RESEARCH ARTICLE | APRIL 21 2023

Improved biosynthesis and characterization of silver nanoparticles using *Laggera crispata (Vahl) Hepper* and *J.R.I* Wood leaves extract

Bulcha Belay Etana; Benny Malengier; M. Dhakshnamoorthy ✉; ... et. al

AIP Conference Proceedings 2743, 020007 (2023)
https://doi.org/10.1063/5.0131853

Improved Biosynthesis and Characterization of Silver Nanoparticles Using *Laggera crispata (Vahl) Hepper* and *J.R.l* Wood Leaves Extract

Bulcha Belay Etana,[1, 2, b)] Benny Malengier,[1] Dhakshnamoorthy M,[2, a)] Wojciech Sitek,[3] and Lieva VanLangenhove[1]

[1]*Department of Materials, Textiles and Chemical Engineering, Ghent University, Ghent, Belgium*
[2]*Faculty of Materials Science and Engineering, Jimma Institute of Technology, Jimma University, Jimma, P.O.Box.378, Oromia, Ethiopia*
[3]*Department of Engineering Materials and Biomaterials, Faculty of Mechanical Engineering, Silesian University of Technology, Konarskiego St. 18 a, 44-100 Gliwice, Poland*

[a)]*Corresponding author: dhakshnamoorthy.mani@ju.edu.et*
[b)]*BulchaBelay.Etana@ugent.be*

Abstract. Biosynthesis of nanoparticles is attractive to researchers because some of the biologically active ingredients in the plants help in green synthesis. In this study, we have developed a single-step, cost-effective, and eco-friendly method for the synthesis of Silver nanoparticles (AgNPs) using the aqueous plant leaves extract of *Laggera crispata (Vahl) Hpper and J.R.l Wood*. AgNPs were biosynthesized using *Laggera crispata (Vahl) Hpper and J.R.l Wood* leaves extract as reducing, capping, and stabilizing agent. The AgNPs began to form just after the addition of aqueous extract of *Laggera* plant to the aqueous solution of Silver nitrate ($AgNO_3$) at room temperature. The biosynthesized AgNPs were characterized by different techniques, such as Fourier Transform Infrared (FTIR) spectroscopy, X-ray Diffraction (XRD), and UV-Visible Spectroscopy. The absorption spectra obtained from UV-Visible spectroscopy showed a sharp Surface Plasmon Resonance (SPR) peak at 451 nm which confirmed the formation of AgNPs. The average particle size of AgNPs was 53 nm as determined by X-ray Diffraction measurement. The FTIR study showed characteristic peaks corresponding to the functional groups that act as capping and stabilizing agent to AgNPs.

INTRODUCTION

In recent years, synthesis of nanomaterials has become an emerging field of research with important role in medicine, drug delivery, and biomedical applications such as bio-imaging, bio-sensing devices, etc. [1–4]. The metal nanoparticles, particularly silver, gold, copper, zinc, magnesium, and titanium have wide range of applications in cosmetics, medicine, and food preservative industries [5–8]. Nowadays, silver nanoparticles (AgNPs) have gained more attention due to their excellent applications in medicine, biomedical, food industry, and smart textile industries [9]. There have been different methods involved in the synthesis of AgNPs, such as wet chemical, sol-gel, hydrothermal, etc. These physical and chemical approaches have many disadvantages, such as toxic waste, high capital cost, toxic by-products which cause environmental pollution. There is another approach of biological method which is eco-friendly and no toxic by-products. The biosynthesis method has been developed and used by many researchers to overcome the limitations of other physical and chemical methods. The biosynthesis of silver nanoparticles (AgNPs) attracted the researchers because of their simple, eco-friendly, increased stability, non-toxicity, and inexpensive method. The biosynthesized AgNPs from plant extracts (leaves, flowers, fruits, peels, seeds) have been widely used

The 5th International Conference on Materials Engineering and Nanotechnology (ICMEN 2021)
AIP Conf. Proc. 2743, 020007-1–020007-7; https://doi.org/10.1063/5.0131853
Published by AIP Publishing. 978-0-7354-4460-7/$30.00

in biomedical fields such as antimicrobial, anti-biofilm, anti-fungal, anticancer, bio-imaging and wound healing [10–12].

The plant extracts contain various phytochemicals which act as reducing agent and capping agent in the synthesis of nanoparticles [13]. This is a bottom-up approach which involves either the reduction or oxidation reaction for the synthesis of AgNPs. The reduction of silver ions into silver metal ($Ag+$ to Ag^0) is mainly due to the presence of phytochemicals such as aldehydes, ketones, amides, and carboxylic acid in the extract. The production capacity and properties of prepared AgNPs are affected by several parameters such as nature, concentration of extract, pH, temperature, etc. [14].

Laggera belongs to a genus of *Asteraceae* family and has mainly three species namely *Laggera alata, Laggera aurita* and *Laggera crispata* distributed in South East Asia and tropical Africa [15]. *Laggera crispata* is a perennial, erect, much branched, strongly aromatic and viscid pubescent herb. The leaves paste of *L. crispata* is used as medicine for the treatment of inflammation and swelling and it also possesses anthelminthic properties. In traditional Chinese medicine, the plant has been used as an anti-inflammatory, antibacterial and anti-leukaemia agent. Recently, great attention has been paid to *Laggera* species because of their diverse chemical components and biological activities [16–18].

Many plant extracts have been used by the researchers from various countries as reducing, capping and stabilizing agent to synthesis silver nanoparticles (AgNPs). However, the literature survey revealed that there are no reports available on *Laggera Crispata (Vahl) Hpper and J.R l Wood* leaves extract in the synthesis of AgNPs. Therefore, in the present study, *Laggera Crispata (Vahl) Hpper and J.R l Wood* leaves extract was used for the biosynthesis of AgNPs.

In earlier studies, Balamurugan et al. synthesized silver nanoparticles by a green method and coated it onto a cotton fabric using ultra sonication processes to test for its antibacterial activity [18]. The modified silver nanoparticles were used with PDDA coating for effective absorbance of the silver nanoparticles on to the cotton fabrics [19–20]. However the application of this coated textile fabric were not stated or mentioned elsewhere in this study. The current research focused on the biosynthesis and characterization of silver nanoparticles (AgNPs). The obtained AgNPs can be utilized in the fabrication of textile based sensors in biomedical applications.

MATERIALS AND METHODS

Materials

In order to obtain biosynthesized AgNPs, fresh leaves of *Laggera crispata (Vahl) Hepper& Wood plant* were collected from Jimma University, Jimma Institute of technology (near work shop center), Jimma, Oromia, Ethiopia. Pure silver nitrate (Sigma Aldrich, 99%), DI water (deionized water, 97%) were acquired from M/s. SIDA Chemical and Equipment suppliers, Addis Ababa, Ethiopia. The analytical grade chemicals were used without further purification. Double distilled deionized water was used for sample preparation.

Preparation of Plant Extract

The fresh green leaves of *Laggera crispata (Vahl) Hepper & Wood plant* (as shown in Figure 1) were washed with water to remove the dust and unwanted particles, and the procedure was repeated for 2–3 times. The cleaned 320g of the leaves were cut into fine pieces and transferred into water bath containing 1000ml distilled water. The mixture was boiled for about 20 min at 60°C and was cooled to room temperature. The acquired leaf extracts were filtered using Whatman filter paper (No. 41) and the collected extract was used in the synthesis of silver nanoparticles as described by U.P. Manik et al. [2].

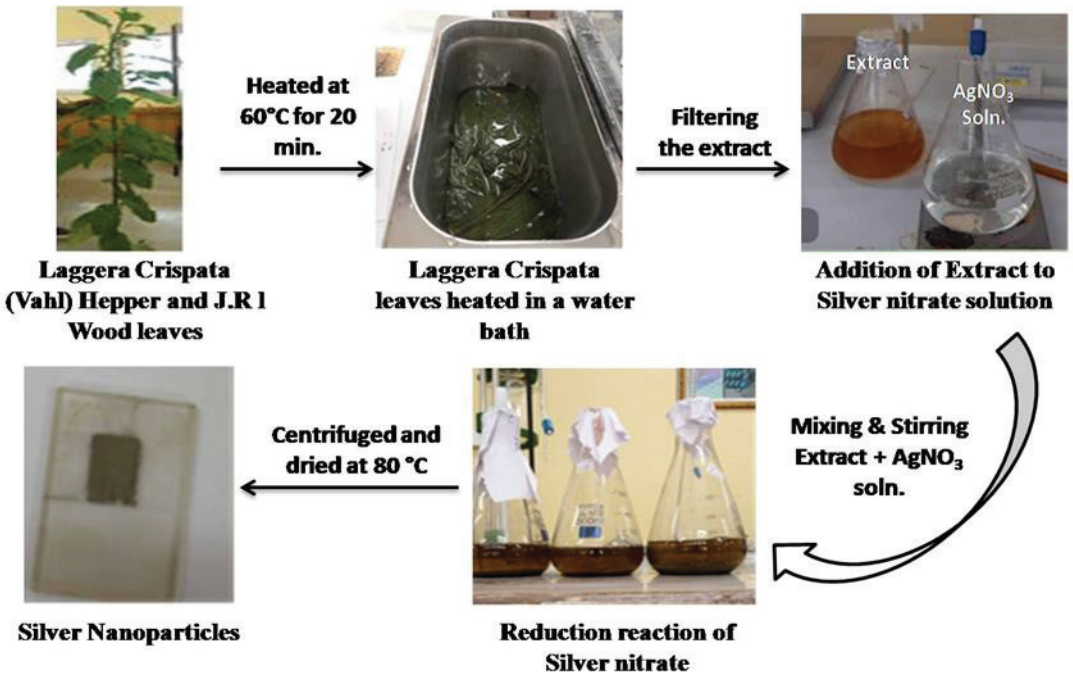

FIGURE 1. Schematic for the green synthesis of Silver nanoparticles (AgNPs)

Green Synthesis of Silver Nano Particle

Silver nanoparticles were obtained through biological reduction of silver nitrate ($AgNO_3$) by the plant extract. In a standard synthesis, the effects of the quantity of leaves extract were analysed. In a 100 ml aqueous solution of $AgNO_3$ (1mM), different volumes of plant extract solution, 100 ml, 200 ml, 300 ml were added (v/v ratio of 1:1, 1:2, 1:3) at room temperature under dark condition (as shown in figure 2).In an alternate experiment, we fixed the plant extract solution volume at 100 ml and added into 100 ml of $AgNO_3$ at different concentrations, 1 mM, 2 mM, and 3 mM separately. In all these cases, we observed an immediate change of the solution colour from light yellowish brown to dark brown. The synthesis was carried out in a 500 ml glass conical flask. Subsequently, the resulting solution was stirred for 20 minutes using a magnetic stirrer (99% rpm, TECNAL TE-0851). Finally, the formed silver nanoparticles were separated by centrifugation at 10000 rpm for 30min in a centrifuge machine (HitachiCF15RN). The precipitate of AgNPs was dried overnight at 80˚C in a vacuum oven.

Characterization

The dried samples of AgNPs were characterized by Fourier transform infrared (FTIR) Spectroscopy (PerkinElmer - Lambda 35) in the wavelength range of 450 - 4000 cm^{-1}. The XRD pattern of dried AgNPs was obtained by a powder X-ray Diffractometer (Bruker d8 Advance, Germany) with the specification of CuKα radiation (λ = 1.5406 Å), 40 kV- 40mA, 2θ/θ scanning mode. The XRD pattern was recorded for the 2θ range of 30° to 80°with a step size of 0.0202. Biosynthesized AgNPs due to the reduction of silver metal ions with aqueous *Laggera crispata (Vahl) Hepper & Wood plant* extract was observed by UV-Visible Spectrometer (Perkin Elmer - Lambda 25) with a resolution of ±1 nm between 300 and 800 nm, and a scanning speed of 200 nm/min.

RESULTS AND DISCUSSIONS

Fourier Transform Infrared (FTIR) spectral measurements were carried out for biosynthesized AgNPs obtained from the reduction of $AgNO_3$ by the addition of *Laggera crispata (Vahl) Hepper& Wood plant* extract with the ratio

of 1:1, 1:2, and 1:3. FTIR spectra were analysed to identify the functional groups of bio-molecules responsible for capping and stabilization of the AgNPs. Figure 2(A) and Figure 2(B) show the FTIR spectra of *Laggera* plant extract and biosynthesized AgNPs respectively.

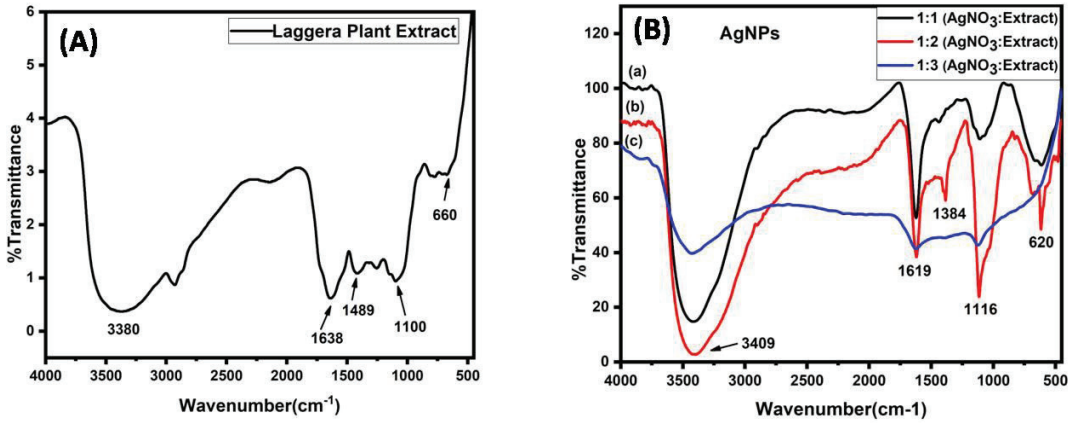

FIGURE 2. FTIR Spectra of (A). *Laggera* plant extract (B). biosynthesized AgNPs from $AgNO_3$ to plant extract ratio of (a). 1:1, (b).1:2, and (c).1:3

FTIR spectrum of *Laggera* plant extract (Figure 2(A)) showed major absorption peaks at 3380, 1638, 1489, 1100, and 660 cm^{-1}. The FTIR spectrum of AgNPs (Figure 2(B)) showed strong absorption bands at 3409, 1619, 1384, 1116, and 620 cm^{-1} which correspond to the functional groups that act as capping agents. A slight shift for the peak at 3380 cm^{-1} to a higher wavelength of 3409 cm^{-1} was observed which indicates the binding of Ag+ ion with O-H group (stretching) of phenolic compound present in the *Laggera* plant extract. The band at 1638 cm^{-1} and 1619 cm^{-1} are assigned to C=C aromatic stretching. The peaks at 1489 and 1384 cm-1 are assigned to -C-O stretching of phenol or tertiary alcohol. The bands at 1100 and 1116 cm^{-1} are attributed to C-N amine stretching and the peaks at 660 and 620 cm^{-1} might be due to the C-H stretching of the aromatic group. The majority of the peaks correspond to the phenolic groups of the polyphenols, tri-terpenoids, alkaloids, steroids, and tannins, adequately present in the plant extract, which help in the formation of capped AgNPs.

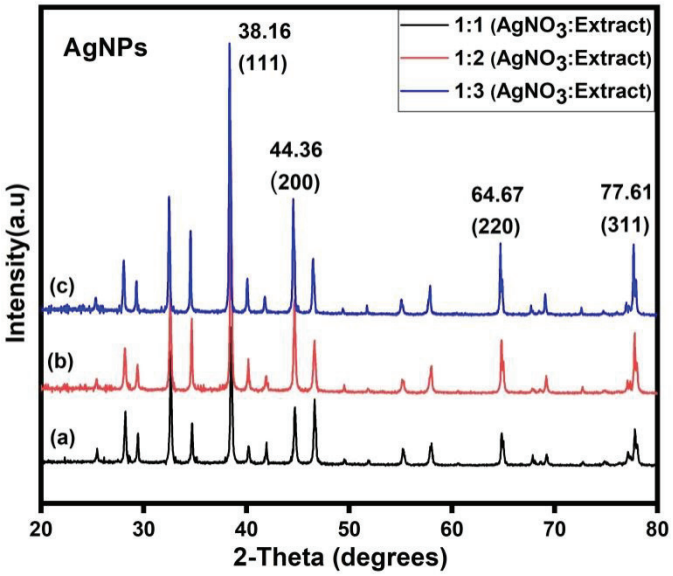

FIGURE 3. XRD of biosynthesized AgNPs from $AgNO_3$ to *Laggera* plant extract ratio of (a). 1:1, (b). 1:2, and (c). 1:3

TABLE 1. XRD pattern results of AgNPs for the ratios (1:1, 1:2, and 1:3) of $AgNO_3$ to *Laggera* plant extract

Position 2θ (°)	Cos θ	FWHM 2θ (°)	d-Spacing (Å)	h, k, l	Particle size (D) nm
Ratio 1:1					
38.16	0.9447	0.1142	2.347	111	76.68
44.36	0.9256	0.0642	2.040	220	42.75
Ratio 1:2					
44.49	0.9256	0.1515	2.034	111	44.42
64.67	0.8445	0.1864	1.440	220	52.69
77.61	0.779	0.1581	1.229	311	67.34
Ratio 1:3					
44.59	0.9256	0.400	2.030	111	42.02
64.79	0.8445	0.1990	1.437	220	49.39

The X-ray diffraction (XRD) pattern of the biosynthesized AgNPs show four diffraction peaks at 2θ values of 38.16, 44.36, 64.67, and 77.61° (Figure 3). These major peaks in the pattern, corresponding to (111), (200), (220), and (311) planes respectively. The XRD study revealed that the resulting particles in the prepared sample are silver nanoparticles having face centered cubic (FCC) crystal structure. There are four additional small peaks in the diffractogram observed at 32.35, 46.38, 54.03 and 57.66°. These peaks might be due to the presence of trace amount $AgNO_3$ which is not fully reduced. The XRD pattern results of AgNPs for the ratios (1:1, 1:2, and 1:3) of $AgNO_3$ to *Laggera* plant extract are shown in Table 1. The particle size (D) of AgNPs are calculated by using Debye-Scherrer formula, $D = 0.9\lambda / \beta \cos\theta$. Where, λ is the wavelength of the X-rays (λ = 1.5406 Å) and β represents the full width at half maximum (FWHM) of a peak. The average of AgNPs synthesized by *Laggera* plant extract is 53 nm. Thus, the XRD analysis proved that the AgNPs with well-defined dimensions could be synthesized by reduction of metal ions using plant extract of *Laggera crispata (Vahl) Hpper and J.R l Wood.*

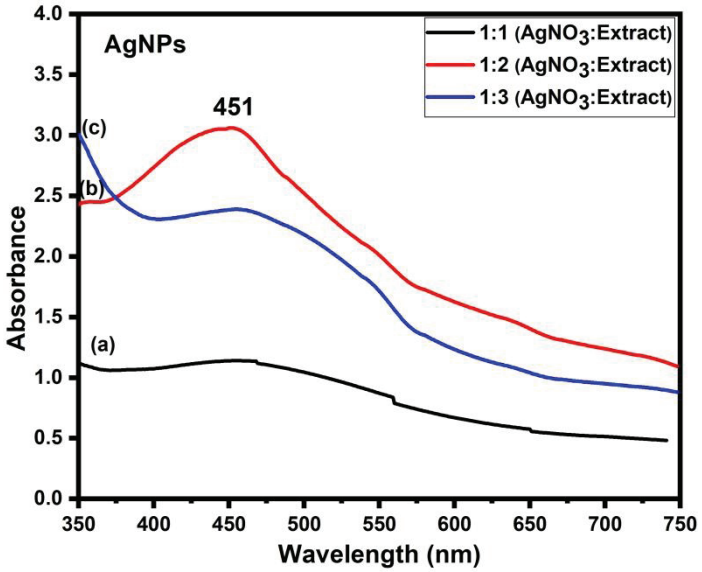

FIGURE 4. UV-Visible spectra of biosynthesized AgNPs from $AgNO_3$ to *Laggera* plant extract ratio of (a). 1:1, (b).1:2, and (c).1:3

Figure 4 shows UV-Visible spectra of biosynthesized AgNPs from $AgNO_3$ to *Laggera Crispata* plant extract ratio of (a). 1:1, (b).1:2, and (c).1:3. The sharp peak was observed at 451 nm which clearly indicates the formation of AgNPs in the solution due to the Surface Plasmon Resonance (SPR) electrons present on the nanoparticle surface. The plasmonic peak is most prominent in case of 1:2 ($AgNO_3$ to plant extract). This corroborates our observation that mixture yields truly nano particles. In this context, we would like to add that plasmonic band of AgNPs are very sensitive to the presence of contaminants. So, we may conclude that AgNPs obtained from 1:2 ratio ($AgNO_3$ to plant extract) may also be applied for analytical method. The 1:3 ratio ($AgNO_3$ to plant extract) mixture also show some plasmonic peak but its intensity is by far less than that observed in case of 1:2 ratio mixture.

CONCLUSION

In conclusion, we have successfully biosynthesized silver nanoparticles (AgNPs) using an aqueous extract of the medicinal plant *Laggera crispata (Vahl) Hpper and J.R.l Wood* leaves. The *Laggera* plant extract acts as a reducing agent for $AgNO_3$. The aqueous solution of $AgNO_3$ was mixed with the plant extract at room temperature and allowed it for overnight to form AgNPs. The formation of AgNPs was confirmed by colour change from light brown/yellow to dark brown and also UV-Visible spectroscopy. The UV-Visible spectrum showed a sharp peak at 451 nm which was due to the presence of surface plasmon resonance (SPR) electrons on AgNPS. The FTIR results found several phytochemicals responsible for the rapid reduction of Ag+ ions, leading to Ag nanoparticles formation. Particularly, hydroxyl groups oxidation of hydrolysate, which likely stimulated the formation of nanoparticles. In XRD data, 2θ positions identify silver crystalline particles having (*hkl*) values, characteristic to face centered cubic (FCC) silver. The average crystalline size was estimated to be 53 nm which confirms that the acquired sample contains nanoparticles. This study of biosynthesized AgNPs using the medicinal plant of *Laggera crispata (Vahl) Hpper and J.R.l* aqueous extract is expected to help in the medical, cosmetic, food, and textile industries.

FUTURE WORK

The authors planned to work on utilizing the biosynthesized AgNPs in smart textiles. The future study of this research will be to develop a textile-based electrode by coating AgNPs/PDMS composite on cotton fabric.

ACKNOWLEDGMENT

The authors would like to thank the Government of Ethiopia (NASCERE Project) for providing the financial support. Also, the authors thank Ghent University, Belgium for the collaborative research facility. The authors would also like to thank, Dr. Prabal Das Gupta, Dr. K. Janarthanan and Dr. Timothy Kwa (JIT, Jimma University) for their valuable suggestions.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

1. M. S. Jabir, Y. M. Saleh, G. M. Sulaiman, N. Y. Yaseen, U. I. Sahib, Y. H. Dewir, M. S. Alwahibi and D. A. Soliman, Nanomaterials **11,** 384 (2021)
2. U. P. Manik, A. Nande, S. Raut and S. J. Dhoble, Results in Materials **6,** 100086 (2020)
3. M. M. Islam Masum, M. M. Siddiqa, K. A. Ali, Y. Zhang, Y. Abdallah, E. Ibrahim, W. Qiu, C. Yan and B. Li, Frontiers in Microbiology **10,** A820 1-18 (2019)
4. R. Bhuvaneswari. J. Xavier1 and M. Arumugam, Journal of Parasitic Disease **41,** 180-187 (2017)
5. S. M. Mousavi, S. A. Hashemi, Y. Ghasemi, A. Atapour, A. M. Amani, A. S. Dashtaki, A. Babapoor and O. Arjmand, Artificial Cells Nanomedical Biotechnology **46,** S855-S872 (2018)
6. N. Krithiga, A. Rajalakshmi, and A. Jayachitra, Journal of Nanoscience, **2015**, 1- 8 (2015)
7. F. Erci, R. C. Koc and I. Isildak, Artificial Cells Nanomedicine Biotechnology **46,** 150-158 (2018)
8. Hemlata, P. R. Meena, A. P. Singh, and K. K. Tejavath, ACS Omega **5,** 5520−5528 (2020)

9. R. Rajan, K. Chandran, S. L. Harper, S. Yun and P. T. Kalaichelvan, Industrial Crops and Products **70,** 356–373 (2015)
10. S. Fahimirad, F. Ajalloueian and M. Ghorbanpour, Ecotoxicology and Environmental Safety **168,** 260–278 (2019)
11. H. F Aritonang, H. Koleangan and A. D. Wuntu, International Journal of Microbiology **2019,** 1-8 (2019)
12. K. K. Bharadwaj, B. Rabha, S. Pati, B. K. Choudhury, T. Sarkar, S. K. Gogoi, N. Kakati, D. Baishya, Z. A. Kari and H. A. Edinur, Nanomaterials **11,** 1999 (2021)
13. M. Khatami, H. Q. Alijani and I. Sharifi, IET Nanobiotechnology, **12** 879–887 (2018)
14. K. Anandalakshmi, J. Venugobal and V. Ramasamy, Applied Nanoscience **6,** 399–408 (2016)
15. R. S. Verma, R. C. Padalia, S. K. Verma, A. Chauhan and M. P. Darokar, Analytical Chemistry Letters **5,** 162–171 (2015)
16. T. Getahun, V. Sharma and N. Gupta, Chemistry & Biodiversity **16,** 1900131 (2019)
17. R. S. Verma, R. C. Padalia, C. S. Chanotiya, A. Chauhan and A. Yadav, Journal of the Serbian Chemical Society **76**, 523–528 (2011)
18. M. Balamurugan, S. Saravanan and T. Soga, Journal of Coating and Technology Research **14,** 735–745 (2017)
19. N. Gokarneshan and K. Velumani, Global Journal of Nanomedicine **2,** 555586 (2017)
20. L. Peng, R. Guo, J. Lan, S. Jiang and X. Wang, Materials Technology **31,** 431–436 (2016)