

Evidence based green synthesis of nanoparticles

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ABSTRACT

Nowadays, nanotechnology has grown to be an important research field in all areas including medicinal chemistry. The size, orientation and physical properties of nanoparticles have reportedly shown to change the performance of any material. For several years, scientists have constantly explored different synthetic methods to synthesize nanoparticles. On the contrary, the green method of synthesis of nanoparticles is easy, efficient, and eco-friendly in comparison to chemical-mediated or microbe-mediated synthesis. The chemical synthesis involves toxic solvents, high pressure, energy and high temperature conversion and microbe involved synthesis is not feasible industrially due to its lab maintenance. Since, green synthesis is the best option to opt for the synthesis of nanoparticles, therefore the nanoparticles were synthesized by using aqueous extract of *Moringa oleifera* and metal ions (such as silver). Silver was of particular interest due to its distinctive physical and chemical properties. *M. oleifera* leaf extract was selected as it is of high medicinal value and it does not require any sample preparation and hence is cost-effective. The fixed ratio of plant extract and silver ions were mixed and kept at room temperature for reduction. The color change from yellow to reddish brown confirmed the formation of nanoparticles. Further, the synthesized nanoparticles were characterized by UV, EPMA, XRD and FTIR data. The antimicrobial activity of synthesized nanoparticle has also been examined in gram positive and gram negative bacteria and encouraging results are in hand.

Keywords: Silver nanoparticles; green synthesis; *M. oleifera*; antimicrobial.

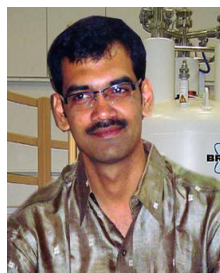


Anamika Mubayi has qualified GATE, CRET and joined D. Phil. under the supervision of Dr. Watal in the Department of Chemistry, University of Allahabad, India. Ms. Mubayi has done her M. S. from the University of Iowa, USA and M. Sc. from CSJM University, Kanpur, India. She has five years of research experience in the field of synthesis and characterization of nanomaterials & catalysts. She has worked as a Senior Research Associate at IIT, Kanpur, India and Research Assistant at the University of Iowa, USA. She has been to Lausanne, Switzerland for a nanotechnology-based training programme. Ms. Mubayi has several publications in National/International journals and has received Poster award in James F. Jakobsen Graduate Conference, University of Iowa, USA. Currently, she is working on plant-mediated synthesis of nanoparticles and their biomedical applications with special reference of Diabetes.



Sanjukta Chatterji has done her D. Phil. from Dept. of Chemistry, University of Allahabad, India in 2011. Dr. Chatterji has done her M. Sc. in Bio-Technology and M. Phil. in Bio-Chemistry. She has availed JRF of National Medicinal Plants Board (NMPB), Government of India, New Delhi, India. Dr. Chatterji has a strong background of Natural Product Technology and bioactivity testing with special reference to antidiabetic, antioxidant, antilipidemic, enzymatic studies *in vivo* and *in vitro*. She has an expert hand in

antimicrobial testing as well. She has a number of good publications to her credit in journals of high repute and has presented her work in various National as well as International conferences.



Prashant Kumar Rai is currently doing post doctorate at All India Institute of Medical Sciences (AIIMS), New Delhi, India. His work is finger printing of Medicinal plants using e Tongue, NMR, and IR spectroscopic techniques. He has done his D. Phil. from Allahabad University, India in 2009 under the supervision of Dr. Watal. He has also synthesized oral Insulin first time in India and the patent is under way. Dr. Rai has two Patent and more than forty

International and National publications inclusive of five book chapters three in "Methods in Molecular Biology Series", and two with Novo Publications, to his credit. Dr. Rai had been to Bethesda, MD, USA, and Basel, Switzerland, for presenting his work. He has worked as a Post Doc Fellow, Department of Chemical Technology, University of Johannesburg, Johannesburg, South Africa in material Sciences (academic year 2010 – 2011). Dr Rai currently serves as lead guest editor in Experimental Diabetes Research, and associate editor in few journals viz. British Journal of Pharmacology and Toxicology, Advance Journal of Food Science and Technology & International Journal of Basic Science and Applied Medical Science.



Geeta Watal, a graduate from BHU, did her post-graduation in Chemistry from the University of Allahabad in 1979. Dr. Watal is actively engaged in Medicinal Research specifically in treating Diabetes and its complications. Ten of her research students have been awarded D. Phil. and six are still working under her able guidance. She had been invited by the American Diabetes Technology Society to Philadelphia, Pennsylvania, USA in Oct.2004, where she presented her work. She had also been to Corvallis, Oregon, USA in July 2005 and to Arlington, Virginia, USA in Aug, 2006 for presenting her work in annual meets of American Society of Pharmacognosy. She has handled a number of projects funded by UGC, ICMR and NMPB. In connection with her collaborative research work, she has travelled to New Paltz campus, New York University, USA in Oct, 2007 and Harvard University, Washington, USA in Oct, 2008. Her National collaborators are from All India Institute of Medical Sciences, New Delhi and Delhi University. Dr. Watal has five Patents for her inventions and more than eighty publications in journals of high repute along with three book chapters in "Advanced Protocols in Oxidative Stress; Methods in Molecular Biology Series" to her credit. Dr. Watal is currently involved in a screening program of herbal extracts from India, and other parts of the globe, for their bioactivities. She has invented a number of highly effective medications by unfolding the mystery involved in the role of phytoelements and plant mediated synthesized compounds at Nano level. Her motto is "Integration of natural medicine into conventional treatments".

Introduction

The prospect of exploiting natural resources for metal nanoparticle synthesis has become to be a competent and environmentally benign approach [1]. Green synthesis of nanoparticles is an eco-friendly approach which might pave the way for researchers across the globe to explore the potential of different herbs in order to synthesize nanoparticles [2]. Silver nanoparticles have been reported to be synthesized from various parts of herbal plants viz. bark of Cinnamom,[3] Neem leaves,[4-5] Tannic acid[6] and various plant leaves [7].

Metal nanoparticles have received significant attention in recent years owing to their unique properties and practical applications [8, 9]. In recent times, several groups have been reported to achieve success in the synthesis of Au, Ag and Pd nanoparticles obtained from extracts of plant parts, e.g., leaves [10], lemongrass [11], neem leaves [12-13] and others [14]. These researchers have not only been able to synthesize nanoparticles but also obtained particles of exotic shapes and morphologies [12]. The impressive success in this field has opened up avenues to develop "greener" methods of synthesizing metal nanoparticles with perfect structural properties using mild starting materials. Traditionally, the chemical and physical methods used to synthesize silver nanoparticles are expensive and often raise questions of environmental risk because of involving the use of toxic, hazardous chemicals [15].

Also, majority of the currently prevailing synthetic methods are usually dependent on the use of organic solvents because of hydrophobicity of the capping agents used [16]. Recently, the search for cleaner methods of synthesis has ushered in developing bio-inspired approaches. Bio-inspired methods are advantageous compared to other synthetic methods as, they are economical and restrict the use of toxic chemicals as well

as high pressure, energy and temperatures [17]. Nanoparticles may be synthesized either intracellularly or extracellularly employing yeast, fungi bacteria or plant materials which have been found to have diverse applications.

Silver nanoparticles (AgNPs) have been proven to possess immense importance and thus, have been extensively studied [18-20]. AgNPs find use in several applications such as electrical conducting, catalytic, sensing, optical and antimicrobial properties [21]. In the last some years, there has been an upsurge in studying AgNPs on account of their inherent antimicrobial efficacy [22]. They are also being seen as future generation therapeutic agents against several drug-resistant microbes [23]. Physicochemical methods for synthesizing AgNPs thus, pose problems due to use of toxic solvents, high energy consumption and generation of by-products. Accordingly, there is an urgent need to develop environment-friendly procedures for synthesizing AgNPs [24]. Plant extracts have shown large prospects in AgNP synthesis [20].

Moringa oleifera (*M. oleifera*) (Family: Moringaceae, English name: drumstick tree) has been reported to be essentially used as an ingredient of the Indian diet since ages. It is cultivated almost all over India and its leaves and fruits are traditionally used as vegetables. Almost all parts of the plant have been utilized in the traditional system of medicine. The plant leaves have also been reported for its antitumor, cardioprotective, hypotensive, wound and eye healing properties [25]. AgNPs synthesized from the aqueous extract of *M. oleifera* leaves in hot condition, have been reported in literature [26]. In the present study, synthesis of AgNPs in cold condition has been reported, reducing the silver ions present in the silver nitrate solution by the aqueous extract of *M. oleifera* leaves. Further, these biologically synthesized nanoparticles were found to be considerably sensitive to different pathogenic bacterial strains tested.

Experimental

Materials

Chemicals used in the present study were of highest purity and purchased from Sigma-Aldrich (New Delhi, India); Merck and Himedia (Mumbai, India). *M. oleifera* leaves were collected locally from University of Allahabad, Allahabad, Uttar Pradesh, India.

Preparation of plant extract

Plant leaf extract of *M. oleifera* was prepared by taking 5 g of the leaves and properly washed in distilled water. They were then cut into fine pieces and taken in a 250 mL Erlenmeyer flask with 100 mL of sterile distilled water. The mixture was boiled for 5 min before finally filtering it. The extract thus obtained was stored at 4 °C and used within a week [7].

Synthesis of silver nanoparticles

The aqueous solution of 1 mM silver nitrate (AgNO₃) was prepared to synthesize AgNPs. 190 mL of aqueous solution

of 1 mM AgNO₃ was slowly added to 10 mL of *M. oleifera* aqueous leaf extract while stirring, for reduction into Ag ions and kept at room temperature for 6 h [7, 27].

UV-Vis spectra analysis

UV-Vis spectrum of the reaction medium recorded the reduction of pure Ag⁺ ions at 6 h after diluting the sample with distilled water. UV-Vis spectral analysis was performed by using UV-Vis double beam spectrophotometer [UV-1700 PharmaSpec UV-Vis Spectrophotometer (Shimadzu)].

XRD (X-ray diffraction) measurement

The AgNP solution was repeatedly centrifuged at 5000 rpm for 20 min, re-dispersed with distilled water and lyophilized to obtain pure AgNPs pellets. The dried mixture of AgNPs was collected to determine the formation of AgNPs by X'Pert Pro x-ray diffractometer (PANalytical BV, The Netherlands) operated at a voltage of 30 kV and a current of 30 mA with CuK α radiation in a θ -2 θ configuration.

EPMA analysis

EPMA-WDS analyses of carbon coated samples were performed using an Electron Probe Micro Analysis JEOL Superprobe (JEOL JXA-8100, Japan) with X-ray wavelength dispersive spectroscopy.

FTIR (Fourier Transform Infrared) analysis

The earlier centrifuged and redispersed AgNPs obtained have removed any free residual biomass. Subsequently, the dried powder was obtained by lyophilizing the purified suspension. The resulting lyophilized powder was examined by Infrared (IR) spectra, recorded on a Bruker Vector-22 Infrared spectrophotometer using KBr pellets.

Bacterial strains of *Klebsiella pneumoniae* (Gram-negative), *Staphylococcus aureus* (Gram-positive), *Pseudomonas aeruginosa* (Gram-negative), *Enterococcus faecalis* (Gram-negative) and *Escherichia coli* (Gram-negative) were clinical isolates obtained from the Department of Biotechnology, All India Institute of Medical Sciences (AIIMS), New Delhi, India and the microbiologist of the department confirmed the identity based on microscopic examination, Gram's character, and biochemical test profile. Bacterial stocks were maintained and stored as 1 ml aliquots at -80°C in *Luria Bertani* (LB) broth for all the five bacterial strains. Bacterial stocks were revived from -80°C and grown in *Luria Bertani* (LB) broth for *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Enterococcus faecalis* and *Escherichia coli*. All cultures were grown overnight at 37°C \pm 0.5°C, pH 7.4 in a shaker incubator (190-220 rpm). Their sensitivity to the reference drug, Streptomycin (Sigma-Aldrich, New Delhi, India) was also checked.

Luria Bertani broth (Himedia), *Luria Bertani* agar (Himedia) standard antibiotic and Streptomycin (Himedia) were used in antimicrobial sensitivity testing. Briefly, *Luria Bertani* (LB) broth/agar medium was used to cultivate bacteria. Fresh overnight cultures of inoculum (50

μ l) of each culture were spread on to LB agar plates. Sterile paper discs of 5 mm diameter (containing 30 μ l of AgNPs) along with the standard antibiotic, Streptomycin containing discs were placed in each plate. Antimicrobial activities of the synthesized AgNPs were determined, using the agar disc diffusion assay method [27].

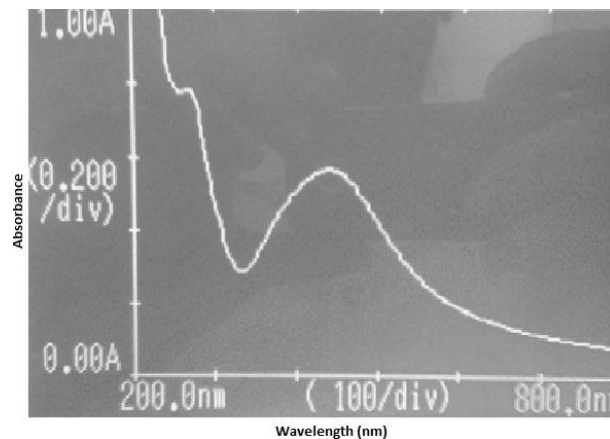


Fig. 1. UV-Vis absorption spectrum of AgNPs synthesized by treating 1mM AgNO₃ solution with *M. oleifera* leaf extract after 6 hrs.

Results and discussion

UV-Visible studies

UV-Vis spectroscopy is an important technique to establish the formation and stability of metal nanoparticles in aqueous solution [28]. The relationship between UV-visible radiation absorbance characteristics and the absorbate's size and shape is well-known. Consequently, shape and size of nanoparticles in aqueous suspension can be assessed by UV-visible absorbance studies.

Fig. 1 depicts the absorbance spectra of reaction mixture containing aqueous silver nitrate solution (1 mM) and *M. oleifera* leaf broth (prepared from 5 g leaf material). The absorption spectra obtained reveal the production of AgNPs within 6 h. On adding the afore-mentioned plant broth to AgNO₃ solution, the solution changed from yellowish orange to brown. The final color turns deep and finally, brownish with passage of time. The intensity of the absorbance was found to increase as the reaction proceeded further.

AgNPs displaying intense yellowish brown colour in water arises from the surface plasmons. This is due to the dipole oscillation arising when an electromagnetic field in the visible range is coupled to the collective oscillations of conduction electrons. It is an established fact that metal nanoparticles ranging from 2 to 100 nm in size demonstrate strong and broad surface plasmon peak [29]. The optical absorption spectra of metal nanoparticles that are governed by surface plasmon resonances (SPR), move towards elongated wavelengths, with the increase in particle size. The absorption band position is also strongly dependent on dielectric constant of the medium and surface-adsorbed species [30].

As postulated by Mie's theory, spherical nanoparticles results in a single SPR band in the absorption spectra. On the other hand, anisotropic particles provide two or more

SPR bands depending on the particle shape [31]. In the present study, a reaction mixture confirms a single SPR band disclosing spherical shape of AgNPs. The reduction of Ag⁺ ions was validated by performing qualitative analysis for free Ag⁺ ions presence with NaCl in the supernatant obtained after centrifugation of the reaction mixture. The AgNPs obtained from the reaction mixture consisting of 1mM AgNO₃ and leaf extract, were purified and further examined. The AgNPs were found to be amazingly stable even after 6 months.

Rapid synthesis of steady AgNPs using leaf broth (20 g of leaf biomass) and 1 mM aqueous AgNO₃ have been reported by Sastry et al. [10] Shivshankar et al. [11] have reported rapid synthesis of stable gold, silver and bi-metallic Ag/Au core shell nanoparticles using 20 g of leaf biomass of 1mM aqueous AgNO₃. Similarly, Pratap et al. [29] reported synthesis of gold and silver nanoparticles with leaf extract using ammonia as a speed-up agent for synthesis of silver. All the above research papers used leaf broth made by boiling finely chopped fresh leaves. This procedure involves incessant agitation of the broth after addition of salt solution.

The reduction of the silver ions is moderately rapid at ambient conditions. This is innovative and interesting to the field of material science as, the evaluated leaf biomass was found to have the capability to reduce metal ions at ambient circumstances. Furthermore, the biomass handling and processing is less rigid since, it does not involve boiling for long hours or successive treatment.

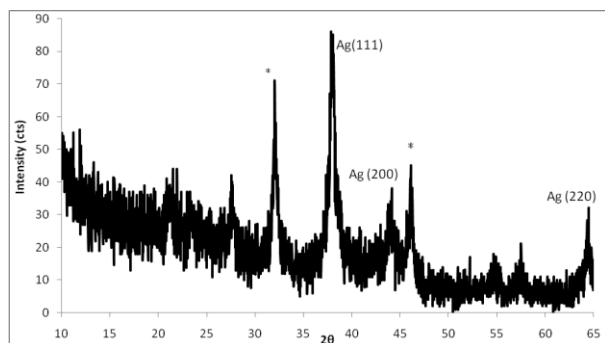


Fig. 2. XRD pattern of the green synthesized AgNPs.

XRD studies

Fig. 2 shows the XRD-spectrum of purified sample of AgNPs. The peaks observed in the spectrum at 2θ values of 38.07°, 44.15° and 64.49° corresponds to 111, 200 and 220 planes for silver, respectively [15].

Some unidentified peaks were also observed near the characteristic peaks. A peak at 46° is possibly due to crystalline nature of the capping agent [32, 10]. This clearly shows that the AgNPs are crystalline in nature due to reduction of Ag⁺ ions by *M. oleifera* leaf extract. The AgNPs were centrifuged and redispersed in distilled water several times before XRD and EPMA analysis, as mentioned earlier. This excludes the possibility of any free compound/protein present that might lead to independent crystallization and thus, resulting in Bragg's reflections. Usually, the particle size is responsible for the broadening of peaks in the XRD pattern of solids [33]. The noise due

to the protein shell surrounding the nanoparticles is visible from the spectrum [34].

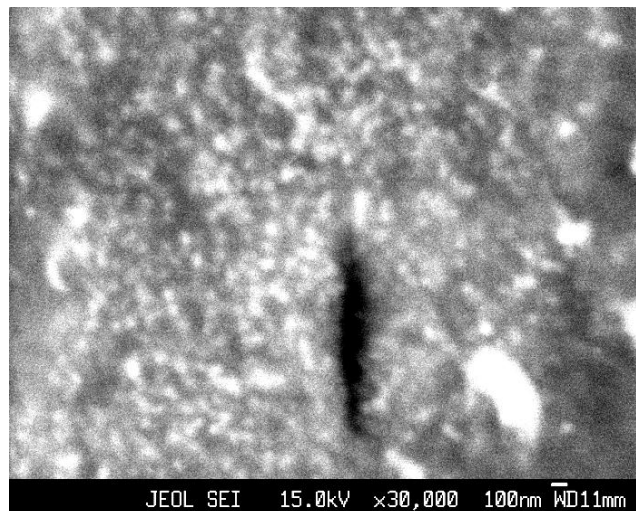


Fig. 3. EPMA micrograph of the AgNPs

EPMA

An EPMA was employed to examine the structure of the nanoparticles that were synthesized. Representative EPMA micrographs of the reaction mixtures comprising 10 ml of *M. oleifera* leaf extract and 1 mM of silver nitrate magnified 30,000 times are shown in Fig. 3. From the figure, it is clear that the AgNPs adhered to nano-clusters.

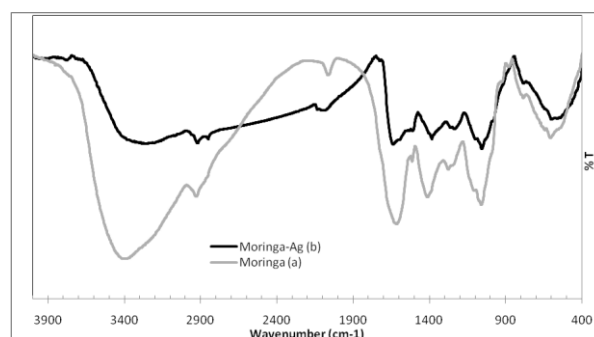


Fig. 4. FTIR spectra of dried powder of (a) *M. oleifera* leaf extract (b) nanoparticles synthesized by leaf extract solution.

FTIR studies

The exact procedures of bio-reduction is not fully comprehended and have reported that the reduction of Ag⁺ to Ag nanoparticles takes place possibly in the presence of the enzyme, NADPH-dependent dehydrogenase [32]. The precise direction in which the electrons are transported is a matter requiring investigation. Moreover, the information concerning environment being responsible for high stability of metal nanoparticles, is not widely available. FTIR investigation of isolated AgNPs free from proteins and water-soluble compounds was performed in this direction. The analysis of IR spectra throws light on biomolecules bearing different functionalities present in fundamental system. Representative spectra (Fig. 4b) clearly show the purified nanoparticles showed the presence of bands due to

O–H stretching ($\sim 3315\text{cm}^{-1}$), aldehydic CH stretching ($\sim 2914\text{ cm}^{-1}$), C=C group ($\sim 1627\text{ cm}^{-1}$) and C–O stretch ($\sim 1043\text{ cm}^{-1}$).

The FTIR spectra of biomass (Fig. 4a) show bands ~ 3403 , ~ 2919 , ~ 2048 , ~ 1604 , ~ 1504 , ~ 1402 , ~ 1270 and $\sim 1045\text{ cm}^{-1}$. The band $\sim 1045\text{ cm}^{-1}$ can be allotted to the ether linkages or -C-O-, 11, 14 (Fig. 4a). The band at $\sim 1045\text{ cm}^{-1}$ largely might be due to the -C-O- groups of the polyols viz. flavones, terpenoids and the polysaccharides present in the biomass [14]. The absorbance band $\sim 1604\text{ cm}^{-1}$ (Fig. 4a) is associated with the stretching vibration of -C=C- or aromatic groups [11, 14]. The band ~ 2048 may be due to C=O stretching vibrations of the carbonyl functional group in ketones, aldehydes, and carboxylic acids. Also, the spectrum (Fig. 4a) also reveals an intense band $\sim 2919\text{ cm}^{-1}$ allocated to the asymmetric stretching vibration of sp^3 hybridized -CH₂ groups [32].

The band $\sim 1627\text{ cm}^{-1}$ (Fig. 4b) is due to amide-I bond of proteins, indicating predominant surface capping species having -C=O functionality which are mainly responsible for stabilization. A broad intense band $\sim 3400\text{ cm}^{-1}$ in both the spectra can be contributed to the N-H stretching frequency arising from the peptide linkages present in the proteins of the extract [32]. The shoulders around the band can be specified as the overtone of the amide-II band and the stretching frequency of the O-H band, possibly arising from the carbohydrates and/or proteins present in the sample. The flattening of the shoulders in Fig 1b indicates decrease in the concentration of the peptide linkages in the solution [32]. The spectra Fig. 4b also shows broad asymmetric band $\sim 2100\text{ cm}^{-1}$ that can be assigned to the N-H stretching band in the free amino groups of AgNPs. The bands of functional groups such as -C-O-C-, -C-O- and -C=O are obtained from the heterocyclic water soluble compounds in the biomass, which as observed in the IR spectra of biomass is in good agreement with the value reported in the literature.

Gold nanoparticles are thoroughly investigated in the polyol synthesis and both oxygen and nitrogen atoms of pyrrolidone unit can assist the adsorption of PVP on to the surface of metal nanostructures to safeguard the synthesized nanoparticles [35]. Similarly, the oxygen atoms here might assist the adsorption of the heterocyclic components on to the particle surface in stabilizing the nanoparticles. It is also obvious from the two spectra of biomass and AgNPs that the flavones lead to the bio-reduction. Flavones could be adhered to the surface of the metal nanoparticles, probably by interaction through π -electrons of carbonyl groups in the absence of other strong ligating agents in adequate concentrations [11].

Antibacterial studies

Antibacterial activity of biogenic AgNPs was evaluated by using standard Zone of Inhibition (ZOI) microbiology assay. The nanoparticles showed inhibition zone against almost all the studied bacteria (Table 1, Fig 5).

Maximum ZOI was found to be 12 mm for *Escherichia coli* (Clinical isolate) and no ZOI for *Pseudomonas aeruginosa*. Whereas, the other three bacterial strains of *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Enterococcus faecalis* showed ZOI of 7, 9

and 6 mm whereas, ZOI of the standard antibiotic, streptomycin obtained against *Escherichia coli* was found to be 11 mm (Table 1).

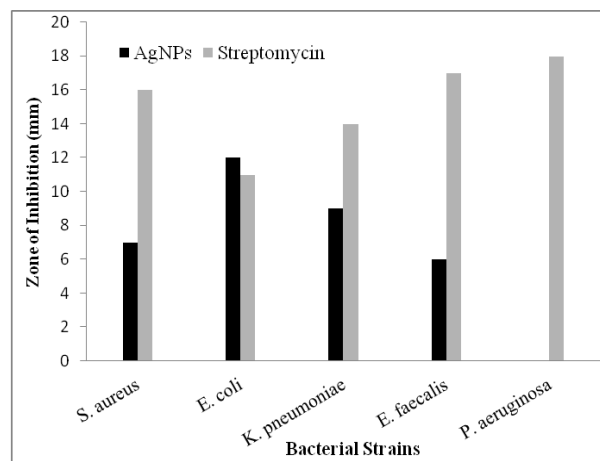


Fig. 5. Antimicrobial activity of silver nanoparticles synthesized from *M. oleifera* leaf extract against microorganism (bacteria).

Table 1. Zone of Inhibition of AgNPs synthesised from *M. oleifera* leaf extract.

Bacterial Strains	Zone of Inhibition (mm)	
	AgNPs	Reference drug*
<i>Staphylococcus aureus</i>	7	16
<i>Escherichia coli</i>	12	11
<i>Klebsiella pneumoniae</i>	9	14
<i>Enterococcus faecalis</i>	6	17
<i>Pseudomonas aeruginosa</i>	-	18

AgNO₃ which is readily soluble in water has been exploited as an antiseptic agent for many decades [36]. Dilute solution of silver nitrate has been used since the 19th century to treat infections and burns [37]. The exact mechanism of the antibacterial effect of silver ions is partially understood. Literature survey reveals that the positive charge on the Ag ion is crucial for its antimicrobial activity. The antibacterial activity is probably derived, through the electrostatic attraction between negatively-charged cell membrane of microorganism and positively-charged nanoparticles [38-40]. However, Sondi and Salopek-Sondi [41] reported that the antimicrobial activity of AgNPs on Gram-negative bacteria was dependent on the concentration of AgNPs and was closely associated with the formation of pits in the cell wall of bacteria. Accumulation of the AgNPs in the pits results in the permeability of the cell membrane, causing cell death. Similarly, Amro et al. [42] suggested that depletion of the silver metal from the outer membrane may cause progressive release of lipopolysaccharide molecules and membrane proteins. This results in the formation of irregularly-shaped pits and hence increases the membrane permeability. Similar mechanism has been reported to be operative by Sondi and Salopek-Sondi [41] in the membrane structure of during treatment with Ag

nanoparticles. Recently, Kim and co-workers [43] have reported that the silver nanoparticles generate free radicals that are responsible for damaging the membrane. They also speculated that the free radicals are developed from the surface of the AgNPs. Lee et al. [44] investigated the antibacterial effect of nanosized silver colloidal solution against padding the solution on textile fabrics. Shrivastava et al. [45] studied antibacterial activity against *E. coli* (ampicillin resistant), and *S. aureus* (multi-drug resistant). They reported that the effect was dose-dependent and was more pronounced against gram-negative organisms than gram-positive ones. They found that the major mechanism through which AgNPs manifest antibacterial properties was either by anchoring or penetrating the bacterial cell wall, and modulating cellular signaling by dephosphorylating putative key peptide substrates on tyrosine residues [45]. Similarly, Chun-Nam and coworkers [46] reported that the AgNPs target the bacterial membrane, leading to a dissipation of the proton motive force resulting in the collapse of the membrane potential. They also proposed that the AgNPs mediated antibacterial effects in a much more efficient physiochemical manner than Ag⁺ ions. The antibacterial efficacy of the biogenic AgNPs reported in the present study may be ascribed to the mechanism described above but, it still remains to clarify the exact effect of the nanoparticles on important cellular metabolism like DNA, RNA and protein synthesis.

Conclusion

The present study represents a clean, non-toxic as well as eco-friendly procedure for synthesizing AgNPs. The capping around each particle provides regular chemical environment formed by the bio-organic compound present in the *M. oleifera* leaf broth, which may be chiefly responsible for the particles to become stabilized. This technique gives us a simple and efficient way for the synthesis of nanoparticles with tunable optical properties governed by particle size. From the of nanotechnology point of view, this is a noteworthy development for synthesizing AgNPs economically. In conclusion, this green chemistry approach toward the synthesis of AgNPs possesses several advantages viz, easy process by which this may be scaled up, economic viability, etc. Applications of such eco-friendly nanoparticles in bactericidal, wound healing and other medical and electronic applications, makes this method potentially stimulating for the large-scale synthesis of other inorganic materials, like nanomaterials. Toxicity studies of *M. oleifera*-mediated synthesized AgNPs are also underway.

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