Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Archives of Applied Science Research, 2013, 5 (3):45-49
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0975-508X
CODEN (USA) AASRC9

Biosynthesis of silver nanoparticles using leaf extract of *Daturaalba* Nees. and evaluation of their antibacterial activity

Abasaheb R. Nalwade[a] and AsawariA. Jadhav[b]

[a]Plant Tissue Culture Laboratory, Annasaheb Awate College, Manchar, Pune (MS), India
[b]Department of Biotechnology, Annasaheb Awate College, Manchar, Pune (MS), India

---

ABSTRACT

Synthesis of silver nanoparticles is achieved through methods such as physical, chemical and biological. Biological method of nanoparticles synthesis is economical and eco-friendly. The plant extract of DaturaalbaNees. incubated with silver nitrate showed gradual change in colour from colourless to reddish brown with increasing intensity. UV-Visible spectrophotometry revealed that peak was obtained at 444 nm. X-Ray Diffraction analysis has shown that silver nanoparticles were crystalline and domain particle size was 28.42 nm. Scanning Electron Microscopy analysis showed aggregates silver nanoparticles and these were spherical in shape. These phytosynthesized nanoparticles were tested for their antibacterial activity. Antibacterial activity of silver nanoparticles was analyzed by measuring the inhibitory zone. It was observed against Chlostridiumdiptheriae.

Keywords: Silver nanoparticles, X-ray Diffraction, Scanning Electron Microscopy, Antibacterial activity

---

INTRODUCTION

Nanotechnology is intensively developed during the last decade and represents one of the most important dimensions in the technological developments of the developed countries. Employment of nanoparticles opens new perspective in electronics, chemical industry, energetic, biology and medicine. Nanotechnology influences all aspects of our life. The field of nanotechnology is the most important area of research in modern material science. Nanoparticles are synthesized by different methods like chemical, physical, sonochemical and bioreduction. Chemical synthetic methods lead to the presence of some toxic chemicals adsorbed on the surface that may have adverse effect in the medical applications.Research community from the field of nanotechnology strongly believes that “Bionanofactories” will overcome the problem, which is coming out with environmental concern. Instead of chemical and physical methods for nanomaterial production, using microbes and plants will help to synthesize the materials in the nano range and in addition, the toxicity of the by-product would be lesser than the others.

*Daturaalba*Nees. has been extensively used in medicine to cure human ailments like asthma, muscle spasm, whooping cough, hemorrhoids, skin ulcers, sciatica etc. In India, it is widely used for the relief of rheumatism and other painful affections. Oil based preparations of this plant are used for all type of wounds from ancient days. Though whole plant has medicinal value, the leaves and seeds alone are recognized as official. The toxicological effects of this plant find its use in the medicinal field.

In the present work, we report the synthesis of silver nanoparticles by reduction of silver ions present in the aqueous solution of silver nitrate by the leaf extract of *Daturaalba*Nees. a common weed in the tropical and sub-tropical regions. The antibacterial activity of synthesized silver nanoparticles has been tested against *Chlostridiumdiptheriae.*

MATERIALS AND METHODS

Synthesis of silver nanoparticles using extract of Daturaalba Nees. Leaves

25 g *Daturaalba* Nees. leaves were washed thoroughly with sterile distilled water and blotted to dryness. Leaves were finally cut and were boiled for 30 min in 80 ml of sterile distilled water. It was filtered through Whatman No. 1 filter paper. Volume of the filtrate was adjusted to 100 ml with sterile distilled water. 10 ml filtered extract was added to 90 ml of 1 mM silver nitrate solution. The reaction was allowed to proceed for 4 h at room temperature.

Characterization of silver nanoparticles

The bioreduction of silver ions was monitored by measuring the UV-Vis spectra of the reaction mediumusing UV-2450 (Simatzu) machine. The silver nanoparticle solution thus obtained was purified by repeated centrifugation at 10,000 rpm for 20 min followed by re-dispersion of the pellet of silver nanoparticles into 10 ml of sterile distilled water. After freeze drying of purified silver nanoparticles, the structure and composition were analyzed by X-Ray diffraction (RIGAKU-D Machine). The crystalline domain size was calculated from the width of XRD peaks using Scherrer's equation.

Scherrer's equation

$D = K\lambda / \beta \cos\Theta$

Where,
$\beta = \pi / 180*FWHM$ (FWHM = Full Width Half Maximum), $\lambda = 1.540598\ A^o$,
$K\lambda = 0.94*1.540598\ A^o = 1.4482$

Scanning Electron Microscopic (SEM) analysis was done using PHILIPS-XL-30SEM machine. Thin films of sample were prepared on a carbon coated copper grid by just dropping a very small amount of the sample on the grid, extra solution was removed using a blotting paper and then the films on the SEM grid were allowed to dry by putting under a mercury lamp for 5 min.

The antibacterial assaywas done on *Chlostridiumdiptheriae*by disc diffusion method. Nutrient agar medium was used to cultivate bacteria. 20 ml molten and cooled medium (Nutrient agar) was poured in sterilized petridishes. The plates were left overnight at room temperature to check for any contamination to appear. *Chlostridiumdiptheriae*was grown on nutrient agar for 24 h. Sterile paper discs of 6 mm diameter were prepared. Three discswere loaded with 30 µl of silver nanoparticles suspended 'hydrosol' and others with 30 µl of drugs were used as apositive control. These plates were incubated at 37 ^{0}C. The diameter of each zone of inhibition was measured after 48 h of incubation.

RESULTS

When 10 ml leaf extract was added to 90 ml 1 mM silver nitrate solution,the reaction started after 30 min of incubation. The formation of silver nanoparticles was confirmed by the appearance of brown colour in the reaction medium and a dark brown colour was formed after 4 h (Fig. 1).For verification of synthesis of nanoparticles, the reaction medium was subjected to UV-Vis spectrophotometer analysis. A peak at 444 nm was obtained.(Fig. 2). Broadening of peak indicated that the particles are polydispersed. XRD analysis showed bragg reflections at (111), (200) and (220) lattice planes in the range of 10 to 70 2Θ value (Fig. 3). The particles domain size was 28.42 nm.

Fig. 1: Photograph of (A) *Daturaalba*Nees. leaf extract B) 1.0 mM $AgNO_3$ solution without leaf extract (C) Colloidal solution of silver nanoparticles

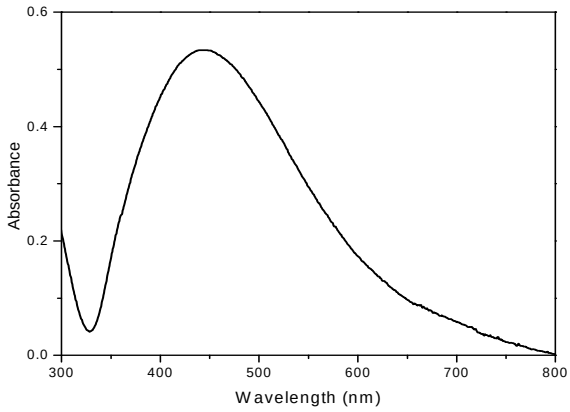

Fig. 2: UV-Visible spectra of silver nanoparticles

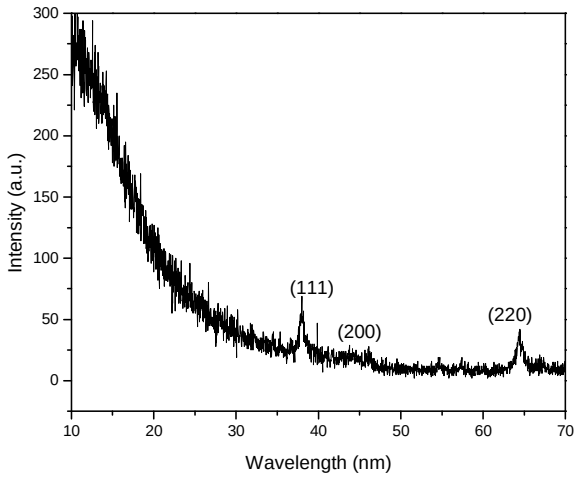

Fig. 3: XRD spectrum of silver nanoparticles

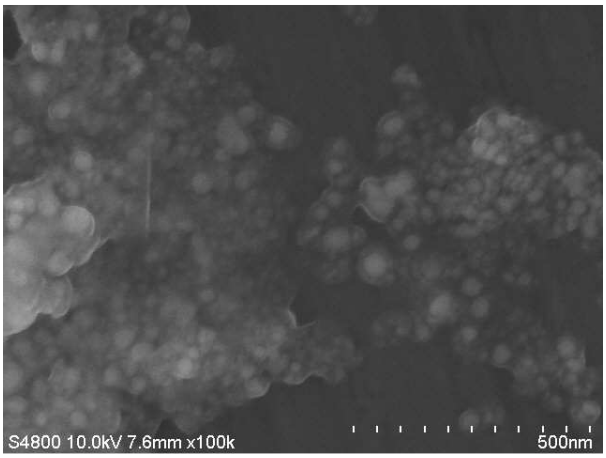

Fig. 4: SEM image of silver nanoparticles synthesized using *Daturaalba*Nees.leaf extract

The SEM image (Figure4) shows the high density silver nanoparticles synthesized by *Daturaalba*Nees. leaf extract. The silver nanoparticles were spherical in shape. Antibacterial activity of *Daturaalba*Nees. leaf derived silver nanoparticles was assessed and found that inhibitory zone for *Chlostridiumdiptheriae*was 20 mm.

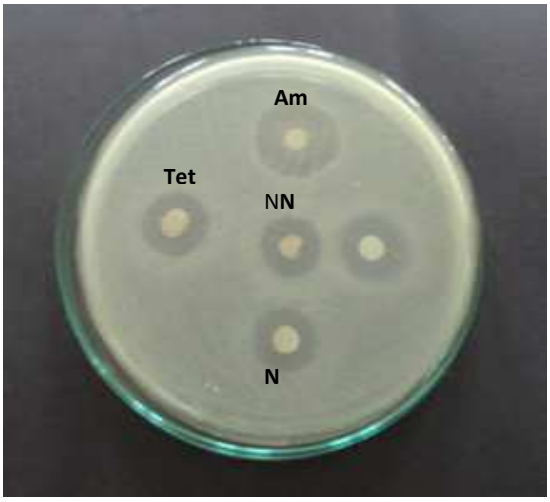

Fig. 5:*Chlostridiundiptheriae*

DISCUSSION

Bioreduction of silver ions present in the aqueous extract was observed by change in colour from yellow to brown. The change in colour was observed due to excitation of surface plasmon vibrations, which indicated the formation of silver nanoparticles [1]. UV-Vis pectrophotometeric analysis is well known to investigate shape and size controlled nanoparticles. UV-Vis spectrograph showed a peak at 444 nm. X-Ray diffraction analysis revealed that nanoparticles are crystalline in nature having diameter of 28.42 nm. SEM image shows the aggregates of silver nanoparticles. Many reports well documented on the biosynthesis of silver nanoparticles using several plant extracts. It wasreported that silver nanopartcilces were synthesized using fruit extract of papaya [2]; stem bark of *Bosvelliaovalifoliolata* [3]; leaf extract of geranium [4]; *Aloe vera* [5]; *Cinnamomum camphora* [6]; *Parthenium* [7]; *Acalypha*[8]; mangosteen [9]; *Andrographis paniculata*[10]; *Clitoriaternata*[11]; *Myricaesculenta*[12]; *Tridaxprocumbens*[13]; *Mallunganudicaulis*[14]; *Ocimum sanctum*[15] and *Sesbania grandiflora*[16].

The mechanism of silver nanoparticles synthesis is not clear. The role of reducing sugars has been speculated for the reduction of silver nitrate to silver nanoparticles [17]. In another study it has been suggested that caffeine and theophylline bring out the reduction process and thus silver nanoparticle synthesis [8]. Natural antioxidants have been reported to have strong reducing ability [28].XRD analysis showed bragg reflections at (111), (200) and (220) lattice planes. The particles domain size was 28.42 nm and nanoparticles were spherical in shape. SEM image showed aggregates of silver nanoparticles (Fig. 4).

Antimicrobial tests were performed against *Chlostridiumdiptheriae.* Antibacterial activity of silver nanoparticles has been reported [2,19,20,3] against many pathogenic bacteria. The presence of nanoparticles in suspension isolated ensures continuous release of ions into the nutrient media. Silver or copper ions released by the nanoparticles may attach to the negatively charged bacterial cell wall and rupture it, thereby leading to protein denaturation and cell death [21].It was proposed that oxygen associates with silver and reacts with the sulphahydryl (=S-H) group on cell wall to form R-S-S-R bonds thus, blocking respiration and causing death of cells [22]. Surface of cell walls of *E. coli* treated with the silver nanoparticles were severely damaged compared to untreated *E. coli*[23].The cell wall ruptures due to silver ions and silver nanoparticles. The attachment of both silver ions and nanoparticles to the cell wall caused accumulation of envelops protein precursors which resulted in dissipation of the proton motive force. Silver nanoparticles also exhibited destabilization of the outer membrane and rupture of the plasma membrane there by causing depletion of intracellular ATP. Our results are consistent with earlier reports [24].

CONCLUSION

Present investigation reveals the bioreduction of aqueous silver ions by the leaf extract of *Datura alba*Nees. has been demonstrated. This green chemistry approach towards the synthesis of silver nanoparticles has many advantages such as simple, it can be easily scaled up, eco-friendly and cost effective. *Datura alba*Nees. is easily available weed on the road sides and waste lands in Maharashtra and synthesized silver nanoparticles have medical and electronic applications.

REFERENCES

[1] P. Mulvney*Langmuir***1996**, 12, 788-800.
[2] D. Jain, H. K. Daima, S. Kachnwaha, S. L.Kothari*Digest J. Nanomater. Biostruct.***2009**, 4, 723-727.
[3] N. Savithramma, M. LingaRao, P. Suvarnalatha Devi *J. Biol. Sci*.**2011**, 11, 39-45.
[4] S. S. Shankar,A. Ahmad, M.Sastry*BiotechnolProg.***2003,** 19, 1727-1631.
[5] S. P. Chandran, M. Chaudhary, R. Pasricha, A. Ahmad, M.Sastry*Biotechnol. Prog.***2006**, 22, 577.
[6]J. Huang, Q. Li, D. Sun, Y. Lu, Y. Su, X. Yang, H. Wang, Y. Wang, W. Shao, N. J. Hong, C. Chen*Nanotechnol.***2007**, 18, 105104-105115.
[7] V. Parashar, R. Parashar, B. Sharma, A. C. Pandey*Digest J. Nanomater.Biostruct.***2009**, 4, 45-50.
[8] C. Krishnaraj, E. G. Jagan, S. Rajasekhar, P. Salvakumar, P. T. Kalaichelvan,N. Mohan *Colloids and Surfaces B***2010**, 76, 50-56.
[9] R. Veerasamy, T. Z. Xin, S. Gunasagaran, T. F. W. Xiang, F. F. C. Yang, N. Jeyakumar, S. A. Dhanraj*J. Saudi Chem. Soc.***2011**, 15(2), 113-123.
[10] S. Sulochana, P. Krishnamoorthy, K. Sivaranjani*J. Pharma. Toxicol.***2012**, 7(5), 251-259.
[11] R. B. Malabadi, G. S. Mulgund, N. T. Meti, K. Nataraja, S. V. Kumar*Res. Pharma.***2012**, 2(4), 10-21.
[12] P. Phanjom, D. E. Zoremi, J. Mazumder, M. Saha, S. B. Baruah*Int. J. Nanosci. Nanotechnol.***2012**, 3(2), 73-79.
[13] T. Dhanalakshmi, S. Rajendran*Archievs Appl. Sci. Res.***2012**, 4(3), 1289-1293.
[14) J. Anarkali, D. Vijayraj, K. Rajathi, S. Sridhar*Archi.Appl. Sci. Res.* **2012**, 4(3), 1436-1441.
[15] C. Ramteke, T. Chakrabarti, B. K. Sarangi, R. A. Pandey*J. Chem.***2013**, 2013, 7.
[16] J. Das, D. M. Paul, P. Velusamy*SpectrochimActa A: Mol. Biomol. Spectroscopy***2013**, 104, 265-270.
[17] S. S. Shankar, A. Rai, A. Ahmad, M. Sastry*Chem. Mater.***2005**, 17(3), 566-572.
[18] Y. Zhou, W. Lin, J. Huang, W. Wang, Y. Gao, L. Lin, Q. Li, L. Lin, M. Du *Nanoscale Res. Lett.***2010**, 5(8), 1351-1359
[19]S. Saha, J. Sarkar, D. Chattopadhyay, S. Patra, A. Chakrabotry, K. Acharya*Digest J. nanometer. Nanostruct.***2010**, 4, 723-727.
[20] N. Prabhu, T. R. Divya,G. Yamuna*Digest J. Nanomater. Biostruct.***2010**, 5, 185-189
[21] Y. E. Lin, R. D. Vidic, J. E. Stout, C. A. McCartney, V. L. Yu *Water Res***1998**, 32, 1997-2000.
[22] V. Sivakumar, B. M. Nagaraja, V. Shashikala, A. H. Padmasri, S. S. Madhavendra, B. D. Raju*J. Mol. Catal. A Chem.*2004, 223, 313-319
[23] K. Cho, J. Park, T. Osaka, S. Park*ElectrochimActa***2005**, 17, 5255-5262
[24] C. N. Lok, C. M. Ho, R. Chen, Q. Y.He, W. Y. Yu, H. Sun, P. K. Tam, J. F. Chiu, C. M. Che. *J. Biol. Inorg. Chem.***2007**, 12, 527.