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Antibacterial Activity of Zinc Oxide Nanoparticle Prepared From Chrysanthemum Denranthema Leaf Extract

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ABSTRACT

Nanoparticle metal oxides represent a new class of important materials that are increasingly being developed for use in research and health-related applications. Highly ionic metal oxides are interesting not only for their wide variety of physical and chemical properties but also for their antibacterial activity. Although the in vitro *antibacterial activity and efficacy of regular zinc oxides have been investigated, little is known about the antibacterial activity of nanoparticles of ZnO. Preliminary growth analysis data suggest that nanoparticles of ZnO have significantly higher antibacterial effects on* Staphylococcus aureus *than do five other metal oxide nanoparticles. In addition, studies have clearly demonstrated that ZnO nanoparticles have a wide range of antibacterial effects on a number of other microorganisms. The antibacterial activity of ZnO may be dependent on the size and the presence of normal visible light. The data suggest that ZnO nanoparticles have a potential application as a bacteriostatic agent in visible light and may have future applications in the development of derivative agents to control the spread and infection of a variety of bacterial strains.*

Key words: *ZnO nanoparticles, Antibacterial, Activity.*

INTRODUCTION

Nanotechnology is a fastest growing field with its huge application in science and technology for the manufacturing new materials at the nano scale level. In the field of nanotechnology, especially the ability of nanotechnology to prepare highly ordered nanoparticles of any shape and size. Nano-materials are also called "a wonder of modern medicine". Nanotechnology is a multidisciplinary scientific field undergoing explosive development. The characters of metal and metal oxide nanoparticles have distinctive feature such as catalytic, optical, magnetic and electrical properties, Nanotechnology is a multidisciplinary scientific field undergoing explosive development. Nanometer - sized particles offer novel structural, optical and electrical properties.[1].

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Nanotechnology come out from the physical, chemical and biological science where new techniques are developed to study and action of single atom and molecules for multiple application in different field of science. In nanotechnology, a nanoparticle defined as a small object that behave like a whole unit The fastest and growing sectors of nano systems in nanotechnology is science and engineering. Nanotechnology is useful in many sectors such as diagnostic techniques, drug delivery, antimicrobial bandages, these green synthesis process are friendly manufacturing process that reduce waste products ,as catalyst for greater efficiency in current manufacturing process by minimizing the use of highly toxic materials, to reduce pollution.[2]. Nanoparticles of zinc oxide can be scatter in industrial coatings to protect wood, plastic, and textiles from exposure to UV rays. Silicon dioxide crystalline nanoparticles are used to fill the gaps between carbon fibers. Silver nanoparticles are used as antibacterial in fabric are used to kill bacteria, making clothing odor-resistant.

MATERIAL AND METHODS

The materials and methods used in the present study entitlrd **"Synthesis of zinc oxide nanoparticle and its antibacterial activity by** *Chrysanthemum dendranthema* leaves.**-**

Materials

All the chemicals such as zinc acetate, distilled water ingredients utilized in this work taken from Research laboratory of chemistry department of SHUATS And the leaves of *Chrysanthemum dendranthema* (plant variety – Koka senkin) plant were collected from horticulture department of SHUATS, Allahabad (U. P) India.

Fig. 1: Plant of Chrysanthemum dendranthema

Methods

Preparation of leaf extract of *Chrysanthemum dendranthema*

Sample Preparation

The leaves were first washed several times with running water and subsequently with distilled water. And both the surface of leaves were sterilized using alcohol by. Leaves were dried at room temperature in shade until all moisture was lost (12–14 days). The dried leaves were cut and grinded for preparing powder.

Fig. 2: Leaves of Chrysanthemum dendranthema

Preparation of extract

Plant extract was prepared by mixing of 5g of *Chrysanthemum dendranthema* plant powder in 50ml of ethanol (95%) in a 250 ml conical flask. The solution was kept for 48 hours in dark and then extract was filtered by whatman filter paper 1. Further Investigation has been carried out from this extract. Figure 2 shows the sample of leaves extract of *Chrysanthemum dendranthema* plant.

Green synthesis of ZnO Nanoparticles

Zinc oxide nanoparticles were synthesized using zinc acetate dihydrate Zn $(CH_3COO)_2{\cdot}2H_2O$. Solution (0.01 M) of zinc acetate was taken and leaves extract was added. The pH of the mixture was maintained.

Fig: 3 plant extract

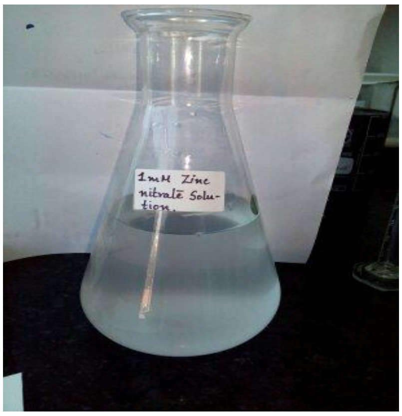

Fig. 4: zinc nitrate salution

Charaterization techniques:

SEM analysis:- Size and structure of the nanoparticles analysed by using Tescan Scanning Electron Microscope machine. Thin films of the sample were prepared on a carbon coated copper grid by just dropping a very small amount of the sample on the grid, extra solution was removed using a blotting paper and then the film on the SEM grid was allowed to dry by putting it under a mercury lamp for 5 min. The morphological features of synthesized zinc oxide nanoparticles from *Chrysanthemum dendrathemum* leaves extract were studied by Scanning Electron Microscope.**:**

Antibacterial activity: Test organism for antibacterial activity

Gram positive (+) bacteria and gram negative (-) bacteria, will be used for the antibacterial activity. The bacterial pathogens were collected from the microbial culture collection bank of department of microbiology and fermentation technology SHUATS.

A test bacterium was grown and maintained on Nutrient agar slant at $37^{o}C$. Following incubation for 5 days, the culture was either utilized for test or stored at $4^{o}C$ for further use. The organism were subculturedonce in every 15 days

Antibacterial activity: Agar well diffusion method

Agar well diffusion method as given by Azam *et al*.[3].

Procedure: Antibacterial activities of the synthesized zinc oxide nanoparticles will be performed against the *Pseudomonas aeruginosa, Bacillus cereus, Proteus vulgaris and Clostridium botulinum.*

These are the Bacterial strains which cause Waterborne disease, The antibacterial test were done by Agar well diffusion method, Azam *et al*.[3] The pure culture of organism were sub-cultured in nutrient broth .The nutrient agar plates were prepared by 25 ml (for one plate) pouring was molten media into sterile petriplates. For bacterial growth, a lawn of culture was prepared by spreading the 100µL fresh culture having each test organism on nutrient agar plates with the help of a sterile glass rod spreader. Plates were left standing for 10 minutes to let the culture get absorbed. Then 6 mm (size) wells (4 well) would be punched into nutrient agar plates for testing nanomaterial antibacterial activity. Using the micro-pipette, 100µl of sample of nanoparticle suspension would be poured in different concentration (25µl, 50µl, 75µl) onto each of wells on the plates. Then take a antibiotic (Ciplox of 250mg), it prepared in 1000ppm. This antibiotic used as positive control. Using the micropipette, 10µl of antibiotic (control) solution would be poured into one well. After incubation for 24 hrs at $37^{o}C$ the plates were observed. If antibacterial activity was present on the plates, it was indicated by an inhibition zone surrounding the well containing the ZnO nanoparticles solution. The zone of inhibition was measured and expressed in millimeters.

RESULT AND DISCUSSION

Conformation of Formation of ZnO NPs

Visual observation

ZnO NPs have attracted great attention because of their superior optical properties. Visual color change is the preliminary test for nanoparticle synthesis. Figure represents the synthesis of ZnO NPs. The color change was noted by visual observation in the bottles that contained $ZnNO_3$ solution with extract. The color of the $ZnNO_3$/extract solution changed from colorless to light yellow. This color change indicates the formation of ZnONP in the solution.

The formation of ZnONP was further confirmed by using ultraviolet-visible spectroscopy (UV-vis), X-ray diffraction (XRD).

X-ray diffraction pattern for ZnO nanoparticles

The XRD pattern of bulk ZnO and nano ZnO were presented in Figure. All the peaks were hexagonal and approximately close to the reported information (jcpds-79-0206). Due to the crystal symmetry and related face velocities, the common crystal habit of ZnO is hexagonal in shape. Also the ZnO NP is the thermodynamically stable crystallographic phase. The width of the peaks in case of ZnO NP has increased due to the quantum size effect. The average particle size was estimated to be 42 nm using Scherer equation (discussed in 'Material and Methods'). These diffraction lines observed at 2θangle 32.8^0, 38.2^0, 55.10° and 65.7^0 respectively, have been indexed as (111), (200), (220) and (311) respectively. XRD patterns were analyzed to determine peak intensity, position and width, full-width at half-maximum (FWHM) data was used with the Scherer formula explained in section materials and method. The typical XRD pattern revealed that the sample contains a mixed phase (cubic and

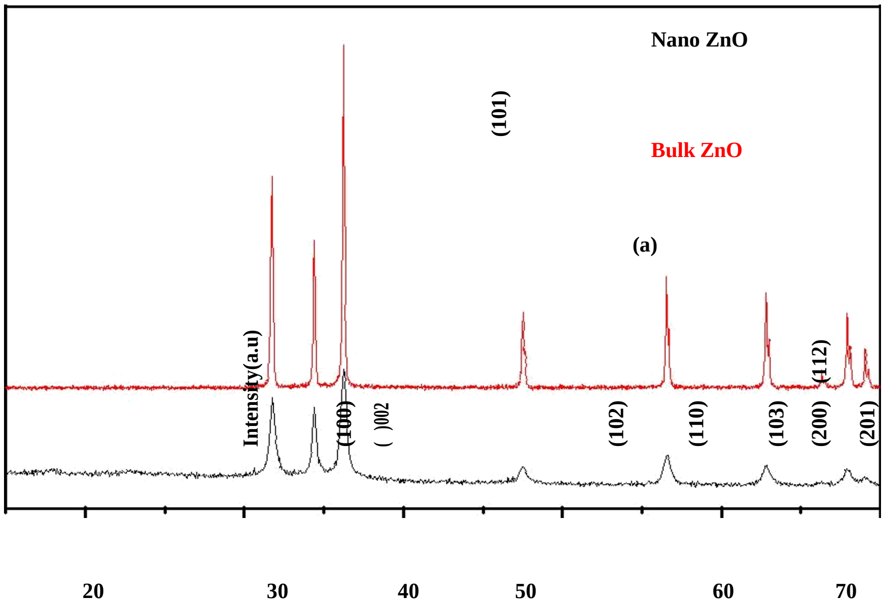

Fig. 5: 2Θ (in degree)

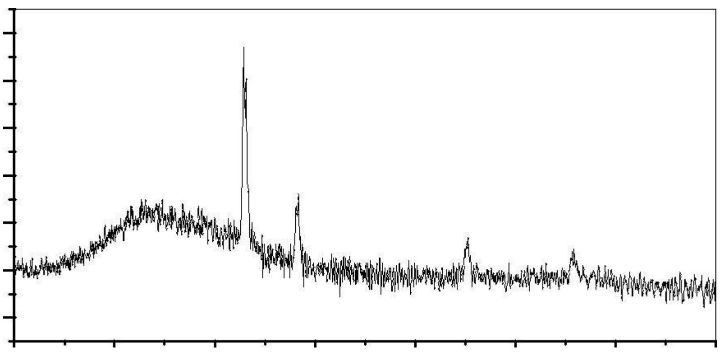

Fig. 6: (a) XRD patterns of ZnO nanoparticles (red) bulk ZnO (black) ZnO synthesized using 0.5% starch. The peaks assigned to diffractions from various planes are of hcp ZnO

SEM Analysis of ZnO

The SEM image of ZnO and Silver nanoparticles synthesized by chemical reduction method and green synthesis process by using 10 % extract and 1mM Zinc nitrate concentration was shown in Figure. It gave a clear image of highly dense ZnO nanoparticles. The SEM image showing silver nanoparticles synthesized using *s* extract confirmed the development of silver nanostructures.

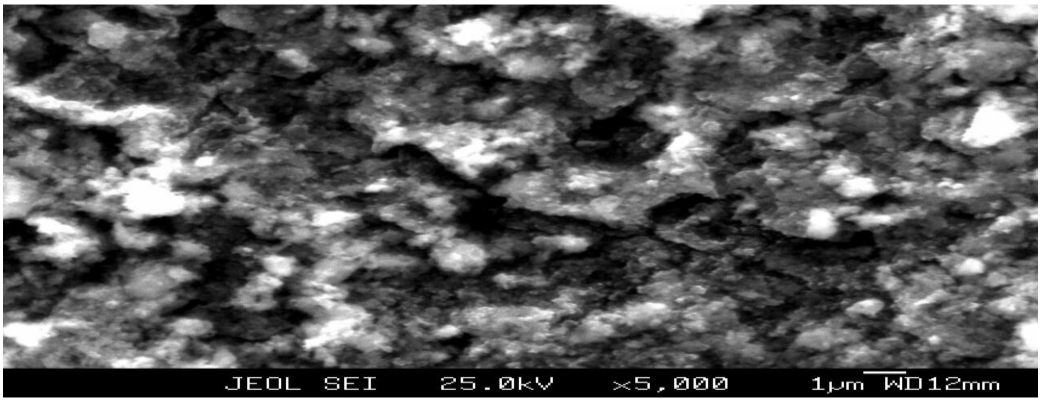

Fig. 7: Sem emage of zinc oxide nano particle

Antibacterial activity of ZnO nanoparticles against bacteria by agar well diffusion method

Concentration µg / µl	Diameter of Inhibition Zone of Inhibition (mm)			
	Pseudomonas aeruginosa	***Clostridium botulinum***	***Proteus vulgaris***	***Bacillus cereus***
2.5	8mm	-	15.5mm	11.5mm
5.0	13mm	-	14mm	13mm
7.5	15mm	-	18mm	14.5mm

*well size= 5mm.

Proteus vulgaris

Bacillus cereus

Pseudomonas aeruginosa

Clostridium botulinum

Fig. 8

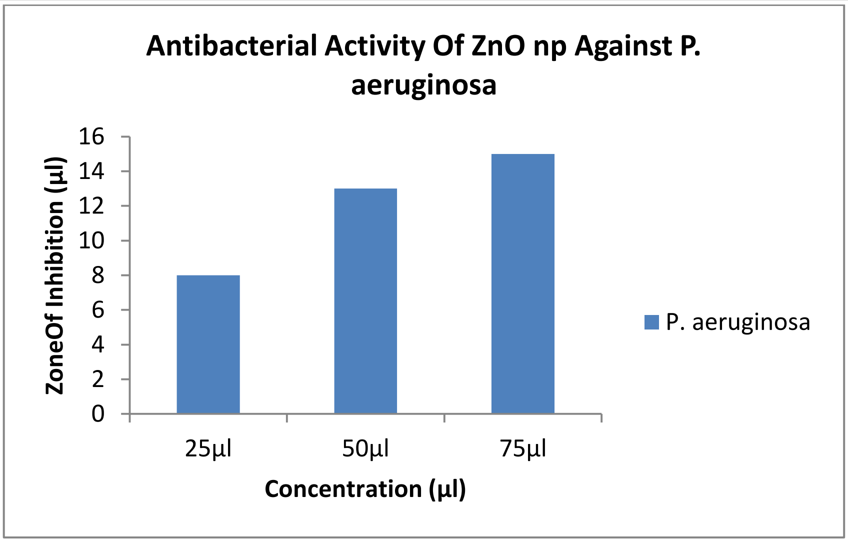

Fig. 9: Graph of antibacterial activity of ZnONPs against *P. aeruginosa*

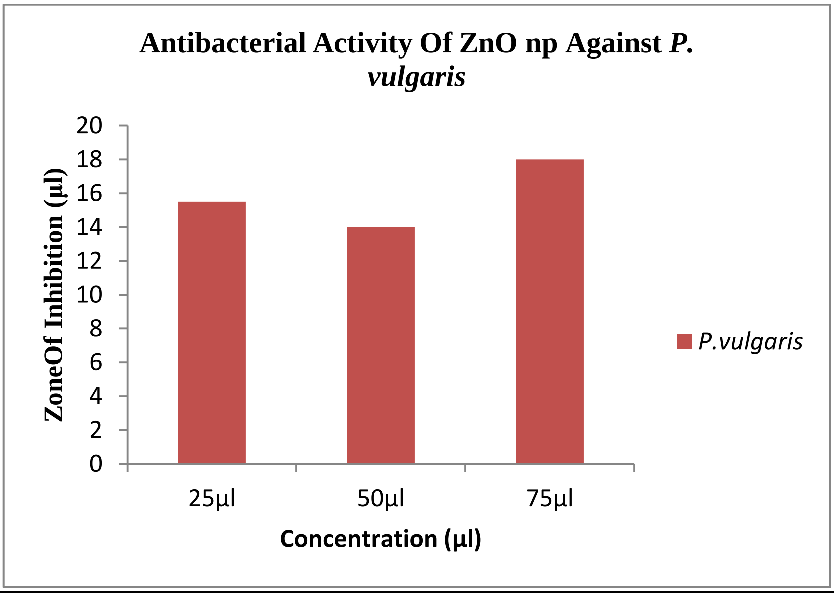

Fig. 10: Graph of antibacterial activity of ZnONPs against *P. vulgaris*

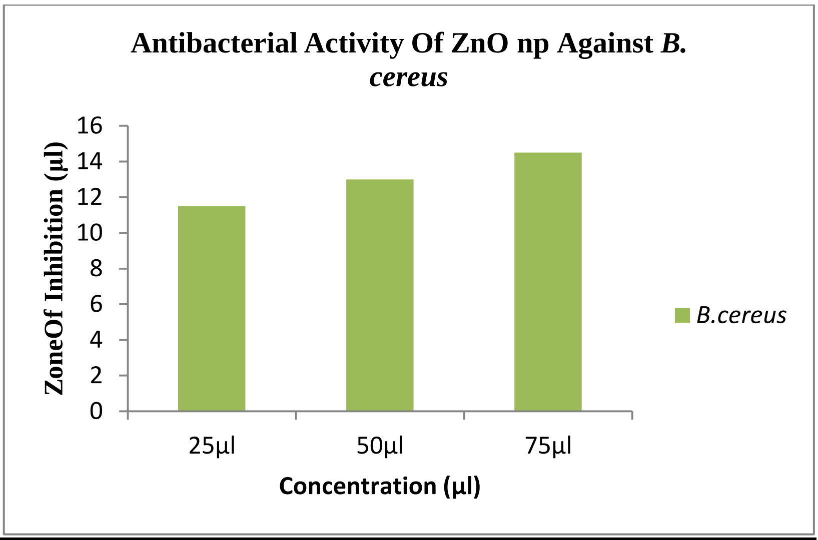

Fig. 11: Graph of antibacterial activity of ZnONPs against *B. cereus*

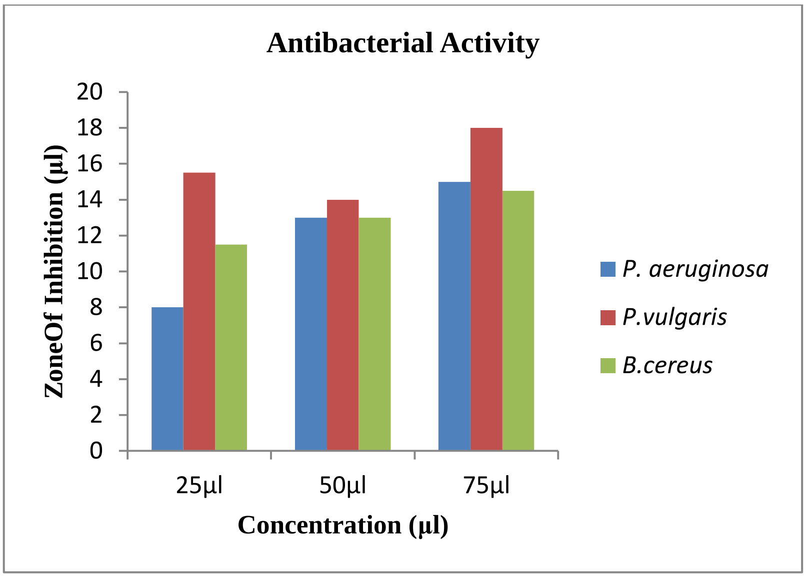

Fig. 12: Comparative Bar graph showing zone of inhibition introduced by different concentration of ZnO nanoparticles *against P. aeruginosa,P. vulgaris, B. cereu*A.

REFERENCES

1. Sharmaila, Chemopreventive effect of quercetin, a natural dietary flavonoid on prostate cancer in in vivo model *U. S. National Library of Medicine* (2014).
2. Haritha, Synthesis and characterization of zinc oxide nanoparticles and its antimicrobial activity against bacillus subtilis and escherichia coli *Rasayan Journal of Chemistry* **4(1):** January 2011 (2011).
3. Azam, Antimicrobial activity of metal oxide nanoparticles against Gram-positive and Gram-negative bacteria: a comparative study *international journal of nanomediciene* **7:** 6003–6009 (2012).
4. Azam, Evaluation of phytosynthesised silver nanoparticles from leaf extracts of *Leucas aspera* and *Hyptis suaveolens* and their larvicidal activity against malaria, dengue and filariasis vectors *Parasite Epidemiol Control*. 2017 Nov; **2(4):** 15–26 (2012).
5. Selvaraj, "Rapid green synthesis of silver nanoparticles from Chrysanthemum indicum L and its antibacterial and cytotoxic effects". *International journal of Nanomedicine v.9.* (2014).
6. Abhulimen, U., Mater. *Res. Soc. Symp. Proc*. **1:** 27 (2005).
7. Agarwal, H., Venkat Rajeshkumar, S., A review on green synthesis of zinc oxide nanoparticle – an ecofriendly approach. *Resoure – Efficient Technologies* (2017).
8. Agarwal, H., Venkat Rajeshkumar, S., Synthesis of zinc oxide nanoparticle using plant leafextract against urinary tract infection pathogen. *Resoure – Efficient Technologies*. 000 (2017) 1-7 (2017).
9. Baxter, J. B. and Aydil, E. S., *Appl. Phys. Lett.* **86:** 3114 (2005).
10. Brayner, R., Ferrari-Iliou, R., Brivois, N., Djediat, S., Benedetti, M. F. and Fievet, F., *Nano Lett.* **6:** 866 (2006).
11. Zhiyun, D., Qianfa, J., "Green synthesis of silver nanoparticles by Chrysanthemum morifolium Ramat, extract and their application in clinical ultrasound gel". *Journal of nanomedicine 1805 – 1815* (2013).
12. Das, D., Nath. B. C., Phukon, P., Kalita, A., Dolui, S., Synthesis Of Zinc Oxide nanoparticles and evaluation Of antioxidant and cytotoxin activity, *Journal of Colloids and Surfaces B; Biointerfaces,* **111:** Pages 556-560 (2013).
13. Dutta, K. R., Nenavathu, P. B. and Gangishetty, K. M., Correlation between defects in capped ZnO nanoparticles and their antibacterial activity. *Journal of*

photochemistry and photobiology. B, Biology. **126C:** 105-111 (2013).

14. Dutta, K. R., Nenavathu, P. B., Gangishetty, K. M., Correlation between defects in capped ZnO nanoparticles and their antibacterial activity. *Journal of photochemistry and photobiology. B, Biology*. **126C:** 105-111 (2013).
15. Santhosh kumar, J., Venkat, S., Rajesh kumar, S., Synthesis of ZnO nanoparticle using plant leaf extract against urinary tract infection pathogen, *Journal of Resource- Efficient Technologies,* **000:** 1-7 (2017).
16. Jing, L., Huiping, M., Pengcheng Gao, R., Jia, Z., Anti-oxidant potential, total phenolic and flavonoid contents of *Rhododendron anthopogonoides* and its protective effect on hypoxia- induced injury in PC12 cell, *BMS complementary and Alternative medicine*,**15:** 287 (2015).
17. Jones, N., Ray, B., Ranjit, K. T. and Manna, A. C. Antibacterial activity of ZnO nanoparticle suspensions on a broad spectrum of microorganisms. *FEMS Microbiology Letters*. **279:** 71–76 (2008).
18. Kaur, C., Kapoor, H. C., Anti-ocidant activity and total phenolic content of some Asian vegetables, Indian Agricultural Research Institute, DOI: 10.1046/j.1365-2621.2002.00552 (2002).
19. Khadeeja, P., Ledwani and Banse, V., Green synthesis of nanoparticles; Their advantages and disadvantages. 1724, 020048 doi: 10.1063/1.4945168 (2016).
20. Kim Hawan, S., Lee, H. S., Ryu, D. S., Choi, S. L., Antibacterial Activity Of Silver nanoparticles Against Staphylococcus aures and Escherichia coli. *Korean J. Microbiol. Biotechnol*, **39(1):** 77–85 (2011).
21. Lakshmi, J. V., Sharath, R., Chandraprabha, M. N., Neelufar, E., Hazra, A. and Patra, M., Synthesis Charterization and evaluation of antimicrobial activity of zinc oxide nanoparticles. *J Biochem Tech* **3(5):** S151-S154 (2012).
22. Manokari, M. S., Mahipal Production of zinc oxide nanoparticle using extracts of *Passiflora edulis Sims. f. flavicarpa Deg.World scientific news.* EISSN 2392-2192. 267-278 (2016).
23. Mohammad, J., Katharina, M., Ashkarran, A. A., Antibacterial Properties Of nanoparticles, *Journal of Biotechnology,* **10:** 1016 (2014).
24. Nagarajan, S. and Kuppusamy, K. A., Extracellular synthesis of zinc oxide nanoparticles using seaweeds of gulf of Mannar, *Journal of Nanobiotechnology* **11:** 1-11 (2013).
25. Nanda, B., Rawat, A. K. S., Naqvi, A. H., Biosynthesis of Stable Antioxidant ZnO Nanoparticles by Pseudomonas aeruginosa Rhamnolipids, *Open Access journal,* **10:** 1371 (2014).
26. Arun1, P., Shanmugaraju1, V., Ramanujam, J. R., Senthil Prabhu, S., Kumaran, E., Biosynthesis of Silver nanoparticles From Corynebacterium sp. And its antimicrobial activity. *International Journal of Current Microbiology and Applied Science.* ISSN: 2319-7692 **2:** Number (2000).
27. Pandiyarasan, V., Shanmugapriya, P., Thanuja, Y. M., Anusuya, T. and Vairavaraja, P., Synthesis and Characterization of ZnO Nanoparticles- A Green Chemistry Approach. *Asian journal of Advance of Basic Science* **1:** 94-101 (2014).