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Antibacterial efficacy of silver nanoparticles synthesized from *Alpinia calcarata*, *Melia dubia* and *Solanum nigrum*

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Abstract

In the present study, silver nanoparticles were synthesized from the rhizome extract of *Alpinia calcarata* and the leaves extract of *Melia dubia* and *Solanum nigrum* by a simple and eco-friendly route. Bioreduction of Ag^+ to Ag^0 was observed when the aqueous leaf extract was mixed with 1mM of silver nitrate ($AgNO_3$) and heated to 95 ºC. The development of silver nanoparticles was characterized on the basis of change of colour with the help of UV-VIS spectroscopy. The biosynthesized silver nanoparticles were tested for their antibacterial activity the test cultures of *Staphylococcus aureus* (Gram Positive) and *Klebsiella pneumonia* (Gram Negative) bacteria. The silver nanoparticles showed a clear zone of inhibition against these bacterial strains. This strategy gives a straightforward, simple and less-time consuming technique for synthesizing nanoparticles which could be utilized in several areas of medicine.

Keywords: Silver nanoparticles, UV-VIS, bioreduction, antibacterial activity, zone of inhibition.

INTRODUCTION

The field of nanotechnology is sprouting as a standout amongst the most dynamic areas in the present day material science. It is a study of events on nanometer scale and mainly concerned with synthesis of nanoparticles by controlling matter at the molecular level. Nanoparticles have an expansive surface area to volume ratio which brings about chemical reactivity and biological activity when compared to their bulk materials. In the last two decades, metal nanoparticles have pulled much attention owing to their unique size with shape-dependent physical, chemical, catalytic and biological properties and broad application prospects in modern industries and medicine (Grzegorz Dzido *et al*., 2015). Among the metallic nanoparicles, silver nanoparticles have emerged as an extravagant product due to their hopeful bactericidal effect capable of killing about 650 types of diseases causing microorganisms (Jayshree Annamalai *et al.,* 2016). Compared with other metals, sliver exhibits higher toxicity to microorganisms, while it exhibits lower toxicity to mammalian cells (Shrivastava *et al*., 2007). Due to their medicinal and antimicrobial properties (Virender K. Sharma *et al*., 2009; Peter Logeswari *et al*., 2005; Nafeesa Khatoon *et al*., 2015). AgNPs have been incorporated into more than 200 consumer products including textile fabrics, polymers, dental material, medical devices, burn dressings and cosmetics (Veeraputhiran, 2013; Zhang *et al*., 2016). Silver has been known to have a disinfecting effect and has been found in applications ranging from customary medicines to culinary things (Sinha, 2009). The chief use of silver and silver nanoparticles in medical industry varied from preventing infections to averting colonization of bacteria on catheters (Kamyar Shameli *et al*., 2012). Besides, it is also widely used in textile industry to eliminate microorganisms and in water treatment plant as disinfectants (Feng Zhang *et al*., 2009; Tiwari *et al*., 2008). Eventhough the silver nanoparticles are produced by a variety of strategies, there is still a need for economically practical as well as ecological amicable procedures to avoid the high pressure, temperature, energy and toxic chemicals in the synthesis protocols to evade antagonistic impacts in medical applications. From the recent results, researchers inspired on biological systems to develop benign nanoparticles using bacteria (Malarkodi *et al*., 2013), fungi (Sadowski *et al*., 2008), plant and leaf extracts (Priya Banerjee *et al*., 2014; Ashok Kumar, 2012; Nithya and Ragunathan, 2009) termed as green chemistry approaches.

Worldwide, infectious diseases are the number one cause of death accounting for approximately one-half of all deaths in tropical countries (Vikram Paritala *et al*., 2015). The development of new resistant strains of bacteria to current antibiotics has become a serious public health problem. These bacterial strains with limited life span and reduced susceptibility to antibiotics raise the specter of untreatable bacterial infections and add urgency to the search for new infections fighting strategies. Medicinal plants are the prime and economic source of Indian traditional medicine. They are very useful to humankind since the dawn of civilization to treat many infectious

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diseases. Over 50% of all modern clinical drugs and natural products play an important role in drug development programs in the pharmaceutical industry (Angalaparameswari *et al*., 2012). Phytomedicines derived from plants have shown great promise in the treatment of intractable infectious diseases. Many plants contain microbial inhibitors and they are used as traditional medicines and as sources of wide range of substances which can be useful to treat infectious or other serious diseases (Yinegar and Yewhalaw, 2009). In the present scenario, the utilization of medicinal plants for the synthesis of nanoparticles has emerged as novel antimicrobial agents.

Alpinia calcarata, commonly known as snap ginger, has various therapeutic activities: thermogenic, anti-inflammatory (Asolkar *et al*., 1992), nervinetonic, stimulant, revulsive, carminative, stomachic, disinfectant, expectorant, broncho-dilator and antifungal property (Warrier *et al*., 1996). *Melia dubia,* commonly called as 'Malaivembu' in Tamil, is known for rich and valuable source of bioactive limonoids. Its leaf and root extracts possess antibacterial, antifungal, antimalarial (Chanthuru *et al*., 2014), anticancer, antiviral and a number of other pharmacological activities on humans (Endo *et al*., 2002; Nakagawa *et al*., 2001). *Solanum nigrum,* commonly known as Black night shade and 'Manathakkali' in Tamil, shows medicinal properties like antimicrobial, anti-oxidant, cytotoxicity, antiulcerogenic and hepatoprotective activity (Rajathi *et al*., 2015). But little information is available on the antibacterial activity of silver nanoparticles mediated by these medicinal leaves and rhizome. Hence, in the present article it has been dealt with the antibacterial activity of silver nanopaticles mediated by *Alpinia calcarata, Melia dubi* and *Solanum nigrum*.

MATERIALS AND METHODS

Alpinia calcarata, Melia dubia and *Solanum nigrum* were purchased from the local market of Tiruchirappalli, Tamilnadu (Figs. 1a – 1c). The plant materials were identified and authenticated at the Rapinant Herbium, St. Joseph College (Autonomous), Thiruchirapalli, Tamilnadu.They were washed with distilled water and air dried for 10 days on a clean sheet. Then they were kept in the hot air oven at 60 °C for 24 hours and then ground to a fine powder. Then the aqueous extracts of the plant materials were prepared, evaporated to dryness and weighed (Ammara Hassan *et al*., 2009).

Synthesis of Silver Nanoparticles

1mM of silver nitrate was added to 0.5g of the plant extract and made up to a final solution of 200 ml, and the solution was centrifuged at 15,000 rpm for 20 minutes. The collected pellets were stored at 4 °C. The supernatant was heated at 50 ° to 95 °C. A change in the colour of the solution was observed during the heating process (Jae Yong Song and Beom Soo Kim 2009).

Fig 1. a. *Alpinia calcarata b. Melia dubia c. Solanum nigrum*

UV-VIS Spectral Analysis

In order to monitor the formation of silver nanoparticles, the absorption spectra of synthesized silver nanoparticles were recorded using UV-VIS spectroscopy. A graph of wavelength on X-axis and absorbance on Y-axis was plotted.

Antibacterial activity of synthesized Silver Nanoparticles

Bacterial cultures of *Staphylococcus aureus and Klebsiella pneumonia* were used as antibacterial test organisms. All the Bacterial strains were obtained from Doctor Diagnostics Centre, Thiruchirappalli, Tamil Nadu. Antibacterial activity of the silver nanoparticles synthesized using *Alpinia calcarata, Melia dubia, Solanum nigrum* powder was tested by agar well diffusion method (Murray *et al*., 1995) against *Staphylococcus aureus and Klebsiella pneumonia*. Each strain was swabbed uniformly onto the individual plates using sterile cotton swabs. Wells of 0.5cm were made on nutrient agar plates using gel puncture. Using a micropipette, 50µg/ml, 75µg/ml, 100µg/ml of nanoparticles and solution was poured into each well on all plates. After incubation at 37 °C for 24 hours, the diameter of zone of inhibition was measured in millimeter. Streptomycin was used as positive control in this study.

RESULTS AND DISCUSSION

Synthesis of Silver Nanoparticles

When the rhizome extract of *Alpinia calcarata* and leaf extract of *Melia dubia* and *Solanum nigrum* were mixed with aqueous solution of silver nitrate, it was reduced into silver ion into silver particles. It is followed by the colour change from watery yellow (Figs. 2a to 2c) to yellowish brown colour (Figs. 3a to 3c) which indicates the formation of silver nanoparticles (Sulochana *et al*., 2012; Sandeep Thapa *et al*., 2015). The development of biologically inspired experimental processes for the synthesis of nanoparticles is evolved into an important branch of nanotechnology. Silver nitrate is used as reducing agent as silver has distinctive properties such as good conductivity, catalytic and chemical stability (Bekkeri Swathy, 2014). The plant extracts along with silver nitrate will be watery yellow in colour. But it is well known that silver nanoparticles exhibit yellowish brown colour in aqueous solution due to excitation of surface plasmon vibrations in silver nanoparticles (Ponarulselvam *et al*., 2012). Thus the colour from watery yellow to yellowish brown colour clearly indicates the formation of silver nanoparticles from *Alpinia calcarata*, *Melia dubia* and *Solanum nigrum*. The time duration of change in colour varies from plant to plant. In the present study, all the extracts synthesize silver nanoparticles within 10 min. Savithramma *et al*. (2011) reported that leaves of *Boswellia ovalifoliolata* synthesized silver nanoparticles within 10 min. Green synthesis of silver nanoparticles using *Ocimum* has also been reported (Mallikarjuna *et al*., 2008). Silver nanoparticles synthesized using leaf extract was

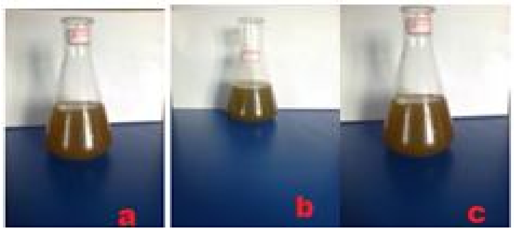

Fig. 2. a. *Alpinia calcarata b. Melia dubia c. Solanum nigrum*

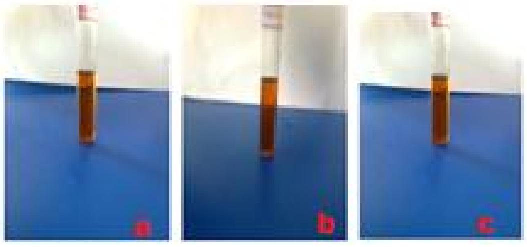

Fig. 3.a. *Alpinia calcarata b. Melia dubia c. Solanum nigrum*

advantageous over bacteria due to long process of maintaining cell cultures.

UV-Vis Spectral Analysis

The Ultraviolet Visible spectrum showed that the Surface Plasmon Resonance (SPR) band was centered at around 415 nm, 426 nm and 433 nm (complete reduction of silver ions) from *Alpinia calcarata, Melia dubia* and *Solanum nigrum* respectively (Figs. 4a to 4c). It is well known that UV-Vis spectroscopy could be exploited to study size and shape controlled nanoparticles in aqueous suspensions (Shrivastava *et al*., 2009). Similarly, the Ultraviolet Visible absorption spectrum of silver nanoparticles synthesized from *Allium cepa* was found to be 413 nm (Benjamin and Bhrathwaj, 2011), 412 nm for *Eucalyptus hybrida* leaf extract (Manish Dubey *et al*., 2009), 430 nm for *Euphorbia hirta* (Elumalai *et al*., 2010), 440 nm for *Catharanthus roseus* (Mukunthan *et al*., 2011).

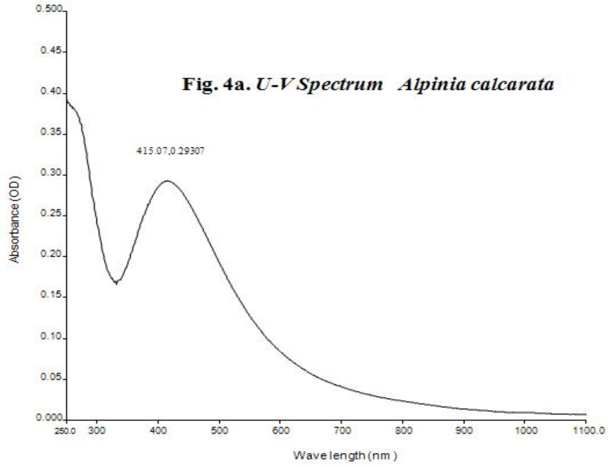

Fig. 4a. *U-V Spectrum Alpinia calcarata*

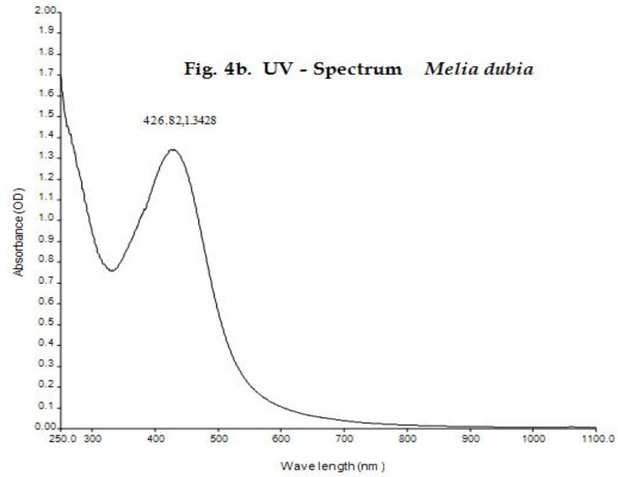

Fig. 4b. UV - Spectrum *Melia dubia*

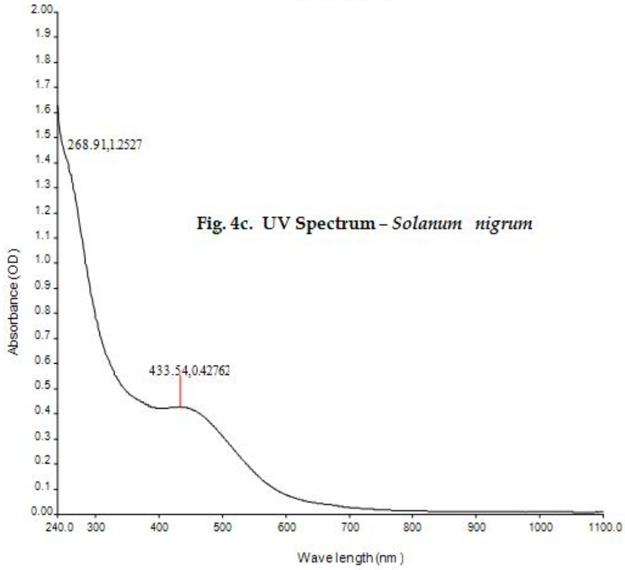

Fig. 4c. UV Spectrum – *Solanum nigrum*

Antibacterial activity of synthesized Silver Nanoparticles

The antibacterial activity of silver nanoparticles of the rhizome extract of *Alpinia calcarata* (Fig. 5a) and leaf extracts of *Melia dubia* (Fig. 5b) and *Solanum nigrum* (Fig. 5C) was investigated against *Staphylococcus aureus* (gram positive bacteria) and *Klebsiella pneumonia* (gram negative bacteria) using agar well diffusion method. Positive control, Streptomycin, was also maintained

Fig. 5a. *Alpinia calcarata* (i) *Staphylococcus aureus* (ii) *Klebsiella pneumonia*

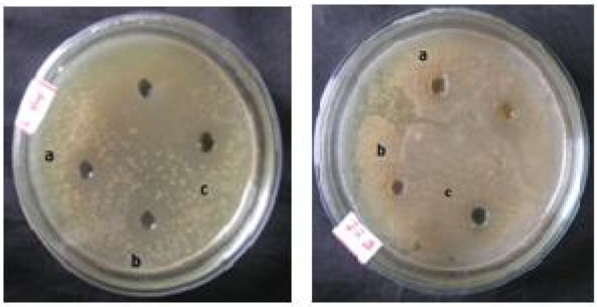

Fig. 5b. *Melia dubia* (i) *Staphylococcus aureus* (ii) *Klebsiella pneumonia*

Fig. 5c. *Solanum nigrum* (i) *Staphylococcus aureus* (ii) *Klebsiella pneumonia*
a – 50 µg/ml ; b – 75 µg/ml ; c – 100 µg/ml ; d – Streptomycin

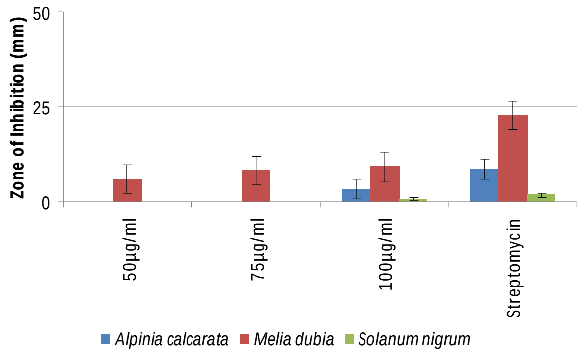

Fig. 6. Antibacterial activity and zone of inhibition (mm) shown by synthesized AgNP from extracts of leaves against *Staphylooccus aureus* (gram positive) using agar well diffusion method.

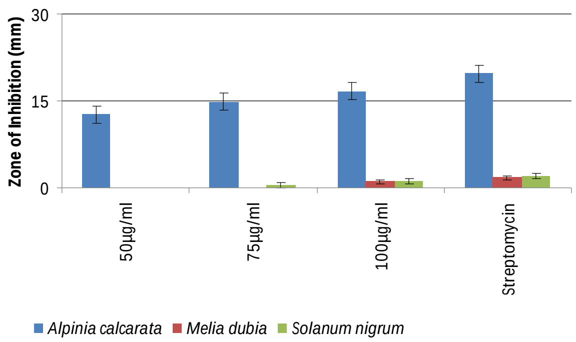

Fig. 7. Antibacterial activity and zone of inhibition (mm) shown by synthesized AgNP from extracts of leaves against *Klebsiella pneumonia* (gram negative) using agar well diffusion method.

Table 1. Antibacterial activity and zone of inhibition (mm) shown by synthesized AgNP from extracts of leaves against *Staphylooccus aureus* (gram positive) using agar well diffusion method.

	Different Concentration (Zone of inhibition in mm)			
	50µg/ml	75µg/ml	100µg/ml	Streptomycin
Alpinia calcarata	-	-	3.4 ± 0.2	8.7 ± 0.3
Melia dubia	6 ± 0.2	8.1 ± 0.3	9.2 ± 0.1	22.7 ± 0.2
Solanum nigrum	-	-	0.8± 0.09	1.7±0.2

Table 2. Antibacterial activity and zone of inhibition (mm) shown by synthesized AgNP from extracts of leaves against *Klebsiella pneumonia* (gram negative) using agar well diffusion method.

	Different Concentration (Zone of inhibition in mm)			
	50µg/ml	75µg/ml	100µg/ml	Streptomycin
Alpinia calcarata	12.7 ± 0.2	14.8 ± 1.1	16.7 ± 0.2	19.7 ± 0.1
Melia dubia	-	-	1.1 ± 0.3	1.8 ± 0.2
Solanum nigrum	-	0.5 ± 0.1	1.2 ± 0.4	2 ± 0.2

in which zone of inhibition was observed and diameter of inhibition zones around each well with silver nanoparticles was recorded (Table.1 and 2, Figure 6 and 7). Silver nanoparticles from *Alpinia calcarata* revealed highest antibacterial activity against *Klebsiella pneumonia* (16.7 mm) at 100 ìg / ml and lowest antibacterial activity (12.7 mm) at 50 ìg / ml concentrations (Table. 2, Figure 7). They showed lowest antibacterial activity against *Staphylococcus aureus* (3.4 mm) at 100 ìg / ml concentration (Table. 1, Figure 6). Leaf extract of *Melia dubia* showed higher antibacterial activity against *Staphylococcus aureus* (9.2 mm) at 100 ìg / ml concentrations and lowest antibacterial activity (6.0 mm) at 50 ìg / ml concentrations (Table1, Figure 6). At the same time, lowest activity was seen against *Klebsiella pneumonia* (1.1 mm) at 100 ìg / ml concentration (Table. 2, Figure 7). Likewise, *Solanum nigrum* revealed lowest antibacterial activity of all against *Klebsiella pneumonia* (0.9 mm) and against *Staphylococcus aureus* (0.8 mm) at 100 ìg / ml concentration (Table. 1 and 2, Figure 6 and 7). In several studies, *Staphylococcus aureus* and *Klebsiella pneumonia* have been used as models to prove that silver nanoparticles could be used as an antimicrobial agent (Prabhu and Poulose, 2012; Doudi *et al*., 2013; Quinteros *et al*., 2016; Rai *et al*., 2012). Likewise many researchers have evaluated the antibacterial efficacy of silver nanoparticles using different leaf extracts. Biosynthesized silver nanoparticles using *Melia azedarach L.* leaf extract were subjected to antibacterial activity against *K. pneumonia, S. aureus, P. aeruginosa* and *Proteus* species at different (10μg / ml, 5μg / ml, 2.5μg / ml) concentrations (Mehmood *et al*., 2013). Highest activity of silver nanoparticles (10μg / ml) was observed against *S. aureus* (11.67± 0.33 mm) and *K. pneumonia* (11.33 ±0.33 mm). Also the anti bacterial activity was assessed on human pathogenic *Bacillus cereus, Escherchia coli, Klebsiella pneumonia* and *Staphylococcus aureus* using silver nanoparticles synthesized by *Ficus microcarpa* which showed zone of inhibition ranging from 10–14 mm at 10, 20, 30, 40 μl / well concentration (Praba *et al*., 2015). The antimicrobial activity of the synthesized nanoparticles using *Andrographis paniculata* (Padamata Sai Sudhakar *et al*., 2014) was tested against four different pathogenic organism *B. Subtilis, E.coli, P. aeruginosa, S. aureus* and *A.niger* in which *P. aeruginosa* exhibited the highest sensitivity to nanoparticles (10 mm) at 1000 μg / ml while *B. Subtilis* was the least sensitive. Antibacterial activity of biogenic silver nanoparticles which showed maximum inhibition zone 12 mm against *E.coli,* and *S. aureus*, *K. pneumoniae* and *Enterococcus faecalis* showed zone of inhibition of 7, 9 and 6 mm respectively (Mubayi *et al*., 2012). Antibacterial effect was size and dose dependent and was more pronounced against Gram negative bacteria than Gram positive bacteria (Singh *et al*., 2008). Silver nanoparticles are highly antimicrobial to several species of bacteria (Ayala-Núñez *et al*., 2009). *Melia dubia* showed highest antibacterial activity against gram positive bacteria and *Alpinia calcarata* against gram negative bacteria. According to the mechanism reported, silver nanoparticles interact with the outer membrane of bacteria, and arrest the respiration and some other metabolic pathway that leads to the death of the bacteria.

CONCLUSION

The present study deals with the synthesis of silver nanoparticles using the rhizome extract of *Alpinia calcarata* and leaf extracts of *Melia dubia* and *Solanum nigrum* with aqueous silver nitrate solution. This approach appears to be cost effective and an alternative to conventional methods of assembling silver nanoparticles. An urgent need in the field of nanotechnology is the development of reliable and eco-friendly processes for the synthesis of metallic nanoparticles. Here, we have reported the green synthesis of silver nanoparticles using the rhizome extract of *Alpinia calcarata* and the leaf extracts of *Melia dubi* and *Solanum nigrum.* This process is quiet fast and low cost. Biologically synthesized silver nanoparticles could be of immense use in medical field for their efficient antibacterial properties. Rhizome extract of *Alpinia calcarata* and the leaf extracts of *Melia dubi* and *Solanum nigrum* produced silver nanoparticles extracellularly and they were highly stable in solution without any impurity. Hence, applications of such eco-friendly silver nanoparticles in bactericidal applications, makes this method potentially exciting for the large-scale synthesis of silver nanoparticles. Moverover synthesized nannoparticles are capable of rendering high antibacterial efficacy and hence have a great potential in the preparation of drugs used against bacterial diseases.

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REFERENCES

Ammara Hassan, Salma Rahman, Farah Deeba, Shahid Mahmud. 2009. Antimicrobial activity of some plant extracts having hepatoprotective effects. *Journal of Medicinal Plants Research*. 3(1):020-023.

Angalaparameswari, S., Mohamed Saleem, T. S., Alagusundaram,M., Ramkanth, S., Thiruvengadarajan. V. S., Gnanaprakash, K., Madhusudhana Chetty, C., Pratheesh, G. 2012. Anti-microbial Activity of Aristolochic Acid from Root of *Aristolochia bracteata*

Retz. *International Journal of Biological and Life Sciences.* 8 (4): 244-247.

Ashok Kumar, D. 2012. Rapid and green synthesis of silver nanoparticles using the leaf extracts of *Parthenium hysterophorus*: A novel biological approach. *International Journal of Research Pharmacy.* 3(1):1169-1173.

Asolkar, L.V., Kakkar, K.K., Chakre, O.J. 1992. Second supplement to glossary of Indian Medicinal Plants with active principles part I (A-K). Publications and informations directorate (CSIR), New Delhi. P. 414.

Awang. K., Lim, C.S., Mohamad, K., Morita, H., Hirasawa, Y., Takeya, K., Thoison, O., Hadi, A.H.A., Ceramicine, A., Walsogyne, A. 2007. Erythrocarpines A-E, new cytotoxic limonoids from *Chisocheton erythrocarpus*. *Bioorg. Med. Chem*. 15:5997-6002. PMid:17576066 https://doi.org/10.1016/j.bmc.2007.05.049

Ayala-Núñez, N.V., Lara Villegas, H.H., Del Carmen Ixtepan Turrent, L. 2009. Silver Nanoparticles Toxicity and Bactericidal Effect Against Methicillin-Resistant *Staphylococcus aureus*: Nanoscale Does Matter. *Nanobiotechnol.* 5(1):2-9. https://doi.org/10.1007/s12030-009-9029-1

Bekkeri swathy. 2014. A Review on Metallic Silver Nanoparticles. *IOSR Journal of Pharmacy.* 4(7): 38-44. https://doi.org/10.9790/3013-0407038044

Benjamin, G. and Bhrathwaj, S. 2011. Biological Synthesis of Silver Nanoparticles from *Allium Cepa* (Onion) & Estimating Its Antibacterial Activity. *International Conference on Bioscience, Biochemistry and Bioinformatics. 35-39.*

Chanthuru, A., Prabhu, M.M., Aysha, O.S., Karthik, R. 2014. Evaluation of leaf and root extracts of *Melia dubia* L.against larvae of *Culex quinquefasciatus* and five important human pathogens. *Biosciences Biotechnology Research Asia*. 11(1):207-210. https://doi.org/10.13005/bbra/1412

Doudi, M., Naghsh, N., Setorki, M. 2013. Comparison of the effects of silver nanoparticles on pathogenic bacteria resistant to beta-lactam antibiotics (ESBLs) as a prokaryote model and Wistar rats as a eukaryote model. *Med. Sci. Monit.Basic. Res.* 19:103–110. PMid:23507904 PMCid:PMC3940702 https://doi.org/10.12659/MSMBR.883835

Elumalai, E.K., Prasad, T.N.V.K., Hemachandran. J., Viviyan Therasa, S., Thirumalai, T., David, E. 2010. Extracellular synthesis of silver nanoparticles using leaves of *Euphorbia hirta* and their antibacterial activities. *J. Pharm. Sci. and Res.* 2(9):549-554.

Endo, T., Kita, M., Shimada, T., Moriguchi, T., Hidaka, T., Matsumoto, R., Hasegawa, S., Omura, M. 2002. Modification of limonoid metabolism in suspension cell culture of *Citrus*. *Plant Biotechnol*. 19:397-403. https://doi.org/10.5511/plantbiotechnology.19.397

Feng Zhang, Xiaolan Wu, Yuyue Chen, and Hong Lin. 2009. Application of Silver Nanoparticles to Cotton Fabric as an Antibacterial Textile Finish. *Fibers and Polymers.* 10(4):496-501. https://doi.org/10.1007/s12221-009-0496-8

Grzegorz Dzido, Piotr Markowski, Anna Ma³achowska-Jutsz, Krystian Prusik, Andrzej, Jarzebski. 2015. Rapid continuous microwave-assisted synthesis of silver nanoparticles to achieve very high productivity and full yield: from mechanistic study to optimal fabrication strategy. *J. Nanopart. Res*. 27(17):1-15. https://doi.org/10.1007/s11051-014-2843-y

Jae Yong Song and Beom Soo Kim. 2009. Rapid biological synthesis of silver nanoparticles using plant leaf extracts. *Bioprocess Biosyst Eng.* 32:79–84. PMid:18438688 https://doi.org/10.1007/s00449-008-0224-6

Jayshree Annamalai, Thangaraju Nallamuthu. 2016. Green synthesis of silver nanoparticles: characterization and determination of antibacterial potency. *Appl Nanosci*. 6:259-265. PMid:26900538 PMCid:PMC4750362 https://doi.org/10.1007/s13204-015-0426-6

Kamyar Shameli, Mansor Bin Ahmad, Seyed Davoud Jazayeri, Parvaneh Shabanzadeh, Parvanh Sangpour, Hossein Jahangirian, Yadollah Gharayebi. 2012. Investigation of antibacterial properties silver nanoparticles prepared via green method. *Chemistry Central Journal*. 6:73-77. PMid:22839208 PMCid:PMC3522570 https://doi.org/10.1186/1752-153X-6-73

Malarkodi, C., Rajeshkumar, S., Paulkumar, K., Gnanajobitha, G., Vanaja, M., Annadurai, G. 2013. Bacterial synthesis of silver nanoparticles by using optimized biomass growth of *Bacillus* sp. *Nanoscience and Nanotechnology*. 3(2):26-32.

Mallikarjuna, K., Narasimha, G., Dillip, G.R., Praveen, B., Shreedhar, B., Sree Lakshmi, C., Reddy, B.V.S., Deva Prasad Raju, B. 2008. Green Synthesis of Silver Nanopartcles Using *Ocimum* Leaf Extract and Their Characterization. Digest *Journal of Nanomaterials and Biostructures*. 6(1):181-186.

Manish Dubey, Seema Bhadauria, B.S., Bhadauria Kushwah, B.S. 2009. Green synthesis of silver nanoparticles from extract of *Eucalyptus hybrid* (Safeda) leaf. *Digest Journal of Nanomaterials and Biostructures*. 4(3):537-543.

Mehmood, A., Murtaza, G., Bhatti, T.M., Kausar, R. 2013. Phyto-mediated synthesis of silver nanoparticles from *Melia azedarach L.* leaf extract: Characterization and antibacterial activity. *Arabian Journal of Chemistry.* doi.org / 10.1016 / j.arabjc.2013.11.046.

Mubayi, A., Chatterji, S., Rai, P.M., Watal, G. 2012. Evidence based green synthesis of nanoparticles. *Adv. Mat. Lett.* 3(6):519-525. https://doi.org/10.5185/amlett.2012.icnano.353

Mukunthan, K.S., Elumalai, E.K., Trupti, N., Patel, V.V., Mody, Rodney Siwale, Ajay Singh. 2011. *Catharanthus roseus*: a natural source for the synthesis of silver nanoparticles. *Asian Pacific Journal of Tropical Biomedicine*. 270-274. https://doi.org/10.1016/S2221-1691(11)60041-5

Murray, P.R., Baron, E.J., Pfaller, M.A., Tenover, F.C., Yolken, H.R. 1995. Manual of Clinical Microbiology, 6th Ed. ASM Press, Washington. P. 15-18.

Nafeesa Khatoon, Razi Ahmad, Meryam Sardar. 2015. Robust and fluorescent silver nanoparticles using Artemisia annua: Biosynthesis, characterization and antibacterial activity. *Biochemical Engineering Journal*. 102:91–97. https://doi.org/10.1016/j.bej.2015.02.019

Nakagawa, H., Duan, H., Takaishi, Y. 2001. Limonoids from *Citrus sudachi*. *Chem. Pharm. Bull.* 49:649-651. PMid:11383626 https://doi.org/10.1248/cpb.49.649

Nithya, R. and Ragunathan, R. 2009. Synthesis of Silver nanoparticles using *Pleurotus sajor caju* and its synthesis of Antimicrobial study. *Digest Journal of Nanomaterials and Biostructures.* 4(4): 623-629.

Padamata Sai Sudhakar, Bala Murali Krishna. K., Sundar, B.S. 2014. Synthesis and Characterization of Silver Nanoparticles using aqueous extract of *Andrographis paniculata* and their Antimicrobial Activities. *The Experiment*. 29(1):1962-1969.

Peter Logeswari, Sivagnanam Silambarasan, Jayanthi Abraham. 2015. Synthesis of silver nanoparticles using plants extract and analysis of their antimicrobial property. *Journal of Saudi Chemical Society*. 19:311-317. https://doi.org/10.1016/j.jscs.2012.04.007

Ponarulselvam, S., Panneerselvam, C., Murugan, K., Aarthi, N., Kalimuthu, K., Thangamani, S. 2012. Synthesis of silver nanoparticles using leaves of Catharanthus roseus Linn. G. Don and their antiplasmodial activities. *Asian Pacific Journal of Tropical Biomedicine*. 574-580. https://doi.org/10.1016/S2221-1691(12)60100-2

Praba, P.S., Vasantha, V.S., Jeyasundari, J., Brightson, Y., Arul Jacob. 2015. Synthesis of plant mediated Silver Nanoparticles using *Ficus microcarpa* and evaluation of their antibacterial activity. *Eur. Chem. Bull.* 4(3):116-120.

Prabhu, S., Poulose, E.K. 2012. Silver nanoparticles: mechanism of antimicrobial action, synthesis, medical applications, and toxicity effects. *Int Nano Lett*. 2(3): 1-10.

Priya Banerjee, Mantosh Satapathy, Aniruddha Mukhopahayay, Papita Das. 2014. Leaf extract mediated green synthesis of silver nanoparticles from widely available Indian plants: synthesis, characterization, antimicrobial property and toxicity analysis. *Bioresources and Bioprocessing*. 1(3):1-10. https://doi.org/10.1186/2228-5326-2-32

Quinteros, M.A., Aiassa Martínez I.M., Dalmasso, P.R., Paez, P.L. 2016. Silver Nanoparticles: Biosynthesis Using an ATCC Reference Strain of *Pseudomonas aeruginosa* and Activity as Broad Spectrum Clinical Antibacterial Agents. *International Journal of Biomaterials.* 1-7. PMid:27340405 PMCid:PMC4906205 https://doi.org/10.1155/2016/5971047

Rai, M.K., Deshmukh, S.D., Ingle, A.P, Gade, A.K. 2012. Silver nanoparticles: the powerful nanoweapon against multidrug-resistant bacteria. *Journal of Applied Microbiology.* 112(5): 841–852. PMid:22324439 https://doi.org/10.1111/j.1365-2672.2012.05253.x

Rajathi, M., Modilal, D., Anandan, R., Sindhu, R., Logeshwari, M.N. 2015. Screening of *Solanum nigrum* for its phytochemical and antimicrobial activity against respiratory tracy pathogens. *International Journal of Pure and Applied*. 3(3):210-215.

Sadowski, Z., Maliszewska, I.H., Grochowalska, B., Polowczyk, I., Kozlecki, T. 2008. Synthesis of silver nanoparticles using microorganisms. *Materials Science-Poland*. 26(2):219-224.

Sandeep Thapa, Rajani Shresth, Anjali Tibrewal, Mukesh Shrestha, Yuvraj, K.C. 2015. Biosynthesis of silver nanoparticles (AgNPs) using different plants and fruits extract and study their antimicrobial activity. *Microbiol. Biotech. Res.* 5(5):32-38.

Savithramma, N., Linga Rao, M., Suvarnalatha Devi. 2011. Evaluation of antibacterial efficacy of biologically synthesized silver nanoparticles using stem barks of *Boswellia ovalifoliolata* Bal. and Henry and *Shorea tumbuggaia* Roxb. *Journal of Biological Sciences*. 11:39-45. https://doi.org/10.3923/jbs.2011.39.45

Shrivastava, S., Bera, T., Roy, A., Singh, G., Ramachandra Rao, P., Dash, D. 2007. Characterization of enhanced antibacterial effect of novel silver nanoparticles. *J. Nanotechnology*. 18:225-103. https://doi.org/10.1088/0957-4484/18/22/225103

Shrivastava, S., Bera, T., Singh, S.K., Singh, G., Ramachandra Rao, P., Dash, D. 2009. Characterization of antiplatelet properties of silver nanoparticles. *ACS Nano*. 3(6):1357-64. PMid:19545167 https://doi.org/10.1021/nn900277t

Singh, M., Singh, S., Prasad, S., Gambhir, I.S. 2008. Nanotechnology in medicine and Antibacterial effect of silver nanoparticles. *Digest Journal of Nanomaterials and Biostructures.* 3(3):115-122.

Sinha, S., Pan, I., Chanda, P., Sen, S.K. 2009. Nanoparticles fabrication using ambient biological resources. *J. Appl. Biosci*. 19:1113-1130.

Sulochana. S., Palaniandi Krishnamoorthy, K., Sivaranjani. 2012. Synthesis of silver nanoparticles using leaf extract of Andrographis paniculata. *Journal of Pharmacology and Toxicology.* 7: 251-258. https://doi.org/10.3923/jpt.2012.251.258

Tiwari, D.K., Behari, J., Sen, P. 2008. Application of Nanoparticles in Waste Water Treatment. *World Applied Sciences Journal*. 3(3):417-433.

Veeraputhiran. 2013. Bio-Catalytic Synthesis of Silver Nanoparticles. *International Journal of ChemTech Research*. 5(5):2555-2562.

Vikram Paritala, Chiruvellab, K.K., Chakradhar Thamminenic, Rama Gopal Ghantad, Arifullah Mohammeda. 2015. Phytochemicals and antimicrobial potentials of mahogany family. *Revista Brasileira de Farmacognosia*. 25:61–83. https://doi.org/10.1016/j.bjp.2014.11.009

Virender, K., Sharma, V.K., Yngard, R.A., Lin, Y. 2009. Silver nanoparticles: Green synthesis and their antimicrobial activities. *Advances in Colloid and Interface Science.* 145(1-2): 83-96. PMid:18945421 https://doi.org/10.1016/j.cis.2008.09.002

Warrier PK, Nambiar VPK and Ramankutty C. Indian medicinal plants. Orient Longman Ltd., Madras.1996

Yinegar, H. and Yewhalaw, D. 2009. Traditional medicinal plant knowledge and use by local healers in Sekoru District, Jimma Zone, South Western Ethiopia. *J. Ethnnobiol. Ethnome.* 3(24):1-7. PMid:17547765 PMCid:PMC1905946 https://doi.org/10.1186/1746-4269-3-24

Zhang, X.F., Liu, Z.G., Shen, W., Gurunathan, S. 2016. Silver Nanoparticles: Synthesis, Characterization, Properties, Applications, and Therapeutic Approaches. *Int. J. Mol. Sci.* 17(9):1534. PMid:27649147 PMCid:PMC5037809 https://doi.org/10.3390/ijms17091534