

Green synthesis and antioxidant activity of silver nanoparticles synthesized using *Ficus benghalensis*

ABSTRACT

Background: Nanoparticles are materials with overall dimensions in the nanoscale, ie, under 100 nm. Silver nanoparticles (AgNPs) have shown excellent bactericidal properties against a wide range of microorganisms. *Ficus benghalensis* is a very large tree and had been considered as effective, economical and safe treatments for curing various diseases.

Aim: The present study is aimed to evaluate the antioxidant activity of AgNPs synthesized using *F. benghalensis*.

Materials and Methods: Bio-mediated synthesis of metal oxide nanoparticles using *F. benghalensis* is a promising alternative to traditional chemical synthesis. The antioxidant activity of AgNPs was synthesized using DPPH radical scavenging assay. 1 gm of *F. benghalensis* mixed with 100 mL of distilled water and boiled in 60-70 degree celsius in the heating mantle for 10-15 minutes. Add filtered using Whatman no. 1 filter paper. 20 milli molar (0.574g) of silver was dissolved in 60mL of distilled water. 40 mL of filtered *F. benghalensis* extract is mixed with 60 mL of silver nanoparticles.

Results: The activity of *F. benghalensis* extract was compared with standard ascorbic acid by measuring absorption intensity in the spectrophotometer at the wavelength of 517 nm. While increasing the concentration (10 μ L, 20 μ L, 30 μ L, 40 μ L, 50 μ L) of *F. benghalensis* extracts, the percentage of inhibition of DPPH also increased.

Conclusion: In this study, a simple, biological and low-cost approach was done for the preparation of silver nanoparticles using *F. benghalensis*. Thus the synthesized *F. benghalensis* mediated silver nanoparticles can be subjected to the various other biological activities such as antioxidant, antibacterial, antifungal and cytotoxic evaluation to know the efficiency of these nanoparticles.

Keywords: Antioxidant; *Ficus benghalensis*; green synthesis; innovative technology; silver nanoparticles

1. INTRODUCTION

Nanoparticles show novel properties which depend on their size, shape and morphology which enable them to interact with plants, animals and microbes(1). Silver nanoparticles (AgNPs) have been proven to have bactericidal properties against a wide range of bacteria (2). They are prepared from different perspectives, often to study their morphology or physical characteristics(3). Biosynthesis of AgNPs involves bacteria, fungi, yeast, and plant extracts(4)(5). Recently, a number of parts of plants such as flowers, leaves and fruits, besides enzymes, have been used for the synthesis of gold and silver nanoparticles (6). The size, morphology and stability of nanoparticles depend on the method of preparation, nature of solvent, concentration, strength of reducing agent and temperature(7). Antioxidant compounds in food play an important role as a health protecting factor(8). Scientific evidence suggests that antioxidants reduce the risk for chronic diseases including cancer and heart disease (9). The potentially important derivatives of oxygen, endorsed as ROS (Reactive Oxygen Species) such as O₂, H₂O₂ and OH radicals are generated within the human body, if there is ROS overproduction the equilibrium is hindered favoring the ROS gain that leads to oxidative stress(10). The ROS readily attack and induce oxidative damage to various biomolecules like proteins, lipids, lipoproteins and DNA (11)(12). This oxidative damage can eventually lead to diabetes mellitus, cancer, atherosclerosis, arthritis, and neurodegenerative diseases. Recently additional interest has been shown in the field of free radical

biology, to avoid the causes of chronic diseases(13). Epidemiological studies have shown that the intake of antioxidants such as Vitamin- C (ascorbic acid) can reduce the risk of coronary heart disease and cancer(14). The use of synthetic antioxidants such as butylated hydroxytoluene, butylated hydroxyanisole, tert-butylhydroquinone and propyl gallate has been negatively perceived by consumers because of its safety and health effects(15). Hence, there is an increased interest and demand in search of natural antioxidants from plant sources. It is well known that many botanical products possess natural antioxidants with high antioxidant activity and investigations on these were initiated based on their uses in traditional herbal medicines(16). The genus *Ficus* includes 750 species in most tropical and subtropical forests throughout the world. The genus is remarkable for its large variation in the habits of its species. *Ficus benghalensis* is a very large tree distributed throughout India(17). It is commonly known as 'Indian Banyan tree' and is considered the holy tree of India. Recent studies reveal that the herbal preparations of different parts of *F. benghalensis* had been considered as most effective and safe treatments for curing various types of diseases in the Indian traditional system and used as medicine. The hanging roots of *F. benghalensis* have been reported as antidiarrheal agents(18). The fruit extract of *F. benghalensis* has been documented for its antitumor and antibacterial activities(19). The plant is used in folk medicine for respiratory disorders and certain skin diseases(20). The bark of *F. benghalensis* has been traditionally used to cure diabetes mellitus and is also used as oral administration of bark extract showed lowering of blood glucose level and enhancement of serum insulin levels in normoglycemic as well as diabetic rats. Blood sugar lowering and serum insulin raising action was also found in a dimethoxy derivative of leucocyanidin 3-O-beta-galactosyl cellobioside and a dimethoxy ether of leucopelargonidin-3-O-alpha-L-rhamnoside isolated from the bark of *F. benghalensis*(21). Bengalenoside, a glucoside isolated from *F. benghalensis*, also showed hypoglycemic activity in normal and alloxan diabetic rabbits. The antioxidant effect of aqueous extract of the bark of *F. benghalensis* has been evaluated in hypercholesterolemic rabbits(22). Our team has extensive knowledge and research experience that has translated into high-quality publications.(12,23–34),(35–39) (40) (41).The aim of the present study is to evaluate the antioxidant activity of silver nanoparticles synthesized using *F. benghalensis*.

2. MATERIALS & METHODS:

2.1. Preparation of the extract

A sample of 1 gm of *F. benghalensis* was mixed with 100 mL of distilled water and boiled in 60-70 degrees Celsius in the heating mantle for 10-15 minutes. Add filtered using Whatman number 1 filter paper. 20 millimolar (0.574 g) of silver was dissolved in 60 mL of distilled water. 40 mL of filtered *F. benghalensis* extract was mixed with 60 mL of silver nanoparticles.

2.2. Synthesis of silver nanoparticles (AgNPs)

Synthesis of AgNPs is done by a green synthesis method, in which the 1 mL of aqueous filtrate of the prepared extract was taken into 100 mL of Erlenmeyer flask. Then the extract was mixed into silver nitrate (AgNO_3) to make the final volume concentration of 1 mM solution. The reaction mixture was exposed to sunlight irradiation conditions until the colour change arose as shown in figure 1. The reaction mixture of seed extracts and AgNPs was subjected to centrifugation at 8,000xg for 15 min; the resulting pellet was washed three times with deionized water and filtered.

2.3. UV Spectrometric analysis of synthesized nanoparticles

The endpoint of the reaction was evaluated by UV–vis spectroscopy. The biologically reduced brown colour solution mixture was scanned by Shimadzu, Lambda UV mini-1240 instrument operated at a resolution of 1 nm. The UV–visible analysis was performed in the absorption wavelength of 200–700 nm periodically for 1 hr in order to observe rapid reduction AgNPs by the action of faeces Bengalis extracts. The distilled water was used as a blank.

2.4. Antioxidant activity

Antioxidant activity is the ability of bioactive constituents to prevent, inhibit, and protect against oxidation of numerous substrates such as DNA and lipid components in living organisms and food products. Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging method was followed for evaluating the antioxidant activity of synthesized nanoparticles. DPPH is a stable free radical in methanol that accepts an electron or hydrogen radical and transforms into a stable diamagnetic molecule. It is commonly employed as a test subject for antioxidative activities. Five distinct extract concentrations were combined with various DPPH concentrations. By comparing the absorbance to a blank containing only DPPH and solvent, the radical scavenging activity was measured. % inhibition was calculated using the below equation:

$$\% \text{inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample} \times 100}{\text{Absorbance of control}}$$

3. RESULTS AND DISCUSSION

The synthesis of biological nanoparticles represents an alternative for the physical and chemical methods of nanoparticle formation. The majority of researchers focused on the green synthesis of nanoparticles for the formation of metal and oxide nanoparticles. The use of plants for the synthesis of nanoparticles is a rapid, low-cost, eco-friendly option and is safe for human use.

3.1 Visual Identification

In this study, the silver nanoparticles were synthesized using *F. benghalensis* extract. At various intervals of incubation time, colour changes in the reaction mixture were noticed. The silver ions are reduced to silver nanoparticles, which are detected by a change in colour (Figure 2). The colour changed from yellow to reddish brown, indicating that silver nanoparticles had synthesized. With increasing incubation time, the brown colour intensity increased. After 24 hours, a dark brown colour appeared, followed by no colour change, indicating that the synthesis of silver nanoparticles was complete. With respect to this current study, studies with similar results were also found. The development of intense yellowish-brown colour owing to the surface plasmon resonance confirmed the synthesis of the silver nanoparticles(42). After 80 mins there was a significant increase in the brown colour which confirms the growth of the silver nanoparticles(43). Colour changes of the reaction mixture 240 min after the bioreduction process, which was recorded by UV-vis spectrophotometer at 400-700 nm. In particular, absorbance in the range of 500-550 nm has been used as an indicator to confirm the reduction of Ag⁺ to metallic Ag(44). In a previous study, silver nanoparticles synthesized using tulsi leaf extract, the colour change was observed from yellow to black-brown(45).

3.2 UV-vis absorption spectrophotometer activity

The UV-vis spectroscopy showed the excitation of the surface plasmon resonance band peak positioned at the wavelength from 250 nm to 650 nm and was observed at various time intervals. The spectrum clearly demonstrates that the absorbance steadily increases as the incubation time is increased. The UV-vis spectroscopic analysis of silver nanoparticles synthesized from the *F. benghalensis* showed absorbance peak at 450 nm (Figure 3). Similarly, a study showed that UV absorption spectra of the silver nanoparticles from methanolic stem extract of *Gymnema sylvestre* was reported to be 443 nm(46).

3.3 Antioxidant activity of silver nanoparticles using *F. benghalensis*:

Antioxidants are an integral part of plants as secondary metabolites which play an important role as free radical scavengers and by converting highly reactive free radicals into less reactive species(47). Different

studies verify the significant role of antioxidants in the reduction of oxidative stress, which provoke us to determine the antioxidant potential of various extracts of *F. benghalensis* (47,48). The antioxidant capability of samples cannot be assessed by a single assay(48).

Therefore, DPPH free radical scavenging, total reduction power and TAC assays were performed to verify the antioxidant potential. DPPH is the stable free radical and antioxidant potential of crude extract was determined on the basis of scavenging of free radical i.e. DPPH. The principle of the assay is based on the conversion of the purple color of the free radical to the yellow color molecule by accepting a hydrogen electron from donor antioxidants present in samples. In the current study, high percentage free radical scavenging activity was shown by *F. benghalensis*. The activity of *F. benghalensis* extract was compared with standard ascorbic acid by measuring absorption intensity in the spectrophotometer at the wavelength of 517 nm. While increasing the concentration (10 μ L, 20 μ L, 30 μ L, 40 μ L, 50 μ L) of *F. benghalensis* extracts, the percentage of inhibition of DPPH also increased. However, the inhibition percentage is directly proportional to the concentration of extract as shown in Figure 4. Previous research works have reported on the various activities exhibited by the nanoparticles synthesized from natural sources such as antibacterial, antioxidant and cytotoxic activity (49–51) In a study, silver nanoparticles were synthesized from aqueous leaf extract of *Cestrum nocturnum* and its antioxidant and antibacterial activities were tested against bacteria and the results confirmed that the silver nanoparticles have more antioxidant activity as compared to vitamin C.(52) In other study, Antioxidant and antibacterial activity of silver nanoparticles is due to the presence of bioactive molecules on the surface (53).

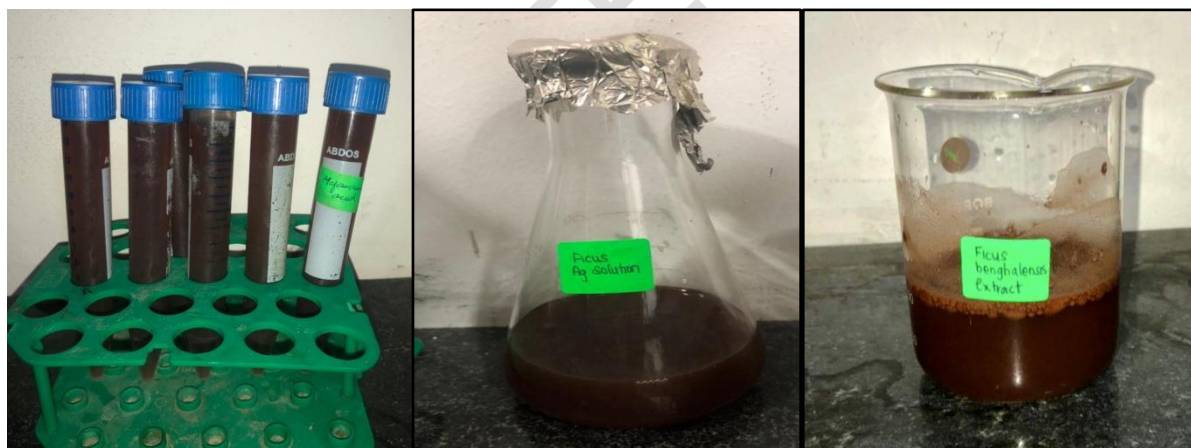


Fig. 1. Synthesis process of silver nanoparticles

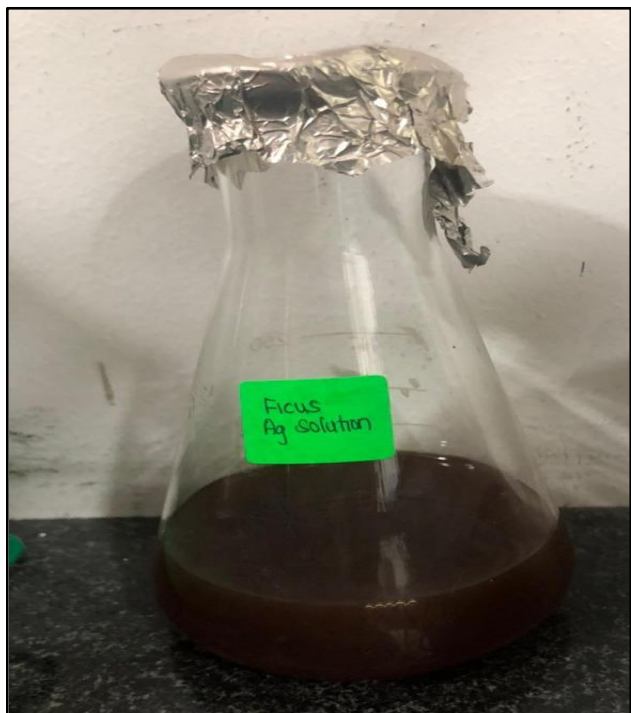


Fig. 2. Reduction of silver ions to silver nanoparticles visually identified by colour changed at various periods of incubation time

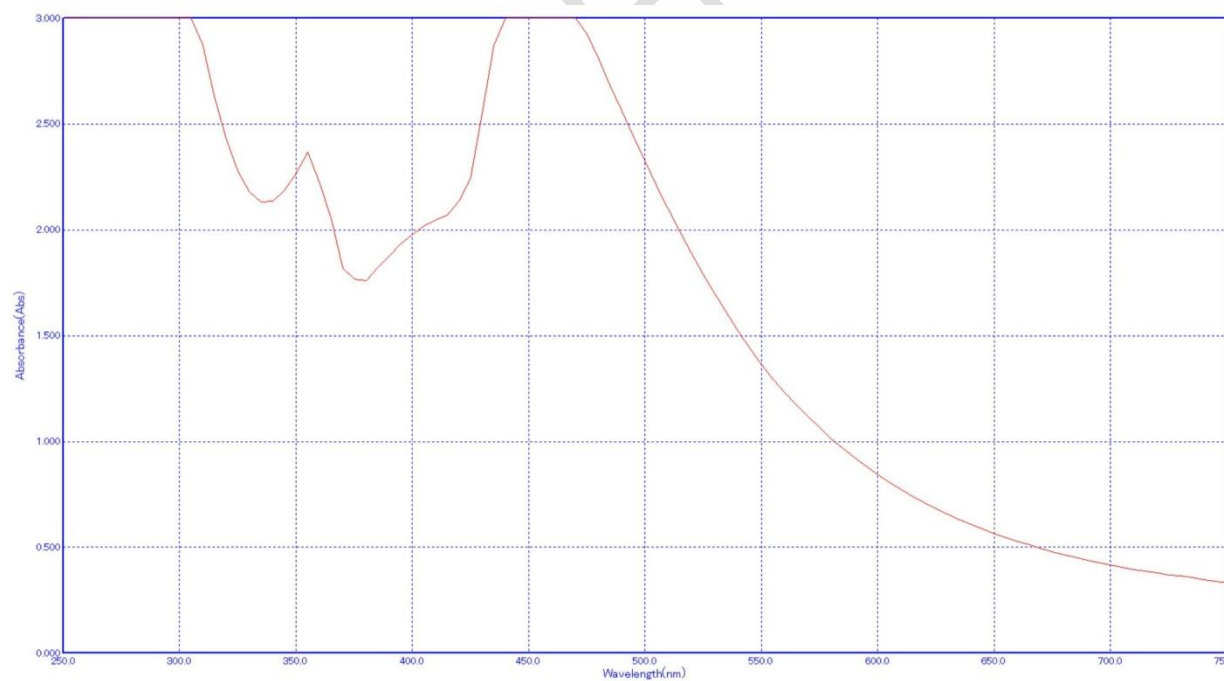


Fig. 3. UV- Vis Spectroscopic analyses of silver nanoparticles synthesized from *F. benghalensis* recorded as function of time

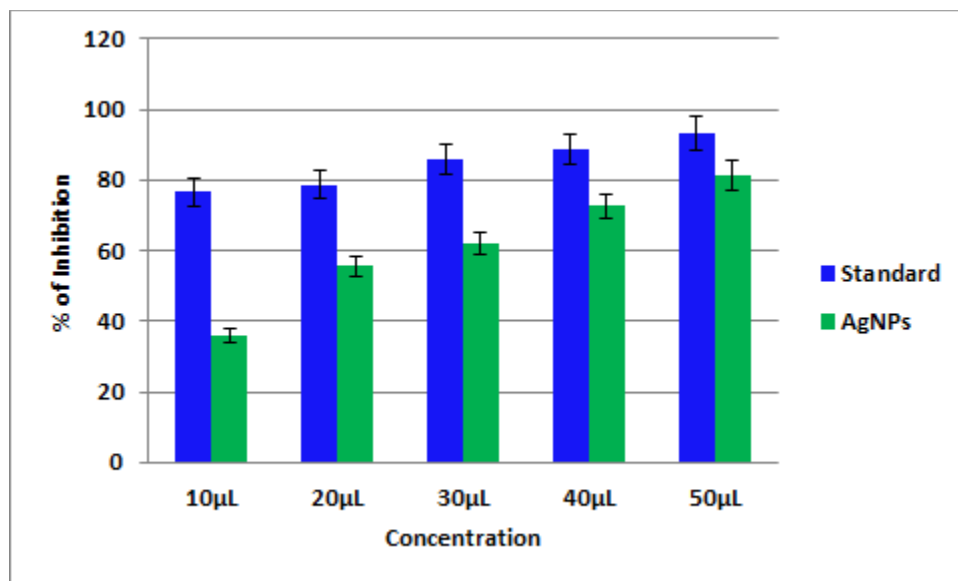


Fig. 4. *F. benghalensis* showed increased DPPH inhibition with increase in concentration of extract. Inhibition was measured after 48 hours of incubation. X- axis shows concentrations in microlitre, Y- axis shows percentage of inhibition.

CONCLUSION

In this study, a simple, biological and low-cost approach was done for the preparation of silver nanoparticles using *F. benghalensis*. Thus the synthesized *F. benghalensis* mediated silver nanoparticles which was shown to have potent antioxidant property can be subjected to the various other biological activities such as antibacterial, antifungal and cytotoxic evaluation to know the efficiency of these nanoparticles. So that they can be developed as a substitute for conventional formulations in the treatment of oral diseases and help in the betterment of life.

CONSENT

It is not applicable

ETHICAL APPROVAL

It is not applicable

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

REFERENCES

1. Shathviha PC, Ezhilarasan D, Rajeshkumar S, Selvaraj J. β -sitosterol Mediated Silver Nanoparticles Induce Cytotoxicity in Human Colon Cancer HT-29 Cells. *Avicenna Journal of Medical Biotechnology*. 2020. Available from: <http://dx.doi.org/10.18502/ajmb.v13i1.4577>
2. Nasim I, Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences, Saveetha University, India. Cytotoxicity and anti-microbial analysis of silver and graphene oxide bio nanoparticles. Vol. 16, *Bioinformation*. 2020. p. 831–6. Available from: <http://dx.doi.org/10.6026/97320630016831>
3. Ramesh A, Varghese S, Jayakumar ND, Malaiappan S. Comparative estimation of sulfiredoxin levels between chronic periodontitis and healthy patients - A case-control study. Vol. 89, *Journal of Periodontology*. 2018. p. 1241–8. Available from: <http://dx.doi.org/10.1002/jper.17-0445>
4. Singh J, Dutta T, Kim K-H, Rawat M, Samddar P, Kumar P. “Green” synthesis of metals and their oxide nanoparticles: applications for environmental remediation. *J Nanobiotechnology*. 2018 Oct 30;16(1):84.
5. Paramasivam A, Priyadharsini JV, Raghunandhakumar S, Elumalai P. A novel COVID-19 and its effects on cardiovascular disease. Vol. 43, *Hypertension Research*. 2020. p. 729–30. Available from: <http://dx.doi.org/10.1038/s41440-020-0461-x>
6. Rajeshkumar S, Sandhiya D. Biomedical Applications of Zinc Oxide Nanoparticles Synthesized Using Eco-friendly Method. *Nanoparticles and their Biomedical Applications*. 2020. p. 65–93. Available from: http://dx.doi.org/10.1007/978-981-15-0391-7_3
7. Ganta SSL, Jeevitha M, Preetha S, Rajeshkumar S. Anti-Inflammatory Activity of Dried Ginger Mediated Iron Nanoparticles. *Journal of Pharmaceutical Research International*. 2020. p. 14–9. Available from: <http://dx.doi.org/10.9734/jpri/2020/v32i2830866>
8. Shunmugam R, Balusamy SR, Kumar V, Menon S, Lakshmi T, Perumalsamy H. Biosynthesis of gold nanoparticles using marine microbe (*Vibrio alginolyticus*) and its anticancer and antioxidant analysis. Vol. 33, *Journal of King Saud University - Science*. 2021. p. 101260. Available from: <http://dx.doi.org/10.1016/j.jksus.2020.101260>
9. Nasim I, Kamath K, Rajeshkumar S. Evaluation of the re-mineralization capacity of a gold nanoparticle-based dental varnish: An in vitro study. Vol. 23, *Journal of Conservative Dentistry*. 2020. p. 390. Available from: http://dx.doi.org/10.4103/jcd.jcd_315_20
10. Rajeshkumar S, Sherif MH, Malarkodi C, Ponnaniakajamideen M, Arasu MV, Al-Dhabi NA, et al.

Cytotoxicity behaviour of response surface model optimized gold nanoparticles by utilizing fucoidan extracted from *Padina tetrastromatica*. Vol. 1228, Journal of Molecular Structure. 2021. p. 129440. Available from: <http://dx.doi.org/10.1016/j.molstruc.2020.129440>

11. Oskam G. Metal oxide nanoparticles: synthesis, characterization and application . Vol. 37, Journal of Sol-Gel Science and Technology. 2006. p. 161–4. Available from: <http://dx.doi.org/10.1007/s10971-005-6621-2>
12. Del Fabbro M, Karanxha L, Panda S, Bucchi C, Nadathur Doraiswamy J, Sankari M, et al. Autologous platelet concentrates for treating periodontal infrabony defects. *Cochrane Database Syst Rev*. 2018 Nov 26;11:CD011423.
13. Paramasivam A, Priyadharsini JV. MitomiRs: new emerging microRNAs in mitochondrial dysfunction and cardiovascular disease . Vol. 43, Hypertension Research. 2020. p. 851–3. Available from: <http://dx.doi.org/10.1038/s41440-020-0423-3>
14. Jayaseelan VP, Arumugam P. Dissecting the theranostic potential of exosomes in autoimmune disorders . Vol. 16, Cellular & Molecular Immunology. 2019. p. 935–6. Available from: <http://dx.doi.org/10.1038/s41423-019-0310-5>
15. Vellappally S, Al Kheraif AA, Divakar DD, Basavarajappa S, Anil S, Fouad H. Tooth implant prosthesis using ultra low power and low cost crystalline carbon bio-tooth sensor with hybridized data acquisition algorithm . Vol. 148, Computer Communications. 2019. p. 176–84. Available from: <http://dx.doi.org/10.1016/j.comcom.2019.09.020>
16. Varghese SS, Ramesh A, Veeraiyan DN. Blended Module-Based Teaching in Biostatistics and Research Methodology: A Retrospective Study with Postgraduate Dental Students . Vol. 83, Journal of Dental Education. 2019. p. 445–50. Available from: <http://dx.doi.org/10.21815/jde.019.054>
17. Vellappally S, Al-Kheraif AA, Anil S, Basavarajappa S, Hassanein AS. Maintaining patient oral health by using a xeno-genetic spiking neural network . *Journal of Ambient Intelligence and Humanized Computing*. 2018. Available from: <http://dx.doi.org/10.1007/s12652-018-1166-8>
18. Sarode SC, Gondivkar S, Sarode GS, Gadail A, Yuwanati M. Hybrid oral potentially malignant disorder: A neglected fact in oral submucous fibrosis . *Oral Oncology*. 2021. p. 105390. Available from: <http://dx.doi.org/10.1016/j.oraloncology.2021.105390>
19. Kavarthapu A, Gurumoorthy K. Linking chronic periodontitis and oral cancer: A review . *Oral Oncology*. 2021. p. 105375. Available from: <http://dx.doi.org/10.1016/j.oraloncology.2021.105375>
20. Website . Available from: Venkatesan J, Singh SK, Anil S, Kim S-K, Shim MS. Preparation, Characterization and Biological Applications of Biosynthesized Silver Nanoparticles with Chitosan-Fucoidan Coating. *Molecules* . 2018 Jun 12;23(6). Available from: <http://dx.doi.org/10.3390/molecules23061429>
21. Venkatesan J, Rekha PD, Anil S, Bhatnagar I, Sudha PN, Dechsakulwatana C, et al. Hydroxyapatite from Cuttlefish Bone: Isolation, Characterizations, and Applications . Vol. 23, Biotechnology and Bioprocess Engineering. 2018. p. 383–93. Available from: <http://dx.doi.org/10.1007/s12257-018-0169-9>
22. Vellappally S, Al Kheraif AA, Anil S, Wahba AA. IoT medical tooth mounted sensor for monitoring teeth and food level using bacterial optimization along with adaptive deep learning neural network . Vol. 135, Measurement. 2019. p. 672–7. Available from: <http://dx.doi.org/10.1016/j.measurement.2018.11.078>

23. Ramesh A, Varghese S, Jayakumar ND, Malaiappan S. Comparative estimation of sulfiredoxin levels between chronic periodontitis and healthy patients - A case-control study. *J Periodontol*. 2018 Oct;89(10):1241–8.
24. Paramasivam A, Priyadharsini JV, Raghunandhakumar S, Elumalai P. A novel COVID-19 and its effects on cardiovascular disease. *Hypertens Res*. 2020 Jul;43(7):729–30.
25. S G, T G, K V, Faleh A A, Sukumaran A, P N S. Development of 3D scaffolds using nanochitosan/silk-fibroin/hyaluronic acid biomaterials for tissue engineering applications. *Int J Biol Macromol*. 2018 Dec;120(Pt A):876–85.
26. Paramasivam A, Vijayashree Priyadharsini J. MitomiRs: new emerging microRNAs in mitochondrial dysfunction and cardiovascular disease. *Hypertens Res*. 2020 Aug;43(8):851–3.
27. Jayaseelan VP, Arumugam P. Dissecting the theranostic potential of exosomes in autoimmune disorders. *Cell Mol Immunol*. 2019 Dec;16(12):935–6.
28. Vellappally S, Al Kheraif AA, Divakar DD, Basavarajappa S, Anil S, Fouad H. Tooth implant prosthesis using ultra low power and low cost crystalline carbon bio-tooth sensor with hybridized data acquisition algorithm. *Comput Commun*. 2019 Dec 15;148:176–84.
29. Vellappally S, Al Kheraif AA, Anil S, Assery MK, Kumar KA, Divakar DD. Analyzing Relationship between Patient and Doctor in Public Dental Health using Particle Memetic Multivariable Logistic Regression Analysis Approach (MLRA2). *J Med Syst*. 2018 Aug 29;42(10):183.
30. Varghese SS, Ramesh A, Veeraiyan DN. Blended Module-Based Teaching in Biostatistics and Research Methodology: A Retrospective Study with Postgraduate Dental Students. *J Dent Educ*. 2019 Apr;83(4):445–50.
31. Venkatesan J, Singh SK, Anil S, Kim S-K, Shim MS. Preparation, Characterization and Biological Applications of Biosynthesized Silver Nanoparticles with Chitosan-Fucoidan Coating. *Molecules* . 2018 Jun 12;23(6). Available from: <http://dx.doi.org/10.3390/molecules23061429>
32. Alsubait SA, Al Ajlan R, Mitwalli H, Aburaisi N, Mahmood A, Muthurangan M, et al. Cytotoxicity of Different Concentrations of Three Root Canal Sealers on Human Mesenchymal Stem Cells. *Biomolecules* . 2018 Aug 1;8(3). Available from: <http://dx.doi.org/10.3390/biom8030068>
33. Venkatesan J, Rekha PD, Anil S, Bhatnagar I, Sudha PN, Dechsakulwatana C, et al. Hydroxyapatite from Cuttlefish Bone: Isolation, Characterizations, and Applications. *Biotechnol Bioprocess Eng*. 2018 Aug 1;23(4):383–93.
34. Vellappally S, Al Kheraif AA, Anil S, Wahba AA. IoT medical tooth mounted sensor for monitoring teeth and food level using bacterial optimization along with adaptive deep learning neural network. *Measurement*. 2019 Mar 1;135:672–7.
35. PradeepKumar AR, Shemesh H, Nivedhitha MS, Hashir MMJ, Arockiam S, Uma Maheswari TN, et al. Diagnosis of Vertical Root Fractures by Cone-beam Computed Tomography in Root-filled Teeth with Confirmation by Direct Visualization: A Systematic Review and Meta-Analysis. *J Endod*. 2021 Aug;47(8):1198–214.
36. R H, Ramani P, Tilakaratne WM, Sukumaran G, Ramasubramanian A, Krishnan RP. Critical appraisal of different triggering pathways for the pathobiology of pemphigus vulgaris-A review. *Oral Dis* . 2021 Jun 21; Available from: <http://dx.doi.org/10.1111/odi.13937>
37. Ezhilarasan D, Lakshmi T, Subha M, Deepak Nallasamy V, Raghunandhakumar S. The ambiguous

role of sirtuins in head and neck squamous cell carcinoma. Oral Dis . 2021 Feb 11; Available from: <http://dx.doi.org/10.1111/odi.13798>

38. Sarode SC, Gondivkar S, Sarode GS, Gadgil A, Yuwanati M. Hybrid oral potentially malignant disorder: A neglected fact in oral submucous fibrosis. Oral Oncol. 2021 Jun 16;105390.
39. Kavarthapu A, Gurumoorthy K. Linking chronic periodontitis and oral cancer: A review. Oral Oncol. 2021 Jun 14;105375.
40. Vellappally S, Abdullah Al-Kheraif A, Anil S, Basavarajappa S, Hassanein AS. Maintaining patient oral health by using a xeno-genetic spiking neural network. J Ambient Intell Humaniz Comput . 2018 Dec 14; Available from: <https://doi.org/10.1007/s12652-018-1166-8>
41. Aldhuwayhi S, Mallineni SK, Sakhamuri S, Thakare AA, Mallineni S, Sajja R, et al. Covid-19 Knowledge and Perceptions Among Dental Specialists: A Cross-Sectional Online Questionnaire Survey. Risk Manag Healthc Policy. 2021 Jul 7;14:2851–61.
42. Madhumitha B, Santhakumar P, Jeevitha M, Rajeshkumar S. Green Synthesis of Selenium Nanoparticle using Capparis decidua fruit extract and its Characterization using Transmission Electron Microscopy And UV- Visible Spectroscopy . Research Journal of Pharmacy and Technology. 2021. p. 2129–32. Available from: <http://dx.doi.org/10.52711/0974-360x.2021.00377>
43. K J, Janani K, Preetha S, Jeevitha, Rajeshkumar S. Green synthesis of Selenium nanoparticles using Capparis decidua and its anti-inflammatory activity . Vol. 11, International Journal of Research in Pharmaceutical Sciences. 2020. p. 6211–5. Available from: <http://dx.doi.org/10.26452/ijrps.v11i4.3298>
44. Menon S, Agarwal H, Venkat Kumar S, Rajeshkumar S. Biomimetic synthesis of selenium nanoparticles and its biomedical applications . Green Synthesis, Characterization and Applications of Nanoparticles. 2019. p. 165–97. Available from: <http://dx.doi.org/10.1016/b978-0-08-102579-6.00008-3>
45. Subha V, Ravindran RSE, Preethi R, Cyril J, Kirubanandan S, Renganathan S. Silver Nanoparticles - Green Synthesis with Aq. Extract of Stems Ipomoea Pes-Caprae, Characterization, Antimicrobial and Anti-Cancer Potential . Vol. 5, International Journal of Medical Nano Research. 2018. Available from: <http://dx.doi.org/10.23937/2378-3664.1410024>
46. Renganathan S, Subha V, Sathya R, Ernest Ravindran RS. Silver nanoparticles from methanolic stem extract of *Gymnema sylvestre* and its characterisation studies. Vol. 7, International Journal of Nano and Biomaterials. 2017. p. 1. Available from: <http://dx.doi.org/10.1504/ijnbm.2017.10010333>
47. Renganathan S, Fatma S, Kalainila P. Green synthesis of copper nanoparticle from passiflora foetida leaf extract and its antibacterial activity. Vol. 10, Asian Journal of Pharmaceutical and Clinical Research. 2017. p. 79. Available from: <http://dx.doi.org/10.22159/ajpcr.2017.v10i4.15744>
48. D H, Hemalatha D, Saraswathi S. Green synthesis of copper oxide nanoparticles using aloe vera extract . Vol. 3, Nanoscale Reports. 2020. Available from: <http://dx.doi.org/10.26524/nr.3.21>
49. Barma MD. Synthesis of Triphala Incorporated Zinc Oxide Nanoparticles and Assessment of its Antimicrobial Activity Against Oral Pathogens : An In-Vitro Study . Vol. 13, Bioscience Biotechnology Research Communications. 2020. p. 74–8. Available from: <http://dx.doi.org/10.21786/bbrc/13.7/14>
50. Mohapatra S, Leelavathi L, I. MA, Adeep KR, S. R. Assessment of Antimicrobial Efficacy of Zinc Oxide Nanoparticles Synthesized Using Clove and Cinnamon Formulation against Oral Pathogens - An In Vitro Study . Vol. 9, Journal of Evolution of Medical and Dental Sciences. 2020. p. 2034–9.

Available from: <http://dx.doi.org/10.14260/jemds/2020/443>

51. Narayanan PM, Wilson WS, Abraham AT, Sevanan M. Synthesis, Characterization, and Antimicrobial Activity of Zinc Oxide Nanoparticles Against Human Pathogens . Vol. 2, BioNanoScience. 2012. p. 329–35. Available from: <http://dx.doi.org/10.1007/s12668-012-0061-6>
52. Keshari AK, Srivastava R, Singh P, Yadav VB, Nath G. Antioxidant and antibacterial activity of silver nanoparticles synthesized by *Cestrum nocturnum* . Vol. 11, Journal of Ayurveda and Integrative Medicine. 2020. p. 37–44. Available from: <http://dx.doi.org/10.1016/j.jaim.2017.11.003>
53. Bhuvaneswari R, Xavier RJ, Arumugam M. Facile synthesis of multifunctional silver nanoparticles using mangrove plant *L.* for its antibacterial, antioxidant and cytotoxic effects. J Parasit Dis. 2017 Mar;41(1):180–7.