Int. J.Nano Dimens. 5(4): 337-340, Autumn 2014
ISSN: 2008-8868

Contents list available at **IJND**
International Journal of Nano Dimension
Journal homepage: *www.IJND.ir*

Chemical synthesis of Zinc Oxide (ZnO) nanoparticles and their antibacterial activity against a clinical isolate *Staphylococcus aureus*

G. Narasimha*
A. Sridevi
B. Devi Prasad
B. Praveen Kumar

Applied Microbiology Laboratory, Department of Virology, Sri Venkateswara University, Tirupati-517502 Andhra Pradesh, India.

Received 06 February 2013
Accepted 10 May 2013

ABSTRACT

In this study, Zinc Oxide nanoparticles (ZnONPs) were synthesized by chemical method and their antibacterial efficiency against a clinical isolate of *Staphylococcus aureus* was studied. The Zinc Oxide nanoparticles have shown a commendable inhibition effect on the growth of most notorious bacterial pathogen *S.aureus*. The nanoparticles size and shapes were characterized by transmission electron microscopy (TEM). The rod shaped zinc nanoparticles were formed as as a result of fast reduction rate of the precursor. The shape controlled nanocrystals possess well defined surfaces and morphologies and showed an excellent antibacterial property against clinical bacterial strain *S. aureus*.

Keywords: *ZnONPs; Chemical synthesis; Characterization; Antibacterial activity; S. aureus.*

INTRODUCTION

Zinc Oxide is one chemical agent known for its ability to kill a wide range of bacteria for long time and it had been used to treat many types of skin infections and their use slowly dissipated and replaced by modern antibiotics. Nevertheless, the chemical induced oxidative stress by Zinc Oxide is an unparalleled phenomenon that any contemporary antibiotics failed to achieve. Zinc Oxide has further advantages those which make it an effective antibacterial drug. Walsh , reported that Zn^{2+} besides being a non-toxic metal is an essential nutrient present virtually in almost every cell is critical for numerous physiological functions and must be consumed in diet to maintain normal health [1]. The toxicity, acute and chronic deficiency of Zn^{2+} due to nutrition factors and several diseases manifested in clinical symptoms those which range from mild to severe and affecting several organ systems including skin [2].

* Corresponding author:
G. Narasimha
Applied Microbiology Laboratory, Department of Virology, Sri Venkateswara University, Tirupati-517502 Andhra Pradesh, India.
Tel +91 9700170539
Fax +91 9700170539
Email dr.g.narasimha@gmail.com

The nanosized Zinc Oxide exhibits deodorizing and antibacterial properties [3] this property pushed them to become a part of the deodorizing and antibacterial cotton fabrics, food packaging [4] and It is photostable chemical used in sunscreen lotions and is also stable under high temperatures and pressures and these properties make it suitable for use in food processing industries and also in food fortification to kill infection spreading pathogens. The Zinc might be absorbed into, being a part of sunscreens and its traces observed in blood and urine samples [5]. Burnett and Wang (2011) reported evidence of systemic absorption of Zinc [6]. The Zinc Oxide is known to act by altering the membrane permeability of the pathogen and by inducing the oxidative stress inside the cell kills the bacterial strain, *Staphylococcus aureus* [7]. It forms a major part of normal skin flora and are abundantly found in anterior nares of the nasal passages and employ an array of immune-evasive strategies, which altogether makes it a successful pathogen [8,9]..According to Wenzel and Perl , 11-32% of the healthy individuals are serving as carriers of the pathogen and it goes up to 25% in the case of hospitalized people [10]. The Methicillin resistant *Staphylococcus aureus* Multi-drug resistant *Staphylococcus aureus* (MRSA) is now the reason for the increased rates of mortality. Our current study is focused on chemical synthesis of ZnO nanoparticles, characterization and determination of their antibacterial activity against human bacterial pathogen *Staphylococcus aureus*.

EXPERIMENTAL

Preparation and Characterization of ZnONPs

A stock solution of Zinc acetate Zn $(CH(CH_3COO)_2.2H_2O$ (0.1M) was prepared in Methanol under constant stirring. To 50mL of the Zinc acetate stock, 25mL of gradient (0.2 to 0.5) NaOH solution prepared in methanol was added slowly under constant mild agitation to attain a P^H between 8 and 11. The resultant reactant solution was autoclaved for 6 – 12 hours, and then allowed to cool naturally at room temperature and finally the white crystals obtained were washed in methanol and dried in a laboratory hot air oven at 60^0C [11]. The synthesized nanoparticles shape and sizes were characterized by Transmission Electron Microscopy (TEM).

Bacterial strain (Staphylococcus aureus)

The *Staphylococcus aureus* stock culture was obtained from SVIMS Hospital, Tirupati and sub cultured on nutrient agar media before their use for our studies.

Antibacterial activity

The agar culture plate containing the *S. aureus* is taken and wells were bored using an agar borer. Then, the Zinc Oxide nano particles were suspended in deionized water to make different concentrations *viz* 100, 500 and 1000 ug/mL and 50 uL/well of this suspension was loaded into agar plates.

RESULTS AND DISCUSSION

The chemically synthesized ZnO nanoparticles shape and size were characterized by Transmission Electron Microscopy. The TEM photograph showing the rod shaped ZnO nanoparticles (Figure 1). It is observed that above 75% of the particles are in 10-25 nm range, and there is a marginal variation in the particle size. In this range, rod shaped structures are also observed. Surprisingly, in this range, no spherical shaped nanoparticles are observed. Rod shaped particles are seen in the lower range of 10-15 nm, but overall contribution of these shapes to the total number of nanoparticles taken for analysis. The length of the short rod-like particles was in the range of 4-6 μm and the diameter was about 0.6 *n*m. TEM results shown in Figure the rod-like particles formed under the higher pH conditions should be related to the high instantaneous concentration of monomer produced in the reaction. With the decrease of pH, the nucleation and growth processes were slowed down synchronously due to the slow reduction rate of the precursor. As a result, formation of the rod shaped particles was suppressed almost completely under low pH value. These demonstrations suggest that it is possible to tune the size of ZnO nanocubes by controlling the experimental conditions.

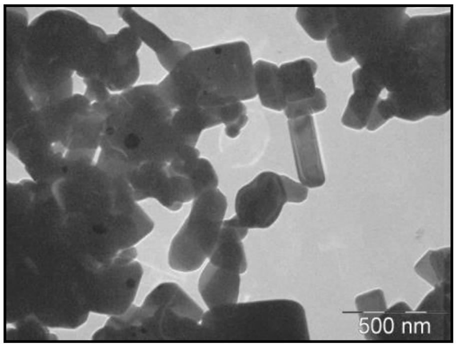

Fig. 1. TEM Photograph of ZnONPs

The efficacy of the ZnO nanoparticles as antibacterial is depicted as a measure of the ZnO of inhibition against bacterial pathogen *S.aureus* in nutrient agar medium at different concentrations of the ZnO and the values are shown in the Table 1 and Figure 2 .With increasing the nanoparticles concentrations the bacterial inhibition also increased with indication of increasing the diameter of bacterial zone of inhibition (Figure 2). Where as in control well (without ZnONPs suspension) the inhibition zone was not formed (Figure 2).

Table 1. Antibacterial activity of ZnONPS at different concentrations

ZnO contration	100 ug/mL	500 ug/mL	1000 ug/mL
Volume loaded per well	50 uL/well	50 uL/well	50 uL/well
Zone of inhibition(cm)	0.65	0.82	1.2

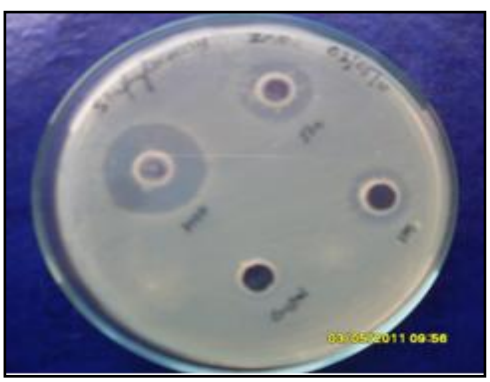

Fig. 2. Bacterial zone of inhibition by ZnONPs (Agar well plate method)

The minimum inhibition concentration (MIC) was formed at nanoparticles concentration at 100ug with 0.65cm inhibition zone. It is an indication of highest efficacy of ZnO nanoparticles against bacterial pathogen *Staphylococcus aureus.*

Zinc Oxide is widely used to treat a variety of other skin conditions, in products such as baby powder and barrier creams to treat diaper rashes, calamine cream, anti-dandruff shampoos and antiseptic ointments [12]. However, the knowledge on the mechanism of action of the ZnO conferring it the antibacterial properties were not clear until recently an evidence suggest that Zinc Oxide alters the permeability of the membrane and induces oxidative stress thus killing the bacteria as shown by Xie Y et al. in *Campylobacter jejuni* [13]. According to his studies Oxidative stress genes *KatA* and *ahpC* were upregulated by 52 and 7 fold respectively and the normal stress gene *dnaK* was upregulated 17 fold suggesting the antibacterial activity of ZnO is induction of stress in *C.jejuni* cells. The effectual nature of nanosized Zinc Oxide compared to normal sized ZnO is not unique to Zinc but a property noticed with other metals like Silver which is due to high surface to volume ratio of nanosized materials [14].

CONCLUSIONS

Zinc Oxide nanoparticles were successfully synthesized chemical process. The ZnO nanoparticles size and shapes were characterized by Transmission Electron Microscope and then rod shaped nanoparticles were observed. These rod shaped Zinc Oxide nanoparticles exhibited an excellent antibacterial property against a bacterial pathogen *Staphylococcus aureus*. Further studies needs on molecular mechanism of rod shaped ZnONPs against bacterial growth inhibition.

REFERENCES

[1] Walsh, C. T., Sandstead, H. H, Prasad, A. S,. Newberne P. M., Fraker P. J., (1994), Zinc: Health effect and research priorities for the 1990s. *Environ. Health Perspec.* 102: 5-46.

[2] Prasad, A. S., (1991), Discovery of human zinc deficiency and studies in an experimental human model. *Am. J. Clin. Nutri*. 53: 403-412.

[3] Nagarajan P., Vijayaraghavan R., (2008), Enhanced bioactivity of ZnO nanoparticles an antimicrobial study. *Sci. and Tech. Adv. Mater*. 9: 035004-8.

[4] Li Q. C., Shui-Lin J., Wan C., (2007), Durability of nano ZnO antibacterial cotton fabric to sweat. *J. Appl. Polym. Sci.*, 103: 412-414.

[5] Brian G., McCall M., Michael K., Laura G., Philip C., Yalchin O., (2010), Small amounts of Zinc from Zinc Oxide particles in sunscreens applied outdoors are absorbed through human skin. *Toxicol. Sci*. 118: 140-149.

[6] Burnett M. E., Wang S. Q,. (2011), Current sunscreen controversies: a critical review. *Photodermatol. Photoimmunol. and Photomed*. 27: 58-67.

[7] Alexander O., (1984), Classics in infectious diseases. "On abscesses". *Rev. Infect. Dis.* 6: 122-128

[8] Kluytmans J., Van Belkum A., Verbrugh H., (1977), Nasal carriage of *Staphylococcus aureus*: Epidemiology, underlying mechanisms, and associated risks. *Clin. Microbiol. Rev*. 10: 505-520.

[9] Alexander M., Cole S. T., Ami O., Dawn Y., Yong-Hwan K., Albert P., Tomas G., (2001), Determinants of *Staphylococcus aureus* Nasal Carriage. *Clin. Diagn. Lab. Immunol*. 8: 1064–1069.

[10] Wenzel R. P., Perl T. M., (2010), The significance of the nasal carriage of *Staphylococcus aureus* and the incidence of postoperative wound infection. *J. Hosp. Infect*. 202: 924-934.

[11] Aneesh P. M., Vanaja K.A., Jayaraj M.K., (2007), Synthesis of ZnO nanoparticles by hydrothermal method. *Proc. of SPIE*. 6639, 66390J-1 – 66390J-9.

[12] Harding F. J., (2007), Breast cancer: cause – Prevention – Cure. *Tekline publishing*. P. 83.

[13] Yanping X., Yiping H., Peter L., Irwin T. J., Xianming Sh., (2011). Antibacterial Activity and Mechanism of Action of Zinc Oxide Nanoparticles against Campylobacter jejuni. *Appl. Environ. Microbiol*. 77: 2325-2331.

[14] Akhavan O., Ghaderi E., (2009), Enhancement of antibacterial properties of Ag nanorods by electric field. *Sci. technol.* & *Advan. Mater*. 10: 015003-6.

Cite this article as: G.Narasimha *et al.*: Chemical synthesis of Zinc Oxide (ZnO) nanoparticles and their Antibacterial activity against a clinical isolate *Staphylococcus aureus*.
Int. J.Nano Dimens. 5(4): 337-340, Autumn 2014