Int. J. Nano Dimens., 10 (2): 217-224, Spring 2019

ORIGINAL ARTICLE

Bio-fabrication of Silver nanoparticles using *Rosa Chinensis* L. extract for antibacterial activities

Sachin Bangale [1,*]***, Suresh Ghotekar*** [2]

[1] *Department of Chemistry, G. M. Vedak College of Science, Tala 402 111, University of Mumbai, Maharashtra, India.*

[2] *Department of Applied Science and Humanities, G. M. Vedak Institute of Technology, Tala 402 111, University of Mumbai, Maharashtra, India.*

Received 11 October 2018; revised 04 December 2018; accepted 14 December 2018; available online 19 December 2018

Abstract

The purpose of this study was to expand a trouble free biological method for the synthesis of silver nanoparticles (AgNPs) using the leaves extract of *Rosa Chinensis* L. to act as reducing and stabilizing agent. Water soluble phytochemicals played a vital role for the reduction silver ions into silver nanoparticles. The leaves extract was exposed to silver ions and the resultant biosynthesized AgNPs characterized by X-ray diffraction (XRD) spectrum showed crystalline structure while morphological shape, average size and the crystalline nature of the nanoparticles were determined by field emission scanning electron microscopy (FESEM), transmission electron microscopic with selected area electron diffraction (TEM-SAED). The elemental analysis displayed strong signal at 3 keV that agrees to silver ions and confirms the presence of metallic silver. FTIR analysis exhibits the possible reducing bio-molecules within the leaf extract. Moreover, AgNPs nanoparticles evinced excellent antibacterial activity against *Staphylococcus aureus* and *Streptococcus pyogenus* bacterial pathogens. The studies describing the synthesis of AgNPs nanoparticles by efficient and green method followed by the investigation of antibacterial activities may be opened new horizons to scientists and researchers for the medicinal purposes.

Keywords: *Agnps; Antibacterial Activities; Green Synthesis; Nanotechnology; Rosa Chinensis L.; TEM.*

How to cite this article

Bangale S, Ghotekar S. Bio-fabrication of Silver nanoparticles using Rosa Chinensis L. extract for antibacterial activities. Int. J. Nano Dimens., 2019; 10 (2): 217-224.

INTRODUCTION

Nowadays, interest of nanobiotechnology is the development of environmentally benign technology for the synthesis of metal/metal oxide nanoparticles with miraculous and boundless applications in the agriculture, cosmetics, defense, environmental safety, food, health and pharmaceutical [1-9]. Metallic nanoparticles exhibit enormous chemical, optical, physical, and thermal properties due to large surface area, spatial confinement and reduced imperfections. The reduction of material size and shape has pronounced effects on the physical properties that may be significantly different from the corresponding bulk material [10]. Among all metals nanoparticles, AgNPs has gained significant boundless interest because of their wide range of application in catalysis [11], sensors [12-14], photonics [15], and photocatalysis [16]. Moreover, AgNPs have great applications in antibacterial [17], antifungal [18], antiviral [19], ant angiogenesis [20], anti-inflammatory [21], etc. and which can be incorporated into cryogenic superconducting material, composite fibres, cosmetic products, electronic components and food industry [20]. AgNPs are competent biological properties, which are almost used in antiseptic sprays, fabrics, topical creams, wound dressing and successfully used in the cancer diagnosis and treatment [21-22].

Hitherto, several chemical and physical methods were employed for the synthesis of AgNPs in the past such as including microwave

* Corresponding Author Email: *bangale_sv@rediffmail.com*

assisted [23], electrochemical [24], chemical reduction method [25], radiation assisted [26], thermal decomposition, chemical and photochemical reactions [27]. However, these methods have some disadvantages like the use of toxic chemicals, need of special instruments, long reaction time, and requirement of external additives during the reaction. But green synthesis protocol is economically affordable, environmentally benign, no need of high temperature or pressure and importantly toxic chemicals. Therein, ecofriendly methods for metal nanoparticles synthesis using the microorganism [28], fungi [29], enzyme [30], plant extracts [31], etc. are given much more attention. Among the several biosynthetic approaches, the use of plant extracts has received much remarkable as they are safe to handle, easily available and have a broad variability of phytochemicals. Moreover, synthesis of functionalized AgNPs using photochemical transformations in test tube play an indispensable role; because the functional groups of various phytochemicals enhance the reduction of silver ions to elemental silver. The phyto constituents such as tannins, carbohydrates, flavonoids, saponins, coumarins, proteins, amino acids, and terpenoids present in the plant extracts play an important role in the synthesis of nanoparticles. The scrutiny of the literature revealed some notable plant parts extract used for facile synthesis of AgNPs. For example *Leucaena leucocephala* L. [17], *Ziziphora tenuior* [31], *Azadirechta indic* [33], *Pistacia atlantica* [33], *Nervalia zeylanica* [34], *Ipomoea digitata* [35], *Caesalpinia pulcherrima* [36], *Hedichium spicatum* [37], *Gymnema sylvestre* [38], *Pomegranate peel* [39], *Buddleja globosa* [40], *Caralluma fimbriata* [41], *Peganum harmala* [42] and unripe fruit of *Annona reticulata* [43] has been already reported.

Herein, we investigate the efficient, cost effective, and safe and ecofriendly green synthesis of AgNPs using leaves extracts of *Rosa chinensis* L. and their antibacterial activity against some human pathogens has been evaluated. The results would help to utilize as synthesized AgNPs effectively in future biomedical concerns.

MATERIALS AND METHODS

Preparation of Rosa chinensis L. extract

The fresh leaves (Fig. 1) of *Rosa chinensis* L. were collected from the campus of G. M. V. College of Science. The collected leaves were washed thoroughly with distilled water and incised into small pieces. About 5 g of finely cut leaves of *Rosa chinensis* L. were weighed and transferred into a washed 250 ml beaker containing 100 ml distilled water, mixed well and boiled for 15 minutes at 80-100 ºC. Mixture was cooled at room temperature and then filter through ordinary filter paper. The extract obtained was filtered through Whatmann number 41 filter paper and the filtrate was collected in 250 ml Erlenmeyer flask. The collected filtrate is used for further synthesis of AgNPs.

Bio-fabrication of AgNPs

The aqueous solution of 0.01 M silver nitrate ($AgNO_3$) was prepared by using double distilled water and used for the synthesis of AgNPs. The leaves extract of *Rosa chinensis* L. Was mixed to 0.01M of $AgNO_3$ solution in 1:9 ratios in a conical flask and color of medium changed to brown within 1 min.The solution turned to brown color, indicating the formation of AgNPs. Then solution was incubated for 24 h at room temperature. The resultant solutions were centrifuged at 8000 rpm for 5 min (20^0C) and the mixture was collected after discarding the supernatant. The collected AgNPs were allowed to dry in a watch glass. A fine shiny black colored material was obtained and this was carefully collected for characterization purposes.

Characterization of the synthesized AgNPs

The powder of green synthesized AgNPs was used for the Fourier transform Infra-red (FT-IR) (JASCO 4100) analysis. Find the exact morphological structures and size of the AgNPs using transmission electron microscopic (TEM) with selected area electron diffraction(SAED)

Fig.1. Leaves of *Rosa Chinensis* L.

analysis is done by using a Philips CM 200 operated at accelerating voltages of 20 and 200 kV. X-ray diffraction (XRD) pattern of AgNPs was obtained using Bruker D8-Advanced Diffractometer (λ=1.54 Å) from which average crystallite size of AgNPs was calculated. The surface morphology, purity and chemical composition study of synthesized AgNPs were carried out by field emission scanning electron microscopy (FESEM) and Energy Dispersive Spectroscopy (EDS) (JEOL JSM-6701).

Phytochemical Screening

Fresh aqueous leave extract of *Rosa chinensis* L. were used for phytochemical screening. Phytochemical screenings were carried out by standard method [44].

Antibacterial activity of synthesized AgNPs

The Minimum inhibition concentration (MIC) of biogenically fabricated AgNPs and leaves extract of *Rosa chinensis* L. were carried out by broth micro-dilution protocol [45]. DMSO was used as diluents to get desired concentration of drugs to test upon standard pathogenic bacterial strains. Serial dilutions were prepared in primary and secondary screening. The control tube containing no antibiotic was immediately subculture by spreading evenly over a plate of medium suitable for the growth of the test bacterial pathogens and incubated overnight at 37 ºC. The MIC of the control bacterial strain was measured to check the accuracy of the drug concentrations. The lowest concentration inhibiting growth of the bacterial pathogen was recorded as the MIC. The amount of growth from the control tube before incubation was compared. Subcultures might evince same number of colonies indicating bacteriostatic, a reduced number of colonies indicating a slow bactericidal activity and no growth if the whole inoculums has been killed. The test must include a second set of the same dilutions inoculated with a bacterial pathogen. A synthesized AgNPs and leaves extract was diluted obtaining 2000 µg/ml concentration, as a stock solution. In primary screening, 500, 250 and 125 µg/ml concentrations of the synthesized AgNPs and leaves extract were taken. The synthesized AgNPs and leaves extract found in this primary screening were further tested in a second set of dilution against all microorganisms. The silver nanoparticles found active in primary screening were similarly diluted to obtain 100, 50, 25, 12.5, 6.250, 3.125 and 1.5625 µg/ml concentrations. The highest dilution showing at least 99 % inhibition is taken as MIC [46].

RESULTS AND DISCUSSIONS

Structural & crystallographic analysis

X-ray diffraction analysis (2θ range is 30º-80º) clearly illustrates crystalline nature of the synthesized AgNPs in (Fig. 2.)The prominent peaks at 2θ=38.10º, 44.40º, 64.50º and 77.40º corresponding to the (111), (200), (220) and (311) Bragg's reflections of the Face Cubic Centered crystal structure (JCPDS card no. 04-0783) of AgNPs, respectively. The average crystallite sizes (D) of AgNPs were calculated by using Debye-Scherrer's equation.

$$D = K\lambda / \beta \cos\theta \quad (1)$$

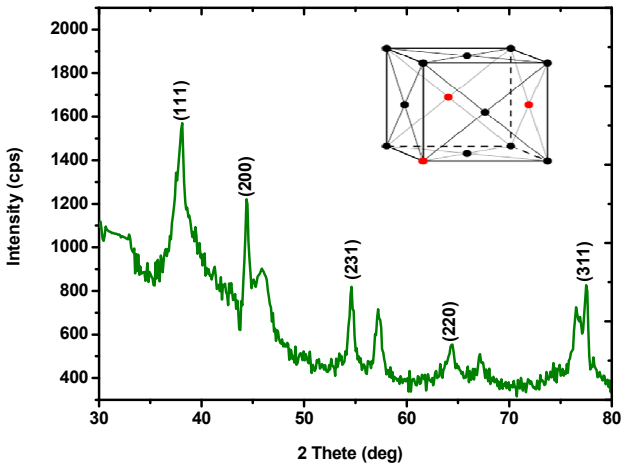

Fig.2. X-ray diffraction profile of biosynthesized AgNPs.

Where, D is the crystal size of synthesized AgNPs (nm), θ is Bragg angle (degrees), λ is the wavelength of the X-ray source used (1.54060 Å), β is the angular width at the half maximum of the diffraction peak (radians) and K is the constant of Debye-Scherrer's equation which is generally, for the spherically grown nanoparticles 0.94. The average crystallite size of the synthesized AgNPs is found 42±5 nm. Thus, XRD pattern clearly illustrated that the AgNPs formed in this study are pure face-centered cubic crystalline in nature and was good agreement with TEM and FESEM results.

FESEM analysis

The acquaintance about surface morphology and crystal size of the synthesized AgNPs has been analyzed by FE-SEM microphotographs. Figs. 3 (a, b, c, d) shows FE-SEM images at different magnifications. It seems that surface is spongy and it can be observed that the average crystal grain size of the quasi-spherical morphology AgNPs was mainly 35-50 nm except for slight agglomeration. This result exceeds the literature result which quasi-spherical shape of AgNPs was prepared by green synthesis method [17].

TEM analysis

To get better understanding of the morphology of AgNPs is shown in TEM images [Figs. 4 (a,b,c,d)]. It indicates the presence of AgNPs with size 25-60 nm which form bead type of aggregation throughout the region, on the contrary the image shows distinct nanoparticles of nearly spherical structure which are correlated well with the XRD results. SAED pattern also shows the spot type pattern which is indicative of the presence of single crystallite particles. No evidence was found for more than one pattern, suggesting the single phase crystalline nature of the material.

EDS analysis

The composition of green synthesized AgNPs has been analyzed by investigating the energy-dispersive X-ray spectroscopy (EDS) and exhibits strong peak at 3eV (Fig. 5) confirms the formation of AgNPs. This quantitative data confirms the purity, elemental composition and formation of AgNPs nanoparticles.

FT-IR studies

FTIR spectroscopic studies were carried out to

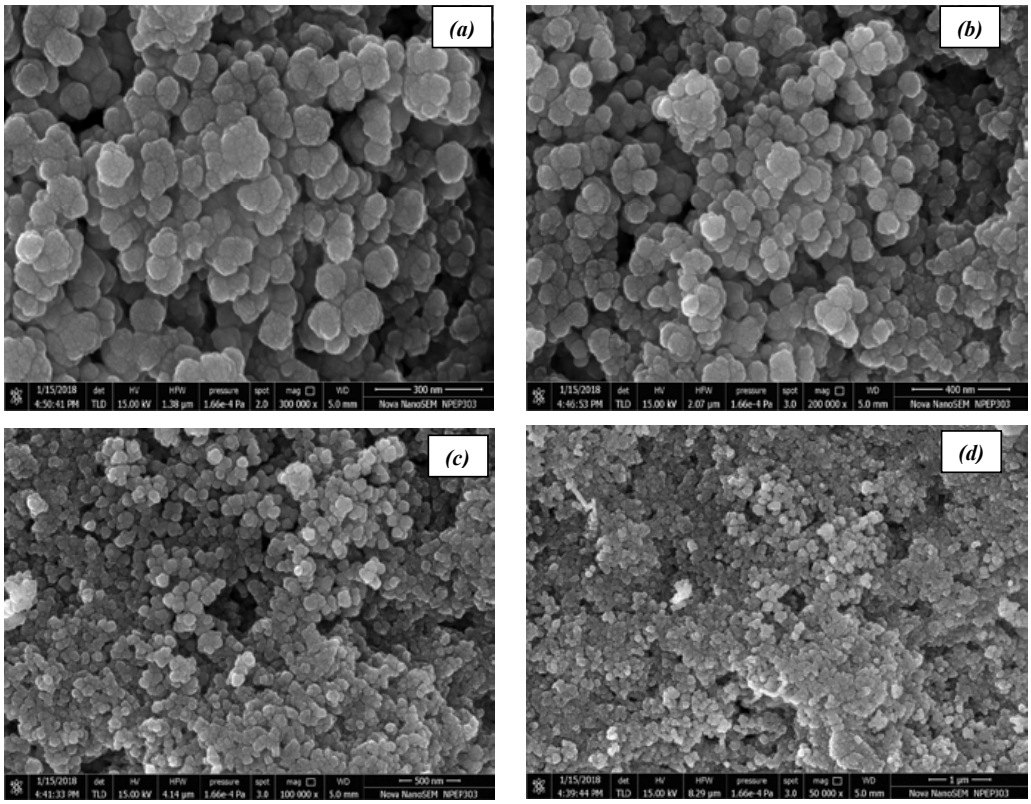

Fig.3. SEM (Fig. a, b, c and d), images of synthesized silver nanoparticles synthesized from *Rosa Chinensis* L.

find out the possible chemical changes present in the extract. The spectra were recorded before and after addition of $AgNO_3$ solution. The broad peaks of the leaves extract and nanoparticles shown in (Fig.6 a) The peak at 3284 cm^{-1} belongs to O-H stretching of phenolic compounds and the bands at 2916 and 2849 cm^{-1} region arising from C-H stretching of aromatic compound were observed. The bands at 1733cm^{-1}areobserved as amide, ester, and acids arise due to carbonyl group stretching vibration. The stretching vibration of -C-C- in aromatic ring causes an absorption peak at 1610 cm^{-1}. The band at 1027 cm^{-1} corresponds to C-OH stretching in leaves extract. Simultaneously,

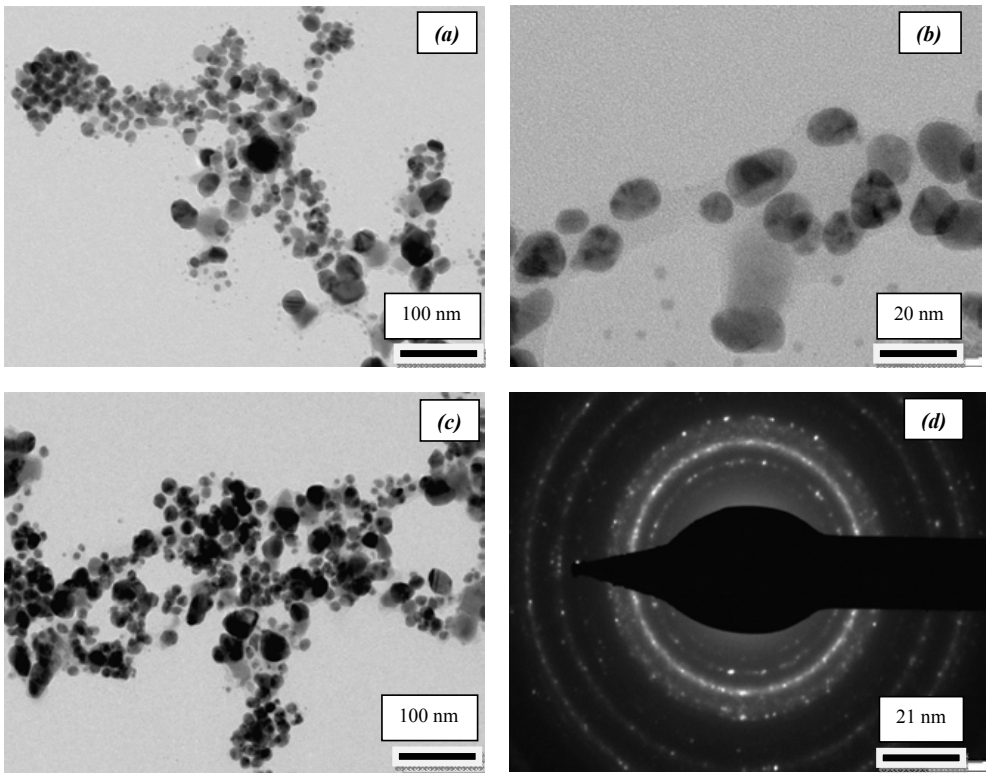

Fig. 4.TEM micrographs of biosynthesized AgNPs (a) AgNPs at 100 nm scale, (b) AgNPs at 20 nm scale (c). AgNPs at 100 nm scale (d) SAED pattern of AgNPs.

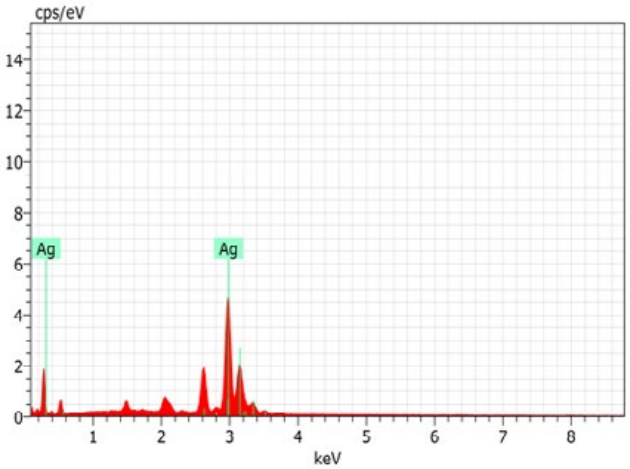

Fig.5. EDS spectrum of biosynthesized AgNPs.

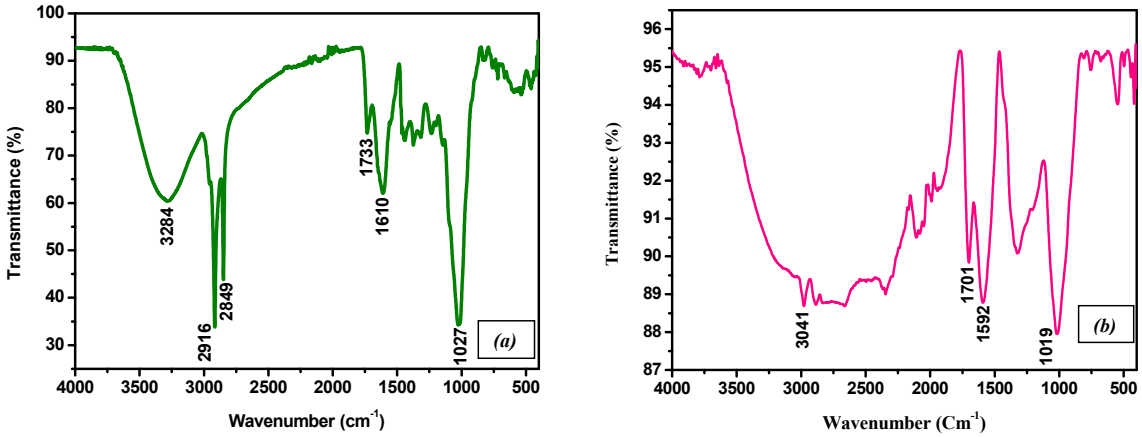

Fig.6. (a) FT-IR spectrum of aqueous leaves extract of *Rosa Chinensis* L., (b) FT-IR spectrum of bio-fabricated AgNPs.

Table 1. Photochemical Screening of aqueous leaves extract of *Rosa chinensis* L.

Phytochemical	Test	Phytochemical	Test
Tannin	+	Flavonoids	+
Protein	-	Emodins	+
Phenols	+	Anthocyanin	-

Table 2. Minimum inhibition concentration of *Rosa chinensis* L. aqueous extract and AgNPs against bacterial pathogens.

Test pathogens	Plant Extract	MIC (µg/ ml) AgNPs	Reference drug
E. coli (MTCC-443)	100	125	100
K. pneumonia (MTCC-109)	200	200	100
P. aeruginosa (MTCC-1688)	125	125	100
S. aureus (MTCC-96)	125	100	250
S. pyogenus (MTCC-442)	125	100	100

in AgNPs (Fig.6 b) the band at 3041 cm^{-1} shows the O-H stretching in phenols and alcohols. The band at 1701cm^{-1}areobserved as amide, ester, and acids arise due to carbonyl group stretching vibration and 1592 cm^{-1} represents the −C-C- in aromatic ring. The strong peak at 1019 cm^{-1} denotes the C-O stretching of phenolic compounds. Thus, mostly phenolic and flavonoid compounds are involved in the biosynthesis of nanoparticles.

Phytochemical screening studies

Table 1 describes the qualitative pharmo-cognostic evaluation of aqueous leaf extract of *Rosa chinensis* L. highlighted the presence of tannins, saponins, coumarins, flavonoids, cardial glycosides, steroids, phenols, carbohydrates, amino acids, etc. which can play a role in reduction and stabilization in the biosynthesis of nanoparticles.

Antibacterial activity of AgNPs

The results of antibacterial activity of the synthesized AgNPs are presented in Table 2. Moderate to good antibacterial activity is observed against some bacteria. Synthesized AgNPs exhibited potent and good antibacterial activity against *S. aureus*, *S. pyogenus* and moderate activity against other bacteria with ampicillin were used as the reference drug.

CONCLUSION

Bio-fabrication of stable AgNPs (using *Rosa chinensis* L. Leaves) was reported in this work. AgNPs were successfully biosynthesized by this facile, rapid, cost effective, environmentally benign, and efficient method, which excludes external reagents. In this process, the plant extract is ascribed to the relative levels of phenols, flavonoids, tannins and emodins which act as reducing as well as capping agents AgNPs. In addition, the biosynthesized AgNPs are found satisfactory antibacterial agents and thus it can be used as potential candidates for various bioengineering applications and will play a vital role in nanobiotechnology.

ACKNOWLEDGEMENT

The author S. V. Bangale are thankful to SAIF IIT Bombay, CIF Savitribai Phule Pune University and Microcare Laboratory, Gujrat for providing the technical, instrumental and biological activities supports. Authors are also thankful to N. G. Vedak and N. S. Yadav for providing necessary facilities and encouragement during research work.

CONFLICT OF INTEREST STATEMENT

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

[1] Chaudhari R. G., Paria S., (2012), Core/Shell nanoparticles: Classes, properties, synthesis mechanisms, characterization and applications. *Chem. Rev*. 112: 2373-2433.

[2] Pansambal S., Deshmukh K., Savale A., Ghotekar S., Pardeshi O., Jain G., Aher Y., Pore D., (2017), Phytosynthesis and biological activities of fluorescent CuO nanoparticles using *Acanthospermum* hispidum L. extract. *J. Nanostruct*. 7: 165-174.

[3] Ahmed S., Ahmad M., Swami B. L., Ikram S., (2016), A review on plant extract mediated synthesis of silver nanoparticles for antimicrobial applications: A green expertise. *J. Adv. Res*. 7: 17-28.

[4] Aher Y. B., Jain G. H., Patil G. E., Savale A. R., Ghotekar S. K., Pore D. M., Pansambal S. S., Deshmukh K. K., (2017), Biosynthesis of copper oxide nanoparticles using leaves extract of *Leucaena leucocephala* L. and their promising upshot against the selected human pathogens. *Int. J. Mole. Clin. Micro*. 7: 776-786.

[5] Ghotekar S., (2019), A review on plant extract mediated biogenic synthesis of CdO nanoparticles and their recent applications. *Asian J. Green Chem*. 3: 187–200.

[6] Sashikiran P., Venkata S. K., Josthna P., Varadarajulu N. Ch., (2018), Biofabrication of silver nanoparticles by leaf extract of *Andrographis serpyllifolia* and their antimicrobial and antioxidant activity. *Int. J. Nano Dimens*. 9: 398-407.

[7] Savale A., Ghotekar S., Pansambal S., Pardeshi O., (2017) Green synthesis of fluorescent CdO nanoparticles using *Leucaena leucocephala* L. extract and their biological activities. *J. Bacteriol. Mycol. Open Access*. 5(5): 00148.

[8] Syedmoradi L., Daneshpour M., Alvandipour M., Gomez F. A., Hajghassem H., Omidfar K., (2017), Point of care testing: The impact of nanotechnology. *Biosens. Bioelectron.* 87: 373–387.

[9] Ghotekar S., Pansambal S., Pagar K., Pardeshi O., Oza R., (2018), Synthesis of $CeVO_4$ nanoparticles using sol-gel auto combustion method and their antifungal activity. *Nanochem. Res*. 3: 189-196.

[10] Albrecht M. A., Evans C. W., Raston C. L., (2006), Green chemistry and the health implications of nanoparticles. *Green Chem*. 8: 417-432.

[11] Bhosale M. A., Bhanage B. M., (2015), Silver nanoparticles: Synthesis, characterization and their application as a sustainable catalyst for organic transformations. *Current Org. Chem*. 9: 1-15.

[12] Bangale S. V., Patil D. R., Bamane S. R., (2011), Nanostructured spinel $ZnFe_2O_4$ for the detection of chlorine gas. *Sensors & Transduc*. 134: 107-119.

[13] Bangale S. V., Khetre S. M., Patil D. R., Bamane S. R., (2011), Simple Synthesis of $ZnCo_2O_4$ nanoparticles as gas-sensing materials. *Sensors & Transduc*. 134: 95-106.

[14] Bangale S. V., Bamane S. R., (2012), Preparation and electrical properties of nanostructured spinel $ZnCr_2O_4$ by combustion route. *J. Mater. Sci: Mater. Electron.* 24: DOI 10.1007/s10854-012-0739-0.

[15] Gould I. R, Lenhard J. R., Muenter A. A, Godleski S. A., Farid S., (2000), Two electron sensitization: A new concept for silver halide photography. *J. Am. Chem. Soc.* 22: 11934-11943.

[16] Paul B., Bhuyan B., Purkayastha D. D., Dhar S. S., (2015), Green synthesis of silver nanoparticles using dried biomass of *Diplazium esculentum* (*retz.*) *sw.* and studies of their photocatalytic and anticoagulative activities. *J. Mol. Liq.* 212: 813-817.

[17] Ghotekar S., Savale A., Pansambal S., (2018), Phytofabrication of fluorescent silver nanoparticles from *Leucaena leucocephala* L. leaves and their biological activities. *J. Water Environ. Nanotechnol*. 3: 95-105.

[18] Wiley B. J., Im S. H., Li Z. Y., McLellan J., Siekkinen A., Xia Y., (2016), Maneuvering the surface plasmon resonance of silver nanostructures through shape-controlled synthesis. *J. Phys. Chem. B*.110: 15666-15675.

[19] Elechiguerra J. L., Burt J. L., Morones J. R., Camacho-Bragado A., Gao X., Lara H. H., (2005), Interaction of silver nanoparticles with HIV-1. *J. Nanobiotechnol*. 5: 3-6.

[20] Gurunathan S., Lee K. J., Kalishwaralal K., Sheikpranbabu S., Vaidyanathan R., Eom S. H., (2009), Antiangiogenic properties of silver nanoparticles. *Biomaterials*. 30: 6341-6350.

[21] Nadworny P. L., Wang J., Tredget E. E., Burrell R. E., (2008), Anti-inflammatory activity of nanocrystalline silver in a porcine contact dermatitis model. *Nanomedicine*. 4: 241-251.

[22] Klaus-Joerger T., Joerger R., Olsson E., Granqvist C., (2001), Bacteria as workers in the living factory: Metal accumulating bacteria and their potential for materials science. *Trends Biotechnol*. 19: 15-20.

[23] Nadagauda M. N., Speth T. F., Varma R. S., (2011), Microwave-assisted green synthesis of silver nanoparticles. *Accounts Chem. Res*. 44: 469-478.

[24] Hirsch T., Zharnikov M., Shaporenko A., Stahl J., Wiess D., Wolfbeis O. S., (2005), Size-controlled electrochemical synthesis of metal nanoparticles on monomolecular templates. *Angew. Chem*. 44: 6775-6778.

[25] Ghotekar S. K., Vaidya P. S., Pande S. N., Pawar S. P., (2015), Synthesis of silver nanoparticles by using 3-methyl pyrazol 5-one (chemical reduction method) and its characterizations. *Int. J. Multidis. Res. Deve.* 2: 419-422.

[26] Cheng Y., Yin L., Lin S., Wiesner M., Bernhardt E., Liu J., (2011), Toxicity reduction of polymer-stabilized silver nanoparticles by sunlight. *J. Phys. Chem*. 115: 4425-4432.

[27] Plante I. J. L., Zeid T. W., Yangab P., Mokari T., (2011), Synthesis of metal sulfide nanomaterials via thermal decomposition of single-source precursors. *J. Mater. Chem*. 20: 6612-6617.

[28] Sunkar S., Nachiyar C. V., (2012), Microbial synthesis and characterization of silver nanoparticles using the endophytic bacterium bacillus cereus: A novel source in the benign synthesis. *Global J. Med. Res*. 12: 43-50.

[29] Vigneshwaran N., Ashtaputre N. M., Varadarajan P. V., Nachane R. P., Paralikar K. M., Balasubramanya R. H., (2007), Biological synthesis of silver nanoparticles using the fungus *Aspergillus flavus*. *Matt. Let.* 61: 1413-1418.

[30] Kowshik M., Ashtaputre S., Kharrazi S., Vogel W., Urban J., Kulkarni S. K., Paknikar K. M., (2003), Extracellular synthesis of silver nanoparticles by a silver-tolerant yeast strain MKY_3. *Nanotechnol*. 14: 95-100.

[31] Sadeghi B., Gholamhoseinpoor F., (2015), A study on the stability and green synthesis of silver nanoparticles using Ziziphora tenuior (Zt) extract at room temperature. *Spectrochi. Acta Part A: Mole. Biomole. Spec.* 134: 310-315.

[32] Ahmed S., Saifullah, Ahmed M., Swami B., Ikram S., (2015), Green synthesis of silver nanoparticles using *Azadirachta indica* aqueous leaf extract. *J. Rad. Res. Appl. Sci*. 9: 1-7.

[33] Sadeghi B., Rostami A., Momeni S. S., (2015), Facile green synthesis of silver nanoparticles using seed aqueous extract of *Pistacia atlantica* and its antibacterial activity. *Spect. Chim. Acta Part A: Mole. Biomol. Spect.*134: 326-332.

[34] Vijayan R., Joseph S., Mathew B., (2018), Green synthesis of silver nanoparticles using *Nervalia zeylanica* leaf extract and evaluation of their antioxidant, catalytic and antimicrobial potentials. *Part. Sci. Tech*. 7: 1-11.

[35] Varadavenkatesan T., Selvaraj R., Vinayagam R., (2018), Dye degradation and antibacterial activity of green synthesized nanoparticles using *Ipomoea digitata* Linn. flower extract. *Int. J. Environ. Sci. Tech*. 3: 1-10.

[36] Moteria P., Chanda S., (2017), Synthesis and characterization of silver nanoparticles using *Caesalpinia pulcherrima* flower extract and assessment of their in vitro antimicrobial, antioxidant cytotoxic and genotoxic activities. *Artif. Cells Nanomed. Biotechnol*. 45: 1556-1567.

[37] Ahmed A., Shah A., (2017), Synthesis of silver nanoparticles using *Hedichium spicatum* extract and their characterization. *J. Adv. Mater*. 1: 1-5.

[38] Pingale S. S., Rupanar S. V., Chaskar M., (2018), Plant-mediated synthesis of silver nanoparticles from *Gymnema sylvestre* and their use in photodegradation of methyl orange dye. *J. Water Environ. Nanotechnol*. 3: 106-115.

[39] Goudarzi M., Mir N., Mousavi-Kamazami M., Bagheri S., Salavati-Niasari M., (2016), Biosynthesis and characterization of silver nanoparticles prepared from two novel natural precursor by facile thermal decomposition method. *Sci. Rep*. 2: 1-13.

[40] Carmona E. R., Benito N., Plaza T., Recio-Sánchez G., (2017), Green synthesis of silver nanoparticles by using leaf extract from the endemic *Buddleja globosa* hope. *Green Chem. Lett. Rev*.10: 250-256.

[41] Pande S. N., Bharati K. T., Wakchure S .K., Ghotekar S. K., Gujrathi D. B., Phatangare N. D., (2015), Green synthesis of silver nanoparticles by *Caralluma Fimbriata* L. and its characterization. *Ind. J. App. Res*, 5: 749-750.

[42] Azizi M., Sedaghat S., Tahvildari K., Derakshi P., Ghaemi A., (2017), Synthesis of silver nanoparticles using *Peganum harmala* extract as a green route. *Green Chem. Lett. Rev.* 10: 420-427.

[43] Ghotekar S. K., Pande S. N., Pansambal S. S., Sanap D. S., Mahale K. M., Sonawane B., (2015), Biosynthesis of silver nanoparticles using unripe fruit extract of *Annona reticulata* L. and its characterization. *World J. Pharm. and Pharm. Sci.* 4: 1304-1312.

[44] Fransworth N. R., (1996), Biological and phytochemical screening of plants. *J. Pharma. Sci*. 55: 225-227.

[45] Rattan A., (2000), Antimicrobials in laboratory medicine. *Churchill B I, Livingstone*, New Delhi. 85–108.

[46] Kamble D. R., Bangale S. V., Ghotekar S. K., Bamane S. R., (2018), Efficient synthesis of $CeVO_4$ nanoparticles using combustion route and their antibacterial activity. *J. Nanostruct*. 8: 144-151.