Asian Journal of Chemistry; Vol. 33, No. 9 (2021), 2049-2052

ASIAN JOURNAL OF CHEMISTRY

https://doi.org/10.14233/ajchem.2021.23295

Single Pot Synthesis of Gellan Gum Coated Silver Nanoparticles and its Antimicrobial Activity

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Received: 9 May 2021; *Accepted*: 12 June 2021; *Published online*: 20 August 2021; AJC-20463

A single pot synthesis of gellan gum coated silver nanoparticles using aniline as a reducing agent has been investigated in present study. The reaction was facile at 80 ºC under reflux condition and the complete reduction of silver ions was noted within 2 h. The resulting gellan gum protected silver nanoparticle was isolated and analyzed with various analytical tools. The antimicrobial activity of the prepared nanocomposite had shown an excellent activity against some selected pathogenic microorganisms.

Keywords: Gellan gum, Silver nanoparticles, Antimicrobial activity.

INTRODUCTION

There is an increasing demand in both academic and industries for many remedial measures have been taken on synthesis of silver nanoparticles. Green synthesis is considered as safe method of synthesizing silver nanoparticles and to maximize the usage of environmental friendly solvents and green chemicals as reducing agent. Silver nanoparticles can be synthesized by various methods including conventional heating [1], microwave method [2], sonochemical method [3], hydrothermal method [4] and thermal pyrolysis method [5]. Many factors involved for synthesis of stable and uniform size nanoparticles including careful selection of capping agent and reaction condition. From small molecules to long chain polymers have been used as stabilizing agent with different chelating functional groups [6,7]. Especially molecules with thiol, dithiocarbamate, sulfide, carboxylic acid and amine terminal functionality are most widely used stabilizing to prevent agglomeration as well as controlling the size and shape of the nanoparticles [8-10]. It is realized that polymers can be used as stabilizing agent because of its low cost and easily dispersed in aqueous medium and nontoxic in nature.

Natural gum has been used as stabilizing agent owing to its availability and water dispersibility. In some cases, gum plays a dual role both reducing agent as well as stabilizing agent depending upon its nature of functional group [11]. In present studies, we have shown that aniline can be used as mild reducing agent for the conversion silver ion into silver nanoparticles under reflux condition in presence of some of natural gum as protecting agent. Recently, various research groups have demon-strated that polyaniline as an important stabilizing for silver nanoparticles for various reasons because of its wider potential applications in various fields including sensor, catalysis, electrocatalysis, anticancer and antimicrobial applications [12-14]. Recent days, polyaniline coated silver nanoparticles have been used for various antimicrobial applications because of its biocompatibility. When the addition of aniline into the silver nitrate containing gum medium, aniline get oxidized and thereby yielded the corresponding aniline oligomer or polyaniline. Because of such redox reaction silver ions reduced into silver nanoparticles and the biopolymer with polyaniline form a stable protection for silver nanoparticles [15-20]. Here, we have chosen gellan gum as stabilizing agent to prepare a stable spherical shaped silver nanoparticle using aniline as a reducing agent under reflux condition. The resulting polymer coated nanoparticles were isolated and then characterized with FT-IR, XRD and SEM techniques. Finally, the antimicrobial activity of the composite was studied with various Gram-negative and Gram-positive microorganisms.

EXPERIMENTAL

Silver nitrite and gellan gum were received from SRL, India. All other solvents used as received from commercial sources.

Instrumental methods: UV-Visible spectral studies were recorded using Shimadzu UV-1800, Japan. FTIR analysis was performed on the samples using the Bruker FT-IR spectrometer in the absorption mode over a range of 4000-400 cm^{-1}. X-ray Diffraction studies were carried out at room temperature on Shimadzu XRD-6000 diffractometer with a graphite filtered Cu*K*α source (0.154 nm), 40 kV, 30 mA. The particle size was analyzed by field emission scanning electron microscopy (FESEM) (FEI Quanta FEG 200).

Experimental procedure: Gellan gum (100 mg) and silver nitrate (15 mg, 5 mM) was dissolved in 20 mL of distilled water followed by addition of 100 µL aniline to above reaction mixture under constant stirring at 80 ºC for 3 h under reflux condition. The resulting nanocomposite was isolated, washed with distilled water and dried.

Antimicrobial studies: Antibacterial activity of gellan gum coated silver nanoparticles was determined by agar well diffusion method using Gram-negative and Gram-positive microorganisms like *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhimurium*, *Klebsiella pneumonia*, *Bacillus cereus*, *Proteus vulgaris* and *Candida albicans*. In this method, pure isolate of each bacterium was subcultured in nutrient broth at 37 ºC for 24 h. About 0.1 % inoculum suspension (10^6 CFU/mL, standardized by 0.5 MacFarland) was swabbed uniformly in sterile Muller-Hinton agar (Hi Media, Mumbai, India). The plates were allowed to dry and a sterile cork borer of diameter 6.0 mm was used to bore wells in the agar plates. Subsequently different concentration of extracts (100 and 25 µg/well) was introduced to wells of the inoculated Muller-Hinton agar plates. Sterile 1 % DMSO served as negative control and ampicillin (10 mcg) was used as a standard. The plates were allowed to stand for 1 h or more for diffusion to take place and then incubated at 37 ºC for 24 h. The zone of observation was recorded to the nearest size in mm [21].

RESULTS AND DISCUSSION

UV-visible spectroscopy analysis: Natural gum coated silver nanoparticles was synthesized by single pot method using aniline as reducing agent. The growth of silver nanoparticles is occurring owing to Oswald repulsion and aggregation of the silver atom during reduction reaction, which was monitored by UV-visible spectral studies. The resulting increasing the absorbance values was measured at different time interval and the resulting increase of absorbance while increasing the growth silver nanoparticles. A stable absorbance value reached after 2 h under reflux condition. Apart from the silver plasmon peak an addition peak was appeared at 280 nm, which is due to unreacted aniline present in the reaction mixture. The plasmon band become flattened and then reached the maximum absorbance value at 2 h of heating. Fig. 1a depicted the UV-visible spectrum silver nanoparticles formed during the reaction and the Fig. 1b represents the different time intervals. A broad silver nanoparticle plasmon band was obtained with a peak position of 480 nm, which indicates a wide distribution of nanoparticles is formed with different sizes. The unreacted aniline and reaction product was then removed by centrifugation and then washing with deionized water.

FT-IR studies: FT-IR spectrum was recorded for the gellan gum with polyaniline composite coated silver nanoparticles and implies that the coexistence of characteristic peaks of PANI as well as gellan gum. The peaks at 3315, 2905, 2748, 1643, 1516, 1409, 1350, 1243, 1134, 1057, 1008, 871, 803, 627 and 548 cm^{-1} are attributed to N-H stretching vibration, aliphatic C-H stretching, C=C stretching of quinoid rings, C=C stretching of benzenoid rings and C-N stretching, respectively. The peak corresponding to the N-Q-N bending vibration of PANI shifted to lower wave number ν_{max} (1057 cm^{-1} from ν_{max} 1134 cm^{-1}) is due to the hydrogen bonding between gellan gum and imine group of the grafted PANI chains. A peak at 803 cm^{-1} is due to C-H out-of-plane bending of 1,4-disubstitutedbenzene ring. Hence, the FTIR spectrum of gellan gum grafted with polyaniline act as a stabilizing agent to protect silver nanoparticles.

XRD analysis: The XRD pattern for the gum coated silver nanoparticles is shown in Fig. 3. The broadening of the XRD peak shape is due to nanocrystallinity of silver and the JCPD pattern is matching with Silver nanoparticles. A peak at 38º is

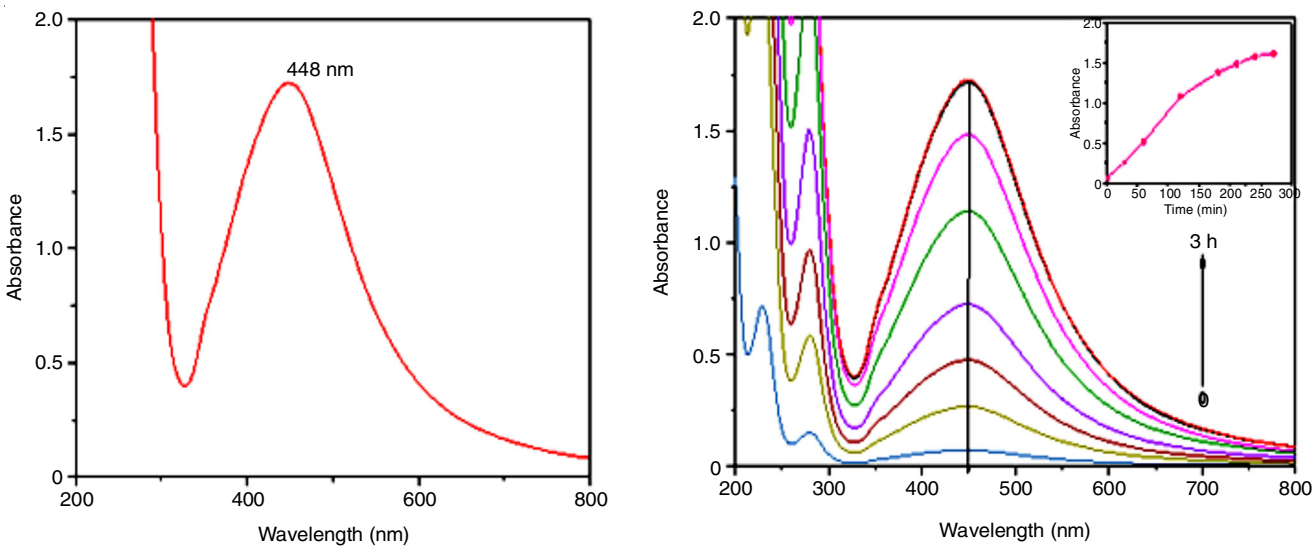

Fig. 1. UV-visible spectroscopy of gellan gum/AgNPs (aniline 100 µL at 80 °C) (a) and their different time interval (b)

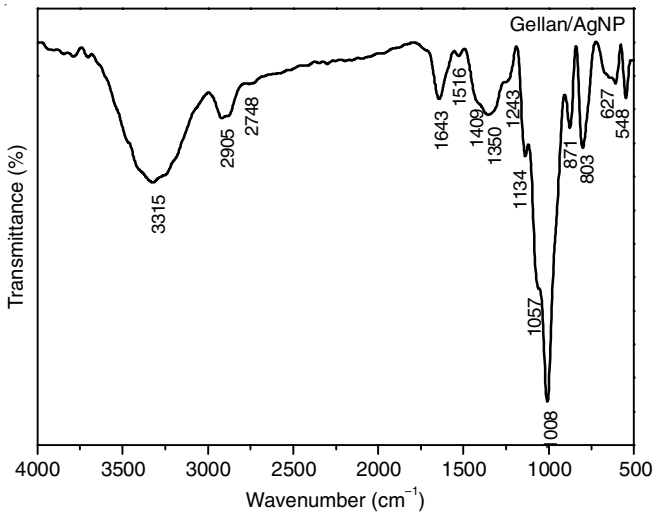

Fig. 2. FT-IR spectrum of gellan gum coated silver nanoparticles

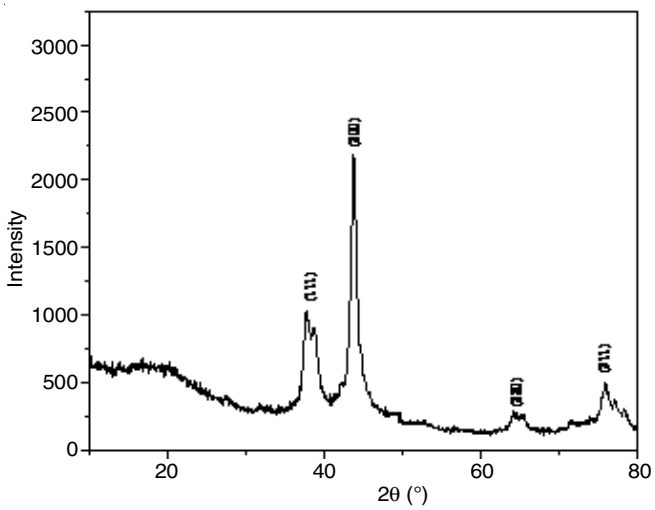

Fig. 3. XRD analysis of gellan gum coated silver nanoparticles

assigned for Ag (111) and a peak at 43º is assigned for Ag(100) and another two less intense peak are corresponding to Ag(222) and Ag(330).

SEM analysis: The SEM images of the gellan gum coated silver nanoparticles are shown in Fig. 4. The bright spotted parts indicate the uniform size distribution of silver nanoparticles

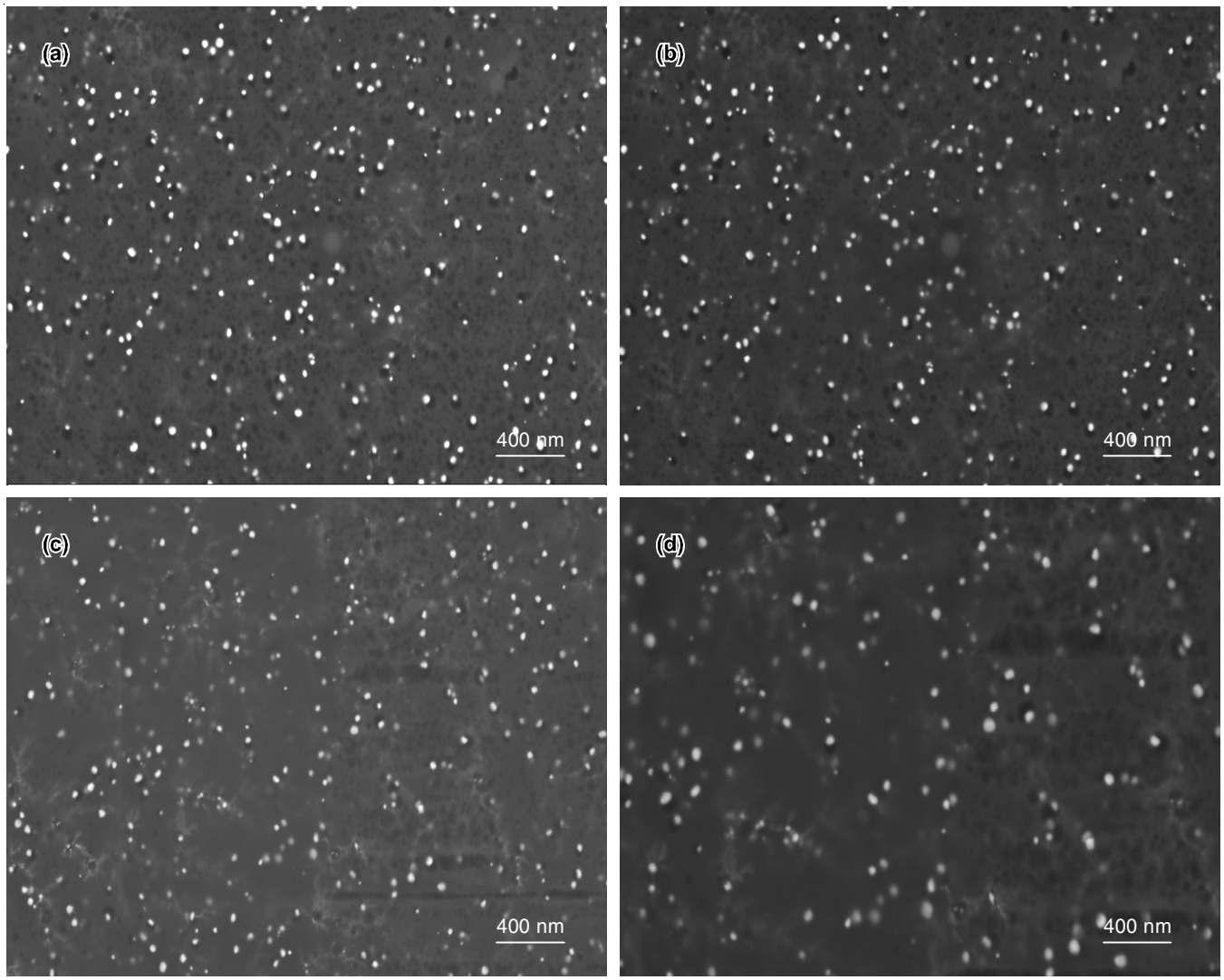

Fig. 4. SEM images of gellan gum coated silver nanoparticles with different magnifications

are shown in different magnificent. The average particle size was found to be 30-40 nm ranges.

Antimicrobial studies: The antimicrobial activity of the gellan gum coated silver nanoparticles was studied against various Gram-negative and Gram-positive microorganisms (Table-1). The antibacterial of silver nanocomposites against Gram-negative and Gram-positive microorganisms *viz*. *E. coli*, *S. aureus*, *S. typhimurium*, *K. pneumonia*, *B. cereus*, *P. vulgaris* and *C. albicans.* Among these *K. pneumonia* and *S. typhimurium* show excellent antimicrobial activity while others are not showing any inhibition zone. From this study, it is proved that gellan gum coated silver nanoparticles have excellent antibacterial activity against some pathogenic microorganism. On the other hand, the antifungal activity of silver nanocomposite was tested and chosen *Candida albicans* as a model system. An excellent antifungal activity was noted for gellan gum coated silver nanoparticles. Thus, the overall studies proved that gellan gum coated silver nanoparticles can be used to some of the pathogenic bacteria and fungal microorganisms.

TABLE-1
MINIMUM INHIBITORY CONCENTRATION OF GELLAN GUM PROTECTED SILVER NANOPARTICLES AGAINST VARIOUS MICROORGANISMS

Organisms	Gellan gum/AgNP		VC	STD
	100 µL	25 µL		
Escherichia coli	–	–	A	R
Staphylococcus aureus	–	–	A	R
Salmonella typhimurium	12	–	A	20
Klebsiella pneumoniae	17	16	A	14
Bacillus cereus	–	–	A	R
Proteus vulgaris	–	–	A	R
Candida albicans	12	–	A	17

VC = Vehicle control (DMSO-75 µL); STD = Standard (Ampicillin 10 mcg); A = Absent; R = Resistance.

Conclusion

A simple, cost-effective and green approach was developed for the synthesis of gellan gum coated silver nanoparticles using aniline as a reducing agent. The synthesized nanoparticles were characterized by UV-Visible spectroscopy, FT-IR, XRD and FE-SEM techniques. The synthesized silver nanocomposites were tested against various Gram-negative and Gram-positive microorganisms. Among the studied microorganisms, *Salmonella typhimurium* and *Klebsiella pneumonia* showed the excellent antimicrobial activity. Moreover, the antifungal activity of the prepared nanocomposite was tested against *Candida albicans* and shows excellent antifungal activity for gellan gum coated silver nanoparticles. From these studies, it is suggested that the present method is simple and greener approach to prepare a gram scale gellan gum coated silver nanoparticles and can be used to treat various bacterial and fungal infections.

ACKNOWLEDGEMENTS

The authors express their sincere thanks to University of Madras and IIT Madras, Chennai, India for providing the instrument facilities. The authors also thank Regional Research Institute of Unani Medicine, Chennai, India to carryout the antimicrobial studies.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

REFERENCES

1. M. Dey, B. Ghosh and T.K. Giri, *Colloids Surf. B Biointerfaces*, **196**, 111341 (2020); https://doi.org/10.1016/j.colsurfb.2020.111341
2. J.S. Karthika, B. Vishalakshi and J. Naik, *Int. J. Biol. Macromol.*, **82**, 61 (2016); https://doi.org/10.1016/j.ijbiomac.2015.10.061
3. S. Dhar, P. Murawala, A. Shiras, V. Pokharkar, A. Shiras and B.L.V. Prasad, *Nanoscale*, **4**, 563 (2012); https://doi.org/10.1039/C1NR10957J
4. P. Dallas, V.K. Sharma and R. Zboril, *Adv. Colloid Interface Sci.*, **166**, 119 (2011); https://doi.org/10.1016/j.cis.2011.05.008
5. R. Zhang, W. Lin, K. Moon and C.P. Wong, *ACS Appl. Mater. Interfaces*, **2**, 2637 (2010); https://doi.org/10.1021/am100456m
6. S. Dhar, E.M. Reddy, A. Shiras, V. Pokharkar and B.L.V. Prasad, *Chem. Eur. J.*, **14**, 10244 (2008); https://doi.org/10.1002/chem.200801093
7. P.V. Asha Rani, G.L.K. Mun, M.P. Hande and S. Valiyaveettil, *ACS Nano*, **3**, 279 (2009); https://doi.org/10.1021/nn800596w
8. J. Jain, S. Arora, J.M. Rajwade, P. Omray, S. Khandelwal and K.M. Paknikar, *Mol. Pharm.*, **6**, 1388 (2009); https://doi.org/10.1021/mp900056g
9. A. Panacek, L. Kvitek, R. Prucek, M. Koláø, R. Veèeøová, N. Pizúrová, V.K. Sharma, T. Nevìèná and R. Zboøil, *J. Phys. Chem. B*, **110**, 16248 (2006); https://doi.org/10.1021/jp063826h
10. P. Velusamy, C.-H. Su, G.V. Kumar, S. Adhikary, K. Pandian, S.C.B. Gopinath, Y. Chen and P. Anbu, *Plos one*, **11**, e0157612 (2006); https://doi.org/10.1371/journal.pone.0157612
11. S. Pandey, G.K. Goswami and K.K. Nanda, *Int. J. Biol. Macromol.*, **51**, 583 (2012); https://doi.org/10.1016/j.ijbiomac.2012.06.033
12. I. Sondi and B. Salopek-Sondi, *J Colloid Interface*, **275**, 177 (2004); https://doi.org/10.1016/j.jcis.2004.02.012
13. A. Hebeish, M. Hashem, M. El-Hady and S. Sharaf, *Carbohydr. Polym.*, **92**, 407 (2013); https://doi.org/10.1016/j.carbpol.2012.08.094
14. C.H. Su, P. Velusamy, G.V. Kumar, K. Pandian and P. Anbu, *J. Mol. Struct.*, **1128**, 718 (2017); https://doi.org/10.1016/j.molstruc.2016.09.045
15. V.K. Sharma, R.A. Yngard and Y. Lin, *Adv. Colloid Interface Sci.*, **145**, 83 (2009); https://doi.org/10.1016/j.cis.2008.09.002
16. M. Rai, A. Yadav and A. Gade, *Biotechnol. Adv.*, **27**, 76 (2009); https://doi.org/10.1016/j.biotechadv.2008.09.002
17. M. Tsuji, M. Hashimoto, Y. Nishizawa, M. Kubokawa and T. Tsuji, *Chem. Eur. J.*, **11**, 440 (2005); https://doi.org/10.1002/chem.200400417
18. F. Fievet, J.P. Lagier and M. Figlarz, *MRS Bull.*, **14**, 29 (1989); https://doi.org/10.1557/S0883769400060930
19. M.N. Nadagouda, T.F. Speth and R.S. Varma, *Acc. Chem. Res.*, **44**, 469 (2011); https://doi.org/10.1021/ar1001457
20. Z. Zhang, W. Shen, J. Xue, Y. Liu, Y. Liu, P. Yan, J. Liu and J. Tang, *Nanoscale Res. Lett.*, **13**, 54 (2018); https://doi.org/10.1186/s11671-018-2450-4
21. M.I. Okeke, C.U. Iroegbu, E.N. Eze, A.S. Okoli and C.O. Esimone, *J. Ethnopharmacol.*, **78**, 119 (2001); https://doi.org/10.1016/S0378-8741(01)00307-5