Research Paper
Impact Factor: 6.089
Peer Reviewed Monthly Journal
www.ijmdrr.com

IJMDRR
E- ISSN –2395-1885
ISSN -2395-1877

CHARACTERIZATION AND GREEN SYENTHESIS OF SILVER NANO PARTICLES FROM SALVIA OFFICINALIS LEAF EXTRACT

P.Wilson* S.Venkateshwari**

**Research Scholar Department of Physics, Government Arts College, Udhagamandalam, Tamilnadu , India.*

***Assistant Professor, Department of Physics, Government Arts College, Udhagamandalam, Tamilnadu , India.*

Abstract

In this work silver nanoparticles were prepared by green synthesis from salvia L leaves extract. These particles was investigated for its morphology,particles size,chemical composition using Scanning Electron Microscopy(SEM) UV-Vis,FTIR. The result of XRD confirmed that the prepared silver particle size to be 20 nm. It is also retain the medical values for the antimicrobial activity.

Keywords- Green Syenthesis;Silver Nitrate;Medicinal Plant.

Introduction

Among Nanomaterials,silver nanoparticles are playing a major role in the field of nanotechnology and biology medicine due to their attractive physiochemical properties and in the use of Nano medicine[1]. Nanomaterial's can be useful in such areas as solar energy conversion catalysis, medicine and water treatment [2]. There are many approaches for the synthesis of silver nanoparticles such as thermal decomposition, electrochemical the use of microwave and green chemistry methods[3]. In comparison with chemical and physical methods, green synthesis has many advantages it can be cost effective, environmental friendly and easily scaled up for large-scale synthesis. The use of plant extracts to produce nanoparticles is one of environmental friendly green processes [4]. Nanoparticles are produced from plant extract, because of their medicinal properties, that could be used in drugs, targeted drug delivery and cosmetic applications[5]. Salvia is derived from the Latin word salvare, "to heal," and for centuries salvias have been valued for their medicinal and culinary qualities. Salvias, commonly known as sages, grow throughout the world. *Salvia officinal is* has been used since ancient times for snakebites, increasing women's fertility, and more[6].It was also used for hair care, insect bites and wasp stings, nervous conditions, mental conditions, oral preparations for inflammation of the mouth, tongue and throat, and also to reduce fevers[7]. The present study is carried over by green synthesis process in the preparation of silver nano particles from *salvia officinal is* leaf using reflux method[8].

Materials and Method

Silver Nitrate($AgNO_3$) was purchased from Spectrum Reagents and Chemical Pvt.Ltd Edayar, Cochin, India. Fresh and healthy leaves of *salvia officinal is(sage)* is collected from The Medicinal Plant Development Area in Dodapetta (MPDA) Nilgiris north forest division Tamilnadu India

A. Preparation Of Leaf Extract

The collected leaves were washed with distilled water to remove the dust particles. The cleaned fresh leaves were cut into small pieces and put into the round bottomed flask and is refluxed with 200ml of distilled water. The refluxing time was around 2 hours (Fig 2a). In the initial stage the solution was white where the leaves were embedded. After refluxing the color changes to pale yellow color. The extract was filtered with Whatmann no 1filter paper to collect the sedimented particles . The particles were shade dried and stored in cool place for further characterization studies.

Fig 2 (a)
Reflux method

Fig 2 (b)
salvia officinalis leaf

Fig .3

Colour change of the solution pale yellow to dark brown after the addiction of Silver Nitrate to the refluxed solution

B. Synthesis Of Silver Nanoparticles

Solution of 0.01mM of ($AgNO_3$) silver nitrate was prepared using distilled water. Salvia offcinalis extract and silver nitrate solution were taken in the ratio of 1:2 respectively. This mixture was stirred at room temperature at 400 rpm around 20 minutes until the color changes. This was then kept at rest until the precipitate completely settled down. The precipitate was centrifuged at 3000 rpm for 30 minutes for further settlement. The precipitate was collected and washed using distilled water. The precipitate was shade dried until the moisture is gone[9].

C. Characterization of Silver Nanoparticles

All the Characterization studies were done at SAIF cochin. The green synthesis of silver Nano particles was confirmed using UV-Visible spectrophotometer. UV-Visible absorption NIR spectrometer Agilent Cary 5000 in the region between 200nm to 3000nm was used to determine absorption of the salvia officinalis leaf extract. The dried salvia offcinalis sample were recorded into FT-IR spectrophotometer analysed and find the functional groups of the given samples using thermo nicolet avtar 370 range between $4000cm^{-1}$to$400cm^{-1}$.XRD patterns were recorded from powder X-raydiffractometer (BrukerD8Advance) operated at 30Kv and spectrum was recorder by cukα radition with wave length 1.506 A^0 in the range 200-800nm to calculate the particle size . SEM study was carried out to investigate the shape and size of the AgNPs using joel 6390LA and Energy Dispersive X-ray(EDX) on oxford XMX N operated at 0.5 kV to30 kV at magnification 300000 and EDAX Resolution at 136 Ev.

III. Result And Discussion

A. Uv–Vis Spectral Studies And Analysis

When the salvia officinalis extract was mixed with the aqueous solution of the silver nitrate, the change in colour was noticed from pale yellow to dark brown which is shown fig (3). The change is colour is due to the particle size reduction of silver ion. The silver nanoparticles exhibit some brown colour in the aqueous solution [10]. The presence of nanoparticles was confirmed by obtaining a spectrum in the visible range 200nm to 800nm using UV-Visible spectrophotometer. From this analysis specific absorption peak was found at around 431 nm, [11] which is a blue shift and confirms the Ag particles produced are in the nanoscale range fig (4**).** It is known that when the surface Plasmon vibration in silver nano particles are excited, and the size reduction takes place.

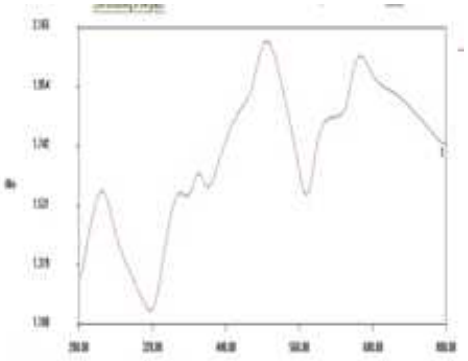


Fig 4. UV–vis spectra of silver nanoparticles

B. Ftir Analysis

FTIR spectroscopy was used to characterize and identify the chemical composition of the salvia officinalis extract mixed with silver nitrate solution. The peak at 3550 cm^{-1}revealed the NH_2 [12]groups and 3415 cm^{-1} [11,12,13]confirming the OH Absorbtion 2924 cm^{-1} there by confirming the CH_3 [14]and CH_2 in aliphatic compounds and CH antisymmeteric stetching [15].1618 cm^{-1} showing the C=O alkenes groups[16].

The band 1382 cm^{-1} shows the SO_2 sulfonyl chlorides and SO_2 antisymmeteric stretch[17] 615 cm^{-1} shows the naphthalenes and in plane ring deformation[18] 468 cm^{-1} shows that naphthalenes and out of plane ring bending[19,10].

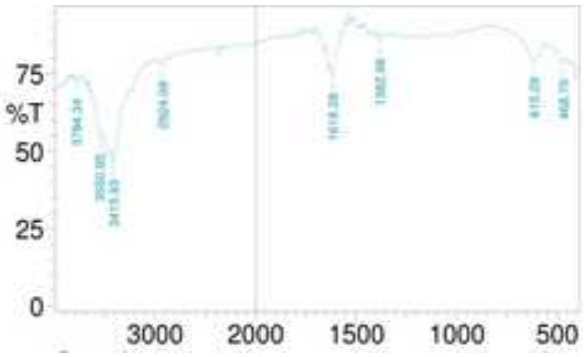


Fig 5. *FTIR spectra of silver nanoparticles*

C. Xrd Analysis

The phase identification and crystalline structures of the nanoparticles were characterized by Xray powder diffraction. the particle size or grain size of the particles was determined using scherrer formula

$$d = \frac{0.9\lambda}{\beta\cos\theta}$$

where *d* is the mean diameter of the nanoparticles, λ is wavelength of x-ray radiation source, β is the angular FWHM of the xrd peak at the diffraction angle θ [20].Figure(6) shows the Xrd pattern of silver nanoparticles obtained using *salviaofficinalis* leaf extract. The distinct peaks were 38.04 ,44. 26,64. 44,77.34,observed [21]. The average crystallite size is calculated using scherrer equation with the peaks obtained. The average size of synthesized particle is calculated to be 20 nm. (111)[21],(200)[24] (220)[22,24] (311)[23] indexed angle for the corresponding peaks respectively for the crystalline plane of silver particle obtained.

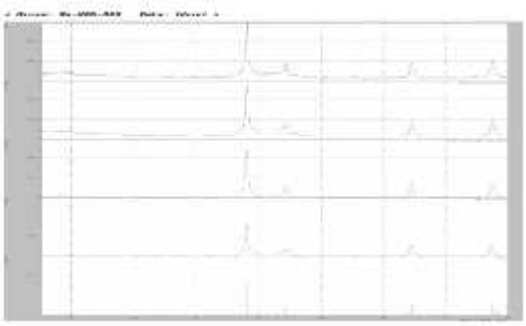


Fig 6 . XRD spectra of silver nanoparticles

D. Sem Analysis

SEM technique is employed to determine the surface morphology and the topography of synthesized silver nanoparticles[9]. SEM image exhibited that the biosynthesized silver nanoparticles are mostly the spherical in shapes. The size of the nanoparticles were with in the range of 20-50nm. It is also noticed that the nanoparticles are in direct contact with each other. The capping agent noticed in the nanoparticles gives the stabilization for the particle. The caping agent may be due to the sediments in the leaf extract.

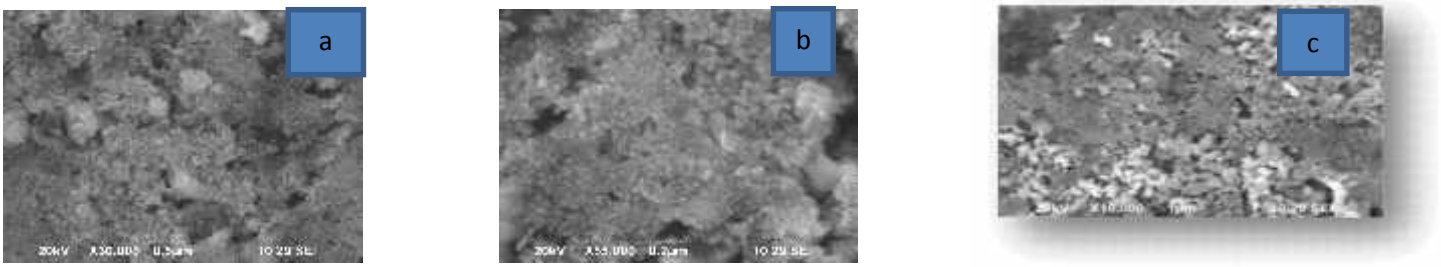


Fig 7. SEM analysis of (a) silver nano particles in 0.5 µm (b) silver nano particles in 0.2µm (c) silver nano particles in 1 µm

E. Edax

Energy dispersive X-ray spectrometers take advantage of the photon nature of light. In the X-ray range the energy of a single photon is just sufficient to produce a measurable voltage pulse X-ray, the output of an ultra low noise preamplifier connected to the low noise are a statistical measure of the corresponding quantum energy. The EDX graph says (fig 6) that Ag is the main component in the prepared nanoparticles which confirms the Ag nanoparticles .The other particles may be the bio components present in the leaf. Metallic silver nanocrystals generally show typical optical absorption peak approximately at 3 keV due to surface plasmon resonance [25,26].

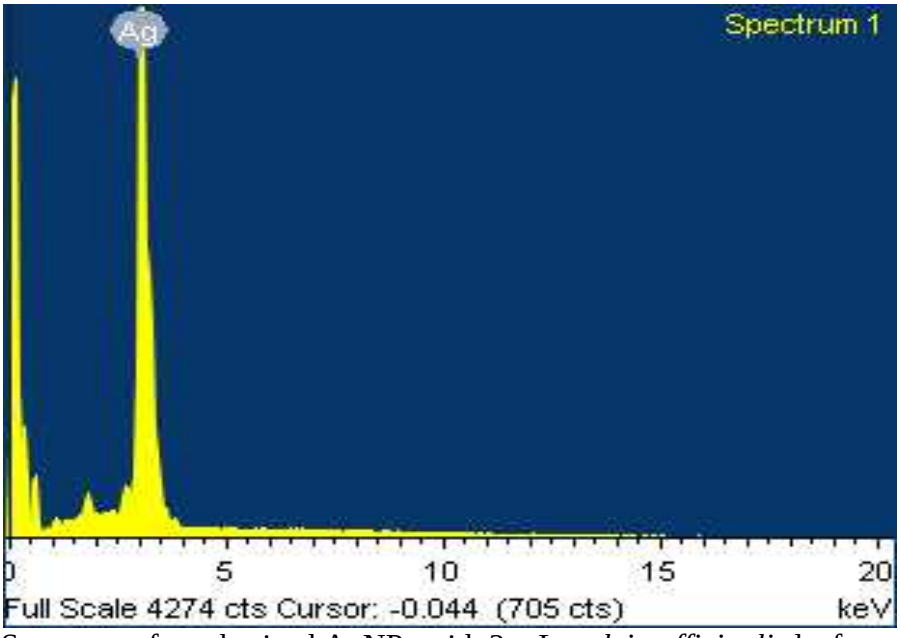


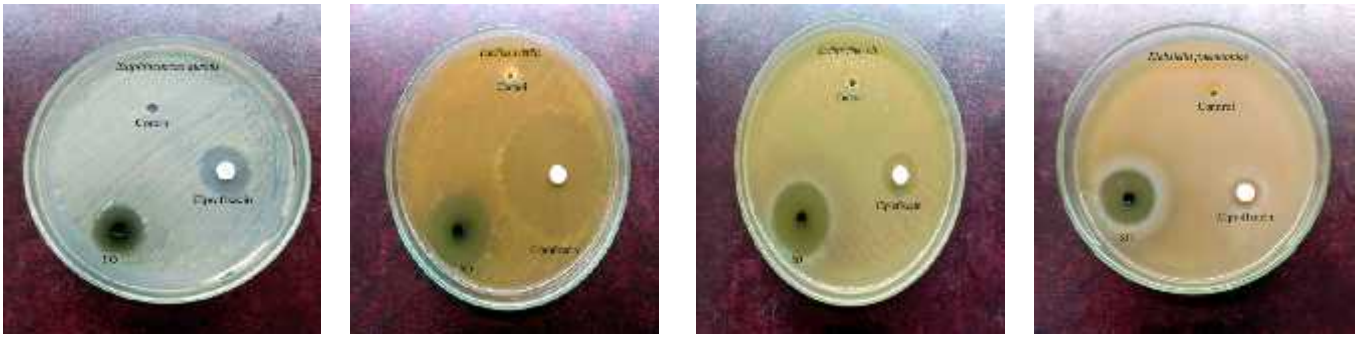
Fig 8. EDX Spectrum of synthesized AgNPs with 2 mL *salvia officinalis* leaf extracts solution

IV. Antimicrobial Studies

A. Antibacterial

In the recently revealed writing, it has been accounted for that the silver nanoparticles shows antibacterial activity not simply against Gram positive and Gram negative microorganisms however additionally against multidrug safe (MDR) microscopic organisms[27]. In the present work, the antibacterial adequacy of S.O plant leaves remove integrated silver nanoparticles was concentrated through well diffusion method technique by utilizing the microorganisms, Staphylococcus aureus ,Bacillus subtilis, Klebsiella pneumonia, Escherichia coli at the fixations 100 µg/ml[28]. The microbes development was very repressed by these orchestrated silver nanoparticle arrangement unequivocally demonstrative of productive antibacterial activity. Also, it was accounted for that the zone of inhibition of S.O plant leaves extricate intervened incorporated silver nanoparticles was more when contrasted with standard medication (**Ciprofloxacin)** and S.O plant leaves separate[29]. Consequently, the noticed outcomes from the examination obviously showed that the S.O plant leaves remove intervened blended silver

nanoparticles perhaps will be used as a possible antibacterial specialist[30]. The bacterial activity of synthesized Ag nanoparticles against four bacteria such as E.coli and S- aureus B- subtilis K- pneumonia showed a clear inhibition zone shown in fig (9) and table (1)[31][32][33][34].



A B C D

Figure 9.Bacterial activities of synthesized Ag nanoparticles against (a) S.aureus (b) ***Bacillus subtilis (c) Klebsiella pneumonia (d) Escherichia coli***

Table 1 The results of antibacterial activity with zone of inhibition.

	Microorganisms	**Control**	**SO**	**Ciprofloxacin**
		Zone of inhibition in mm		
1.	***Staphylococcus aureus***	-	17	15
2.	***Bacillus subtilis***	-	22	40
3.	***Klebsiella pneumoniae***	-	20	12
4.	***Escherichia coli***		20	16

B. Antifungal

The salvia officinalis AgNPs were tested for antifungal activity by disc diffusion method against the test organisms Aspergillus niger and Aspergillus flavus and Candida albicans Penicillium sps [35]. The sterilized Sabouraud dextrose agar (SDA) medium plates were prepared and overnight grown C. albicans culture was spread with the use of sterile cotton swab. A. niger and A. flavus cultures were spread on potato dextrose agar (PDA)[36]. The antifungal activity was also evaluated against the silver nitrate (1 mM) and salvia officinalis leaf extract as control and antibiotic Ketoconazole(25 lg) as a standard. Sterile discs of 6 mm diameter were soaked in SNPs, dried plant extract and silver nitrate solution (1 mM)[37]. After spreading, the test organisms on plates, discs were dispensed onto the surface of the inoculated agar plate. Each disc was pressed down to ensure full contact with the agar surface[38]. The plates were incubated at 27^0C after the placing of discs. After the overnight incubation, each plate was examined for the proper growth.

The diameter of the zones of complete inhibition as judged by the un aided eye was measured, including the diameter of the disc. Zones were measured to the nearest whole millimeter, using sliding calipers, which is held on the back of the inverted petri plates for the measurement[39].

A b c d

Figure 10.Antifungal activity against (a) A. niger, (b) A. flavus and (c) C albicans. (d) Penicillium sps

Table 2 **The results of antifungal activity with zone of inhibition**

S.No.	Microorganisms	Control	SO	Ketoconazole
		Zone of inhibition in mm		
1.	Aspergillus niger	-	15	11
2.	Aspergillus flavus	-	16	12
3.	Candida albicans	-	19	12
4.	Penicillium sps	-	18	11

Conclusion

Green synthesis of stable silver nanoparticles using salvia officinalis leaf extract at room temperature was reported in this study. Synthesis was found to be efficient in terms of reaction time as well as stability of the synthesized nanoparticles which exclude external stabilizers/reducing agents. It proves to be an eco-friendly, rapid green approach for the synthesis providing a cost effective and an efficient way for the synthesis of silver nanoparticles. Benefits of using plant extract for synthesis is that it is energy efficient, cost effective, protecting human health and environment leading to lesser waste and safer products. This eco-friendly method could be a competitive alternative to the conventional physical/chemical methods used for synthesis of silver nanoparticle and thus has a potential to use in biomedical applications. XRD reveals that the particles produced were in the average size of 22nm. UV analysis also supports the reduction of particles into the nano size. FTIR gives the vibration ,stretching and stability in the formation of $AgNO_3$ nano particles Synthesis .SEM shows the shape of $AgNo_3$ and EDAX shows its composition.. The silver nanoparticles show significant activity against studied bacterial and fungal species. Our results confirm that biosynthesized AgNPs has given excellent antifungal activity against salvia officinalis silver nanoparticles.

References

1. Hemali Pdalia, Pooja,Moteriya Sumitra Chanda Green synthesis of silver nanoparticles from marigold flower and its synergistic antimicrobial potential Arabian Journal of Chemistry (2015) 8, 732–741
2. M.Ramya1 and M Sylvia Subapriya1 Green synthesis of silver nanoparticles *ISSN 2278 – 5221 www.ijpmbs.com Vol. 1, No. 1 July 2012*

3. [3].JingChen, Jing Wang,Xin Zhang,Yeling Jin Microwave-assisted green synthesis of silver nanoparticles by carboxymethyl cellulose sodium and silver nitrate Materials Chemistry and Physics 108 (2008) 421–424
4. K. Mallikarjuna, G. Narasimha, G.R.Dillip, B. Praveen, B. Shreedhar, C. Sree lakshmi, B. V.S. Reddy, B.Devaprasad Raju Green synthesis of silver nanoparticles using ocimum leaf extract and their characterization Vol. 6, No 1, January-March 2011, p. 181 – 186
5. A. Singh,D. Jain, M. K. Upadhyaya N. Khandelwal, H. N.Verma Green synthesis of silver using Mexicana extract and evaluation of their Antimicrobial activities Vol. 5, No 2, 2010,p.483-489.
6. Sarkar, P.Kumbhakar*, A. K. Mitra Green synthesis of silver nanoparticles and its optical Vol. 5, No 2 , May 2010, p. 491 – 496.
7. Manjeet Singh , I. Sinha ,R.K. Mandal Role of pH in the green synthesis of silver nanoparticlesMaterials Letters 63 (2009) 425–427
8. Soheyla Honary, Hamed Barabadi, Eshrat Gharaei-Fathabad1 and Farzaneh Naghibi Green Synthesis of Nanoparticles Induced by the Fungus Penicillium citrinum Tropical Journal of Pharmaceutical Research February 2013; 12 (1): 7-11
9. Muhammad Rafique, IqraSadaf, M. Shahid Rafique & M. Bilal Tahir A review on green synthesis of silver nanoparticles and their applications To link to this article: https://doi.org/10.1080/21691401.2016.1241792
10. G.Geoprincy1,B.n.vidhya srri2,u.poonguzhali2,n.nagendra gandhi1,s.renganathan1 view on Green synthesis of silver nanoparticles Asian Journal of Pharmaceutical and Clinical Research Vol 6, Suppl 1, 2013.
11. Jayshree Annamalai1,Thangaraju Nallamuthu Green synthesis of silver nanoparticles: characterization and determination of antibacterial potency Appl Nanosci (2016) 6:259–265 DOI 10.1007/s13204-015-042.
12. Akl M Awwad1,Nidá M Salem2 and Amany O Abdeen1 Awwad et al.Green synthesis of silver nanoparticles using carob leaf extract and its antibacterial activity International Journal of Industrial Chemistry 2013, 4:29 http://www.industchem.com/content/4/1/29
13. Babita Baruwati, Vivek Polshettiwar and Rajender S.Varma Glutathione promoted expeditious green synthesis of silver nanoparticles in water using microwaves *First published as an Advance Article on the web 5th May 2009* DOI: 10.1039/b90218.
14. Chao Wang, Yeon Ju Kim, Priyanka Singh, Ramya Mathiyalagan, Yan Jin & Deok Chun Yang Green synthesis of silver nanoparticles by Bacillus methylotrophicus, and their activity Artificial Cells, Nanomedicine, and Biotechnology,44:4,1127-1132, DOI:10.3109 21691401.2015.1011805
15. Seyyed Mojtaba Mousavi, Seyyed Alireza Hashemi, Younes Ghasemi, Amir Atapour, Ali Mohammad Amani, Amir Savar Dashtaki, Aziz Babapoor & Omid Arjmand (2018) Green synthesis of silver nanoparticles toward bio and medical applications: review study, Artificial Cells, Nanomedicine, and Biotechnology, 46:sup3, S855-S872, DOI: 10.1080 /21691 401.2018.1517769.
16. Shakeel Ahmed, Saifullah, Mudasir Ahmad, Babu Lal Swami & Saiqa Ikram (2016) Green synthesis of silver nanoparticles using Azadirachtaindica aqueous leaf extract, Journal of Radiation Research and Applied Sciences, 9:1, 1-7, DOI: 10.1016/j.jrras.2015.06.006.
17. Anal K. Jha & K. Prasad (2010) Green Synthesis of Silver Nanoparticles Using Cycas Leaf, International Journal of Green Nanotechnology: Physics and Chemistry, 1:2, P110P117, DOI:

10.1080/19430871003684572 To link to this article: https:// doi.org/ 10.1080/ 1943087 1003 68 45 72

18. Selvaraj Arokiyaraj, Savariar Vincent, Muthupandian Saravanan, Yoonseok Lee, Young Kyoon Oh & Kyoung Hoon Kim (2017) Green synthesis of silver nanoparticles using Rheumpalmatum root extract and their antibacterial activity against Staphylococcusaureus and Pseudomonasaeruginosa, Artificial Cells, Nano medicine, and Biotechnology, 45:2, 372-379, DOI: 10.3109/21691401.2016.1160403 To link to this article: https://doi.org/ 10.3109 /21691401. 2016.1160403
19. P.P.N. Vijay Kumara, S.V.N. Pammib, Pratap Kolluc, K.V.V. Satyanarayanad, U. Shameema Green synthesis and characterization of silver nanoparticles using Boerhaavia diffusa plant extract and their antibacterial activity,
20. Seyedeh Masumeh Ghaseminezhada, Sepideh Hamedib, Seyed Abbas Shojaosadatib Green synthesis of silver nanoparticles by a novel method: Comparative study of their properties,∗Carbohydrate Polymers 89 (2012) 467–472
21. Antariksh Saxena , R.M.Tripathi ,Fahmina Zafar , Priti Singh Green synthesis of silver nanoparticles using aqueous solution of Ficus benghalensis leaf extract and characterization of their antibacterial activityMaterials Letters 67 (2012) 91–94.
22. Muhammad Ovais,Ali Talha Khalil, Abida Raza, Muhammad Adeeb Khan, Irshad Ahmad, Nazar Ul Islam, Muthupandian Saravanan, Muhammad Furqan Ubaid, Muhammad Ali1 & Zabta Khan Shinwar Green synthesis of silver nanoparticles via plant extracts: beginning a new era in cancer theranostics 10.2217/nnm-2016-0279 © 2016 Future Medicine Ltd ISSN 1743-5889.
23. Naba Kumar Mondal, Arnab Chowdhury, Uttiya Dey, Priyanka Mukhopadhya, Soumendranath Chatterjee, Kousik Das, Jayanta Kumar Green synthesis of silver nanoparticles and its application for mosquito control Datta1Asian Pac J Trop Dis 2014; 4(Suppl 1): S204-S210 journal homepage: www.elsevier.com/locate/apjtd
24. Qian Suna, Xiang Caia,b, Jiangwei Li a, Min Zhengb, Zuliang Chenb,c, Chang-Ping Yua, Green synthesis of silver nanoparticles using tea leaf extractand evaluation of their stability and antibacterial activity Colloids and Surfaces A: Physicochem. Eng. Aspects 444 (2014)
    a. 226– 231
25. P. Magudapatty, P. Gangopadhyayrans, B.K. Panigrahi, K.G.M.Nair, S. Dhara, Electrical transport studies of Ag nanoparticles embedded in glass matrix, Physica B 299 (2001) 142–146.
26. Vivek Dhand a,b,c,, L. Soumya a, S. Bharadwaj d, Shilpa Chakra a, Deepika Bhatt b,1, B ,Sreedhar e Green synthesis of silver nanoparticles Using Coffea arabica seed extract and its antibacterial activity Materials Science and Engineering C 58 (2016) 36–43.
27. V. Sridhara, K. Pratima, G. Krishnamurthy, and B. Sreekanth, “Vegetable assisted synthesis of silver nanoparticles and its antibacterial activity against two human pathogens,” *Asian J.Pharm. Clin. Res.*, vol. 6, no. 2, pp. 53–57, 2013
28. K. Ali, B. Ahmed, S. Dwivedi, Q. Saquib, A. A. Al-Khedhairy, and J. Musarrat, “Microwave accelerated green synthesis of stable silver nanoparticles with Eucalyptus globulus leaf extract and their antibacterial and antibiofilm activity on clinical isolates,” *PLoS One*, vol. 10, no. 7, pp. 1–20, 2015, doi: 10.1371/journal.pone.0131178.
29. J. R. Nakkala, R. Mata, A. K. Gupta, and S. R. Sadras, “Biological activities of green silver nanoparticles synthesized with Acorous calamus rhizome extract,” *Eur. J. Med. Chem.*, vol. 85, pp. 784–794, 2014, doi: 10.1016/j.ejmech.2014.08.024.

30. R. S. Patil, M. R. Kokate, and S. S. Kolekar, "Bioinspired synthesis of highly stabilized silver nanoparticles using Ocimum tenuiflorum leaf extract and their antibacterial activity," *Spectrochim. Acta - Part A Mol. Biomol. Spectrosc.*, vol. 91, pp. 234–238, 2012, doi: 10.1016/j.saa.2012.02.009.
31. A. Nabikhan, K. Kandasamy, A. Raj, and N. M. Alikunhi, "Synthesis of antimicrobial silver nanoparticles by callus and leaf extracts from saltmarsh plant, Sesuvium portulacastrum L.," *Colloids Surfaces B Biointerfaces*, vol. 79, no. 2, pp. 488–493, 2010, doi: 10.1016/j.colsurfb.2010.05.018.
32. P. Pourali, B. Yahyaei, and S. Afsharnezhad, "Bio-Synthesis of Gold Nanoparticles by Fusarium oxysporum and Assessment of Their Conjugation Possibility with Two Types of β-Lactam Antibiotics without Any Additional Linkers," *Microbiol. (Russian Fed.*, vol. 87, no. 2, pp. 229–237, 2018, doi: 10.1134/S0026261718020108.
33. R. Mariselvam, A. J. A. Ranjitsingh, A. Usha Raja Nanthini, K. Kalirajan, C. Padmalatha, and P. Mosae Selvakumar, "Green synthesis of silver nanoparticles from the extract of the inflorescence of Cocos nucifera (Family: Arecaceae) for enhanced antibacterial activity," *Spectrochim. Acta - Part A Mol. Biomol. Spectrosc.*, vol. 129, pp. 537–541, 2014, doi: 10.1016/j.saa.2014.03.066.
34. R. Geethalakshmi and D. V. L. Sarada, "Synthesis of plant-mediated silver nanoparticles using Trianthema decandra extract and evaluation of their anti microbial activities," *Int. J. Eng. Sci. Technol.*, vol. 2, no. 5, pp. 970–975, 2010.
35. P. K. Sharma and P. Singh, "Antibacterial and antifungal activity of piperazinylbenzothiazine," *Der Pharma Chem.*, vol. 8, no. 5, pp. 191–193, 2016.
36. Dellavalle, "Antifungal Activity of Medicinal Plant Extracts Against," *Chil. J. Agric. Res.*, vol. 71, no. June, pp. 231–239, 2016.
37. A. Panáček *et al.*, "Antifungal activity of silver nanoparticles against Candida spp.," *Biomaterials*, vol. 30, no. 31, pp. 6333–6340, 2009, doi: 10.1016/j.biomaterials.2009.07.065.
38. S. Jebril, R. Khanfir Ben Jenana, and C. Dridi, "Green synthesis of silver nanoparticles using Melia azedarach leaf extract and their antifungal activities: In vitro and in vivo," *Mater. Chem. Phys.*, vol. 248, no. December 2019, 2020, doi: 10.1016/j.matchemphys.2020.122898.
39. R. W. Raut, V. D. Mendhulkar, and S. B. Kashid, "Photosensitized synthesis of silver nanoparticles using Withania somnifera leaf powder and silver nitrate," *J. Photochem. Photobiol. B Biol.*, vol. 132, pp. 45–55, 2014, doi: 10.1016/j.jphotobiol.2014.02.001.