

Biogenic synthesis and characterization of silver nanoparticles using aqueous extract of *Lepidium sativum* assessing its antibacterial and antioxidant properties.

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Abstract:

In every step of research, along with engineering in medical fields, metallic nanoparticles are used and the scientists continue to attract them to explore new proportions for each one of them, typically due to their appropriate small sizes. Silver nanoparticles have achieved a special emphasis among many noble metal nanoparticles. In this present research study we used green synthesis for silver nanoparticles (NPs) and characterization for assessment of antibacterial and antioxidant activities. *Lepidium sativum* seeds mediated NPs can be concluded to be effectively used to produce potential antioxidant and antimicrobial AgNPs in Industrial applications.

Keywords: Metallic nanoparticles *Lepidium sativum* seeds, Antibacterial and antioxidant

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I. Introduction

Silver is extensively used in nano systems and employed in various biomedical purposes. Silver nanoparticles have excellent medical and nonmedical properties and applications when compared with other metal nanoparticles. (Ge 2014) The green approach of nanoparticles synthesis possesses reduced or no toxicity and number of plants and herbal extracts has been reported to be involved in such synthesis. (Ahmed 2016) Plant extracts contain number of secondary metabolite which plays a critical role during the nanoparticle synthesis by acting as reducing or capping agents. (Prasad 2014) Studies have shown that silver nanoparticles are highly stable and toxic to bacteria, fungus, and viruses.

A variety of techniques are required for the synthesis of silver nanoparticles such as, chemical reduction, ion sputtering, sol gel, etc. (Mahdi et al., 2015, Padalia et al., 2014, Bindhu and Umadevi, 2015); many of the techniques for synthesis nanoparticle involve the use of toxic chemicals or high energy requirements, which are quite challenging and contain wasteful purifications. (Ahmad, Swami, & Ikram, 2015). It is therefore the obligation to highlight the alternative as a synthetic path, which is not only cost-effective but can be environmentally safe at the same time. Keeping in mind the aesthetic context, the green synthesis is made as a main technique and demonstrates its ability at the top.

Nanoparticles acquire fully novel physical (optical, magnetic and electronic) and chemical properties. High antimicrobial activity and wide range of size and shape, self-assembly and of silver nanoparticles (AgNPs) regarded as vital for their possible role in medical devices, optical devices, catalysis and electronics biotechnologies (Tolaymat 2010, Alvarez-Puebla 2009). Small size, spherical shape and a high surface-to-volume ratio of AgNPs make it possible to interact with the pathogens' cell walls, making them greater antimicrobial activity (Agnihotri 2014).

The use of plant extracts for metallic nanoparticles synthesis has been thoroughly studied as they increase the monodispersed of nanoparticles. Biomolecules such as phenols, polysaccharides, terpenoids, flavones, alkaloids, enzymes, amino acids, proteins, and alcoholic compounds found in plants function as both reducing agents and capping agents that stabilize and regulate the morphology of NPs (Ravindran 2003). In recent studies, green synthesis of metallic NPs using plant extracts has been extensively employed. (Ashok Kumar 2015, Nakkala 2014, Kumar 2011) Considering the vast potentiality of plants as sources this work aims to apply a biological green technique for the synthesis of silver nanoparticles as an alternative to conventional methods.

Lepidium sativum (family, Brassicaceae) is a common plant for its nutritional and medicinal values used by Arab countries and including India. *Lepidium sativum* is known for treating various diseases in ancient India and Saudi Arabia, such as bone fracture recovery, fever, arthritis, and many others. In local traditional

medicine, seed extracts have been used to treat dysentery, gastrointestinal disorders, stomachache, indigestion, febrile fever, and skin disorders. The antimicrobial activity of *L sativum* seed extracts has also been reported against different microbial pathogens.

This work aims to use a biological green approach for the synthesis of silver nanoparticles as an alternative to traditional approaches, taking into consideration the enormous potential of plants as sources. Thus, green synthesis, characterization of silver nanoparticles from *Lepidium sativum* seeds and assessment of antibacterial activities of synthesized silver nanoparticles are recorded in the current research.

II. Methodology

Silver Nanoparticles Preparation

Preparation Of Plant Extract

One gram of *lepidium sativum* seeds were quickly rinsed with RO water, followed by purified water and boiled at 50-60 ° C for 20 min with 100 mL of Milli Q water (Ashok kumaret al., 2014). The extract obtained was filtered into Whatman No. 1 filter paper and the filtrate was collected for further use in the 500 mL Erlenmeyer flask and stored at 4°C. Silver nitrate 0.168 mg (1 mM) was used in 250 mL of Milli Q water to prepare the metal solution (Ankamwaret al., 2005; Bar et al., 2009a).

SYNTHESIS OF SILVER NANOPARTICLES USING SEED EXTRACT

In the Erlenmeyer flask, 1 mM aqueous solution of silver nitrate was taken and the pH was adjusted to 8. The extract was added to the (AgNO₃) silver nitrate solution at room temperature at a ratio of 1:4(v / v). Initial colour shift was detected at a 10-minute period of up to 1 hour, accompanied by UV-vis spectrophotometer examination at a 4-hour period from 4 hours to 24 hours for tentative validation of synthesized AgNPs (Ananda et al., 2011).

Initially, silver nanoparticles were prepared by reducing Ag⁺ to Ag⁰ with 50 mL of *Lepidium sativum* seed extract at its initial pH of 4 and 50 mL of 0.1 mM AgNO₃ solution at room temperature. AgNO₃'s colorless solution plus seed extract greenish yellow turned to dark brown in 10 minutes later, this is a signaling that the silver nanoparticles had been formed (Fig 1).

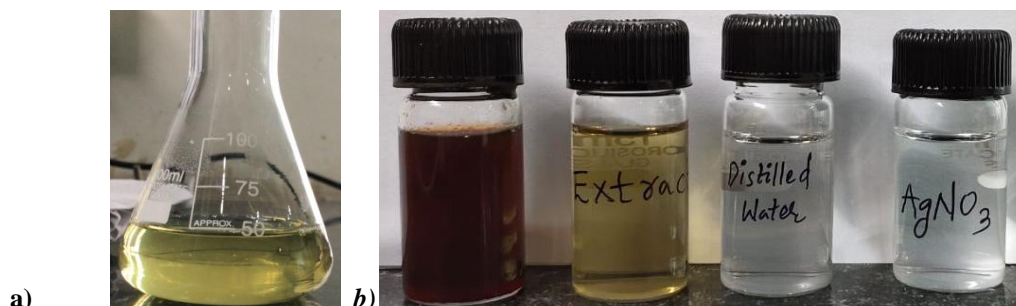


Figure 1.a) *Lepidium sativum* seed extract **b)** Conformation and Formation of silver nanoparticles after adding 0.1 mM AgNO₃ solution in Seed extract.

CHARACTERIZATION OF SILVER NANOPARTICLES

UV-Visible spectrometer

The development of AgNPs during the reduction process was demonstrated by a difference in the colour of the reaction solution which can be visually detected. The approximate definition for the reduction of Ag⁺ is the change in the colour of the reaction solution from a colorless to a dark brown solution. The surface plasma resonance (SPR) peak presence at 404 nm wavelength confirms the development of AgNPs. Spherical particles are responsible for the observed symmetric sharper SPR band with a single peak reported by (Jagajjananiet al., 2013). The addition of fresh *Lepidium sativum* seed extract to 0.1 mM of silver nitrate solution resulted in a brown colour. The highest absorbance at 404 nm wavelength was shown by UV-vis spectrophotometer analysis (Fig 2).

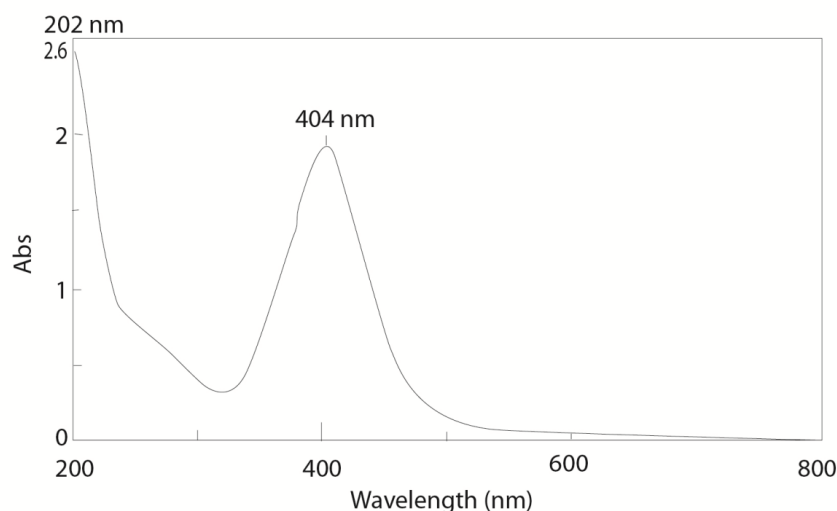


Figure 2. UV-vis Spectrophotometer analysis of synthesized AgNPs of silver nanoparticles after adding 0.1 mM AgNO₃ solution in Seed extract.

Fourier transform infrared spectroscopy (FT-IR)

AgNPs is studied by the FT-IR spectrophotometer to determine the presence of phytochemical coatings on silver nanoparticles. FT-IR spectrum of SC-mediated synthesized AgNPs shows prominent peaks at 3728.56 *cm*⁻¹, 3448.46 *cm*⁻¹, 2924.15 *cm*⁻¹, 2855.26 *cm*⁻¹, 1751.10 *cm*⁻¹, 1641.47 *cm*⁻¹, 1583.39 *cm*⁻¹, 1540.86 *cm*⁻¹, 1458.39 *cm*⁻¹, 1384.49, 1260.79 *cm*⁻¹, 1159.44 *cm*⁻¹, 1101.38 *cm*⁻¹, 1025.26 *cm*⁻¹, 829.76 *cm*⁻¹, 765.00 *cm*⁻¹ and 588.29 *cm*⁻¹. The band at the band at 3728 *cm*⁻¹ can be associated with OH extending amide vibrations. The sharp bands at 2924 and 794 *cm*⁻¹ are C-H and CH amide group respectively alkanes. At 2855 and 1384 *cm*⁻¹ the two medium peaks may be alkanic C-H and CH₃CH. In fingerprint regions 765 and 588, the C-Cl group expands alkyl halides and the C-Br group spans alkyl halides. The reduction, capping and stabilization of silver nanoparticles are involved in these bioactive molecules (**Fig 3**)

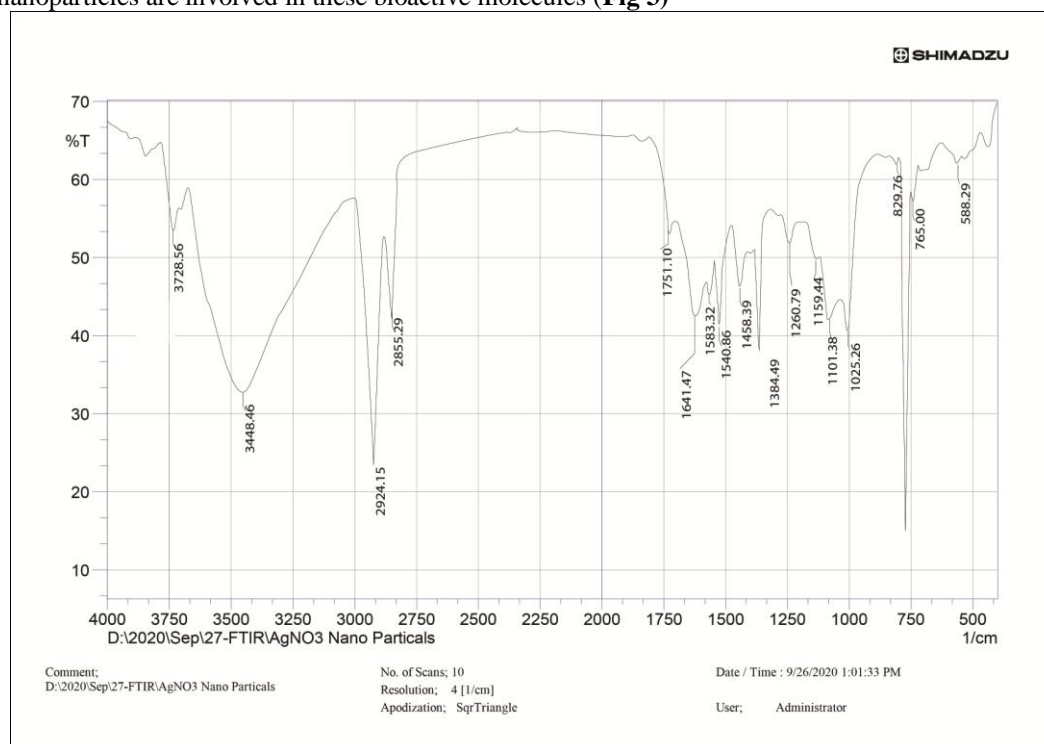


Figure 3. FT-IR spectrum of synthesized AgNPs of silver nanoparticles after adding 0.1 mM AgNO₃ solution in Seed extract.

X-Ray diffraction

X-Ray diffraction pattern of AgNPs synthesized by using SC and AB are as shown in figure 23 and figure 24 respectively. X-RD spectrum of SC mediated synthesized AgNPs shows the clear presence of peaks as per 32.82, 38.82, 44.22 and 64.32. Similarly, X-RD spectrum of AB mediated synthesized AgNPs display peaks at 32.26 and 46.2 which in line with the standard values of JCPDS No.: 04-0783 for silver.

Morphological studies and elemental analysis of SC mediated synthesis of AgNPs

Morphology of SC mediated synthesis of AgNPs was studied by using SEM-EDAX and SEM microscopy and elemental analysis was carried out by using EDAX. SEM images and EDAX spectrum of AgNPs prepared at room temperature, by stirring, heating and at alkaline condition are demonstrated in figure 25 & 26. Comparatively, AgNPs prepared by heating shows higher particle size and are more distributed than AgNPs synthesized at RT and by ST. Further, SEM image of AgNPs synthesized at alkaline condition showed the presence of silver nanoclusters. Morphology of AgNPs was also studied by SEM to get the more clear interpretation regarding the morphology of the green synthesized AgNPs. It was observed that the AgNPs (fig 4) are polymorphic showing triangular, hexagonal, deformed spherical and results are in line with SEM analysis.

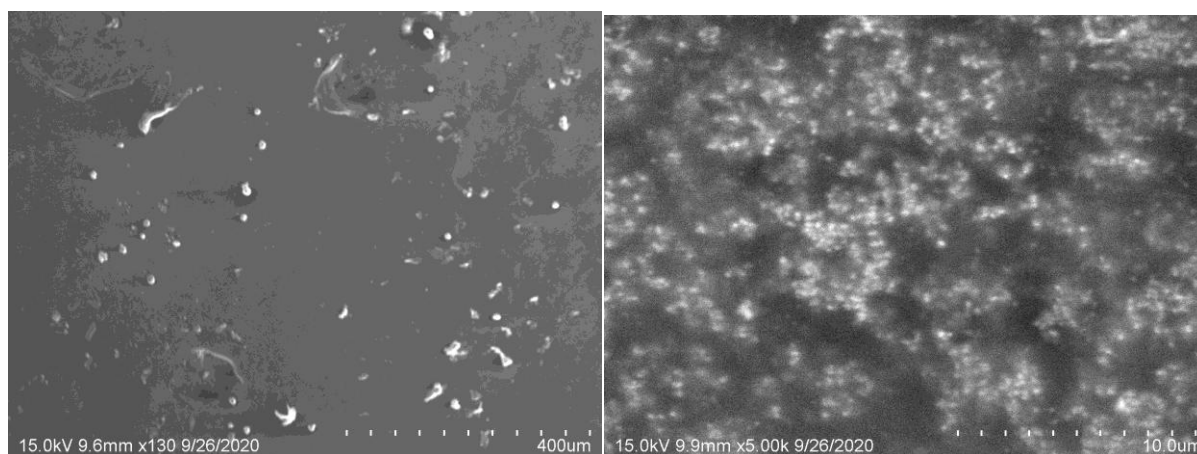
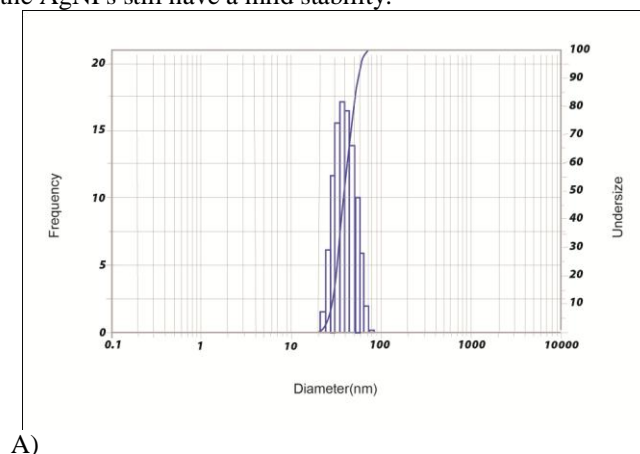


Figure 4: Scanning electron microscope images of synthesized AgNPs of silver nanoparticles after adding 0.1 mM AgNO₃ solution in Seed extract

Zeta potential

Fresh *Lepidium sativum* seed extract mediated silver nanoparticles resulted that the particulate size of green AgNPs with average particle size of 40 nm was from 10 to 60 nm. Zeta potential indicated by seed bioactive metabolites for the stabilisation of green synthesised AgNPs and the capping of AgNPs. The small distribution of particulate size with z-average value of 40.5 nm and low index of Polydistributor 0.31 is evident from (Fig. 6) of the *Lepidium sativum* seed extract AgNPs. From the figure B indicates that the possible value of the capping molecules on the AgNP surface -31.2 suggested that they consist mostly of the groups with a negative charge and that the AgNPs still have a mild stability.



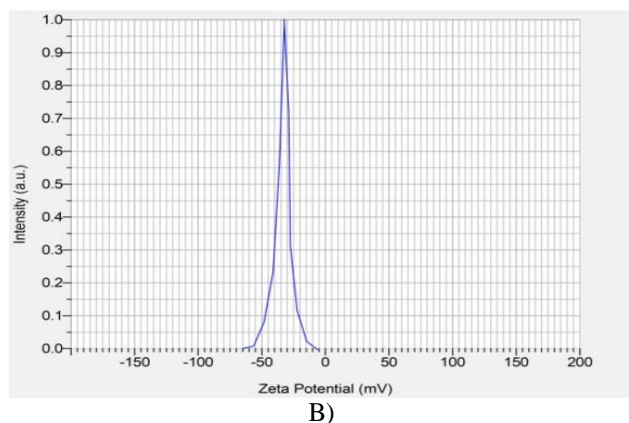


Figure 6: A and B Zeta potential of green synthesized AgNPs silver nanoparticles after adding 0.1 mM AgNO₃ solution in Seed extract

UV VISIBLE SPECTROSCOPY

With a Shimadzu UV spectrophotometer (model UV-1800), UV-visible research was carried out. Colloidal solution OD values (1 ml) were observed at an interval of 4 h, beginning from 4 h to 24 h. Distilled water was used as blank. Using the wavelength of 300 to 800 nm, the absorbance maxima is recognised at various time intervals (Salataet al., 2004; Feldheim and Foss, 2002; Mervatet al., 2012).

FOURIER TRANSFORM INFRA RED SPECTROSCOPY

For the identification of functional groups capping in silver nanoparticles, FTIR spectroscopy analysis of synthesized nanoparticles was performed. The synthesized colloidal solution was centrifuged for 10 min at 8,000 rpm and the pellet was washed with DD water three times. The subsequent suspension was dried completely and examined by FTIR (Perkin Elmer Spectrum-one, USA) using a freeze dryer (Jaidevet al., 2010). The samples were mixed with KBr powder and pelletized to conduct FTIR, and the spectrum results were collected using Perkin Elmer with a wavelength range between 4000 cm⁻¹ and 400 cm⁻¹ after drying (Feldheim and Foss, 2002; Mittal et al., 2014).

X-RAY DIFFRACTION ANALYSIS OF GREEN SYNTHESIZED SILVER NANOPARTICLES

The crystalline nature of S-AgNPs has been examined by X-ray diffraction (XRD) analysis. The S-AgNP powder was coated on the XRD grid and examined using the GE X-ray diffraction method XRD 3003 TT with 1.5406 Å wavelengths at a scale of 10 ° –80 °. This research was carried out at department of biochemistry, Osmania University, Hyderabad.

SCANNING ELECTRON MICROSCOPY OF GREEN SYNTHESIZED SILVER NANOPARTICLES

The surface morphology of nanoparticles was studied by Scanning Electron Microscopy (SEM) and the dry nanoparticle samples were prepared by falling over a carbon-coated grid and allowed to dry prior to measurement on the Hitachi S-3400 N. SEM instruments were operated at an accelerated voltage at 20 kV

ENERGY DISPERSIVE SPECTROSCOPY ANALYSIS OF GREEN SYNTHESIZED SILVER NANOPARTICLES

The presence of elemental silver was confirmed through energy dispersive spectroscopy (EDS). EDX analysis was carried out for dried nanoparticles exposed were coated on to carbon film and performed on Scanning electron microscopy instrument equipped with Thermo EDX attachments (Najimuet al., 2014). Thin films of the sample were prepared on a carbon coated copper grid by just dropping a very small amount of the sample on the grid, and then the film on the SEM grid were allowed to dry by putting it under a mercury lamp for 5 min (Nalawadeet al., 2014).

ZETA POTENTIAL

The particle size distribution of AgNPs was evaluated using colloidal solutions were subjected for zeta potential analysis. Data obtained were analyzed using Zetasizer software (Sukirthaet al., 2012).

ANTI BACTERIAL ACTIVITY

The antibacterial potential of biofunctionalized AgNPs bacterial and fungal by using agar well diffusion according to standard methods (NCCLS 2000). About 20 mL of molten and cooled nutrient agar media was poured into sterilized Petri dishes. The plates were left overnight at room temperature to allow any contamination to appear. The discs were placed on Muller Hinton agar plates inoculated with each of the previously mentioned microorganisms and 20 µg/mL of Ag-NPs and fruit extracts were placed on inoculated agars. The test plates were incubated at 37 °C for 24 h. Later on the incubation period, the zone of inhibition (in

millimeter diameter) was noted and tabulated. Streptomycin was used as an antibacterial standard against all pathogens. Experiments were performed in triplicate

III. Results And Discussion

ANTIBACTERIAL ACTIVITY

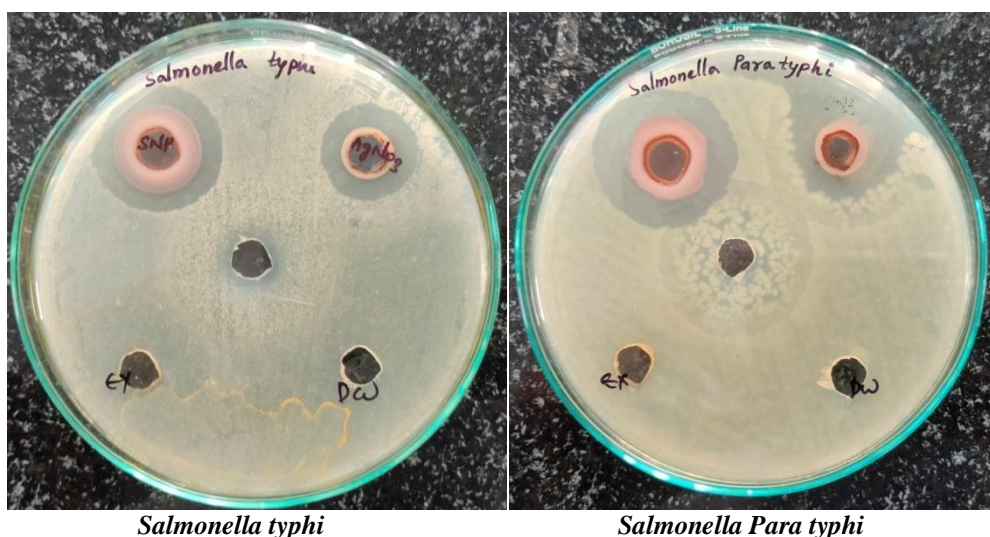
In this study, the antibacterial activity of the spice blend AgNPs was evaluated against two Gram-positive and two Gram-negative bacterial species. The biosynthesized Ag-NPs were effective against *Streptococcus faecalis*, *Bacillus cereus*, *Escherichia coli* and *Shigella flexneri* (Figure 6), with a notable MIC at 6.25 mg/mL for the four organisms.

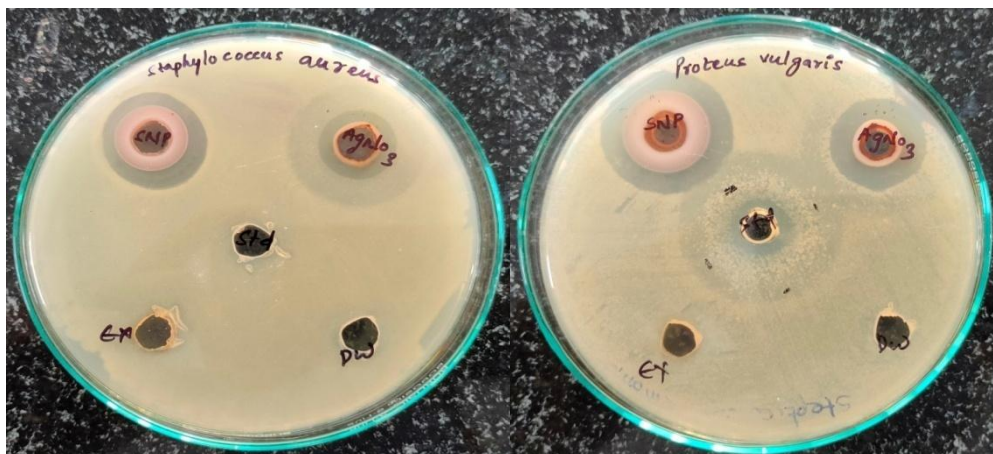
A significant number of antimicrobial agents, such as silver ions and inorganic sources, have been used and continue to be used in diverse industries as antimicrobial agents. However, multi-drug tolerance restricts these chemical antimicrobial agents. The efforts have also concentrated on alternative methods of countering microbial drug resistance, which is why biosynthesized AgNPs have been evaluated.

In this present research investigation, the antibacterial activity of the synthesized AgNPs from *Lepidium sativum* aqueous extracts were tested against six Gram-positive and six Gram-negative bacterial cultures. The antibacterial efficacy of nanoparticles was observed maximum against the five bacterial strains tabulated table (1) Figure (7). Ofloxacin antibiotic were used as a standard in each Petri dish. The zone of inhibition were exhibited with 31 mm against *Bacillus subtilis*, 30 mm against *Bacillus stearothermophilus* followed by 25 mm against *Proteus vulgaris*, *Salmonella Para typhi* with 24 mm, *Salmonella typhi* with 23 mm, *Staphylococcus aureus* with 19 mm, *Pseudomonas aeruginosa* with 21mm, *Micrococcus luteus* with 26 mm, *klebsiella pneumoniae* with 21 mm, *E.Coli* with 29 mm, *Bacillus megaterium* with 19mm and *Bacillus cereus* with 21 mm. In addition to silver nanoparticles, biologically active compounds in the *Lepidium sativum* aqueous extract may be responsible for improved antibacterial activity of green synthesised nanoparticles.

Test Organisms	Silver nanoparticles	AgNo3	Standard	DW	Extract
Zone of inhibition in mm					
<i>Salmonella typhi</i>	23	18	09	ND	ND
<i>Salmonella Para typhi</i>	24	16	20	ND	ND
<i>Staphylococcus aureus</i>	19	10	06	ND	ND
<i>Proteus vulgaris</i>	25	15	17	ND	ND
<i>Pseudomonas aeruginosa</i>	21	16	30	ND	ND
<i>Micrococcus luteus</i>	26	17	10	ND	ND
<i>klebsiella pneumoniae</i>	21	09	22	ND	ND
<i>E.Coli</i>	29	11	06	ND	ND
<i>Bacillus stearothermophilus</i>	30	09	07	ND	ND
<i>Bacillus subtilis</i>	31	18	07	ND	ND
<i>Bacillus megaterium</i>	19	19	08	ND	ND
<i>Bacillus cereus</i>	21	08	07	ND	ND

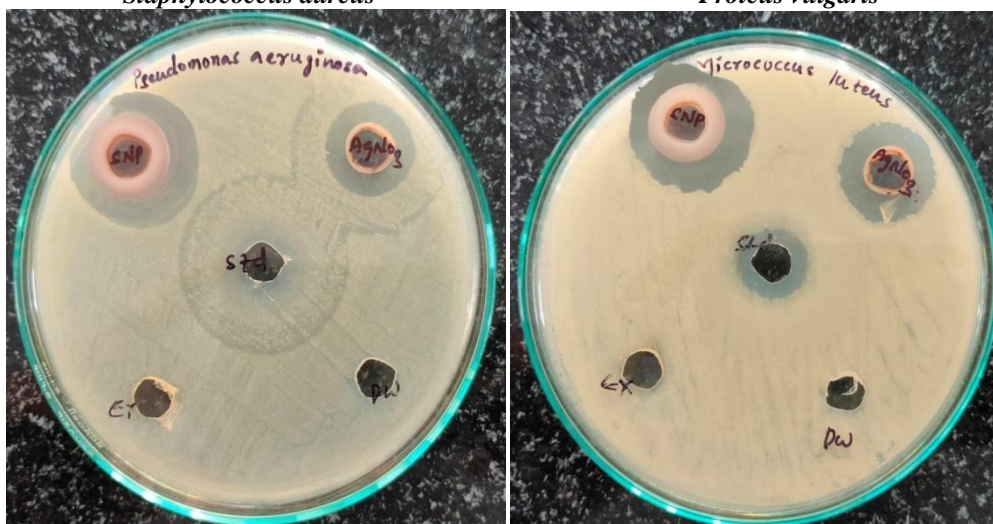
Table 1 antibacterial activity of the synthesized AgNPs from *Lepidium sativum* aqueous extracts tested against six Gram-positive and six Gram-negative bacterial, ND-Not detected, DW-Distilled water, AgNo3-Silver nitrate





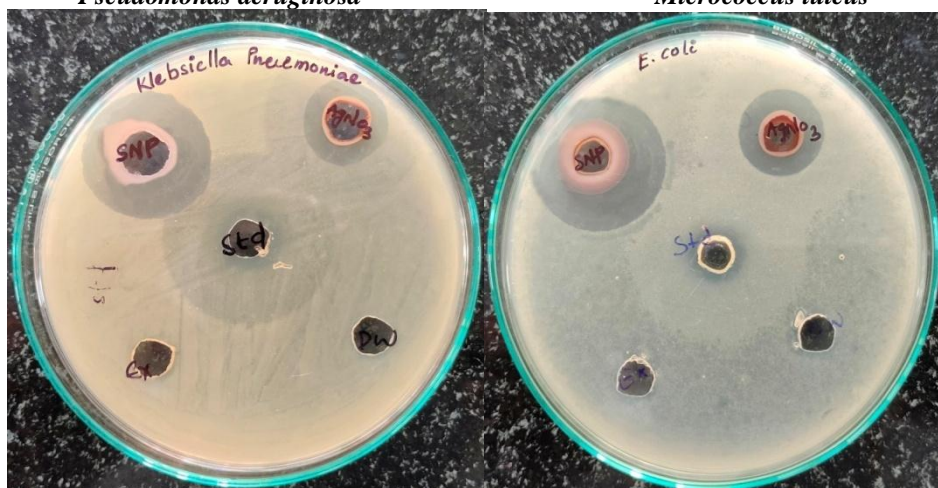
Staphylococcus aureus

Proteus vulgaris



Pseudomonas aeruginosa

Micrococcus luteus



klebsiella pneumoniae

E.Coli

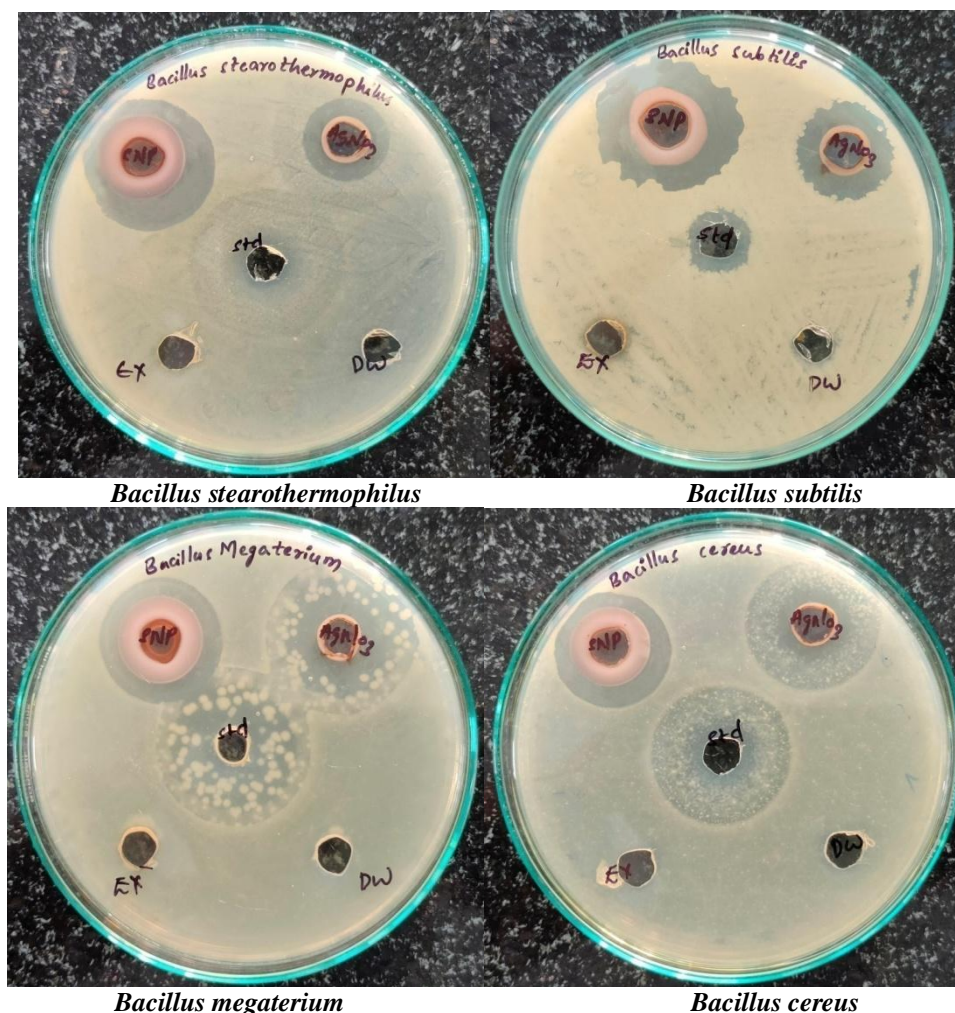


Figure 7: Antibacterial activity of *Lepidium sativum* seed mediated Silver nanoparticles

ANTIOXIDANT ACTIVITY

DPPH

Free radical scavenging activity of green synthesized silver nanoparticles The 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) free radicals scavenging potential of the silver nanoparticles were determined. Different concentrations of silver nanoparticles with (20, 40, 60, 80, and 100 µg / mL) and standard vitamin C have been taken in various test tubes. 1 mL of 2.5 mM of freshly prepared DPPH was dissolved in methanol and added to the test tubes. The solution was eventually incubated in a dark in place for 30 minutes. Stable DPPH absorbance was recorded at 517 nm. The standard solution of vitamin C was prepared using the same method. Free radical scavenging activity was represented as an inhibitory percentage determined using the DPPH radical scavenging activity equation (Kanipandian et al., 2014).

The antioxidant effect of *Lepidium sativum* seed mediated Silver nanoparticles was analysed by DPPH assay, Nitric oxide assay and Reducing power assays. DPPH is a neutral free radical that accepts donor hydrogen or electron and becomes reduced from violet to yellow by colour changes. In the present study the DPPH radical quench ability of *Lepidium sativum* seed mediated Silver nanoparticles were tested using standard Ascorbic acid as a positive control in this assay. The DPPH radical scavenging activity of *Lepidium sativum* seed Silver nanoparticles increased dependently on concentration and also increased over time. The dose-dependent decolonization of purple DPPH radical to yellow DPPH molecules by the sample with the percentage of activity was observed at five different concentrations. The highest Inhibition percentage of *Lepidium sativum* seed mediated silver nanoparticles was $53.29 \pm 0.64\%$ in $100 \mu\text{g/ml}$ and the extract with $49.01 \pm 0.64\%$, while the standard ascorbic acid activity was $53.54 \pm 0.20\%$. Compared to the standard, the nitrite oxide radical scavenging activity of *Lepidium sativum* seed mediated silver nanoparticles was more and *Lepidium sativum* seed extract was moderate scavenging activity **Table 2. Fig 8**

Concentrations	Percentage of inhibition DPPH
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µg/ml	Ascorbic acid	SNP	EXT
20	7.42±0.27	11.46±0.23	4.36±0.23
40	17.84±0.39	26.03±0.42	19.24±0.42
60	36.78±0.33	35.06±0.12	29.42±0.12
80	39.79±0.11	49.58±0.31	39.12±0.31
100	53.54±0.20	53.29±0.64	49.01±0.64

Table 2 : Antioxidant DPPH assay of *Lepidium sativum* seed mediated Silver nanoparticles

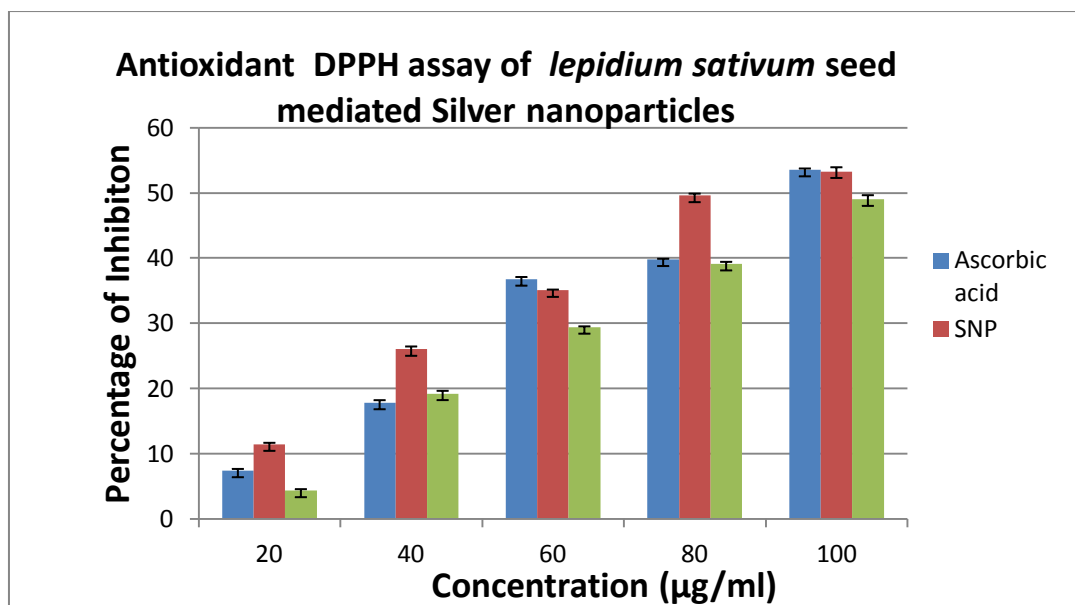


Figure 8: Antioxidant DPPH assay of *Lepidium sativum* seed mediated Silver nanoparticles

It is well known that in various inflammatory processes, nitric oxide has an significant role. Sustained production levels of this radical are directly tissue-toxic and lead to vascular collapse (Hazra, 2008).The highest Inhibition percentage of *Lepidium sativum* seed mediated silver nanoparticles was 69.43±0.35% in 100µg/ml and the extract with 46.34±0.62%, while the standard ascorbic acid activity was 41.54±0.26%. Compared to the standard, the nitrite oxide radical scavenging activity of *Lepidium sativum* seed mediated silver nanoparticles was more and *Lepidium sativum* seed extract was moderate scavenging activity **Table 3.Fig 9**

Concentrations µg/ml	Percentage of inhibition NO		
	Ascorbic acid	SNP	EXT
20	5.14±0.29	15.56±0.32	6.58±0.42
40	12.83±0.38	29.12±0.71	18.11±0.78
60	26.39±0.36	46.12±0.42	26.21±0.62
80	31.78±0.17	56.62±0.56	39.31±0.41
100	41.54±0.26	69.43±0.35	46.34±0.62

Table 3: Antioxidant NO assay of *Lepidium sativum* seed mediated Silver nanoparticles

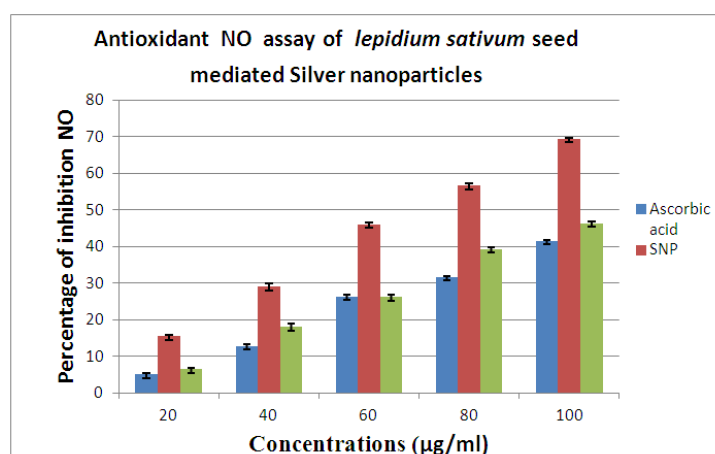


Figure 9: Antioxidant NO assay of *Lepidium sativum* seed mediated Silver nanoparticles

Since the reducing power activity of the compounds could serve as a significant indicator of the antioxidant potential, we assessed this property by measuring the ability of the extracts to transform Fe³⁺ to Fe²⁺ and to donate an electron. The highest Inhibition percentage of *Lepidium sativum* seed mediated silver nanoparticles was $66.9 \pm 1.5\%$ in 100 $\mu\text{g/ml}$ and the extract with $46.4 \pm 1.5\%$, while the standard ascorbic acid activity was $61.54 \pm 0.20\%$. Compared to the standard, the Reducing powerscavenging activity of *Lepidium sativum* seed mediated silver nanoparticles was more and *Lepidium sativum* seed extract was moderate scavenging activity **Table 4, Fig 10**

Concentrations $\mu\text{g/ml}$	Percentage of inhibition Reducing power assays		
	Ascorbic acid	SNP	EXT
20	10.14 ± 0.27	17.1 ± 1.7	19.9 ± 4.3
40	20.83 ± 0.39	29.6 ± 2.4	13.7 ± 3.2
60	36.39 ± 0.33	33.3 ± 2.3	29.9 ± 2.6
80	48.78 ± 0.11	45.6 ± 1.7	38.1 ± 2.3
100	61.54 ± 0.20	66.9 ± 1.5	46.4 ± 1.5

Table 4: Antioxidant reducing power assays of inhibition *Lepidium sativum* seed mediated Silver nanoparticles

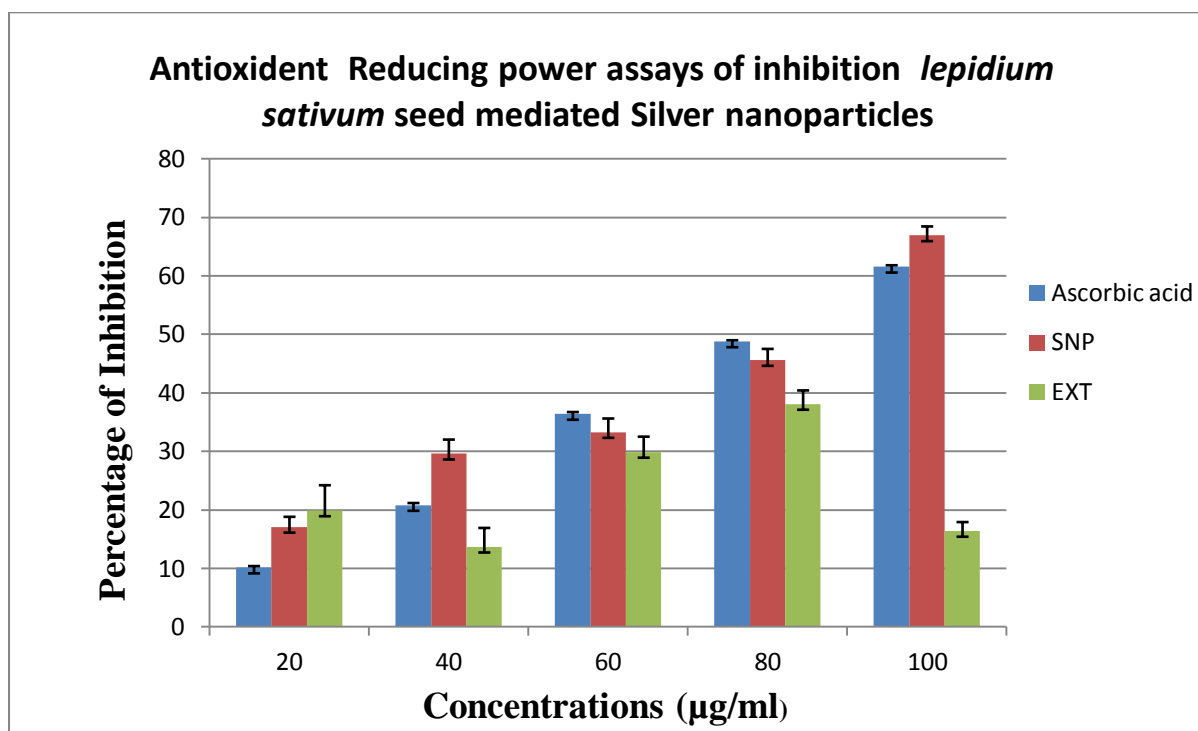


Figure 10: Antioxidant reducing power assays of inhibition *Lepidium sativum* seed mediated Silver nanoparticles

IV. Discussion

Generally, three different techniques, including physical, chemical, and biological processes, have been used to synthesis nanoparticles. Gurav 1994, Kruis 2000, Magnusson 2000, Schmidt 1988. In the present study *Lepidium sativum* seed extract mediated nanoparticles were synthesized. AgNO₃'s colorless solution and seed extract greenish yellow, turned to dark brown in color 10 minutes later; this is a signaling that the silver nanoparticles had been formed Marslin 2018. Gurunathan 2009 reported that we can control the shape, size, and monodispersity of the nanoparticles using plant extracts as reducing agents. The benefits of biological techniques are the accessibility of a broad variety of biological resources, reduced time requirements, high density, stability and ready solubility of prepared nanoparticles in water Thakkar 2010. The surface plasma resonance (SPR) peak presence at 425 nm confirms the development of AgNPs. Spherical particles are responsible for the observed symmetric sharper SPR band with a single peak reported by (Jagajjanani et al., 2013). The addition of fresh *Lepidium sativum* seed extract to 0.1 mM of silver nitrate solution resulted in a brown colour. The highest absorbance at 425 nm was shown by UV-vis spectrophotometer analysis.

Antibacterial activity of silver ions is reported early in the report to suggest that silver ions can prevent diagnostic diarrhoea by binding them to the thiolite group and contributing to cellular death (Liauet et al., 1997, Feng et al . 2000). The present research work evidenced the antibacterial potentiality with *Lepidium sativum* seed extract mediated silver nanoparticles. Krishnaraj et al. (2010) examined the biosynthesis of silver

nanoparticles and their activity on waterborne bacterial pathogens. **Balasubramanian et al., (2015)** studied antimicrobial activity against pathogens such as *Enterococcus faecalis*, *Staphylococcus aureus* (gram-positive), *Pseudomonas aeruginosa*, *E. coli* (gram-negative) by disc diffusion. The antioxidant activity of synthesised nanoparticles using the DPPH assay was analysed by **Kharat and Mendhulkar (2016)** and the antioxidant potential of photosynthesized nanoparticles was observed (**Kharat and Mendhulkar, 2016**). There are few studies on the biosynthesized AgNPs' antioxidant activity. In *Fraxinus excelsior* leaf extract (**Parveen 2016**), *Elephantopus scaber* (**Kharat 2016**), *Terminalia* species leaf extract (**El-Rafie 2014**), *Cleistanthus collinus* (**Kanipandian 2014**), the synthesis, characterization and antioxidant activities of AgNPs were recorded. The presence of functional groups on the surface of silver nanoparticles allows these properties of silver nanoparticles to occur.

V. Conclusion

An successful eco-friendly and inexpensive method has been achieved for the green synthesis of silver nanoparticles (AgNPs) by *Lepidium sativum* seed extract. Biosynthesized AgNPs is spherical in form, by antibacterial activity studies the nanoparticle shows more inhibition on gram positive bacteria and DPPH antioxidant studies shows more reducing power with increase in concentration of AgNp extract. This green biosynthesis approach is a non-toxic alternative to conventional chemical and physical approaches and will be suitable for large-scale biological processing and prospective therapies.

References:

- [1]. Ge L, Li Q, Wang M, Ouyang J, Li X, Xing MM, et al. Nanosilver particles in medical applications: Synthesis, performance, and toxicity. *Int J Nanomedicine*. 2014;9:2399–407.
- [2]. Ahmed S, Ahmad M, Swami BL, Ikram S. A review on plants extract mediated synthesis of silver nanoparticles for antimicrobial applications: A green expertise. *J Adv Res*. 2016;7:17–28. [PMC free article] [PubMed]
- [3]. Prasad R. Synthesis of silver nanoparticles in photosynthetic plants. *J Nanopart* 2014. 2014:8.
- [4]. Ravindran, P.; Fu, J.; Wallen, S.L. Completely Green Synthesis and Stabilisation of Metal Nanoparticles. *J Am Chem Soc* 2003, 125, 13940–13941. doi: 10.1021/ja029267j
- [5]. Albrecht, M.A.; Evans, C.W.; Raston, C.L. Green Chemistry and the Health Implications of Nanoparticles. *Green Chem* 2006, 8,417–432. doi: 10.1039/b517131h
- [6]. Ashok Kumar, S.; Ravi, S.; Kathiravan, V.; Velmurugan, S. Synthesis of Silver Nanoparticles Using *A. indicum* Leaf Extract and their Antibacterial Activity. *Spectrochim Acta Part A: Mol Biomol Spectrosc* 2015, 134,34–39. doi: 10.1016/j.saa.2014.05.076
- [7]. Nakkala, J.R.; Mata, R.; Gupta, A.K.; Sadras, S.R. Green Synthesis and Characterization of Silver Nanoparticles Using *Boerhaaviadiffusa* Plant Extract and their Antibacterial Activity. *Indus Crop Prod* 2014, 52,562–566. doi: 10.1016/j.indcrop.2013.10.050
- [8]. Kumar, V.; Yadav, S.K. Synthesis of Stable, Polyspherical Silver and Gold Nanoparticles Using Leaf Extract of *Lonicera japonica* L. *Int J Green Nanotechnol* 2011, 3,281–291. doi: 10.1080/19430892.2011.633474 [Taylor & Francis Online],
- [9]. Tolaymat, T.M.; Badawy, A.M.E.; Genaidy, A.; Scheckel, K.G.; Luxton, T.P.; Suidan, M. An Evidence-based Environmental Perspective of Manufactured Silver Nanoparticle in Synthesis and Applications: A Systematic Review and Critical Appraisal of Peer-reviewed Scientific Papers. *Sci Total Environ*. 2010, 408, 999–1006. doi: 10.1016/j.scitotenv.2009.11.003
- [10]. Alvarez-Puebla, R.A.; Aroca, R.A. Synthesis of Silver Nanoparticles with Controllable Surface Charge and their Application to Surface-enhanced Raman Scattering. *Anal Chem* 2009, 81,2280–2285. doi: 10.1021/ac8024416
- [11]. Agnihotri, S.; Mukherji, S.; Mukherji, S. Size-controlled Silver Nanoparticles Synthesized over the Range 5–100 nm Using the Same Protocol and their Antibacterial Efficacy. *RSC Adv* 2014, 4,3974–3983. doi: 10.1039/C3RA44507K ()
- [12]. S. Mahdi, M. Taghdiri, V. Makari, M. Rahimi-Nasrabadi Procedure optimization for green synthesis of silver nanoparticles by aqueous extract of *Eucalyptus oleosa*
- [13]. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 136 (2015), pp. 1249-1254.
- [14]. H. Padalia, P. Moteriya, S. Chanda Green synthesis of silver nanoparticles from marigold flower and its synergistic antimicrobial potential *Arabian Journal of Chemistry* (2014), 10.1016/j.arabjc.2014.11.015
- [15]. M.R. Bindhu, M. Umadevi Antibacterial and catalytic activities of green synthesized silver nanoparticles *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 135 (2015), pp. 373-378
- [16]. S. Ahmed, M. Ahmad, B.L. Swami, S. Ikram Plants extract mediated synthesis of silver nanoparticles for antimicrobial applications: a green expertise *Journal of Advance Research* (2015), 10.1016/j.jare.2015.02.007
- [17]. Gurav A.S., Kodas T.T., Wang L.M., Kauppinen E.I., Joutsensaari J. Generation of nanometer-size fullerene particles via vapor condensation. *Chem. Phys. Lett*. 1994;218:304–308. doi: 10.1016/0009-2614(93)E1491-X.
- [18]. Kruis F.E., Fissan H., Rellinghaus B. Sintering and evaporation characteristics of gas-phase synthesis of size-selected PbS nanoparticles. *Mater. Sci. Eng. B*. 2000;69:329–334. doi: 10.1016/S0921-5107(99)00298-6
- [19]. Magnusson M.H., Deppert K., Malm J.O., Bovin J.O., Samuelson L. Size-selected gold nanoparticles by aerosol technology. *Nanostruct. Mater*. 1999;12:45–48. doi: 10.1016/S0965-9773(99)00063-X.
- [20]. Schmidt-Ott A. New approaches to in situ characterization of ultrafine agglomerates. *J. Aerosol Sci*. 1988;19:553–563. doi: 10.1016/0021-8502(88)90207-8.
- [21]. Marslin, K. Siram, Q. Maqbool et al., “Secondary metabolites in the green synthesis of metallic nanoparticles,” *Materials*, vol. 11, no. 6, p. 940, 2018
- [22]. Gurunathan S., Kalishwaralal K., Vaidyanathan R., Venkataraman D., Pandian S.R., Muniyandi J., Hariharan N., Eom S.H. Biosynthesis, purification and characterization of silver nanoparticles using *Escherichia coli*. *Colloids Surf. B Biointerfaces*. 2009;74:328–335.
- [23]. Thakkar K.N., Mhatre S.S., Parikh R.Y. Biological synthesis of metallic nanoparticles. *Nanomedicine*. 2010;6:257–262. doi: 10.1016/j.nano.2009.07.002.
- [24]. sing SEM, we can probe the morphology of particles and derive a histogram from the images by either by measuring and counting the particles manually, or by using specific software .