Antimicrobial Activity of Zinc Oxide and Zinc Oxide-Zinc Sulphide Core Shell Nanoparticles

Phyu Phyu Myint[1], Myat Kyaw Thu[2]

ABSTRACT

This research concerns the preparation, characterization and antimicrobial activity of zinc oxide nanoparticles. In the present work, zinc oxide nanoparticles were synthesized from zinc acetate sources using sodium hydroxide as a precipitating agent by the co-precipitation method. ZnO nanoparticles were synthesized from the precursors of Zn $(CH_3COO)_2.2H_2O$ and NaOH by the co-precipitation method. In these processes the concentration of solutions was prepared as a three solution (0.1 M) by mixing $ZnCl_2$ and Na_2S solution. The prepared ZnO nanoparticles were covered with ZnS using a solution-based chemical method at a low temperature. The characterization processes (antimicrobial activity) were performed. In this research, ZnO-ZnS core shell nanoparticles were characterized by antimicrobial activity at the temperature 300˚C and 500˚C. The antimicrobial activity of prepared ZnO nanoparticles on six microorganisms (*Bacillus subtilis, Candida albicans, Escherichia coli, Pseudomonas fluorescens, Staphylococcus aureus, and Salmonella typhi)* was investigated by the Agar well diffusion method. It was found that high activity on *Escherichia coli, m*edium activity on *Bacillus subtilis, Candida albicans* & *Staphylococcus aureus* and low activity on *Pseudomonas fluorescens* & *Salmonellatyphi.*

Keywords: ZnO nanoparticles and ZnO-ZnS core shell nanoparticles, antimicrobial activity, agar well diffusion method

INTRODUCTION

In nature, pure zinc oxide is a white powder, but in nature it occurs as the rare mineral zincite. (Hernandezbattez, *et.al.,* 2008). The most common use of ZnO nanoparticles is in sunscreen, which usually contains manganese and other impurities that confer a yellow to red color. Zinc Oxide is an amphoteric oxide. It is nearly insoluble in water, but it will dissolve in most acids, such as hydrochloric acid. Zinc Oxide is widely used in different areas because of its unique photocatalytic, electrical, electronic, optical, dermatological, and antibacterial properties (Alessio, *et al.,* 2008). Zinc oxide (ZnO) nanoparticles (NPs) as a cheap nontoxic semiconductor with a wide direct band gap are a promising material for different applications such as photocatalysts, photodetectors, gas sensors, piezoelectric sensors and ultraviolet lasers and can be synthesized in different ways such as by the chemical vapor phase method, thermal evaporation, the vapor-solid technique, the hydrothermal method and the co-precipitation method. Recently, it was found that surface coating with different semiconductors can dramatically change the properties of ZnO nano-crystallites. In recent years, much effort has been devoted to the development of core–shell structured materials on the nanometer scale. The shell usually acts as a barrier between the core and the environment and can alter the charge, functionality, and reactivity of the surface. It can also change the stability and dispersive ability of the core material. Furthermore, the electrical, catalytic, optical, or magnetic properties of the core material can be improved by covering it with another material. Zinc oxide is one of the important shell-forming materials because of its wide band gap, it is known to be nontoxic, biosafe, and biocompatible, and it can be easily prepared through chemical solution processes including sol-gel, hydrothermal synthesis, and electrochemical deposition. ZnO is a versatile material and has been used for its catalytic, electrical, optoelectronic, and photochemical properties (Rakgalakane, *et al.,* (2011).

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MATERIALS AND METHODS

Preparation of Zinc Oxide Nanoparticles

Firstly a (0.5M) zinc acetate dihydrate solution was prepared by dissolving 10.95 g of ($ZnAc_2.2H_2O$) in 100 mL of distilled water. Then 1 M was added to this solution. The mixture solution was stirred for 2 hours until a white precipitate was formed. Then the solution was centrifuged. The precipitate was washed with deionized water and acetone at least three times and it was heated at 75 ºC. The dried sample was then collected and calcined in a muffle furnace at, 300 ºC for 1 h. A pale brown color powder was obtained and it was carefully collected and packed for further use.

Preparation of ZnO-ZnS Core Shell Nanoparticles

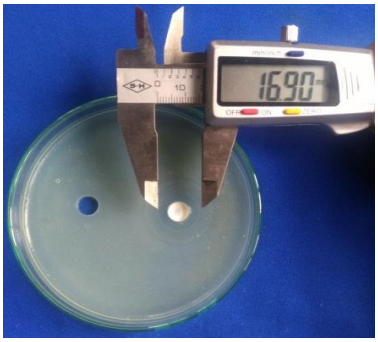
ZnO nanoparticles were covered with ZnS using a simple chemical method. 0.3 g of the as-grown ZnO nanoparticles with 50 mL isopropanol was sonicated for 10 min. After adjusting the pH to 10 using ammonium hydroxide, a solution of Na_2S was added to the mixture drop wise. Then it was kept under continuous stirring at 60 ºC for 2 h. Finally, in order to achieve the formation of ZnO-ZnS core shell nanoparticles, a solution of 0.05 M $ZnCl_2$ was added drop wise into the above mixed solution and stirred for 1 h. Afterwards, the product was washed with deionized water and acetone and was dried at 70 ºC. In order to investigate the effect of the shell thickness on the structural properties of ZnO-ZnS core shell nanoparticles, the samples were synthesized with various concentrations of the solution (0.1 M) calcined at 300 and 500 ºC. (Azar Sadollahkhain *et al.*, 2014)

Antimicrobial Activity (Well Diffusion Assay)

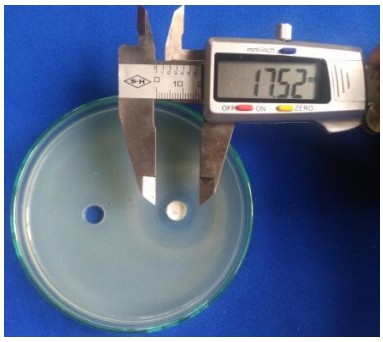
Isolated bacterial strains grown on nutrient agar were inoculated into 50 mL conical flasks containing 10 mL of sterile growth medium. Test organisms were *Aspergillus flavors*, *Bacillus subtilis*, *Candida albicans*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. 0.3 mL of test organisms was added to assay medium, then poured into plates. After solidification, well impregnated with broth samples were applied to the test plates and these plates were incubated for 24-36 h at 30 °C. After 24-36 h, clear zones (inhibitory zones) surrounding the test well indicate the presence of bioactive compounds which inhibit the growth of test organisms.

RESULTS AND DISCUSSION

Antimicrobial Activity of Zinc oxide Nanoparticles at 300 ºC

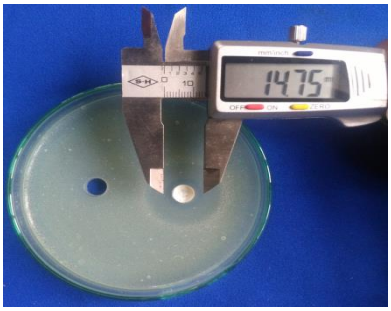


(i) Bacillus pumilus

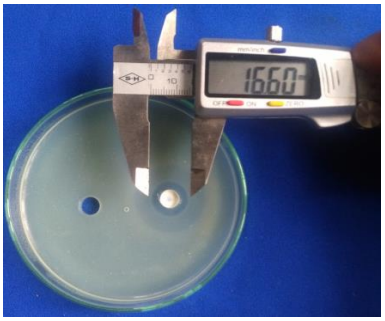


(ii) *Candida albicans*

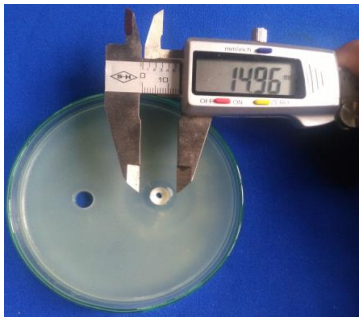
(iii) *Escherichia coli*



(iv)Pseudomonas fluorescens



(v) *Staphylococcus aureus*



(vi) *Salmonella typhi*

Figure 1. Antimicrobial Activity of ZnO Nanoparticles on Six Microorganisms

Table 1. Inhibition Zone Diameters of ZnO Nanoparticles on Six Microorganisms

Microorganisms / Sample	Inhibition Zone Diameter (mm)					
	1	2	3	4	5	6
ZnO Nanoparticles	16.90 (++)	17.52 (++)	22.94 (+++)	14.75 (++)	16.60 (++)	14.96 (++)

Agar well = 8mm

10 mm ~ 14 mm (+) – mild activity

15 mm ~ 19 mm (++) – medium activity

20 mm ~ above (+++) – high activity

1. *Bacillus subtilis* 2. *Candida albicans* 3. *Escherichia coli*

4. *Pseudomonas fluorescens* 5. *Staphylococcus aureus* 6. *Salmonella typhi*

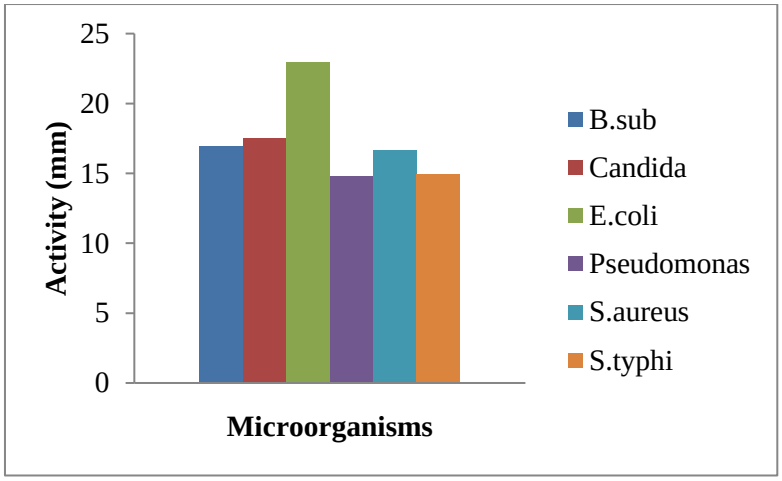


Figure 2. Comparison of Test Organisms by ZnO Nanoparticles at 300 ^{o}C

Antimicrobial Activity of ZnO Nanoparticles at 300 ^{o}C

The antimicrobial activity of ZnO nanoparticles was investigated using gram positive and gram negative bacterial strains, *Bacillus subtilis, Staphylococcus aureus, Pseudomonas fluorescens, Candida albicans, Escherichia coli* and *Salmonella typhi*, respectively. The measurable zone diameter, including the well diameter, shows the degree of antimicrobial activity.

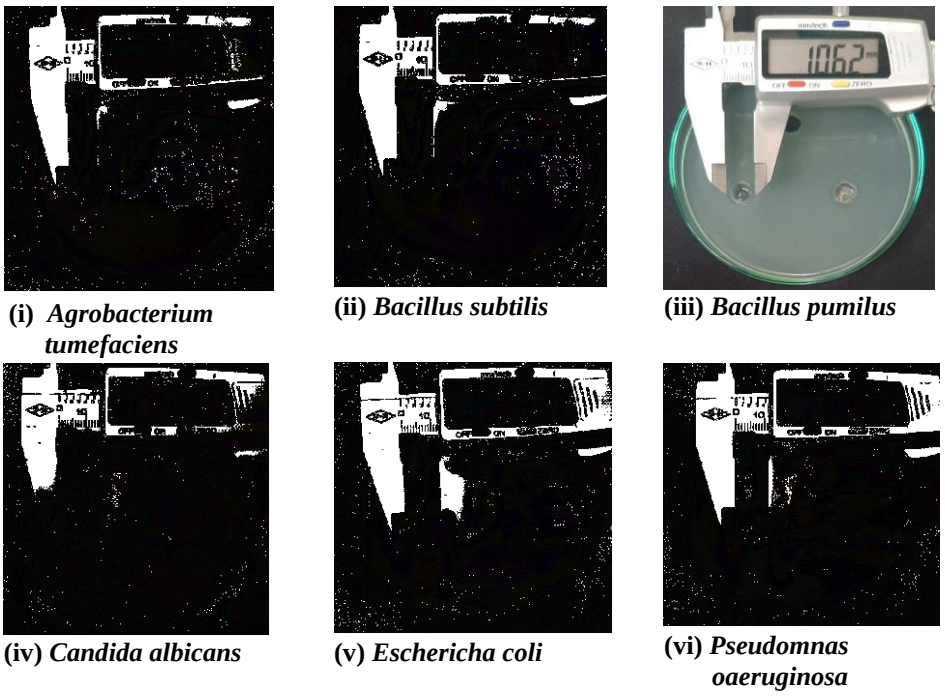
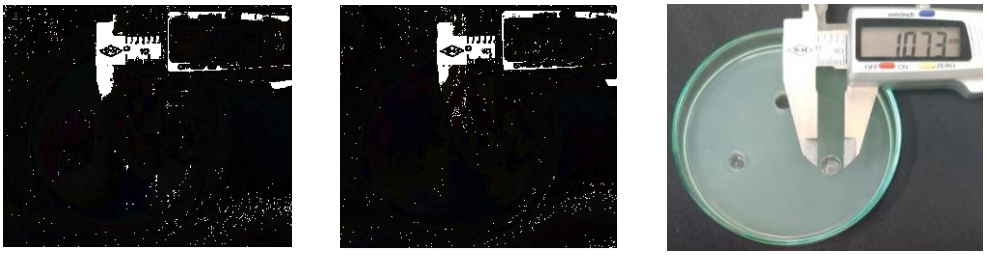


Figure 3. Antimicrobial activity of 0.1 M ZnO-ZnS core shell nanoparticles at 300 ^{o}C

(i) *Agrobacterium tumefaciens* (ii) *Bacillus subtilis* (iii) *Bacillus pumilus*

(iv)*Candida albicans* (v) *Escherichia coli* (vi) *Pseudomonas aeruginosa*



(i) *Agrobacterium tumefaciens* (ii) *Bacillus subtilis* (iii) *Bacillus pumilus*

(iv) *Candida albicans* (v) *Escherichia coli* (vi) *Pseudomonas aeruginosa*

Figure 4 . Antimicrobial activity of 0.1 M ZnO-ZnS core shell nanoparticles at 500ºC
(i) *Agrobacterium tumefaciens* (ii) *Bacillus subtilis* (iii) *Bacillus pumilus*
(iv)*Candida albicans* (v) *Escherichia coli* (vi) *Pseudomonas aeruginosa*

Table 2 Antimicrobial Activity of (0.1M) ZnO-ZnS Core Shell Nanoparticles

Test Organisms	**ZnO-ZnS Core Shell Nanoparticles**	
	Inhibition Zone Diameter (mm)	
	300 ºC	**500 ºC**
Agrobacterium tumefaciens	10.18	15.54
Bacillus subtilis	12.04	12.36
Bacillus pumilus	10.62	10.73
Candida albicans	14.99	15.52
Escherichia coli	12.32	13.48
Pseudomonas aeruginosa	11.28	13.59
Staphylococcus aureus	-	-

C = control

Agar well ~ 8mm

8.1 mm ~ 14.9 mm = Less effective

15 mm ~ 19.9 mm = Intermediately effective

20 mm above = Highly effective

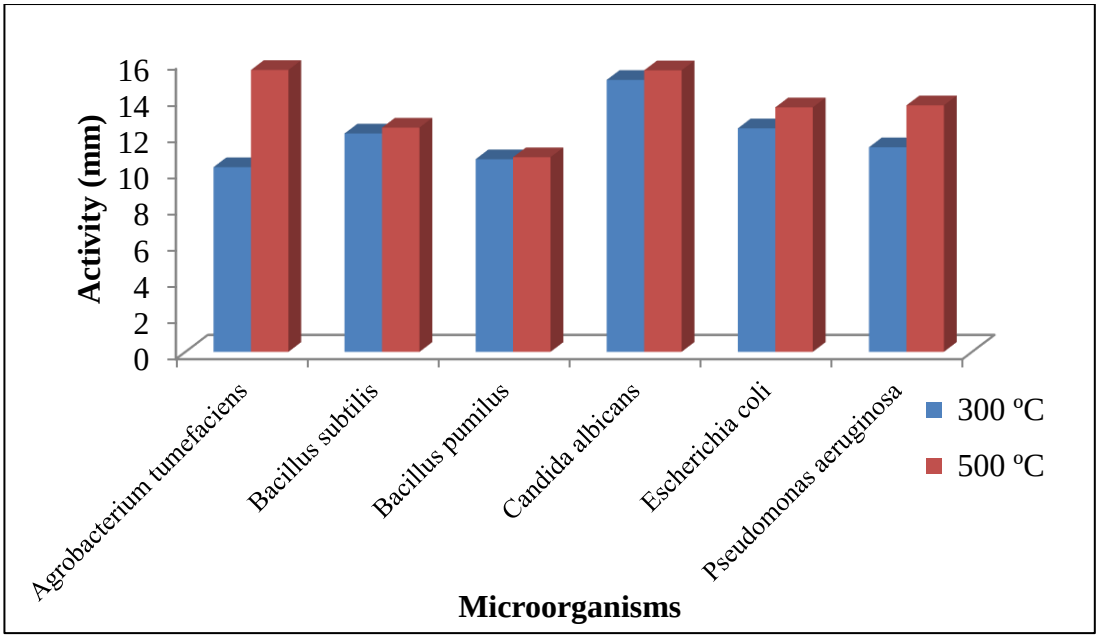


Figure 5. Comparison of microorganisms by ZnO-ZnS core shell nanoparticles at 300 ºC and 500 ºC

Antimicrobial Activity of ZnO-ZnS Core Shell Nanoparticles at 300 and 500 ºC

The antimicrobial activity of ZnO-ZnS Core Shell Nanoparticles was investigated using gram positive and gram negative bacterial strains, *Agrobacterium tumefaciens, Bacillus subtilis, Bacillus pumilus, Candida albicans, Escherichia coli* and *Pseudomonas aeruginosa*, respectively. The measurable zone diameter, including the well diameter, shows the degree of antimicrobial activity.

CONCLUSION

In this research work, ZnO powder was synthesized by the co-precipitation method. In future, this method of synthesizing ZnO powder could also be extended to other industrially important metal oxides. Antimicrobial Activity, high activity on *Escherichia coli, m*edium activity on *Bacillus subtilis, Candida albicans and Staphylococcus aureus,* low activity on *Pseudomonas fluorescens* and *Salmonella typhi.* According to antimicrobial activity the prepared zinc oxide nanoparticles can be used as antimicrobial agents. Especially, the antimicrobial activity of zinc oxide nanoparticles was found to have high activity against *Escherichia coli*. At the temperatures of 300 and 500 ºC, it was observed that the high activity on *Agrobacterium tumefaciens and Candida albicans, m*edium activity on *Escherichia coli* and *Pseudomonas aeruginosa,* and low activity on *Bacillus subtilis* and *Bacillus pumilus.* According to antimicrobial activity the prepared ZnO-ZnS Core Shell Nanoparticles can be used as antimicrobial agents. Especially, the antimicrobial activity of ZnO-ZnS Core Shell nanoparticles was found to have high activity against *Agrobacterium tumefaciens*.

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