citrus sinensis peel</td>
<td>Ag</td>
<td>35±2 nm (at 25 °C), 10±1 nm (at 60 °C); spherical</td>
<td>Kaviya et al. (2011b)</td>
</tr>
<tr>
<td>Coleus amboinicus Lour</td>
<td>Ag</td>
<td>25.8±0.8 nm</td>
<td>Subramanian (2012)</td>
</tr>
<tr>
<td>Coleus aromaticus</td>
<td>Ag</td>
<td>44 nm</td>
<td>Vanaja and Annadurai (2012)</td>
</tr>
<tr>
<td>Curcuma longa</td>
<td>Ag</td>
<td></td>
<td>Sathishkumar et al. (2010)</td>
</tr>
<tr>
<td>Cymbopogon sp. (lemongrass)</td>
<td>Au</td>
<td>200–500 nm; spherical, triangular</td>
<td>Kesharwani et al. (2009)</td>
</tr>
<tr>
<td>Datura metel</td>
<td>Ag</td>
<td>16–40 nm; quasi-linear superstructures</td>
<td>Ahmmad et al. (2010)</td>
</tr>
<tr>
<td>Desmodium triflorum</td>
<td>Ag</td>
<td>5–20 nm</td>
<td>Ahmmad et al. (2010)</td>
</tr>
<tr>
<td>Diopros kaki</td>
<td>Pt</td>
<td>15–19 nm</td>
<td>Song et al. (2010)</td>
</tr>
<tr>
<td>Dioscorea bulbifera</td>
<td>Au</td>
<td>11–30 nm spheres</td>
<td>Ghosh et al. (2011)</td>
</tr>
<tr>
<td>Eclipta prostrata</td>
<td>Ag</td>
<td>35–60 nm; triangles, pentagons, hexagons</td>
<td>Rajakumar and Abdul Rahuman (2011)</td>
</tr>
<tr>
<td>Emblica officinalis</td>
<td>Ag, Au</td>
<td>10–20 nm Ag; 15–25 nm Au</td>
<td>Ankamwar et al. (2005)</td>
</tr>
<tr>
<td>Eucalyptus hybrid</td>
<td>Ag</td>
<td>50–150 nm</td>
<td>Dubey et al. (2009)</td>
</tr>
<tr>
<td>Euphorbiaeae latex</td>
<td>Cu/Ag</td>
<td>18 nm Ag, 10.5 nm Cu</td>
<td>Patil et al. (2012b); Valodkar et al. (2011)</td>
</tr>
<tr>
<td>Garcinia mangostana (mangosteen leaf)</td>
<td>Ag</td>
<td>35 nm</td>
<td>Veerasamy et al. (2010)</td>
</tr>
<tr>
<td>Gelidiella agrestis</td>
<td>Ag</td>
<td>22 nm</td>
<td>Vivek et al. (2011)</td>
</tr>
<tr>
<td>Jatropha curcas L. taxt</td>
<td>Pb</td>
<td>10–12.5 nm</td>
<td>Joglekar et al. (2011)</td>
</tr>
<tr>
<td>Memecylon edule</td>
<td>Ag, Au</td>
<td>20–50 nm; triangular, circular, hexagonal</td>
<td>Elavazhagan and Arunachalam (2011)</td>
</tr>
<tr>
<td>Melia azedarach</td>
<td>Ag</td>
<td></td>
<td>Sukirti et al. (2011)</td>
</tr>
<tr>
<td>Mentha piperita (peppermint)</td>
<td>Ag, Au</td>
<td>5–150 nm; spherical</td>
<td>Ali et al. (2011); Parashar et al. (2009a)</td>
</tr>
<tr>
<td>Moringa oleifera</td>
<td>Ag</td>
<td>57 nm</td>
<td>Prasad and Elumalai (2011)</td>
</tr>
<tr>
<td>Mucuna priuens</td>
<td>Au</td>
<td>6–17.7 nm; spherical</td>
<td>Arulkumar and Sabesan (2010)</td>
</tr>
<tr>
<td>Musa paradisaic</td>
<td>Ag</td>
<td>20 nm</td>
<td>Bankar et al. (2010)</td>
</tr>
<tr>
<td>Nelumbo nucifera (lotus)</td>
<td>Ag</td>
<td>25–80 nm; spherical, triangular</td>
<td>Santhoshkumar et al. (2011)</td>
</tr>
<tr>
<td>Parthenium leaf</td>
<td>Au</td>
<td>50 nm; face-centered cubic</td>
<td>Parashar et al. (2009b)</td>
</tr>
<tr>
<td>Pogostemon cablinus</td>
<td>Ag</td>
<td>25–30 nm; spherical</td>
<td>Raghunandan et al. (2009)</td>
</tr>
<tr>
<td>Pyras sp. (pear fruit extract)</td>
<td>Au</td>
<td>200–500 nm; triangular, hexagonal</td>
<td>Ghodake et al. (2010)</td>
</tr>
<tr>
<td>Rhododendron dauricum</td>
<td>Ag</td>
<td>25–40 nm; spherical</td>
<td>Mittal et al. (2012)</td>
</tr>
<tr>
<td>Rosa rugosa</td>
<td>Ag, Au</td>
<td>30–60 nm Ag; 50–250 nm Au</td>
<td>Dubey et al. (2010a)</td>
</tr>
<tr>
<td>Sesuvium portulacastrum</td>
<td>Ag</td>
<td>5–20 nm; spherical</td>
<td>Nabihjan et al. (2010)</td>
</tr>
<tr>
<td>Svetenia mahogani (mahogany)</td>
<td>Ag/Au bimetallic</td>
<td>50 nm Ag at pH 7; 20 nm Ag and 100 nm Au at pH 12.5; 50 nm Au/Ag bimetallic at pH 12.5</td>
<td>Mondal et al. (2011)</td>
</tr>
<tr>
<td>Syzygium cumini</td>
<td>Ag</td>
<td>29–92; spherical</td>
<td>Banerjee (2011); Kumar et al. (2010)</td>
</tr>
<tr>
<td>Tanacetum vulgare (tansy fruit)</td>
<td>Ag, Au</td>
<td>16 nm Ag, 11 nm Au</td>
<td>Dubey et al. (2010b)</td>
</tr>
<tr>
<td>Terminalia catappa</td>
<td>Au</td>
<td>10–35 nm</td>
<td>Ankamwar (2010)</td>
</tr>
<tr>
<td>Trachyspermum copticum</td>
<td>Ag</td>
<td>6–50 nm</td>
<td>Vijayaraghavan et al. (2012)</td>
</tr>
<tr>
<td>Vitex negundo L.</td>
<td>Au</td>
<td>10–30 nm; face-centered cubic</td>
<td>Zargar et al. (2011)</td>
</tr>
</tbody>
</table>

Extract of rhizome of Dioscorea batatas has been used to synthesize silver nanoparticles (Nagajyothi and Lee, 2011). The nanoparticles were found to be antimicrobial against the yeasts C. albicans and Saccharomyces cerevisiae. Narayanan and Sakthivel (2010) reported the synthesis of silver nanoparticles using a leaf extract of Coleus amboinicus. The concentration of the extract mixed with the silver salt influenced the size and shape of the nanoparticles formed (Narayanan and Sakthivel, 2010). Nanoparticles of triangular, decahedral, hexagonal and spherical shapes could be produced by varying the concentration of the extract (Narayanan and Sakthivel, 2010).

Babu and Prabu (2011) synthesized 35 nm silver nanoparticles using a flower extract of Calotropis procera. Kouvaris et al. (2012) used a leaf extract of Arbutus unedo to produce nanoparticles with a narrow size distribution. Baskaralingam et al. (2012) used a leaf extract of Calotropis gigantea to produce silver nanoparticles. These
nanoparticles had antibacterial activity against *Vibrio alginolyticus* (Baskaralingam et al., 2012).

Ravindra et al. (2010) produced silver nanoparticles within cotton fibers loaded with silver ions. Leaf extract of *Eucalyptus citriodora* (neelagiri) and *Ficus bengalensis* (marri) plants were used for the synthesis (Ravindra et al., 2010). The average size of the nanoparticles was ~20 nm. The cotton fibers loaded with the silver nanoparticles were shown to be antibacterial towards *E. coli* (Ravindra et al., 2010).

Patil et al. (2012a,b) produced highly stabilized silver nanoparticles (25–40 nm) using a leaf extract * Ocimum tenuiflorum*. The particles were antibacterial towards Gram-negative and Gram-positive bacteria. Krishnaraj et al. (2010) synthesized silver nanoparticles (20–30 nm) using a leaf extract of *Acalypha indica*. The nanoparticles were shown to be antimicrobial against water borne pathogens such as *E. coli* and *Vibrio cholerae*.

Spherical silver nanoparticles (40–50 nm) have been produced using a leaf extract of *Euphorbia hirta* (Elumalai et al., 2010). Silver nanoparticles produced using peel extract of *Citrus sinensis* were found to have a broad spectrum antibacterial activity (Kaviya et al., 2011b). The particles formed at 60 °C had an average size of around 10 nm but reducing the reaction temperature to 25 °C increased the average size to 35 nm (Kaviya et al., 2011b).

Kandasamy et al. (2012) used a leaf extract of the *Prosopis chilensis* (L.) tree to produce silver nanoparticles that were active against *Vibrio* species in the shrimp *Penaeus monodon*. Kumar et al. (2010) and Banerjee (2011) used extracts of *S. cumini* leaves and seeds to produce silver nanoparticles. The nanoparticles produced using the seed extract were found to have stronger antioxidant properties in vitro than the original extract (Banerjee, 2011), suggesting a concentration of the polyphenolic antioxidants on the surface of the particles by adsorption.

Vijayaraghavan et al. (2012) reported a one-step synthesis of silver nano/microparticles using *Trachyspermum ammi* and *Papaver somniferum* extracts. The extracts of *T. ammi* produced nanoparticles (87–988 nm) of various triangular shapes and the extract of *P. somniferum* resulted in spherical shaped microparticles (3–8 μm). Li et al. (2007a,b) used extracts of *Capsicum annum* leaf to produce silver nanoparticles and amorphous selenium/protein composite nanoparticles.

A leaf extract of *Cassia auriculata* has been used to synthesize spherical and triangular gold nanoparticles (15–25 nm) within 10 min at room temperature (Kumar et al., 2011). Raghunandan et al. (2010) produced irregularly shaped gold nanoparticles using an extract of dried clove (*Syzygium aromaticum*) buds. Reduction and stabilization of gold nanoparticles were ascribed to the flavonoids present in the extract, Parida et al. (2011) reported the synthesis of gold nanoparticles mediated by an extract of *Allium cepa*. The particles had an average size of 100 nm and could be internalized by MCF-7 breast cancer cells via endocytosis (Parida et al., 2011). Kasthuri et al. (2009a) used a dilute phyllanthin containing extract derived from the plant *Phyllanthus amarus*, to produce hexagonal and triangular gold nanoparticles from HAuCl₄. Increasing the concentration of the extract led to the formation of spherical nanoparticles (Kasthuri et al., 2009a).

Narayanan and Sakhthivel (2008) produced gold nanoparticles using a leaf extract of *Coriandrum sativum* (coriander). The particles ranged in size from about 7 to 58 nm and had diverse shapes (spherical, triangular, decahedral). Edison and Sethuraman (2012) used an aqueous extract of *Terminalia chebula* to produce gold nanoparticles with sizes ranging from 6 to 60 nm. These nanoparticles were active against both Gram-positive *S. aureus* and Gram-negative *E. coli*.

Liu et al. (2012) synthesized gold nanoparticle using extracts of *Chrysanthemum* and tea beverages. A nanoparticle based assay was developed for quantifying the antioxidant properties of teas (Liu et al., 2012). Daisy and SaiPriya (2012) synthesized gold nanoparticles (55–98 nm) using an aqueous extract of *Cassia fistula*. Extracts of *C. fistula* bark are known to be hypoglycemic. Gold nanoparticles made using the extract were found to be superior to the extract as hypoglycemic agents in rats for the management of diabetes mellitus (Liu et al., 2012). Clearly, the particles concentrated the hypoglycemic agent from the extract on their surfaces.

Castro et al. (2011) reported on the use of sugar beet pulp as an effective reductant for making gold nanowires at room temperature. The nanoparticles formed initially later joined to form chains and nanowires. The formation of the nanowires and nanorods depended on the conditions of the reaction, particularly the pH.

The synthesis of gold and silver nanoparticles using leaf extracts of *P. graveolens* and *Azadirachta indica* was reported by Shankar et al. (2003, 2004), respectively. Bimetallic Ag–Au core–shell nanoparticles could be made by the reduction of aqueous Ag⁺ and Au⁺ ions using *Azadirachta indica* leaf extract (Shankar et al., 2004). Presence of
Gold and silver nanoparticles have been produced using extracts of *Aloe vera* (Chandran et al., 2006) and *Camellia sinensis* (Vilchis-Nestor et al., 2008). The optical properties of the nanoparticles depended on the initial concentration of the metal salts and the *C. sinensis* extract (Vilchis-Nestor et al., 2008). Caffeine and theophylline found in *C. sinensis* extract may have contributed to reduction of the metal ions and formation of the nanoparticles.

Kasthuri et al. (2009b) reported the ability of apiin found in the leaf extract of henna to reduce ions to gold and silver nanoparticles. Secondary hydroxyl and carbonyl groups of apiin were responsible for the reduction. The size and shape of the nanoparticles could be controlled by changing the concentration of apiin (Kasthuri et al., 2009b). Dwivedi and Gopal (2011) used extracts of *Chenopodium album* leaves to produce silver and gold nanoparticle. The particles had quasi-spherical shapes and were in the size range of 10–30 nm.

Production of spherical and triangular shaped silver and gold nanoparticles using fruit extract of *Tanacetum vulgare* has been reported (Dubey et al., 2010a). A FTIR study revealed that the carbonyl measurement of the extract did not affect the size and shape of the nanoparticles. The zeta potential of the silver nanoparticles was shown to vary with pH: a low zeta potential at strongly acidic pH (Dubey et al., 2010a). A larger particle size could be achieved by reducing the pH of the reaction (Dubey et al., 2010a).

Njagi et al. (2011) used an aqueous extract of sorghum bran to produce nanoparticles of iron and silver at room temperature. Valodkar et al. (2011) synthesized nanoparticles (5–10 nm) of silver and copper using latex of *Euphorbiaceae*. These nanoparticles exhibited excellent bactericidal activity towards both Gram-negative and Gram-positive bacteria (Valodkar et al., 2011). Velayutham et al. (2011) used a leaf extract of *Catharanthus roseus* to synthesize nanoparticles of titanium dioxide. The particles were of irregular shape and ranged in size from 25 to 110 nm. Suspensions of these nanoparticles were adulticidal and larvicidal against the hematopha-

Huang et al. (2011) reported a one-step synthesis of Au–Pd core–shell nanoparticles using an aqueous solution of bayberry tannin at room temperature. The tannin preferentially reduced the Au$^{3+}$ ions to Au nanoparticles when a mixture of Au$^{3+}$ and Pd$^{2+}$ was contacted with the tannin (Huang et al., 2011). Subsequently the gold nanoparticles served as seeds for the growth of a Pd shell.

Amaranth et al. (2012) reported the antibacterial activity of palladium nanoparticles and their stabilization by chitosan and grape polyphenols. Palladium nanoparticles could be synthesized using coffee and tea extracts (Nadagouda and Varma, 2008). The nanoparticles were in the size range of 20–60 nm (Nadagouda and Varma, 2008). Sathishkumar et al. (2009a) produced palladium nanoparticles using an extract of cinnamon (*C. zeylanicum*) bark as the reducing agent. In this process, the reaction pH, the temperature and the concentration of the extract did not affect the size and shape of the nanoparticles.

Palladium nanoparticles (75–85 nm) have been synthesized using an extract of *Annona squamosa* L. peel (Roopan et al., 2011). Palladium nanoparticles having an average size of 15 nm were synthesized using a leaf extract of soybean *Glycine max* (Kumar et al., 2012). Lee et al. (2011) synthesized copper nanoparticles (40–100 nm) using magnolia leaf extract. These copper nanoparticles had antimicrobial activity against *E. coli* and were toxic to human adenocarcinomic alveolar basal epithelial cells (A549 cells).

5. Applications of nanoparticles

Nanoparticles synthesized by the various methods have been used in diverse in vitro diagnostic applications (Chen et al., 2012; Doria et al., 2012; Fortina et al., 2007; Yous et al., 2011). Both gold and silver nanoparticles have been commonly found to have broad spectrum antimicrobial activity against human and animal pathogens (Ali et al., 2011; Arulkumar and Sabesan, 2010; Duran et al., 2007; Jain et al., 2009; Kandasamy et al., 2012; Kim et al., 2007; Krishnaraj et al., 2010; Lara et al., 2011; Patil et al., 2012a;b; Prasad and Elumalai, 2011; Ravindra et al., 2010; Sathishkumar et al., 2009b; Saxena et al., 2011; Seil and Webster, 2012; Singh et al., 2010; Singhal et al., 2011). Silver nanoparticles are already widely used as antimicrobial agents in commercial medical and consumer products (Rai et al., 2009; Ravindra et al., 2010). Fig. 5 shows the general applications of metal nanoparticles in biological field. 

![Fig. 5. Types of metal nanoparticles and their applications in biotechnology.](image-url)
Silver nanoparticles are larvicidal against filariasis and malaria vectors (Rajakumar and Abdul Rahuman, 2011; Santhoshkumar et al., 2011) and have been found to be active against plasmodial pathogens (Poranurvel et al., 2012) and cancer cells (Fortina et al., 2007; Ravindra et al., 2010; Subramanian, 2012; Sukirtha et al., 2011). Antifungal effects of silver nanoparticles have been demonstrated (Vivek et al., 2011).

Extensive literature exists on the mechanisms of antimicrobial action of silver and gold nanoparticles (Chaloupka et al., 2010; Cui et al., 2012; Li et al., 2010). Silver ion is highly toxic to most microorganisms (Jung et al., 2008) and at least one mode of antimicrobial action of nanoparticles is through a slow release of silver ions via oxidation within or outside the cell. Silver nanoparticles are known to affect the permeability of membranes of microbial and other cells (Li et al., 2010). Nanoparticles are known to inactivate proteins and interfere with the replication of DNA (Chaloupka et al., 2010). The applications of metal nanoparticles synthesized from plant extracts are summarized in Table 2.

Applications of nanoparticles are emerging in crop protection and agriculture (Khot et al., 2012; Nair et al., 2010; Perez-de-Luque and Industri, 2011) and have been found to be active against plasmodial pathogens (Poranurvel et al., 2012) and cancer cells (Fortina et al., 2007; Ravindra et al., 2010; Subramanian, 2012; Sukirtha et al., 2011). Antifungal effects of silver nanoparticles have been demonstrated (Vivek et al., 2011).

6. Concluding remarks

The use of plant extracts for making metallic nanoparticles is inexpensive, easily scaled up and environmentally benign. It is especially suited for making nanoparticles that must be free of toxic contaminants as required in therapeutic applications. The plant extract based synthesis can provide nanoparticles of a controlled size and morphology. In medicine, nanoparticles are being used as antimicrobial agents in bandages, for example. Applications in targeted drug delivery and clinical diagnostics are developing.

Acknowledgment

AKM gratefully acknowledges the Council for Scientific and Industrial Research (CSIR), New Delhi, India, for the award of a Senior Research Fellowship in support of this work.

References


Kandasamy K, Alikunhi NM, Manickaswami G, Nabikhan A, Ayyavu G. Synthesis of silver

Huang JL, Li QB, Sun DH, Lu YH, Su YB, Yang X, et al. Biosynthesis of silver and gold

Haverkamp RG, Marshall AT. The mechanism of metal nanoparticle formation in

Han G, Ghosh P, Rotello VM. Functionalized gold nanoparticles for drug delivery.

Gan PP, Li SFY. Potential of plant as a biological factory to synthesize gold and

G h o s h S , P a t i l S , A h i r e M , K i t t u r e R , J a b g u n d e A , K a l e S , e t a l . S y n t h e s i s o f g o l d

Gold nanoparticles produced in a


Luangpipat T, Beattie IR, Chisti Y, Haverkamp RG. Gold nanoparticles produced in a


Li WR, Xie XB, Shi QS, Zeng HY, Ou-Yang YS, Chen YB. Antibacterial activity and mech-

Lu H, Xu H, Chen ZS, Chen G. Biosynthesis of nanoparticles by microorganisms and their applications. J Nanomater 2011.


Mondal S, Roy N, Laskar RA, SK, Basu S, Mandal D. Biogenic synthesis of Ag, Au and bi-


Mukherjee P, Ahmad A, Mandal D, Senapati S, Sainkar SR, Khan MI. Fungus-


