Section A-Research paper

SEAWEED SILVER NANOPARTICLES: CHARACTERIZATION, GREEN SYNTHESIS, AND BIOACTIVITY ANALYSIS

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Abstract

An important area of nanotechnology is the development of experimental procedures for the synthesis of nanoparticles that are biologically inspired. New environmentally friendly "green" synthesis techniques are being developed to address the rising demand for commercial nanoparticles. In this study, *Ulva fasciata* sea weed extract was used to synthesize stable silver nanoparticles (AgNPs). The synthesis of AgNPs and the reduction of Ag ions in the media are both observed using a UV-Vis spectrometer. AgNPs that have been synthesized have had their morphology studied using XRD and SEM. The peaks in the XRD pattern correspond to the metallic silver's face-centered-cubic (FCC) structure. Silver nanoparticles are discovered to have an average grain size of 6.35 nm. TGA/DTA results showed that desorption of chemisorbed water was associated with exothermic reaction and weight loss. The carbonyl, hydroxyl, amine, and protein functional groups that form a layer atop AgNPs and stabilize them in a medium were identified using FTIR. Moreover, the antibacterial activity of silver nanoparticles was done and the results were phenomenal with large inhibition zones.

Key words- Silver nanoparticles, *Ulva fasciata*, Face-centered-cubic structure, Thermal analysis.

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1. Introduction

Scientists' interest in research based on advanced nanomaterials of noble metals like silver has increased over the past few decades due to their study of catalytic activity, optical properties, electronic properties, antibacterial properties, and magnetic properties [1-9] as well as their use in a variety of fields like the production of biomaterials, biochemistry, medical, and pharmaceutical products. In order to synthesize nanoparticles, chemical, physical, and biological approaches have been devised; however, chemical and physical procedures produce harmful toxic byproducts and are very expensive [10, 11]. There has been a search for an approach that is low-cost, safe, and dependable as well as "eco-friendly" to synthesize stable metal nanoparticles with controlled size and form. A number of plant extracts, including *Ocimum Sanctum* [12], Petroselinum crispum [13], *Murraya koenigii* [14], and *Coriandrum Sativum* [15], have recently established the unique processes known as green/biosynthesis for the synthesis of metal nanoparticles.

Nature has developed a number of methods for the synthesis of inorganic materials with nano and micro length scales, which have helped to build a relatively new and mostly unexplored field of study centered on the biosynthesis of nanomaterials. The green chemistry principles are compatible with bio-organism-based synthesis. Utilizing safe, non-toxic, and environmentally friendly reagents, green synthesis of nanoparticles can be executed. Because they are synthesized in a single step, nanoparticles synthesized utilizing biological or green technology have a variety of different natures, higher stability, and suitable dimensions.

Plants offer a better synthesis platform for nanoparticles since they include natural capping agents and are free of hazardous compounds [16-18]. *Ulva fasciata* extracts are used for the current investigation since it has a number of pharmacological properties. For instance, the antibacterial activity of certain metal nanoparticles, such as silver colloids, is directly correlated with their size, i.e., the higher the antibacterial activity, the smaller the silver nuclei. In addition, these nanoparticles' catalytic activity is influenced by their size, structure, shape, size distribution, and chemical-physical environment. Controlling size and size distribution is thus a crucial task. In general, changing the synthesis processes, reducing agents, and stabilizers is frequently used to obtain precise control of form, size, and size distribution of nanoparticles. With the use of *Ulva fasciata* extract and the green synthesis method, the reduction of water soluble silver ions to silver nanoparticles is done in this work.

2. Material and Methods

2.1 Synthesis of silver nanoparticles

$AgNO_3$ (99.99%) in aqueous solution at 0.01M concentration was used to synthesis AgNPs in a green way. 45 mL of 0.01M $AgNO_3$ aqueous solution was mixed with 5 mL of *U. fasciata* broth and left to react at room temperature. When the reaction mixture's color changes from translucent yellow to dark brown over a range of time intervals, the formation of AgNPs was confirmed. The reduction mechanism of the silver ion into nanoparticles in solution was examined using UV-vis spectral spectroscopy, which involved collecting the AgNPs solution. The AgNPs solution was centrifuged, and the extra liquid was removed by evaporating it in a dryer, producing silver nano powder that was dark in color.

2.2 Characterization

Using UV-vis spectrometers (Shimadzu UV-1800, Japan) at room temperature and with a resolution of 1 nm, optical absorbance between 190 and 800 nm was regularly observed in samples of bio-reduced AgNPs solution. A study of infrared spectra using the Fourier transform was utilized to locate probable biomolecules and functional groups. A Perkin Elmer spectrometer with a resolution of 1 cm-1 and a wavelength range of 500–4000 cm^{-1} was used to measure FT-IR spectra. Cu-K radiation with a wavelength of 0.10287 nm was used for the X-ray diffractometer measurement in order to look at the crystalline structure, behavior, and quality of the manufactured AgNPs powder. The scanning was carried out in a 2 radial range between 30° and 80° at a rate of 0.02/min, with a 2s time constant. The Debye-Scherer equation was used to determine the size of AgNPs. By using a scanning electronic microscope, the surface shape and particle size of produced AgNPs were studied. TGA/DTA was used to analyze the weight loss and reaction type of synthesized AgNPs in the temperature range of room temperature to 1000^oC. Al_2O_3 was utilized to heat the samples at a rate of 20^oC/min in an environment of air.

2.3 Antibacterial activity

One Gram (+) ve (*Bacillus*) and one Gram (-) ve (*Escherichia coli*) bacteria were used as test microorganisms, obtained from IMTEch Chandigarh. The antibacterial properties of the *Ulva fasciata* extracts and of the AgNPs produced using the *Ulva fasciata* extracts were successfully analysed. For assessing *Ulva fasciata* mediated AgNPs containing solution, the agar well diffusion method was used. The wells were

filled with solutions containing silver nanoparticles at different concentrations (10 µl, 20 µl and 30 µl). The microbial cultures were used to swab the plates that contained nutrient agar media. The plates were incubated for 24 to 48 hours at 37°C. Ampicillin was used as the standard for comparison. Then, against each type of test microorganism, the highest zone of inhibition was seen and measured for analysis.

3 Results and Discussion

3.1 Ultraviolet visible spectroscopy

Fig.1 depicts the reaction between *Ulva fasciata* extract and silver nitrate solution as a function of time while using water as the solvent. The synthesis of additional AgNPs is indicated by a 20-minute visual color change from translucent to light brown, which was supported by FT-IR and UV-vis spectrometer examination. Due to a longer reaction time after 70 minutes, the hue significantly changed to dark brown, accelerating the formation of silver nanoparticles. It is widely known that surface Plasmon vibration causes silver nanoparticles to appear yellowish brown [19, 20]. Fig. 2 displays the UV-vis spectrometer-measured absorbance peak of produced AgNPs at various time intervals. The peak at 316 nm is related to AgNPs' absorbance. With more reaction time, the strength of the 316 nm absorption peak also increased.

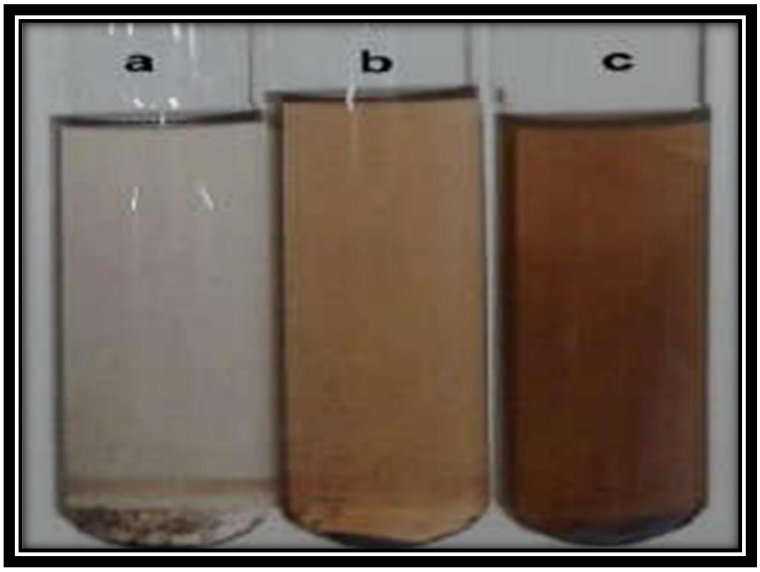

Fig.1. Synthesized silver nanoparticles- (a) at 10 min (b) 20 min (c) 70 min

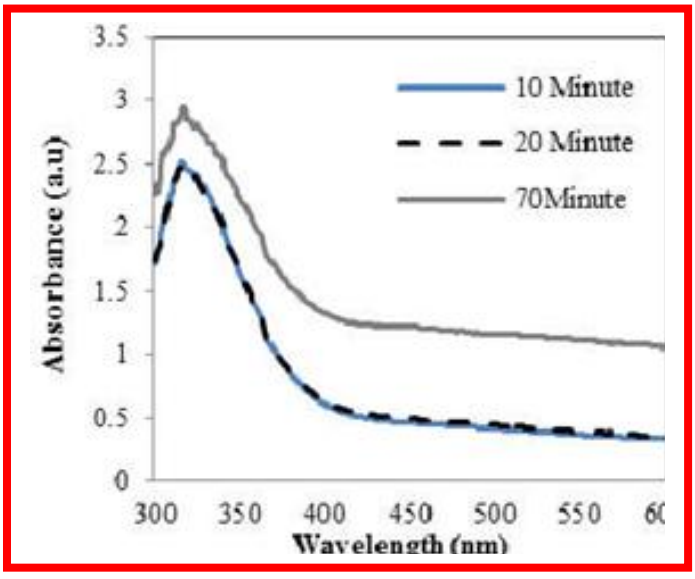

Fig. 2. UV spectrum of synthesized silver nanoparticles at varied time intervals

3.2 FTIR analysis

A clear FT-IR spectra of produced AgNPs is seen in Fig. 3 at wavelengths of 663.52 cm^{-1}, 1401.31 cm^{-1}, 1642.41 cm^{-1}, and 3361.02 cm^{-1}. Absorption peaks at 663.52 cm^{-1} for halogen compounds, 1401.31 cm^{-1} for alcohol and phenols, and 1642.41 cm^{-1} for tertiary amides are associated with C-Cl stretching. Alcohols and phenols have O-H stretching designated at 3361.02 cm^{-1}. The ability of carbonyl groups in proteins and amino acid residues to bind metal is stronger, and these groups may synthesis a coating over metal

nanoparticles (such as the capping of silver nanoparticles) to prevent agglomeration and so stabilize the medium, according to an IR spectroscopic investigation [21, 22]. These findings imply that biological molecules play dual roles in the aqueous medium's production and stabilization of silver nanoparticles.

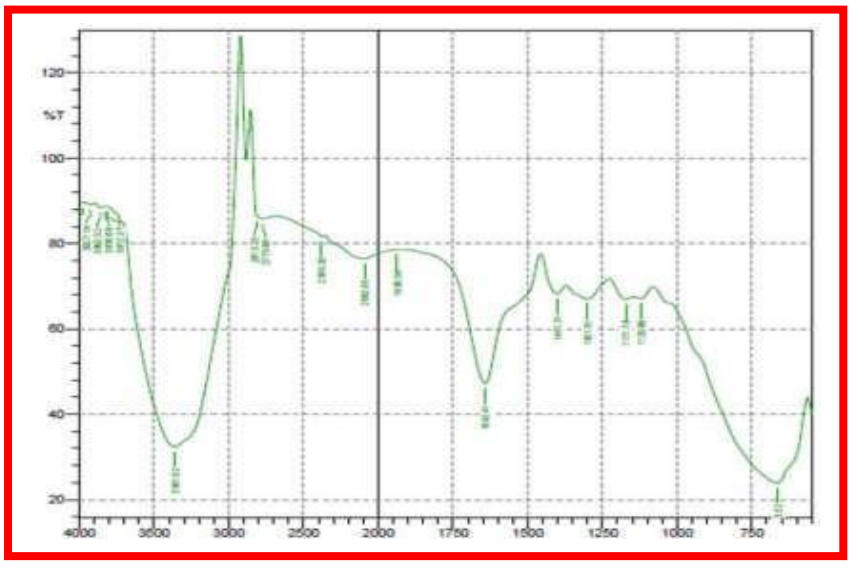

Fig.3. FT-IR spectrum of synthesized AgNPs using *Ulva fasciata* extract

3.3 X-ray diffraction

The XRD was used to determine the crystal structure of AgNPs, and the results showed peaks at 37.90°, 44.05°, 64.25°, and 77.20°, which correspond to the (111), (200), (220), and (311) face-centered cubic structures of silver, respectively. These results were compared to those on the standard powder diffraction card of the Joint Committee on Powder Diffraction Standards (JCPDS), silver file No. 04-0783. A few strong unassigned peaks that were elevated at 32.00, 45.95, 54.60, and 57.20 were also observed. These Bragg peaks may have been caused by bioorganic proteins or chemicals found in *Ulva fasciata* extract. Using full widths at half maximum (FWHM) measurements, the Debye-Scherer equation was utilized to determine the average particle size [23-24]. Approximately 5.78 nm was the calculated average particle size. The Debye-Scherer formula is $\tau = K\lambda / \beta\cos\theta$, where, τ, K, λ, β and θ represents the particle size, dimensionless shape factor, x-ray wavelength, line broadening at half the maximum intensity and Braggs angle respectively.

3.4 SEM analysis

The SEM image of synthesized silver nanoparticles is shown in Fig. 4. According to SEM image, the majorities of silver nanoparticles has spherical shapes with smooth surfaces, is evenly distributed, and are arranged in close, compact clusters. Using cutting-edge software called "IMAGEJ," the average particle size was discovered to be approximately 6.45 nm, and the obtained particle size is nearly identical to XRD.

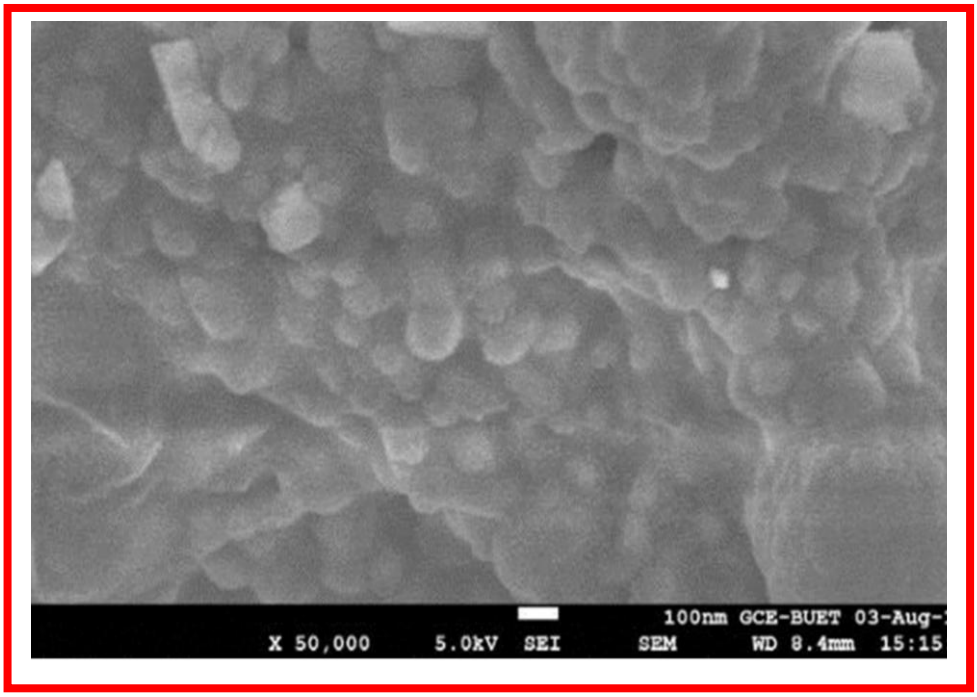

Fig. 4. SEM image of synthesized AgNPs using *Ulva fasciata* extract

3.5 Thermogravimetric-Differential Scanning Calorimetry analysis

AgNPs powder's TGA-DSC spectra are shown in Fig. 5. The TGA spectrum shows that AgNPs samples lose weight most frequently between 200 and 500 ^{o}C, with a small amount below 200 ^{o}C and above 800 ^{o}C. Nearly no weight loss occurs in the 550°C to 800°C temperature range. It can be generally attributed to the evaporation of organic molecules and water. DSC spectra show two exothermic peaks that rise between 250°C and 350°C and 400°C and 500°C, respectively.

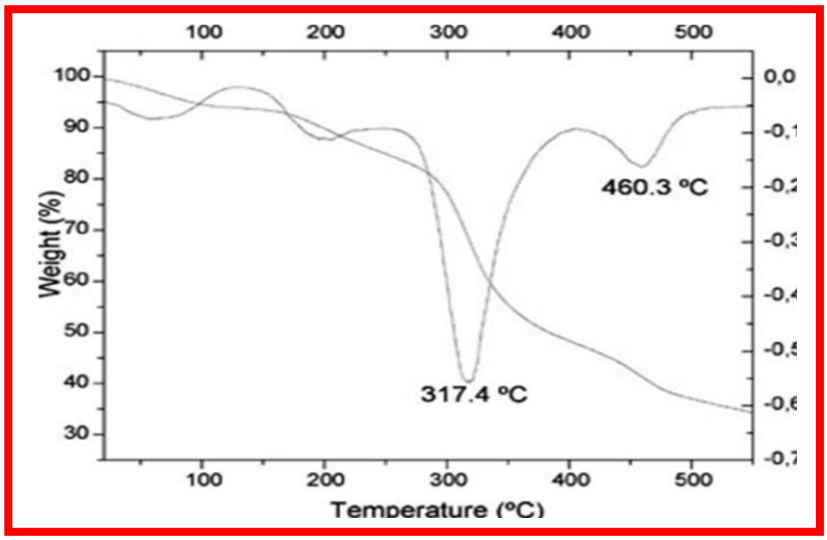

Fig. 5. TGA-DSC spectrum of synthesized AgNPs using *Ulva fasciata* extract

3.6 Antibacterial activity

E. coli and *Bacillus* colonies were used in this study to examine the antibacterial property of AgNPs and the results were shown in Fig.6 and 7 respectively. The outcomes are displayed in Table 1. The obtained inhibition zones show that the test sample's antibacterial activity is at its peak. The antibacterial potential of AgNPs is also supported by findings from earlier studies. AgNPs made from banana leaf extract have the highest zone of bacterial inhibition for gram positive *Bacillus*, which can be inferred from the fact that these particles were the smallest in diameter compared to those made from neem and tulsi leaf extracts, which both had comparable antimicrobial properties.

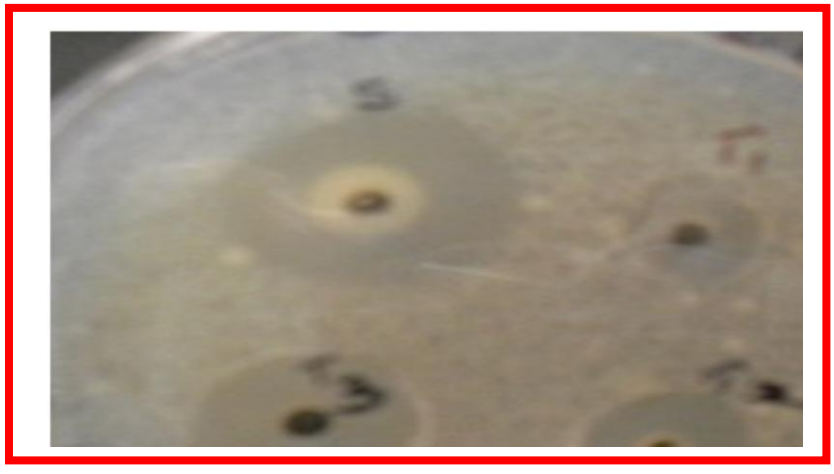

Fig. 6. Antibacterial activity of Ag nanoparticles against *E. coli*

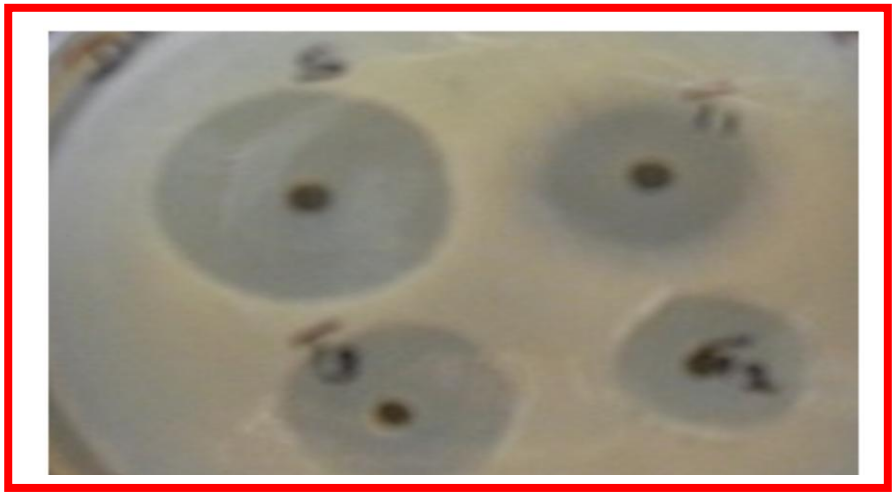

Fig. 7. Antibacterial activity of Ag nanoparticles against *Bacillus*

Table 1. Antibacterial activities of Ag nanoparticles synthesized using *U. fasciata* extracts

Bacterial culture	Zone of inhibition (mm)			
	T1	T2	T3	S
Escherichia coli	8	7	11	14
Bacillus	5	7	9	9

4 Conclusion

Ulva fasciata extract was found to synthesize AgNPs quickly with a definite size and shape and it was environmentally benign. The UV-vis spectrum at 316 nm revealed that AgNPs started to form within 10 min and had a greater production yield at 70 min after the addition of extract to silver nitrate. It was discovered that AgNP synthesis grew over time. The silver's FCC structure is attributed to the XRD peaks. The developed AgNPs had a spherical form, and an XRD study revealed that the particle size was 5.78 nm, which was further supported by a SEM investigation. The majority of TGA/DTA weight loss occurs between 200 and 500 ^{o}C, with just a small amount below 200 ^{o}C and over 800 ^{o}C, and the reaction was exothermic. The biological components that produce and stabilize silver nanoparticles in the aqueous media were identified using the FT-IR spectrum. Also, the nanoparticles exhibited antibacterial activity against *E. coli* and

Bacillus. In conclusion, green synthesis provides a productive and environmentally responsible way to synthesis silver nanoparticles.

References

[1] Song, J.Y.; and Kim, B.S. (2009). Rapid biological synthesis of silver nanoparticles using plant leaf extracts. Bioprocess and Biosystems Engineering, 32(1), 79-84.

[2] Santos, K.D.O.; Elias, W.C.; Signori, A.M.; Giacomelli, F.C.; Yang, H.; and Domingos, J.B. (2012). Synthesis and catalytic properties of silver nanoparticle–linear polyethylene imine colloidal systems. The Journal of Physical Chemistry C, 116(7), 4594-4604.

[3] De Gaetano, F.; Ambrosio, L.; Raucci, M.G.; Marotta, A.; and Catauro, M. (2005). Sol-gel processing of drug delivery materials and release kinetics. Journal of Materials Science: Materials in Medicine, 16(3), 261-265.

[4] Prammitha R,; Jeice AR, ; Jayakumar, K. (2023) Review of green synthesized TiO_2 nanoparticles for diverse applications, Surfaces and Interfaces,39 102912

[5] Crabtree, J.H.; Burchette, R.J.; Siddiqi, R.A.; Huen, I.T.; Hadnott, L.L.; and Fishman, A. (2003). The efficacy of silver-ion implanted catheters in reducing peritoneal dialysis-related infections. Peritoneal Dialysis International, 23(4), 368-374.

[6] Rathi.,V. H. ; Jeice. A.R (2023) Green fabrication of titanium dioxide nanoparticles and their applications in photocatalytic dye degradation and microbial activities. Chemical Physics Impact 6, 100197.

[7] Królikowska, A.; Kudelski, A.; Michota, A.; and Bukowska, J. (2003). SERS studies on the structure of thioglycolic acid monolayers on silver and gold. Surface science, 532, 227-232.

[8] Prammitha. R;, Jeice. A.R. ;, Jayakumar, K; (2023) Influences of calcinations temperature on titanium dioxide nanoparticles synthesized using *Averrhoa carambola* leaf extract: in-vitro antimicrobial activity and UV-light catalyzed degradation of textile wastewater, Biomass conversion and biorefinery,13

[9] Jeice A. R. ; Jayam. Sr. G. ; Wilson K.S.J (2016) Polaronic effects on diamagnetic susceptibility of a hydrogenic donor in nanostructures. Indian J. Phys. 90(7):805-809 DOI 10.1007/s12648-015-0816-3

[10] Phanjom, P.; and Ahmed, G. (2015). Biosynthesis of silver nanoparticles by Aspergillus oryzae (MTCC No. 1846) and its characterizations. Nanoscience and Nanotechnology, 5(1),14-21.

[11] Vinod, V.T.P.; Saravanan, P.; Sreedhar, B.; Devi, D.K..; and Sashidhar, R.B. (2011). A facile synthesis and characterization of Ag, Au and Pt nanoparticles using a

natural hydrocolloid gum kondagogu (Cochlospermum gossypium). Colloids and Surfaces B: Biointerfaces, 83(2), 291-298.

[12] Singhal, G.; Bhavesh, R.; Kasariya, K..; Sharma, A.R.; and Singh, R.P. (2011). Biosynthesisof silver nanoparticles using Ocimum sanctum (Tulsi) leaf extract and screening its antimicrobial activity. Journal of Nanoparticle Research, 13(7), 2981-2988.

[13] Roy, K..; Sarkar, C.K..; and Ghosh, C.K. (2015). Plant-mediated synthesis of silver nanoparticles using parsley (Petroselinum crispum) leaf extract: spectral analysis of the particles and antibacterial study. Applied Nanoscience, 5(8), 945-951.

[14] Christensen, L.; Vivekanandhan, S.; Misra, M.; and Mohanty, A.K. (2011). Biosynthesis of silver nanoparticles using murraya koenigii (curry leaf): an investigation on the effect of broth concentration in reduction mechanism and particle size. Advanced Materials Letters, 2(6), 429-434.

[15] Sathyavathi, R.; Krishna, M.B.; Rao, S.V.; Saritha, R.; and Rao, D.N. (2010). Biosynthesis ofsilver nanoparticles using Coriandrum sativum leaf extract and their application in nonlinearoptics. Advanced science letters, 3(2), 138-143.

[16] Chithra, V.; and Leelamma, S. (1997). Hypolipidemic effect of coriander seeds (Coriandrum sativum): mechanism of action. Plant Foods for Human Nutrition, 51(2), 167-172.

[17] De Almeida, M.E.; Bion, F.M.; and Guerra, N.B. (2003). In vivo antioxidant effect of aqueous and etheric coriander (Coriandrum sativum L.) extracts. European Journal of LipidScience and Technology, 105(9), 483-487.

[18] Ramadan, M.; and Mörsel, J.T. (2002). Oil composition of coriander (Coriandrum sativum L.) fruit-seeds. European Food Research and Technology, 215(3), 204-209.

[19] Mahmudin, L.; Suharyadi, E.; Utomo, A.B.S.; and Abraha, K. (2015). Optical properties of silver nanoparticles for surface plasmon resonance (spr)-based biosensor applications. Journal of Modern Physics, 6(08), 1071-1076.

[20] Mulvaney, P. (1996). Surface plasmon spectroscopy of nanosized metal particles. Langmuir,12(3), 788-800.

[21] Balaji, D.S.; Basavaraja, S.; Deshpande, R.; Mahesh, D.B.; Prabhakar, B.K.; and Venkataraman, A. (2009). Extracellular biosynthesis of functionalized silver nanoparticles by strains of Cladosporium cladosporioides fungus. Colloids and surfaces B: Biointerfaces, 68(1), 88-92.

[22] Mandal, S.; Phadtare, S.; and Sastry, M. (2005). Interfacing biology with nanoparticles. Current Applied Physics, 5(2), 118-127.

[23] Tripathy, A.; Raichur, A.M.; Chandrasekaran, N.; Prathna, T.C.; and Mukherjee, A. (2010). Process variables in biomimetic synthesis of silver nanoparticles by aqueous extract of Azadirachta indica (Neem) leaves. Journal of Nanoparticle Research, 12(1), 237-246.

[24] Bao Q, Zhang D, Qi P (2011) Synthesis and characterization of silver nanoparticle and graphene oxide nanosheet composites as a bactericidal agent for water disinfection. J Colloid Interface Sci 360:463–470.