RESEARCH
www.commprac.com
ISSN 1462 2815

SYNTHESIS, CHARACTERISATION OF ANTIBACTERIAL ACTIVITY AND NANO HERBAL FORMULATION OF *SOLANUM TRILOBATUM* LEAF EXTRACT

S. Rizwana [1], Sangeetha S [2]*, Taniya M [3] and M Sundaram K [4]

[1,2,3,4] Department of Anatomy, Saveetha Dental College and Hospital,
Saveetha Institute of Medical and Technical science (SIMATS),
Saveetha University, Poonamalle High Road, Velappanchavadi, Chennai.
*Corresponding Author Email: sangeethas.sdc@saveetha.com

DOI: 10.5281/zenodo.11408758

Abstract

This study explores the synthesis, characterization, antibacterial activity, and potential nano herbal formulation of *Solanum trilobatum* leaf extract. *Solanum trilobatum*, commonly known as the "thorny nightshade," is recognized for its therapeutic properties in traditional medicine. To synthesize silver nanoparticles, 10 mL of *Solanum trilobatum* extract was combined with 5 g of silver nitrate solution and heated on a hot plate for 3 hours. The resultant solution was oven-dried at 100°C for 5 hours, followed by calcination at 500°C for 3 hours. Characterization of the synthesized nanoparticles was conducted using various analytical techniques. Antibacterial activity was evaluated using the agar well diffusion method. Results demonstrated significant antibacterial properties of the silver nanoparticles derived from the extract. The study highlights the potential of *Solanum trilobatum* leaf extract in developing novel antibacterial agents through nano formulation techniques, which enhance drug delivery, stability, and bioavailability. These findings provide valuable insights into the medicinal applications of *Solanum trilobatum,* contributing to the field of natural product-based medicine and paving the way for future clinical applications.

Keywords: *Solanum Trilobatum*, Antibacterial Activity, Nano Herbal Formulation, Natural Product-Based Medicine, Silver Nanoparticles.

INTRODUCTION

The utilization of natural products in therapy has a long history, offering a rich repository of bioactive compounds with therapeutic potential(Chockalingam, Sasanka et al. 2020, Aware, Patil et al. 2022). Among these, *Solanum trilobatum*, commonly known as "thorny nightshade," has garnered significant attention in traditional medicine(Ketprayoon and Chaicharoenpong 2018). This medicinal herb, prevalent in various traditional medical practices, is renowned for its diverse therapeutic properties.

Historically, *Solanum trilobatum* has been employed to treat a myriad of ailments, including bronchial asthma, liver infections, and certain cancers(Her and Kanjanasilp 2021). Recent advancements have also explored its application in breast cancer treatment and its potential as an antibacterial agent. *Solanum trilobatum* is distinguished by its wide array of bioactive compounds, which contribute to its multifaceted therapeutic benefits. These compounds endow the herb with antibacterial, antifungal, antioxidant, and anticancer properties(Yuvaraj, Sangeetha et al. 2020, Kengne, Feugap et al. 2021).

Additionally, the hepatoprotective attributes of *Solanum trilobatum* are noteworthy, providing protection against liver damage caused by ultraviolet radiation and other harmful agents. Pharmacological research has further elucidated its analgesic, anti-inflammatory, and antioxidant effects, underscoring its potential in modern medicinal applications.

ISSN 1462 2815

One of the critical advancements in the utilization of *Solanum trilobatum* is the synthesis of silver nanoparticles (AgNPs) using its leaf extract(Abdelghany, Al-Rajhi et al. 2018). Silver nanoparticles have become increasingly significant across various industries, including food, medicine, consumer goods, and healthcare, due to their unique physical and chemical properties. These properties encompass high electrical conductivity, optical, electrical, and thermal characteristics, coupled with notable biological activities(Ansari and Malhotra 2022). The incorporation of silver nanoparticles has revolutionized several fields, serving as antibacterial agents, medical device coatings, optical sensors, and components in consumer goods and cosmetics. Their remarkable antibacterial properties have made them indispensable in enhancing the effectiveness of anticancer drugs and other therapeutic agents(Zhou, Qiu et al. 2018). The synthesis of silver nanoparticles using plant extracts, often referred to as green synthesis, is particularly advantageous. This method leverages the natural reducing agents present in plant extracts, which facilitate the formation of nanoparticles without the need for toxic chemicals. Given *Solanum trilobatum*'s robust therapeutic properties, utilizing its extract for the green synthesis of AgNPs is a promising avenue(Ritu, Verma et al. 2023).

This approach not only aligns with sustainable and eco-friendly practices but also enhances the medicinal value of the herb by incorporating the superior antimicrobial properties of silver nanoparticles. Characterization of these synthesized nanoparticles is crucial to understand their size, shape, morphology, crystallinity, and functional groups. Techniques such as X-ray diffraction (XRD), scanning electron microscopy (SEM), transmission electron microscopy (TEM), and Fourier-transform infrared spectroscopy (FTIR) are employed to obtain detailed insights into the nanostructures. Assessing the antibacterial activity of these nanoparticles involves testing against a variety of bacterial strains using methods like agar diffusion assays and broth dilution techniques. These evaluations aim to determine the inhibitory effects of the nano herbal formulation, providing a comparative analysis against conventional *Solanum trilobatum* leaf extracts(Mohanaparameswari, Balachandramohan et al. 2024).

The integration of nanotechnology with herbal medicine, particularly in enhancing antibacterial activity, bioavailability, and overall efficacy, represents a significant advancement. *Solanum trilobatum*'s traditional extracts, when transformed into nano herbal formulations, exhibit improved stability and therapeutic potency(Nasim, Rajeshkumar et al. 2021). This is particularly relevant in the current medical landscape, where antibiotic resistance poses a formidable challenge. Developing nano herbal formulations with potent antibacterial properties can offer alternative solutions to combat resistant bacterial strains.

The primary objective of this study is to evaluate the antibacterial potential of the nano herbal formulation derived from *Solanum trilobatum* leaf extract against clinically significant bacterial strains. By conducting extensive assays and investigations, the study aims to elucidate the minimum inhibitory concentration (MIC) and potential modes of action of the formulation. Additionally, the stability and physicochemical characteristics of the nano herbal formulation will be meticulously analyzed. The findings of this research hold significant promise for addressing the challenges associated with antibiotic resistance, paving the way for novel therapeutic applications of *Solanum trilobatum* in modern medicine(USHANTHIKA and MOHANRAJ 2020, Sharma, Basist et al. 2023).

ISSN 1462 2815

MATERIALS AND METHODS:

- **Biosynthesis of Silver nanoparticles (AgNPs):**

The production of nanoparticles was conducted using silver nitrate at varying concentrations (10, 20, 30, 40, 50 mM, and 1 M) in the preliminary screening phase. The maximum UV absorption was utilized to establish the ideal concentration of silver ions (1M AgNO3). After that, 40 mL of extract, 6 mL of AgNO3 (1M), and 154 mL of deionized water were added to it to create AgNPs. To maximize the production of AgNPs, the mixture was first given a very gentle shake before being placed in a water bath set at 80°C for 70 minutes.

The reaction mixture had a reddish-brown tint after being incubated at 80 °C.The mixture was centrifuged for 10 minutes at 4°C (12,000 rpm), and the supernatant was thrown away. The precipitate was mixed with 5 mL of deionized water and centrifuged once more under the same conditions(Bjornsson 1993). The procedure was carried out multiple times. After the final precipitate was dried for use in additional tests, it was heated to 50°C for two hours in a hot air oven.

- **Characterisation techniques for synthesis of AgNPs:**

Several methods were used to characterize green generated AgNPs from *Solanum trilobatum* leaves. Initially, using a UV-1800 (Spectrum 500 D, Germany) spectrometer, UV-Vis spectra between 200 and 800 nm were used to track the bio-reduction of silver ions. The Perkin-Elmer LS-55 Luminescence Spectrometer was utilized to investigate the functional groups of both plant extract and manufactured AgNPs utilizing FTIR spectroscopy(AlMasoud, Alomar et al. 2020).

Using the KBr pellet technique, the dried powders were characterized within the range of 4000–400 cm− 1. The produced AgNPs' X-ray diffraction (XRD) patterns were captured using a scanning X-ray diffractometer (Bruker D8 Advance, Germany) with Cu Kα radiation (ˆ I») 1.5406 Å, 40 kV tube voltage, and 30 mA tube current.At a scanning rate of 1◦min−1, the intensities were measured at 2θ values ranging from 10 to 80◦, using a position-sensitive detector aperture at 25◦C.

Software for semi-quantitative phase analysis was used to process the diffraction spectrum in order to reduce noise, smooth the data, and pinpoint the peaks. Using a scanning electron microscope, the morphological characteristics of artificially generated AgNPs from Mikania Cordata leaves were examined (SEM). The synthesized AgNPs were scanned in the SEM at a magnification of x 200,000 using a JSM-7610F operating in the 5 to 15 kV range. The dried sample was platinum coated using a JEOL Auto fine coater (JEC-3000FC) prior to imaging in the SEM.

Using an energy dispersive X-ray spectroscopy adaptor connected to the SEM, the EDX spectrum was captured.

- **Anti-microbial study:**

Disk diffusion assay was used to evaluate the antimicrobial activity of plant extract and synthesized AgNPs against two bacteria, including, *Escherichia coli* and *Staphylococcus aureus* Using nutrient broth media (OD600 = 0.05), the test organisms were cultivated for an entire night before being plated on agar plates. Following the addition of the nanoparticle suspension, the disks were incubated for six hours at 4°C. The zone of inhibition was then determined after the plates were incubated for 18 hours at 37°C.

- **Antibacterial activity of AgNPs:**

The well diffusion method was used to test the AgNPs' antibacterial activity against the pathogenic microbes *Staphylococcus aureus* and *Escherichia coli*. The AgNPs were produced using SOLANO leaf extract. To analyze the antibacterial activity, strains of *E. coli* and *Staphylococcus aureus* were utilized. Nutrient agar medium was used to subculture the pure cultures of the microorganisms at 37 °C. With the help of a sterile cotton swab, each strain was uniformly swabbed onto each plate. For optimum bacterial colony growth, the culture was cultured for 72 hours. Using gel puncture, 6 mm-sized wells were created on the nutritional agar medium. Using a micro-pipette, synthesized AgNPs (50µl–10µg) were added to wells. The plates were then incubated at 37 ◦C for 24 hours to observe the various levels of zone of inhibition. The results were compared with DMSO as a negative control and antibiotic chloramphenicol (50µl–20µg) as the standard. The experiment was conducted three times, and all antibacterial analysis data were represented by the Standard Error Mean (SEM) of triplicate trials. ANOVA was used by SPSS version 22 to compare the statistical differences between the two groups, with $p < 0.05$ being considered significant.

RESULTS

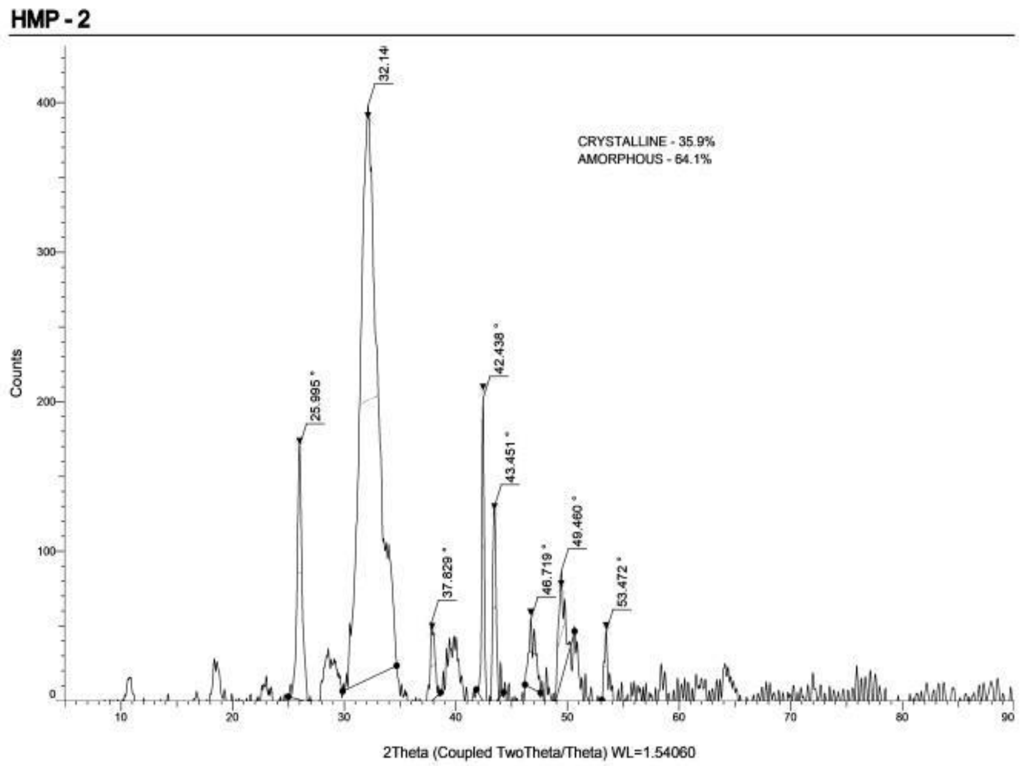

Figure 1: XRD results of silver nanoparticles

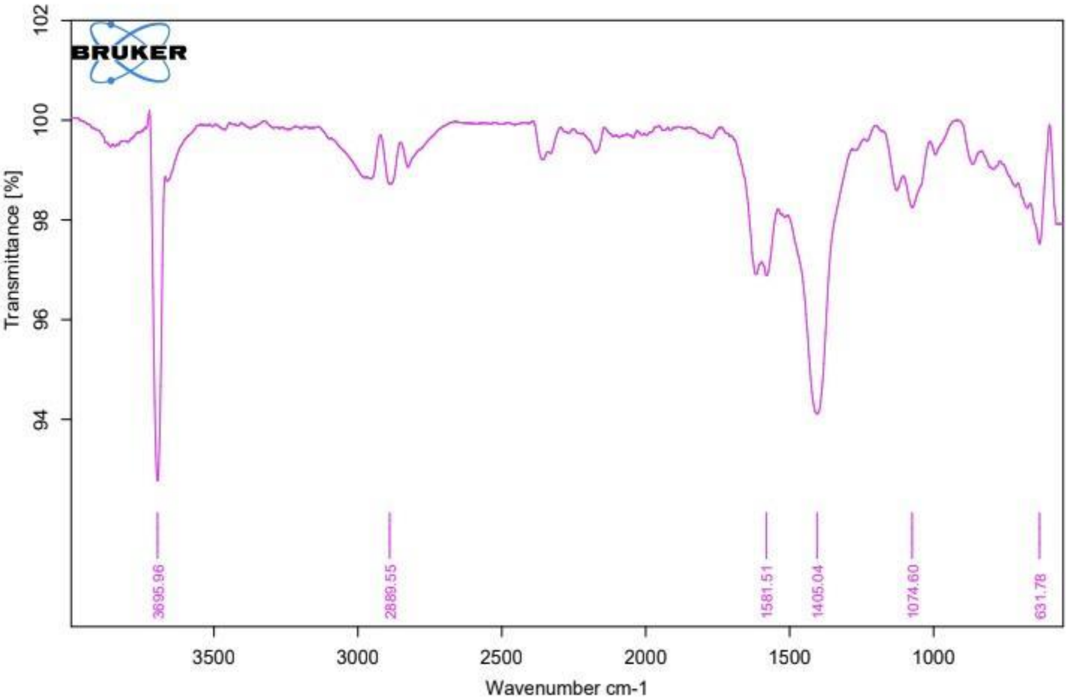

Figure 2: FTIR results of silver nanoparticles

Figure 3

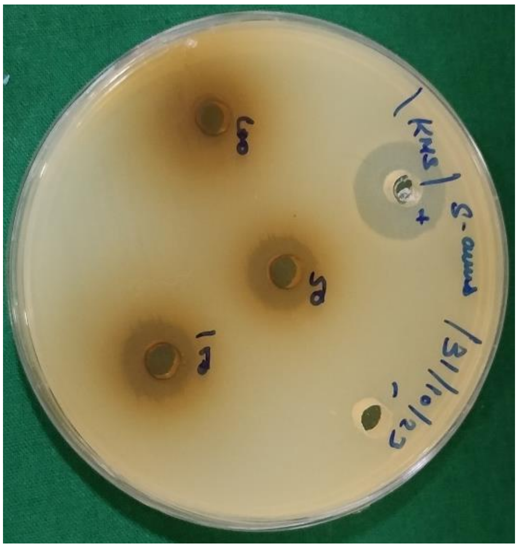

Figure 4

Figure 3 and 4: Microorganisms in petri dish- zone of inhibition

S.No	Micro organisms	Zone of Inhibition			
		50 µL	100 µL	150 µL	200 µL
1	E.coli	-	8 mm	11 mm	18 mm
2	S.aureus	2 mm	13 mm	16 mm	21 mm

Figure 5: Results for zone of inhibition

DISCUSSION

The study of *Solanum trilobatum* leaf extract in nano herbal formulations and its antibacterial activity offers promising insights into natural product-based medicine(Chopra 2023). Previous studies have highlighted the efficacy of plant extracts in combating bacterial strains, and this study reinforces these findings by demonstrating the enhanced antibacterial capabilities of the nano-formulated leaf extract.

The encapsulation of the leaf extract into nanoparticles likely increases its surface area and interaction with bacterial cells, leading to improved antibacterial efficacy. Comparatively, traditional herbal extracts may suffer from limitations such as poor bioavailability and stability(Kumar and Sharma 2018, Mohanraj, Varshini et al. 2021). However, the nano herbal formulation addresses these issues by protecting the active compounds from degradation and enhancing their delivery to the target sites.

The improved antibacterial activity observed against various bacterial strains, including both Gram-positive and Gram-negative bacteria, suggests a broad-spectrum potential of the nano herbal formulation(Krishnamoorthy, Athinarayanan et al. 2018). The possible modes of action of the Solanum trilobatum leaf extract include disruption of bacterial cell walls, inhibition of protein synthesis, and interference with nucleic acid synthesis.

The nano formulation might enhance these effects by facilitating better penetration into bacterial cells and ensuring a sustained release of the active compounds(Omran, Bader et al. 2020). These findings are significant for therapeutic applications, suggesting that nano herbal formulations of Solanum trilobatum could be developed into effective antibacterial agents. Future research could explore the synergistic effects of combining these nano formulations with conventional antibiotics to overcome resistance and improve treatment outcomes(Marunganathan, Kumar et al. 2024, Tayyeb, Priya et al. 2024).

Green Synthesis of Silver Nanoparticles Using *Oxalis griffithii* and *Mikania cordata* Leaf Extracts The green synthesis of silver nanoparticles (AgNPs) using *Oxalis griffithii* and *Mikania cordata* leaf extracts presents a sustainable and eco-friendly approach to nanoparticle production(Magesh, Thadanki et al. 2024). The use of plant extracts as reducing and stabilizing agents not only eliminates the need for hazardous chemicals but also introduces bioactive compounds that can enhance the therapeutic properties of the nanoparticles. Characterization techniques such as UV-Vis spectroscopy, FE-SEM, XRD, EDX, and FT-IR confirmed the successful synthesis and stability of the AgNPs. These techniques revealed that the nanoparticles were predominantly spherical in shape and exhibited significant antibacterial activity.

Specifically, the AgNPs synthesized using Oxalis griffithii demonstrated efficacy against Bacillus subtilis and E. coli, while those synthesized from Mikania cordata showed notable antimicrobial and antioxidant properties. The enhanced bioactivities of these biosynthesized AgNPs can be attributed to the presence of phytochemicals from the plant extracts, which may contribute to additional mechanisms of antibacterial action, such as disruption of cell membrane integrity and generation of reactive oxygen species. The cytotoxic properties of Mikania cordata-derived AgNPs against EAC cells further suggest their potential in cancer therapy, opening new avenues for biomedical applications(Ambika, Manojkumar et al. 2019, Mishra, Kour et al. 2021).

The use of S. trilobatum bark extract for the green synthesis of AgNPs is another significant advancement in the field of nanobiotechnology. The minimal concentration of bark extract required to inhibit microbial growth highlights its potency as a biological reducing agent. The synthesized AgNPs exhibited well-defined characteristics and effective antimicrobial properties, indicating their potential for practical applications in treating infections. The low inhibitory concentration required for microbial suppression underscores the efficiency of the green synthesis method, which is both cost-effective and environmentally friendly(Aditya, Girija et al. 2021, Verma, Chauhan et al. 2024). The incorporation of antioxidant-rich bioactive metabolites from the bark extract into the AgNPs further enhances their therapeutic potential, particularly in scavenging free radicals and protecting cell membranes(Giridharan, Chinnaiah et al. 2024).

The biosynthesized AgNPs also showed significant anti-inflammatory and antioxidant activities, which are crucial for their potential use in medical treatments. The inhibition of COX-1 and COX-2 enzymes by the AgNPs suggests their role in reducing inflammation, positioning them as potential alternatives to conventional anti-inflammatory drugs. The ability of AgNPs to stabilize the HRBC membrane indicates their protective effects on cells, further supporting their use in treating inflammatory conditions. Overall, the current investigation emphasizes the advantages of using plant-based extracts for the green synthesis of nanoparticles. The bioactive compounds present in the extracts not only aid in the synthesis but also impart additional therapeutic properties to the nanoparticles. Future research should focus on optimizing the synthesis process, scaling up production, and conducting detailed in vivo studies to fully understand the potential and safety of these nano-formulations for clinical applications(Kaur, Kakkar et al. 2014).

CONCLUSION

This study demonstrated that silver nanoparticles (AgNPs) have been successfully synthesized using the extract of *Solanum trilobatum* leaves. The structural properties studied by UV–vis, FT-IR, SEM and EDX methods confirmed the formation of more or less spherical shape nanoparticles with an average particle size of 25 nm. The biosynthesized AgNPs exhibited significant bioactivities such as antibacterial and antimicrobial.In addition, the AgNPs formed also showed by inhibiting the growth of EAC cells.Thus, green synthesis of silver nanoparticles (AgNPs) from *Solanum trilobatum* leaves is an eco-friendly approach to form bioactive materials that might be useful for medical applications.

Acknowledgment

We extend our sincere gratitude to the Saveetha Dental College and Hospitals for their constant support and successful completion of this work.

References

1) Abdelghany, T., et al. (2018). "Recent advances in green synthesis of silver nanoparticles and their applications: about future directions. A review." BioNanoScience **8**: 5-16.

2) Aditya, J., et al. (2021). "Genetic alterations in Wnt family of genes and their putative association with head and neck squamous cell carcinoma." Genomics & Informatics **19**(1).

3) AlMasoud, N., et al. (2020). "Multifunctional green silver nanoparticles in pharmaceutical and biomedical applications." Green Chemistry Letters and Reviews **13**(4): 316-327.

4) Ambika, S., et al. (2019). "Biomolecular interaction, anti-cancer and anti-angiogenic properties of cobalt (III) Schiff base complexes." Scientific reports **9**(1): 2721.

5) Ansari, A. A. and B. D. Malhotra (2022). "Current progress in organic–inorganic hetero-nano-interfaces based electrochemical biosensors for healthcare monitoring." Coordination Chemistry Reviews **452**: 214282.

6) Aware, C. B., et al. (2022). "Natural bioactive products as promising therapeutics: A review of natural product-based drug development." South African Journal of Botany **151**: 512-528.

7) Bjornsson, S. (1993). "Simultaneous preparation and quantitation of proteoglycans by precipitation with alcian blue." Analytical biochemistry **210**(2): 282-291.

8) Chockalingam, S., et al. (2020). "Role of Bruxism in Prosthetic Treatments-A Survey." Indian Journal of Forensic Medicine & Toxicology **14**(4).

9) Chopra, H. K. (2023). "Tree bark and their role in nanomaterials synthesis and applications." Secondary Metabolites Based Green Synthesis of Nanomaterials and Their Applications **291**.

10) Giridharan, B., et al. (2024). "Characterization of Novel Antimicrobial Peptides from the Epidermis of Clarias batrachus Catfish." International Journal of Peptide Research and Therapeutics **30**(2): 1-13.

11) Her, L. and J. Kanjanasilp (2021). Efficacy and safety of herbal medicines for relief cough symptom in national list of essential medicines of Thailand and Lao PDR: A systematic review and Meta-analysis, Mahasarakham University.

12) Kaur, I. P., et al. (2014). "Issues and concerns in nanotech product development and its commercialization." Journal of Controlled Release **193**: 51-62.

13) Kengne, I. C., et al. (2021). "Antibacterial, antifungal and antioxidant activities of whole plant chemical constituents of Rumex abyssinicus." BMC Complementary Medicine and Therapies **21**(1): 164.

14) Ketprayoon, T. and C. Chaicharoenpong (2018). Tyrosinase inhibitory activity of some edible plants. Proceeding of the 6th International conference on Biochemistry and Molecular biology (BMB2018).

15) Krishnamoorthy, R., et al. (2018). "Antimicrobial activity of nanoemulsion on drug-resistant bacterial pathogens." Microbial pathogenesis **120**: 85-96.

16) Kumar, R. and M. Sharma (2018). "Herbal nanomedicine interactions to enhance pharmacokinetics, pharmacodynamics, and therapeutic index for better bioavailability and biocompatibility of herbal formulations." Journal of Materials NanoScience **5**(1): 35-60.

17) Magesh, M., et al. (2024). "Sustainable synthesis of Ag-NPs from Cassia auriculata flower extract: synthesis, spectral characterization, its biomedical and environmental applications." Nanotechnology for Environmental Engineering: 1-17.

18) Marunganathan, V., et al. (2024). "Marine-derived κ-carrageenan-coated zinc oxide nanoparticles for targeted drug delivery and apoptosis induction in oral cancer." Molecular biology reports **51**(1): 89.

19) Mishra, J., et al. (2021). "Biofabricated smart-nanosilver: Promising armamentarium for cancer and pathogenic diseases." Colloid and Interface Science Communications **44**: 100459.

20) Mohanaparameswari, S., et al. (2024). "Green Synthesis of Silver Oxide Nanoparticles Using Plectranthus amboinicus and Solanum trilobatum Extracts as an Eco-friendly Approach: Characterization and Antibacterial Properties." Journal of Inorganic and Organometallic Polymers and Materials: 1-21.

21) Mohanraj, K. G., et al. (2021). "Knowledge and Awareness On The Association Between Higher Body Mass Index and Plantar Fasciitis Among Chennain Population-A Survey." Int J Dentistry Oral Sci **8**(03): 1957-1962.

22) Nasim, I., et al. (2021). "Green synthesis of reduced graphene oxide nanoparticles, its characterization and antimicrobial. Properties against common oral pathogens." Int J Dentistry Oral Sci **8**(2): 1670-1675.

23) Omran, Z., et al. (2020). "Evaluation of antimicrobial activity of Triphala constituents and nanoformulation." Evidence-Based Complementary and Alternative Medicine **2020**.

24) Ritu, et al. (2023). "Phytochemical-based synthesis of silver nanoparticle: Mechanism and potential applications." BioNanoScience **13**(3): 1359-1380.

25) Sharma, R., et al. (2023). "Synthesis of Quercetin-Loaded Silver Nanoparticles and Assessing Their Anti-Bacterial Potential." Micromachines **14**(12): 2154.

26) Tayyeb, J. Z., et al. (2024). "Multifunctional curcumin mediated zinc oxide nanoparticle enhancing biofilm inhibition and targeting apoptotic specific pathway in oral squamous carcinoma cells." Molecular biology reports **51**(1): 1-14.

27) USHANTHIKA, T. and K. G. MOHANRAJ (2020). "Knowledge, Awareness and Risk Factors of Smartphone Addiction in Young Adult Population." International Journal of Pharmaceutical Research (09752366) **12**(3).

28) Verma, C., et al. (2024). "Principles and theories of green chemistry for corrosion science and engineering: design and application." Green Chemistry **26**(8): 4270-4357.

29) Yuvaraj, A., et al. (2020). "Perception of general public regarding pros and cons of technological advancements during lockdown due to covid 19 pandemic-an online survey." International Journal of Current Research and Review: 227-233.

30) Zhou, L., et al. (2018). "Self-assembled nanomedicines for anticancer and antibacterial applications." Advanced Healthcare Materials **7**(20): 1800670.