Research Article

# Synthesis and screening of antibacterial activity of bionanoparticles against *Enterococcus faecalis*

**A. Shamaa Anjum, R. V. Geetha***

**ABSTRACT**

**Aim:** Nanotechnology is one of the most active fields in researches nowadays in modern material science and technology. The present leads to the synthesis of *Aloe vera* silver bionanoparticles and studying its antimicrobial property against *Streptococcus mutans* and *Enterococcus faecalis*. **Materials and Methods:** Siver bio nano particles were synthesized used *Aleo vera* extract and they were characterized using ultra violet – visible spectroscopy. The anti microbial property of the synthesized bio nano particles were screened by agar well diffusion technique and the zone of inhibition were recorded and compared with positive control amoxycillin. **Results:** The antibacterial activity of silver nanoparticles with *A. vera* extract was moderate compared to that of control – amoxicillin. **Conclusion:** Silver bionanoparticles of *A. vera* extract were found to be less effective than the control amoxicillin. Thus, by increasing the concentration of *A. vera* extract in the nanoparticle, the effectiveness of *A. vera* as an antibacterial agent can be assessed in future.

**KEY WORDS:** *Aloe vera*, *Enterococcus faecalis*, Silver bionanoparticles, *Streptococcus mutans*

## INTRODUCTION

Nanotechnology[1] is enabling technology that deals with nanometer-sized objects. Nanodentistry is the application of nanoparticles and dental nanorobots for diagnosis and treatment, improving comprehensive oral health. These nanoparticles are different in chemical and biological reactivity[2] and offer unique physicochemical properties, such as ultra-small sizes, large surface area/mass ratio, and increased chemical reactivity.[3] These advantages are used to design highly specific materials and devices to interact at the subcellular and molecular level of the human body to achieve maximal therapeutic efficacy with minimal side effects.[4]

Enterococci are ubiquitous low GC percent Gram-positive bacteria. The digestive microbiota in humans include the two clinically significant species *E. faecalis* and *E. faecium* and they are also members of the natural microflora of various fermented food products.[5,6] *E. faecalis* is the most common microorganism found in failed/infected root canals of primary and permanent teeth.[7-9] It is very resistant and can survive even in 100–10,000 folds in starvation stage.[7]

Resistance of microorganisms to antibiotics is steadily rising and reports show that a number of recognized antibacterial agents in existence have demonstrated resistance by different species of microorganism.[10] Due to this, synthesis or extraction of compounds such as nanoparticles with antibacterial properties is essential as they have promising applications in the fight against the ever-growing number of antibacterial-resistant pathogenic microorganisms which pose a threat to human and animal health.[11] Enterococci are mainly responsible for bacteremia, urinary tract infections, endocarditis, and wound infections and[12] with *E. faecalis* accounting for nearly 60–80% of all enterococcal infections.[12,13]

*Streptococcus mutans* change the environment of the oral flora, enabling fastidious organisms to colonize and cause the formation of dental plaques.[14] It is normally present in low numbers in the plaque of affected individuals. When salivary flow decreases,

Access this article online

Website: jprsolutions.info | ISSN: 0975-7619

Department of Microbiology, Saveetha Dental College, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai, Tamil Nadu, India

***Corresponding author:** R. V. Geetha, Department of Microbiology, Saveetha Dental College, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai - 600 077, Tamil Nadu, India. Tel.: +91-9710456203. E-mail: rvgeetha2015@gmail.com

Received on: 22-03-2019; Revised on: 27-05-2019; Accepted on: 29-05-2019

the pH of the plaque drops, leading to the selection of aciduric (acid-tolerant) bacteria such as *S. mutans*.[15] This sequence of events indicates that *S. mutans* is involved in the initiation of decay.

The present study is aimed at synthesis and screening of antibacterial activity of bionanoparticles against *E. faecalis* and *S. mutans*.

## MATERIALS AND METHODS

### Test Organisms

The bacterial strains used were *S. mutans and Enterococcus faecalis*. The organisms were obtained from the Department of Microbiology, Saveetha Dental College and Hospitals, Chennai, India.

### Green synthesis of Aleo vera Silver Nanoparticles

About 2 ml stock solution of *Aloe vera* extract was slowly added into 20 ml of 1 mM solution of silver nitrate under continuous stirring for 20 min. The concentration of *A. vera* extract to $AgNO_3$ was maintained at 1:10 ratio to control the shape and size of the nanoparticles. The solution was incubated for 24 h at room temperature. Colorless solution changed into pale yellow color initially and after 24 h, color changed from pale yellow to reddish-brown which indicates the formation of silver nanoparticles. Finally, nanoparticles were lyophilized to obtain a fine powder and characterized by ultraviolet (UV)-visible spectroscopy.

## METHODOLOGY

The bionanoparticles were prepared in the following concentrations in sterile water: 2.5 mg/ml, 5 mg/ml, and 10 mg/ml so that 100 µl of extract of different concentrations delivers 250 µg, 500 µg, and 1000 µg, respectively. The screening of antibacterial activity of the bionanoparticle was carried out using the agar well diffusion method. The bacterial strain was inoculated into nutrient broth and incubated at 37°C overnight. The culture was then adjusted to 0.5 McFarland turbidity standard. Lawn culture of the test organism was made on the Muller-Hinton agar (MHA-HiMedia M1084) plates using sterile cotton swab and the plates were dried for 15 min. A sterile cork borer was then used to make wells (6 mm diameter) for different concentrations of the extracts 100 µl of the varying concentrations (250 µg/ml, 500 µg/ml, and 1000 µg/ml) of the extracts which were introduced into the wells with the help of micropipettes. The culture plates were allowed to stand on the working bench for 30 min for pre-diffusion and were then incubated in upright position at 37°C for 24 h. After 24 h, antibacterial activity was determined by the measurement of diameter of zones of inhibition (mm). Standard antibiotic discs of amoxicillin (30 mcg/disc) were used as positive control. All the tests were done in triplicate to minimize the test error.

## RESULTS

Silver nanoparticles with *A. vera* extract were prepared and evidenced by UV absorption spectra.

| Extract concentration | *Streptococcus mutans* | *Enterococcus faecalis* |
|---|---|---|
| 250 µg/ml | 10 | 9 |
| 500 µg/ml | 14 | 13 |
| 1000 µg/ml | 23 | 19 |
| Control: Amoxicillin | 25 | 23 |

## DISCUSSION

In this *in vitro* study, the antibacterial efficacy of silver nanoparticles with *A. vera* extract and amoxicillin against *S. mutans* and *E. faecalis* was done and the zone of inhibition was compared. The zone of inhibition was found to be increasing as the concentration of the extract increased. The antibacterial activity of silver nanoparticles with *A. vera* extract was moderate compared to that of control – amoxicillin. The antibacterial activity of *A. vera* silver nanoparticles against *S. mutans* was found to be more effective than against *E. faecalis*.

Similar studies were carried out on with the aqueous leaf extract of *Azadirachta indica* by Shakeel Ahmed *et al*., 2016.[16] Studies were also carried out on neem extracts by Kumar *et al*.[17] Similar results were obtained in a study done by Manojkanna *et al*.[4] on neem extract against *E. faecalis* and *Candida albicans*.

## CONCLUSION

Silver nanoparticles of *A. vera* extract against *E. faecalis* and *Streptococcus faecalis* are less effective than amoxicillin. Thus, by increasing the concentration of *A. vera* extract in the nanoparticle, the effectiveness of *A. vera* as an antibacterial agent can be assessed in future.

## REFERENCES

1. Feynman R. There's plenty of room at the bottom. Science 1991;254:1300-1.
2. Kishen AS. Nanotechnology in Endodontics Current and Potential Clinical Applications. Cham, Switzerland: Springer Science+Business Media; 2015.
3. Cohen ML. Nanotubes, nanoscience, and nanotechnology. Mater Sci Eng C 2001;15:1-11.
4. Manojkana K, Chandana CS, Gayathri R, Priya VV, Geetha RV. Synthesis and characterization of silver nano particles from *Plectranthus ambionicus* extract and its antimicrobial activity against *Enterococcus faecalis* and *Candida albicans*. J Pharm Sci Res 2017;9:2423-5.
5. Franz CM, Holzapfel WH, Stiles ME. Enterococci at the crossroads of food safety? Int J Food Microbiol 1999;47:1-24.
6. Giraffa G. Functionality of enterococci in dairy products. Int J

Food Microbiol 2003;88:215-22.
7. Bhardwaj A, Ballal S, Velmurugan N. Comparative evaluation of the antimicrobial activity of natural extracts of *Morinda citrifolia*, papain and *aloe vera* (all in gel formulation), 2% chlorhexidine gel and calcium hydroxide, against *Enterococcus faecalis*: An *in vitro* study. J Conserv Dent 2012;15:293-7.
8. Hugar SM, Mistry LN, Hogade S, Badkar CM. An *in vitro* comparative evaluation of efficacy of desinfective ability of garlic oil (Lasuna), Clove leafe oil (Lavang) and autoclaving on endodontic K files tested against *Enterococcus feaecalis*. Int Ayurvedic Med J 2015;3:2277-84.
9. Bazvand L, Aminozarbian MG, Farhad A, Noormohammadi H, Hasheminia SM, Mobasherizadeh S, *et al.* Antibacterial effect of triantibiotic mixture, chlorhexidine gel, and two natural materials propolis and *Aloe vera* against *Enterococcus faecalis*: An *ex vivo* study. Dent Res J (Isfahan) 2014;11:469-74.
10. Raffi M, Mehrwan S, Bhatti TM, Akhter JI, Hameed A, Yawar W, *et al*. Investigations into the antibacterial behavior of copper nanoparticles against *Escherichia coli*. Ann Microbiol 2010;60:75-80.
11. Usman MS, El Zowalaty ME, Shameli K, Zainuddin N, Salama M, Ibrahim NA, *et al.* Synthesis, characterization, and antimicrobial properties of copper nanoparticles. Int J Nanomedicine 2013;8:4467-79.
12. Bar K, Wisplinghoff H, Wenzel RP, Bearman GM, Edmond MB. Systemic inflammatory response syndrome in adult patients with nosocomial bloodstream infections due to enterococci. BMC Infect Dis 2006;6:145.
13. Peel T, Cheng AC, Spelman T, Huysmans M, Spelman D. Differing risk factors for vancomycin-resistant and vancomycin-sensitive enterococcal bacteraemia. Clin Microbiol Infect 2012;18:388-94.
14. Marsh PD. Microbiology of dental plaque biofilms and their role in oral health and caries. Dent Clin North Am 2010;54:441-54.
15. Marcotte H, Lavoie MC. Oral microbial ecology and the role of salivary immunoglobulin A. Microbiol Mol Biol Rev 1998;62:71-109.
16. Ahmed S, Ahmad SM, Swami BL, Ikram S. Green synthesis of silver nanoparticles using *Azadirachta indica* aqueous leaf extract. J Radiat Res Appl Sci 2016;9:1-7.
17. Kumar NK, Vazhacharickal PJ, Mathew JJ, Joy J. Synthesis of silver nano particles from neem leaf (*Azadirachta indica*) extract and its antibacterial activity. CIB Tech J Biotechnol 2015;4:20-31.

Source of support: Nil; Conflict of interest: None Declared