

Synthesis of Zinc Oxide Nano Particles from Aloe Barbadensis for Medical Application

J. Shanmugapriya, P. Monisha,
A. Nandhini, K. Praveena

Abstract:- Nano sized ZnO particles of specific morphology were synthesized using the plant extract of Aloe Vera. In modern science nanotechnology is an ablaze field for researchers. Nanoparticles having a size of 1-100 nm in one dimension are used significantly concerning medical chemistry Atomic physics and all other known fields. Nanoparticles are used immensely due to its small size, orientation, physical properties.

Which are reportedly shown to change the performance Of any other material which is in contact with these tiny particles. The biological approach is the most emerging approach of preparation. The green synthesis was done by using the aloe Vera extract and zinc oxide. A fixed ratio of plant extract to metal ion was prepared and the color change was observed which proved the formation of nanoparticles. The nanoparticles Were characterized by UV- Vis spectrophotometer, FTIR analysis, XRD, SEM .

Keywords: Zinc oxide nanoparticles, Ultraviolet visible spectroscopy, Scanning electron microscopy, Antibacterial activity, Fourier transform infrared spectroscopy

INTRODUCTION

Nano materials have attracted tremendous interest due to their noticeable performance in electronics, optics, and photonics. nano materials are typically classified into three groups: 0-dimensional, 1-dimensional, 2dimensional, 0-dimensional nano structures, referred to as quantum dots or nanoparticles with an aspect ratio near unity, have been extensively used in biological applications. Nanotechnology emerges from the physical, chemical, biological and engineering sciences where new techniques are being developed to probe and maneuver single atoms and molecules for multiple applications in different field of scientific world.

Nanotechnology is the design, fabrication and application of nanostructures or nanomaterials and the fundamental understanding of the relationships between physical properties or phenomena and material dimensions. It is a new field or a new scientific domain. Nanotechnology also promises the possibility of creating nanostructures of metastable phases with non- conventional properties including superconductivity and magnetism. Another very important aspect of nanotechnology is the miniaturization of current and new instruments, sensors and machines that will greatly impact the world we live in. Examples of possible miniaturization are computers with infinitely great power that computer algorithms to mimic human brains,

biosensors that warn us at the early stage of the onset of disease and preferably at the molecular level and target specific drugs that automatically attack the diseased cells on site, nanorobots that can repair internal damage and remove chemical toxins inhuman bodies, nanoscaled electronics that constantly monitor our local environment. Nanomaterials have properties that are significantly different and considerably improved relative to those of their coarser-grained counter parts.

Nanoparticles are the simplest form of structures with sizes in the nm range. In principle any collection of atoms bonded together with a structural radius of < 100 nm can be considered a nanoparticle. These can include, e.g., fullerenes, metal clusters (agglomerates of metal atoms), large molecules, such as proteins, and even hydrogen bonded assemblies of water molecules, which exist in water at ambient temperatures. Nanoparticles are very commonplace in nature for instance proteins exist in almost all biological systems, metal oxide nanoparticles are easily produced, etc.

Zinc oxide is an inorganic compound with the formula ZnO. ZnO is a white powder that is insoluble in water, and it is widely used as an additive in numerous materials and products. A pure ZnO is a white powder, but in nature it occurs as the rare mineral zincite. Zinc oxide is an amphoteric oxide.it is nearly insoluble in water, but will dissolve in most acids. ZnO has high refractive index, high thermal conductivity, binding, antibacterial and UV protection properties. Aloe vera is succulent plant species of the genus aloe. An evergreen perennia, it originates from the Arabian Peninsula but grows wild in tropical climates around the world and is cultivated for agricultural and medicinal uses. The species is also used for decorative purposes and grows successfully indoors as a potted plant.

Aloe Barbadensis is the scientific name of Aloe Vera, it is a hardly, perennial, tropical, drought resistance, succulent plant belonging to the Liliaceae family and is very cactus like in its characteristics which, historically has been used for variety of medical purposes.

1.6 SCIENTIFIC CLASSIFICATION

Kingdom	-	Plantae
Clade	-	Angiosperms
Clade	-	Monocots
Order	-	Asparagales
Family	-	Asphodelaceae
Subfamily	-	Asphodeloideae
Genus	-	Aloe
Species	-	A.vera

BLOCK DIAGRAM

Green synthesis ZnO NPs



A Schematic diagram for the formation of ZnO NPs using Aloe vera leaf extract

1.7 BINOMIAL NAME (Aloevera)



ALOE VERA

GREEN SYNTHESIS OF ZnO NPS MATERIALS AND METHODS

EXTRACTION OF THE PLANT MATERIAL

All the chemical reagent used in this experiment were of analytical grade. The Aloe Vera leaves were collected. The fresh plant materials were washed with running tap water and shade dried. Then Aleogel broth extract at different concentrations were prepared with distilled water and the volume was made upto 100ml. the collected extracts were stored and then taken up for further investigations.



ALOE VERA GEL EXTRACT (10g)

SYNTHESIS OF ZnO NANOPARTICLES

Preparation of zinc oxide NPs or the synthesis of NPs, 10 g of aloe vera gel – extract was taken and boiled at 50°-60° C by using a stirrer heater. Then, 8.8461 g of zinc nitrate was added to the solutions as temperatures reached at 60°C. This mixture was then boiled until it converted to a deep yellow coloured suspension. This paste was then collected in a ceramic crucible and heated in an air heated furnace at 60°C for 2h. A light white coloured powder

was obtained and this powder was carefully collected and sent for different characterizations.



a) Boiling of aloe vera gel with Distilled water (50°-60°C)

**b) After boiling of aloe vera gel Again
boiled the solution at 60°C**



Zinc nitrate

$$=294.87 \times 0.3 \times 100 / 1000 = 8.8461\text{g}$$



**Deep yellow coloured
suspension**



ZINC OXIDE NANOPARTICLES

RESULTS AND INTERPRETATION

Characterization of zinc oxide nanoparticles

The sample is characterized by

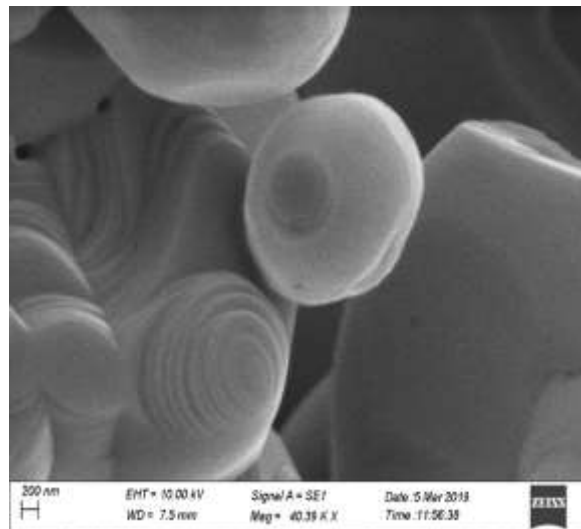
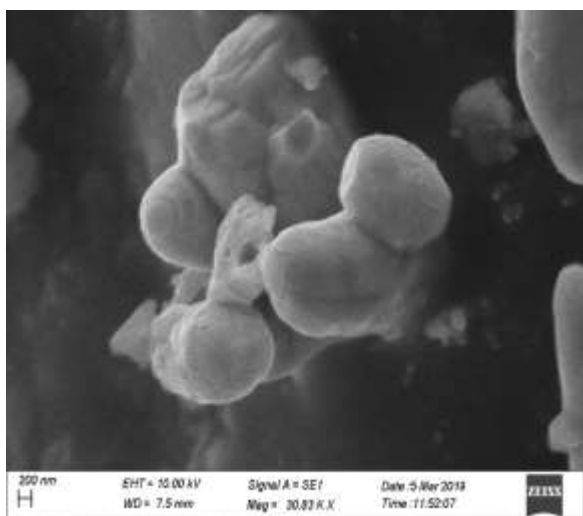
1. Scanning electron microscopy (SEM),
2. UV-Visible spectroscopy (UV-Vis) and
3. Fourier transform infrared spectroscopy (FTIR).

SCANNING ELECTRON MICROSCOPY (SEM) ANALYSIS

The morphology of the synthesized ZnO NPs was examined using SEM. Samples for SEM analysis was prepared by the Nano-composites on carbon-tab. The copper tab was subjected to SEM analysis by the instrument Carl Zeiss 55 model.

SEM is one of the promising techniques for the topography study of the sample and it gives important information regarding the shape and size of the nanoparticles. The surface morphology of the ZnO NPs synthesized using *Aloe Vera* leaf extract. The entire SEM image clearly shows the average size is **30-50 nm** for ZnO NPs. The ZnO NPs is **spherical and hexagonal** shape.

SEM images of the ZnO NPs using *Aloe vera* leaf extract.



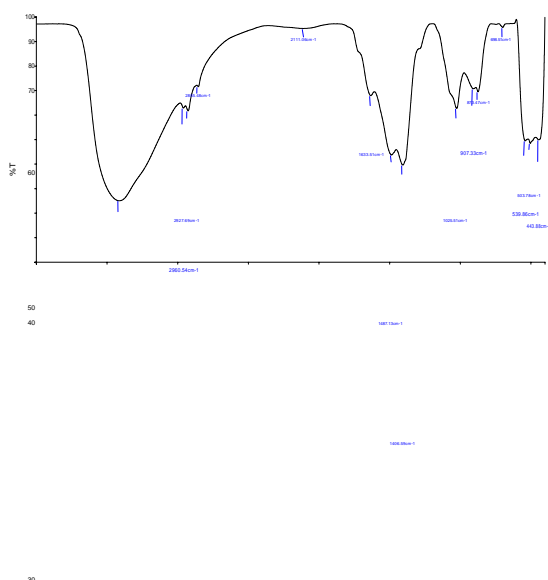
FOURIER TRANSFORM-INFRARED SPECTROSCOPY (FT-IR) ANALYSIS

A Perkin-Elmer Fourier Transform Infra-Red (FT-IR) spectrometer was used in transmission mode and the corresponding spectra were recorded in the range of $4000-400\text{ cm}^{-1}$ using the KBr pellet

technique for ZnO NPs.

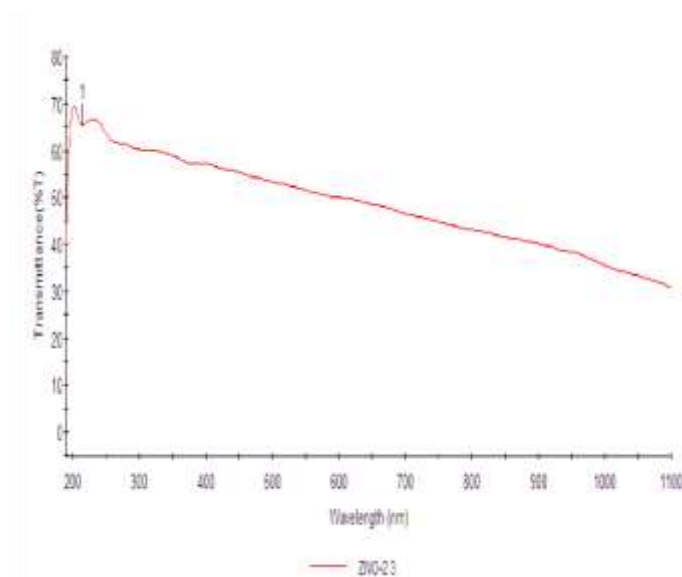
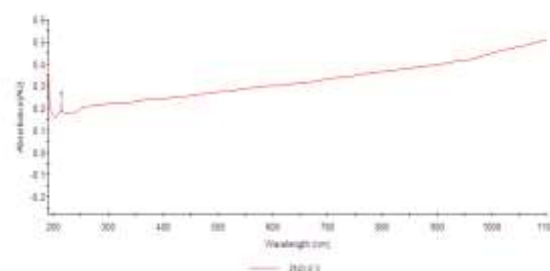
The broad absorption in the frequency band $3750-3000\text{ cm}^{-1}$ is assigned to O-H stretching from residual alcohols, water and Zn-OH. The peaks at 2860 and 2950 cm^{-1} are due to symmetric and asymmetric C-H bonds. At 2852 and 2922 cm^{-1} , the sharp intensity of the symmetry and asymmetric stretching mode of the CH_2 groups observed for ZnO NPs. The absorption bands are caused by the vibration in CO_3^{2-} between 400 and 1800 cm^{-1} . The absorption bands are observed at 1712 cm^{-1} for ZnO NPs. The C=C stretching group around 1643 cm^{-1} is

responsible for *Aloe Vera L.* capping on the ZnO samples. The symmetric stretching vibration of C=O are observed at 1434 cm^{-1} . The Zn-O stretching bands appear at 440 cm^{-1} for the ZnO sample.



The absorption spectra of ZnO NPs sample was studied in the range between 200 and 1100nm by Lambda 35 spectrometer.

The absorbance of the sample is depended on several factors such as band gap, oxygen deficiency, surface roughness and impurity centers. The excitonic peak is observed around **220nm** for ZnO NPs.



Name	No.	Peak(nm)	Peak(AU)	No.	Valley(nm)	Valley(AU)
------	-----	----------	----------	-----	------------	------------

ZNO-2	1	214.4	0.184			
-------	---	-------	-------	--	--	--

Name	No.	Peak(nm)	Peak(%T)	No.	Valley(nm)	Valley(%T)
ZNO-2	1	214.4	65.484			

Anticancer assay

MTT ASSAY FOR CELL CYTOTOXICITY

PRINCIPLE

MTT (3-(4, 5 dimethylthiazol-2-yl)-2, 5-diphenyl tetrazolium bromide) assay, is based on the ability of a mitochondrial dehydrogenase enzyme of viable cells to cleave the tetrazolium rings of the **pale yellow MTT and form a dark blue colored formazan crystals** which is largely impermeable to cell membranes, thus resulting in its accumulation within healthy cells. Solubilization of cells by the addition of detergents (DMSO) results in the liberation of crystals which are solubilized. The number of surviving cells is directly proportional to the level of formazan product created. The color can be quantified using a multi-well plate reader.

MATERIALS REQUIRED

DMEM(Dulbecco's Modified Eagle Medium) liquid medium, Fetal Bovine Serum (FBS) and antibiotic solution were from Gibco (USA), DMSO (Dimethyl sulfoxide) and MTT (3-(4,5 dimethylthiazol-2-yl)-2,5-diphenyl tetrazoliumbromide)(5 mg/ml) were from Sigma, (USA), 1X PBS(phosphate buffered saline) was from Himedia, (India). 96 well tissue culture plate and wash beaker were from Tarson (India).

PROCEDURE

Cell culture

MCF-7 (Human breast carcinoma cells) cell line were cultured in liquid medium (DMEM) supplemented 10% Fetal Bovine Serum (FBS), 100 u/ml penicillin and 100 µg/ml streptomycin, and maintained under an atmosphere of 5% CO₂ at 37°C.

MTT Assay

The ZnO sample was tested for in vitro cytotoxicity, using MCF-7 cells by 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. Briefly, the cultured MCF-7 cells were harvested by trypsinization, pooled in a 15 ml tube. Then, the cells were plated at a density of 1×10^5 cells/ml cells/well (200 µL) into 96-well tissue culture plate in DMEM medium containing 10 % FBS and 1% antibiotic solution for 24-48 hour at 37°C.

The wells were washed with sterile PBS and treated with various concentrations of the ZnO sample in a serum free DMEM medium. Each sample was replicated three times and the cells

were incubated at 37°C in a humidified 5% CO₂ incubator for 24 h. After the incubation period, MTT (20 µL of 5 mg/ml) was added into each well and the cells incubated for another 2-4 h until purple precipitates were clearly visible under an inverted microscope.

Finally, the medium together with MTT (220 µL) were aspirated off the wells and washed with 1X PBS (200 µl). Furthermore, to dissolve formazan crystals, DMSO (100 µL) was added and the plate was shaken for 5 min. The absorbance for each well was measured at 570 nm using a micro plate reader (Thermo Fisher Scientific, USA) and the percentage cell viability and IC₅₀ value was calculated using GraphPad Prism 6.0 software

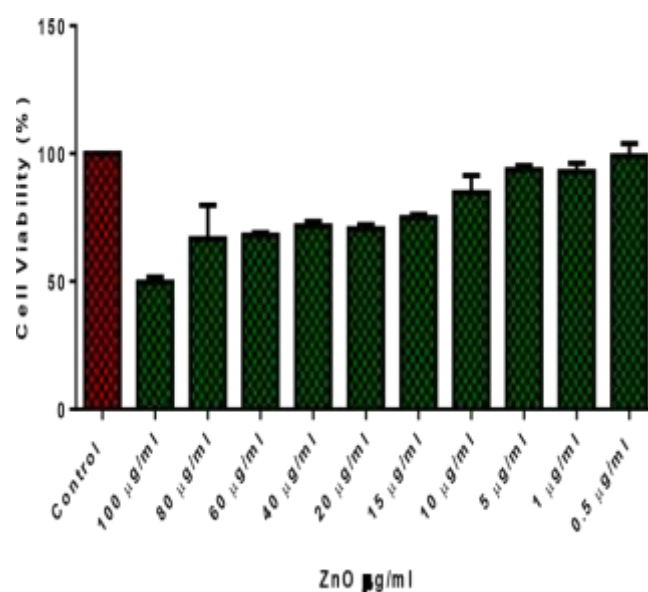
(USA)

Anticancer properties

The ZnO was tested for in vitro cytotoxicity, using MCF-7 (Human Breast cancer cells) cell line were incubated with different (0.5-100 µg/ml) concentrations of ZnO as shown in Fig. 3. In MTT assay, the IC₅₀ value of 22.23 µg/ml (evaluated

after 24h) of ZnO samples against MCF-7 cells was ($p \leq 0.05$ P value <0.01).

ANTICANCER PROPERTY ZnO NP



CONCLUSION

Nanoparticles present an extremely attractive platform for the diverse range of applications. The single step process for biosynthesis of nanoparticles provided by it attracts more researches to go for future development in the area of electrochemical sensor, biosensor, medicine, healthcare and agriculture.

In this review, ZnO nanoparticles synthesized by using Aloe Vera and various studies of that ZnO nanoparticles are articulated. The synthesis of nanoparticles in nano biotechnology area has augmented its importance to create ecofriendly; cost effective, stable nanoparticles.

REFERENCES

- [1] Agarwal S and Sharma TR, 2011. Multiple biological activities of Aloe barbadensis (A. vera): an overview. Asian Journal of Pharmacy and Life Science 1(2) 195-205.
- [2] Ayeshamariam, M. Kashif, V. S. Vidhya, M. G. V. Sankaracharyulu, V. Swaminathan, M. Bououdinaf, G. M. Jayachandran, 2014.
- [3] Biosynthesis of (ZnO– Aloe Vera) Nano composites and Antibacterial/ Antifungal Studies. Journal of Optoelectronics and Biomedical Materials Vol. 6, Issue 3, 85 - 99
- [4] Bunyapraphatsara N, Yongchaiyudha S, Rungpitarangsi V and Chokechaijaroenporn O, 1996. Antidiabetic activity of Aloe vera L. juice. Phytomedicine 3: 245– 248.
- [5] Elizabeth Varghese and Mary George, 2015. Green Synthesis of Zinc Oxide Nanoparticles, International Journal of Advance Research in Science and Engineering, Vol. No.4, Issue No.01.
- [6] Hoshino A., K. Fujioka, T. Oku, 2004.—Quantum dots targeted to the assigned organelle in living cells, Microbiology and Immunology, vol. 48, no. 12, pp. 985– 994
- [7] Jain N, Bhargava A, Tarafdar J C, Singh S K and Panwar J A 2013 J. Appl. Microbiological. Biotechnol. 97 859
- [8] Sutradhar P, Saha M and Maiti D 2014 J. Nanostruct. Chem. 4 86
- [9] Chandran S P, Chaudhary M and Pasricha R 2006 Biotechnol. Prog. 22 577
- [10] Susan A, Ahmada B Mansor, Farideh Namvar and Rosfarizan Mohamad 2013 Materials 6 5942
- [11] Waltera Michael G, Rudineb Alexander B and Wamser Carl C 2010 J. Porphyrin Phthalocyanines 14 759
- [12] P. Mukherjee, R. Bhattacharya, N. Bone, Y. K. Lee, C. R. Patra, S. Wang, L. Lu, C. Secreto, P. C. Banerjee, M. J. Yaszemski, N. E. Kay and D. Mukhopadhyay, J. Nanobiotechnol., 2007, 5, 4.
- [13] S. Kotthaus, B. H. Gunther, R. Hang and H. Schafer, IEEE Trans. Compon., Packag., Manuf. Technol., Part A, 1997, 20, 15–20. [14] G. Cao, Nanostructures and Nanomaterials: Synthesis, Properties and Applications, Imperial College Press, London, 2004.
- [15] W. Zhang and G. Wang, New Chem. Mater., 2003, 31, 42– 44. 36
- T. Klaus-Joerger, R. Joerger, E. Olsson and C. G. Granqvist, Trends Biotechnol., 2001, 19, 15–20.
- [16] K.-H. Cho, J.-E. Park, T. Osaka and S. G. Park, Electrochim. Acta, 2005, 51, 956–960.
- [17] N. Dura'n, P. D. Marcato, S. De, I. H. Gabriel, O. L. Alves and E. Esposito, J. Biomed. Nanotechnol., 2007, 3, 203–208.
- [18] A. Ahmad, P. Mukherjee, S. Senapati, D. Mandal, M. I. Khan, R. Kumar and M. Sastry, Colloids Surf., B, 2003, 28, 313–318.
- [19] M. Jose-Yacaman, J. A. Ascencio, H. B. Liu and J. L. GardeaTorresdey, J. Vac. Sci. Technol., B, 2001, 19, 1091–1103.
- [20] T. Osaka, T. Matsunaga, T. Nakanishi, A. Arkaki, D. Niwa and H. Iida, Anal. Bioanal. Chem., 2006, 384, 593–600.

- [21] S. Sun, H. Zeng, D. Robinson, S. Raoux, P. M. Rice, S. X. Wang and G. Li, J. Am. Chem. Soc., 2004, 126, 273–279.
- [22] R. Herrera-Becerra, C. Zorrilla, J. L. Rius and J. A. Ascencio, Appl. Phys. A: Mater. Sci. Process., 2008, 91, 241–246.
- [23] E. al din. Haratifar, H. R. Shahverdi, M. Shakibaie, K. MollazadehMoghaddam, M. Amini, H. Montazeri and A. R. Shahverdi, J. Nanomater., 2009, 962021.
- [24] V. Armendariz, M. Jose-Yacaman, A. Duarte Moller, J. R. PeraltaVidea, H. Troiani, I. Herrera and J. L. Gardea-Torresdey, Rev. Mex. Fis. Suppl., 2004, 50, 7–11.
- [25] J. L. Gardea-Torresdey, K. J. Tiemann, J. G. Parsons, G. Gamez and M. Jos'e-Yacam'an, Adv. Environ. Res., 2002, 6, 313–323. 61
- [26] V. Armendariz, I. Herrera, J. R. Peralta-Videa, M. Jos'e-Yacam'an, H. Troiani, P. Santiago and J. L. Gardea-Torresdey, J. Nanopart. Res., 2004, 6, 377–382.
- [27] S. S. Shankar, A. Ahmad, R. Pasricha and M. Sastry, J. Mater. Chem., 2003, 13, 1822–1826.
- [28] J. Huang, L. Lin, Q. Li, D. Sun, Y. Wang, Y. Lu, N. He, K. Yang, X. Yang, H. Wang, W. Wang and W. Lin, Ind. Eng. Chem. Res., 2008, 47, 6081–6090.
- [29] T. Harris and R. Bali, J. Nanopart. Res., 2007, 10, 691–695.
- [30] R. G. Haverkamp, A. T. Marshall and D. van Agterveld, J. Nanopart. Res., 2007, 9, 697–700.