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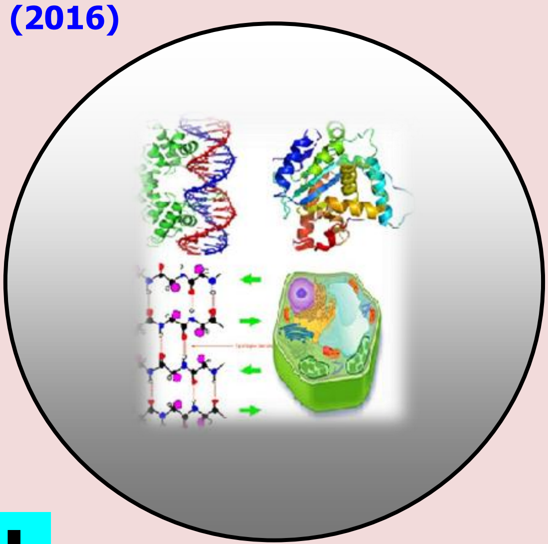

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RESEARCH PAPER

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Synthesis of Silver Nanoparticles from Plant Extracts-A Review on Greener Approach

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ABSTRACT

Plant mediated green synthesis of nanoparticles have captured the attention of scientists all over the world. The process of synthesis has had a huge impact on environment as it reduces the use of toxic chemicals and eliminates risk of harmful byproducts. Metallic nanoparticles, due to their small size, find uses in various fields of science and technology with special application in medicine and antimicrobial activity.Among many nanoparticles, silver nanoparticles (AgNPs)have attained special focus owing to its simplicity and rapid rate of synthesis.Its antimicrobial activity can be attributed to its ability to penetrate cell wall and interrupt cell metabolism. Conventionally thesenanoparticles are synthesized using chemicals.However, greener approach of synthesis addresses the concern for environment. This review explores the various methods of synthesis of AgNPs using huge plant diversity, their characterization and antimicrobial activity.
Keywords:Metallic Nanoparticles, AgNPs, Antimicrobial Activity and Green Synthesis.

INTRODUCTION

Nanomaterials in nanotechnology have become inseparable part of everyday life. Nanomaterials have been synthesized using noble metals as well as transition metals since ages. They are being widely used in various fields of science and technology like purification of water by gold and AgNPsapplication in medicine, engineering, entertainment, fabrics, biosensor materials (Pradeep et al., 2009, Jiang et al., 2004, Hirano et al., 2003, Yeo et al., 2003, Ren et al., 2002). Silver nanoparticles [AgNPs] find wider use in medicine because they are nontoxic to humans but inhibit the growth of micrórganisms (Jeong et al., 2005). Silver belongs to the family of noble metals which has been popular because of its brightness and shining appearance. It has been used in making utensils and jewelry since ages. Silver having standard reduction potential $Ag^+|Ag$=0.80 V is highly inactive and can exist without any oxidative effects of oxygen or water. It is an electropositive element which can easily be reduced. At nanoscale, silver particles show remarkable reactivity because of size-quantization effect according to which reduction in number of atoms reduces the reduction potential (Henglein, 1989).

Different synthetic methods can be employed to produce AgNPs of various shapes and size which can be controlled by adjusting certain reaction condition. Chemical method of synthesis involves reduction of silver ions in aqueous or non aqueous solutions, electrochemical reduction, microwave/ultrasonic assisted synthesisetc (Yin et al., 2002, Zhu et al., 2004, Wang et al., 2003, Zhou et al., 2004, Yin et al., 2004). The advantage of using chemical methods lies in the fact that bulk production of nanoparticles is possible in short time. However, the tendency of nanoparticles to accumulate, affects their properties. Therefore it is important to focus on the size of the nanoparticles. Chemical methods have also been associated with environmental hazards.

The techniqueslike microwave, irradiation, and photo inducement are the physical methods used to prepare AgNPs. These methods are rapid because of increase in reaction rate but smaller sizes are difficult to achieve. A biological method of preparing AgNPs is of greater interest because of its environment friendly nature. It involvesenzymatic and non enzymatic reduction approach. The aim of this review is to discuss ecofriendly approachof synthesis of AgNPs using plant extracts. It also explores the various characterization techniques used for the study of structure and size of AgNPs. The review also investigates the application of the AgNPs against microbes.

Comprehensive Review on AgNPs

Devina et al., 2010, described the greener method of synthesis of silver nanoparticle using algae and microwave irradiation in water. Four strains of normal marine microalgae *C. calcitrans, C. salina, I. galbana and T. gracilis* were identified and subjected to various methods of synthesis of AgNPs. The change in colour from colorless to dark brown suggested the formation of nanoparticles which was further confirmed by the appearance of absorption peak at 420nm. Formation of nanoparticles is attributed to the metabolites excreted by the culture which may have acted as reducing agent. These nanoparticles arefound to be stable over a range of one month. SEM studies reveal the formation of silver nanoparticle of size 71.9nm and 53.1 nm. Gnanadesigan et al., 2011, synthesized AgNPs using Rhizophora mucronata (*R. mucronata*) leaf extract. Brownish yellow solution, on treatment of plant extract with silver nitrate aqueous solution, confirms the formation of AgNPs. Average size of the synthesized nanoparticle observed through AFM topography were 60-95nm. FTIR results identify biomolecules flavonoids, triterpenoids and polyphenols responsible for the stabilization of the synthesized AgNPs. Biosynthesized nanoparticles were tested in many concentrations against Aeaegypti and Cxquinque fasciatus larvae and were reported to posses maximum larvicidal activity at concentration 5mg/L and 10mg/L. This may be attributed to denaturation of sulfur or phosphorus containing proteins. These nanoparticles cause denaturation of organelles or enzymes thereby reducing cell permeability finally leading to cell death.

Fathima Stanley Rosarin et al., 2012, use amla *P. emblica* fruit extract for the synthesis of AgNPs. The extract was used to reduce 100mL aqueous solution of 1mM $AgNO_3$. The change in colour to brownish orange indicated the formation of AgNPs which is further confirmed by the appearance of SPR bands between 428 to 438 nm. SEM studies reveal the formation of spherical and cubic shapes AgNPs with average particlesize distribution 188 nm. Cytotoxic effects of these nanoparticles were studied on human laryngeal carcinoma cell line (Hep2). It was observed that the nanoparticles interfered with mitochondrial functions leading to apopsis.

Elemike Elias Emekaet al., 2013 synthesized AgNPs using fresh pineapple leaf extract using water as a solvent (ratio 1:10 w/v). The resulting solution is added to 1 mM aqueous $AgNO_3$ solution heated at 70°C. The appearance of brownish colour followed by surface plasmon bands at 440–460 nm confirms the formation of AgNPs. FTIR results reveal the involvement of sugars like sucrose, glucose etc. in bio reduction of $AgNO_3$. Kinetic studies confirm the faster rate of reaction and stability of the nanoparticles. The synthesized nanoparticles were successfully tested against *S. aureus, S. pneumoniae*, *P. mirabilis* and *Esherichiacoli*. It inhibited the growth of bacteria possibly through penetration into their cellwalls causing cellular damage. The article also suggests the presence of optical properties in these AgNPs by UV studies.

Anjali Rawani et al., 2013, synthesized AgNPs using aqueous extracts from fresh leaves, dry leaves and green berries of *Solanum nigrum* L. Synthesized AgNPs are tested against larvae of Culex *quinque fasciatus* and *Anopheles stephensi* and four human pathogens and five fish pathogens respectively. The synthesized nanoparticles were spherical to polyhedral in shape with average size of 56.6 nm. These were characterized using UV-vis spectroscopy, TEM images, X-ray and FTIR. Inhibitory action of these nanoparticles was observed against pathogens mentioned above and was attributed to the association of these particles with sulphur and phosphorus present in cell membrane of the pathogens. No environmental hazard was found by using these nanoparticles on the pathogens.

Synthesis of AgNPs using methanolic extract was first time done by Ghassan Mohammad Sulaiman et al., 2013. The leaves of *Eucalyptus chapmaniana* (*E. chapmaniana*) were extracted using soxhlet extraction method and methanol as solvent.Aqueous solution of silver nitrate was used to prepare nanoparticles which were characterized using UV-visible and XRD studies. Silver surface plasmon resonance band was observed at 413 nm. XRD studies reveal that structure of nanoparticle is FCC.

The efficacy of AgNPs was tested against *Escherichia coli* (*E. coli*), *Pseudomonas aeruginosa (P. aeruginosa), Klebsiellapneumoniae (K. pneumoniae), Proteus volgaris (P. volgaris)* (Gram negative), *Staphylococcus aureus (S. aureus) (Grampositive)* and *Candida albicans (C. albicans*) (Yeast). The inhibitory action of biosynthesized AgNPs against microbes was dependent on dose and time of incubation. Cytotoxic effects of AgNPs were tested against human leukemia cell line (HL-60) and were attributed to its physiochemical interaction with functional groups of proteins.

Geethalakshmi, et al., 2013 used the saponin obtained from leaf extracts of *T. decandra (Aizoaceae*), which is a popular herb of south India, to prepare gold and AgNPs. Dried leaves of T. decandra were extracted using methanol as solvent. To the leaf extract 1 mL of 1 mM aqueous $HAuCl_4$ and $AgNO_3$ solutions were added to make Au and Ag nanoparticles. The change in color from yellow to dark ruby and orange indicates the formation of Au and Ag nanoparticles respectively. Au nanoparticles showed strong absorption at 440-580nm while AgNPs showed absorption at 260-380nm. FTIR spectra confirm the formation of gold and AgNPs. Cubical, hexagonal and spherical nanoparticles of gold ranging in size 37.7 and 79.9 nm were identified with the help of SEM. AgNPs obtained were spherical in shape having 17.9 nm and 59.6 range diameters. Further, EDX studies confirm the presence of gold and AgNPs. Surface Plasmon Resonance for gold nanoparticles was observed at 0.2 and 2.2 keV whereas, it was observed at 0.2,3, 3.3 and 3.7 keV for AgNPs. The antibacterial activity of synthesized gold and AgNPs were tested against different bacteria, *namely S. aureus, S. faecalis, E. faecalis, E. coli, P. aeruginosa, P. vulgaris, B. subtilis, Y. enterocol-itica, K. pneumoniae* and a *fungus, C. albicans*. Due to the smaller size and increased surface area AgNPs were reported to be more effective then gold nanoparticles against the mentioned pathogens.

Kanayairam Velayutham et al., 2013, synthesized AgNPs using aqueous bark extract of *Ficusracemosa* (F. racemosa). The appearance of brown color solution on addition of 1mM silver nitrate (AgNO3) to the aqueous bark extract indicated the formation of AgNPs. A peak at 425nm in UV visible spectra revealed the formation of nanoparticles. As revealed by SEM the average size of these nanoparticles is reported to be 250.60 nm and they are cylindrical and rod shaped. The larvicidal activity of AgNPs was tested against fourthinstar larvae of filariasis vector, *Culexquinque fasciatus (Cx. quinquefasciatus)* and *Japanese encephalitis vectors, Culexgelidus (Cx. gelidus)*. The extract was found to be most effective against the larvae of Cx. Quinque fasciatus and Cx. Gelidus.

Gabriele E. Schaumann et al., 2014, compared the biological effects of AgNP and TiO_2 NP on ground water and drinking water. AgNPis found to be useful in filter feeders where as TiO_2 NP had strong effect on bacterial biofilms.

H. Joy Prabu et al., 2015, synthesized AgNPs using aqueous extracts of Indian herbal plants *Tragiainvolucrata*, *Cymbopogon citronella, Solanumver bascifolium* and *Tylophoraovata* aqueous extractsas a reducing agent. The extract was added to 60 ml of 10^{-3} M $AgNO_3$ solution at ambient room temperature. The dark brown color solution indicates the formation of AgNPs. The solution was centrifuged and suspended pellets were purified with ethanol, dried and ultrasonicateto obtain AgNPs. The suggested mechanism, by FTIR studies, for the formation of AgNPs involves the oxidation of flavonoids there by causing reduction of Ag^+. The SPR signals were obtained in the range of 400-500nm. The XRD and FESEM studies reveal the formation of AgNPs of size range between 28 nm to 45 nm. They are further proved to be polydispersed and spherical in shape.

Bonnia et al., 2016, reported biosynthesis of AgNPs using leaf extract of ‘*Polygonum Hydropiper’* via greener approach. The extract was added to 90 ml of 0.01M silver nitrate solution dropwise. The appearance of brown colorand surface plasmon resonance (SPR) in the range 400-460 nmshows the formation of AgNPs.

Siriporn Phongtongpasuk et al., 2016, synthesizes AgNPs using dragon fruit peel extract with water (ratio 1:20 w/v) as reducing agent for 10 mM silver nitrate ($AgNO_3$) solution. The solution was centrifuged at 12,000 rpm for 15 min at 4 ^{o}C. The phytochemical analysis and the effect of pH was studied on biosynthesis of nanoparticles. It was observed that the color of the reaction mixture was pH dependent, absorption peaks were obtained in the range of 457to 431 nm. As the pH was increased hypsochromic shift of theabsorption bands was observed. TEM studies revealed the spherical shaped polydispersed AgNPs with average diameter being 26.2±8.2 at pH: 3.35, 25.7±8.7 at pH4.35 and 25.3±7.9 nm at pH 5.35. In XRD presence of silver nanoparticle is indicated by the peak at 32.5. FTIR studies confirm the reduction of Ag^+ to Ag^0. The nanoparticles were found to be quite effective against *E. coli*, *P. aeruginosa* and *S. aureus.*

N. Hanumanta Rao et al., 2016 synthesized AgNPs using crude methanolic root extracts of *Diospyrospani culata.* The effect of metal ion concentration (silver acetate) and plant extract concentration on the formation of brown color AgNPs was studied.

The change in color from green to brown and SPR band in between 400 to 500nm confirmed the formation of AgNPs. XRD, FEG-SE,TEM studies revealed the average size of synthesized nanoparticles is 17 nm. Antibacterial and antifungal studies were conducted and the studies indicated the silver nanoparticle concentration dependent inhibition of bacteria. The synthesized nanoparticles showed promising activity against all the tested fungal strains.

Ravichandran et al., 2016, reported the greener method of synthesis of silver nanoparticles from leaf extract of *Artocarpusaltilis*. 0.01 M AgNO3 solution was added to the aqueous leaf extract and brown colored nanoparticles were obtained. Spherical shaped nanoparticles of average size of 34 nm were confirmed and characterized by UV-VIS spectroscopy, FESEM-EDX, TEM, FTIR and XRD. FTIR spectrum suggested the involvement of polyphenols of leaf extract in formation of AgNPs. The inhibitory action of these nanoparticles on bacteria was comparable to streptomycin and on fungus it was similar to amoxicillin.

R. Sriranjani et al., 2016, synthesis AgNPs from aqueous extract of C. phlomidis leaf using silver nitrate (90 ml, 1 mM) solution. The persistent reddish brown colour indicated the presence of biosynthesized AgNPs. The size morphology and composition of synthesized nanoparticles was studied by UV–visible spectroscopy, Scanningelectron microscope (SEM), transmission electron microscope (TEM), and energy dispersive X-ray analysis (EDAX) and FTIR. In UV-VIS spectral analysis the SPR band at 450nm. TEM analysis confirmed the crystalline nature of nanoparticle with diameter range 10-15nm.The synthesized nanoparticles were found to possess excellent anticancer activity and potential antioxidant activity. Abou Talibet al., 2016, prepared orange-red color silver sol by simple biomimetic chemical reduction method using lotus leaf extract and silver nitrate in absence and presence of shape-directing, cross-linking and stabilizing agent cetyltrimethyl ammonium bromide (CTAB), at room temperature. A broad shoulder at 500 nm was observed in UV–visible spectral studies which are attributed to larger particle size. The TEM image indicated the arrangement of AgNPs in the form of necklace-like structure. The addition of optimum concentration of CTAB along with plethora proteins helped in tuning the morphology of AgNPs. S.K. Verma et al., 2016, synthesized cross-linked stimuli-responsive hybrid polymer-encapsulated AgNPs using Poly (N-isopropylacrlamide co Sulphonic) and leaf extract of *Calotropis gigentea*. UV-VIS wavelength scan (300 to 700 nm) clearly shows absorbance peak at 420 nm for yellow colored hybrid nanoparticles. On the basis of Dynamic light scattering (DLS), small angle x-ray scattering (SAXS) and UV-VIS studies authors hypothesized that small peak at 20°C is due to covalently bonded silver particles to polymer network. These hybrid particles displayed promising antibacterial properties against *Salmonella typhimurium* SL4522. Kofi J. Brobbey et al., 2017, synthesized silver nanoparticles and incorporated these AgNPs into paper surface using a flame pyrolysis procedure known as Liquid Flame Spray. The synthesized particles were characterized by using SEM and XPS measurements and results reveal homogeneous monolayer of silver nanoparticles on the surface of paper. The results also demonstrated the antibacterial properties against *E. coli*. The technique can be used on a larger scale for production of antibacterial paper. Saxena Varsha et al., 2018, synthesized silver nanoparticles using leaf extract of *Vincarosea. M*icrowave mediated extraction and conventional extraction methods have been compared. The study concludes that microwave assisted extraction of leaves is economic and time efficient. The leaf extract and nanoparticles were subjected to antibacterial activity against *S.mutans* which revealed that nanoparticles were very effective against these bacteria.

CONCLUSION

Nature has blessed us with immense diversity of medicinal plants. Nanoparticles using plant extracts have charmed the scientist into exploring their medicinal scope.Metal nanoparticles find extensive use in many fields of medicine and science. The present study aimed at focusing the role of various plant materials in synthesizing silver nanoparticles. AgNPs have wide range of application especially in the field of biomedicine. These can be synthesized by chemical as well as physical methods. However, greener methods of synthesis is the call of the day as it is cost effective; provide healthier work places, protect human health and environment by producing less waste. This review summarizes the ecofriendly and rapid methods of synthesis of AgNPs using wide variety of plants. The study also highlights the effect of silver nanoparticles against different microorganisms which are responsible for various diseases giving us gainful insight into pharmacological potential of these nanoparticles. It further reveals different methods used for characterization of nanoparticles. The similar approach can be extended to the synthesis of nanoparticles using different metals for varied applications.

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