Tropical Plant Research
An International Journal
© Society for Tropical Plant Research

ISSN (E): 2349 – 1183
ISSN (P): 2349 – 9265
1(2): 55–59, 2014

Research article

A biomimetic approach towards synthesis of Zinc oxide Nanoparticles using *Hybanthus enneaspermus* (L.) F. Muell.

M.S. Shekhawat[1]*, C. P. Ravindran[1] and M. Manokari[2]

[1] Biotechnology Lab. Postgraduate Department of Plant Science, Mahatma Gandhi Govt. Arts College, Mahe, Pondicherry, India
[2] Biotechnology Unit, K.M. Centre for Post Graduate Studies, Pondicherry, India
*Corresponding Author: smahipal3@gmail.com [Accepted: 22 August 2014]

Abstract: We report green approach for the synthesis of Zinc oxide (ZnO) nanoparticles from the aqueous extracts of leaves, stem and roots of *Hybanthus enneaspermus* (L.) F. Muell in present study. *H. enneaspermus* plant has been used in traditional system of medicines in India, but so far it has not been tested for synthesis of Zinc oxide nanoparticles. The nanoparticles were synthesized using Zinc Nitrate hexahydrate [$Zn(NO_3)_2.6H_2O$] solution. The Zinc Nitrate solution was exposed to the herbal extracts, which was reduced and the nanoparticles were synthesized. The formation of nanoparticles was confirmed by the color change of the reaction mixture. The synthesized ZnO nanoparticles were characterized by UV-Vis spectrophotometeric analysis. The reaction mixtures leaf extract showed absorption peak at 300 nm, stem extract at 290 nm and root extract at 288 nm.
Keywords: Green synthesis - Zinc oxide nanoparticles - *Hybanthus enneaspermus*.

[**Cite as:** Shekhawat MS, Ravindran CP & Manokari M (2014) A Biomimetic Approach towards Synthesis of Zinc oxide Nanoparticles using *Hybanthus enneaspermus* (L.) F. Muell. *Tropical Plant Research* 1(2): 55–59]

INTRODUCTION

Nanoparticles are of great scientific interest, and they represent a bridge between the bulk materials and the molecules. Earlier, the nanoparticles were studied because of their size-dependent physical and chemical properties but now they have entered in commercial exploration with huge applications (Shearer *et al.* 2000). The intrinsic properties of metal nanoparticles are due to their size, composition and morphology (Dickson & Lyon 2000). These nano size metal oxides have been attracted researchers by their ability to withstand under harsh condition and safe to mankind (Fu *et al.* 2005).

Zinc oxide nanoparticles (ZnONPs) are versatile elements with various biomedical properties like, antimicrobial (Rajendran *et al*. 2010), antibacterial activities (Zhang *et al.* 2010) etc. ZnO nanoarticles are among the top most photocatalysts which are used in disinfecting waste water and to decompose pesticides, herbicides (Zhao *et al.* 2010) etc. Biosynthesis of ZnO nanoparticles and their use in various fields are reported by many authors (Bagabas *et al.* 2013, Malarkodi *et al.* 2013).

Biogenesis of Zinc oxide nanoparticles using plant parts was reported in *Corriandrum sativum, Acalypa indica* (Gnanasangeetha & Sarala 2013a,b), *Aloe barbadensis* (Sangeetha *et al.* 2011), *Morinda pubescens, Passiflora foetida* (Shekhawat *et al.* 2014a,b) etc.

Hybanthus enneaspermus (L.) F. Muell. belongs to family Violaceae, is commonly called Ratanpurus, Sthalakamala, Gem for men, Spade flower etc. (Kirtikar & Basu 1975). Traditionally *H. enneaspermus* plant is used to treat malaria, diabetes, male sterility, gonorrhea, urinary tract infection, jaundice, Cholera etc. (Sarita *et al.* 2004, Patel *et al.* 2011, Kheraro & Bouquet 1950, Pushpangadan & Atal 1984, Gopal & Shah 1985). This plant is also used to improve memory and to treat asthma, tuberculosis, eye diseases etc. (Udayan & Indira 2009).

The aqueous extract of *H. enneaspermus* was used for the biosynthesis of silver nanoparticles by Sripriya *et al.* (2013). As per the literature survey no data were available regarding the biogenesis of Zinc oxide nanoparticles using *H. enneaspermus*. The present study intended to synthesize and characterize ZnO nanoparticles from various extracts of this valuable medicinal plant for the first time.

MATERIAL AND METHODS

Collection of plant material and preparation of broth solutions

Received: 19 May 2014 Published online: 31 August 2014

Figure 1. *Hybanthus enneaspermus* (L.) F. Muell. in natural habitat.

Hybanthus enneaspermus is an important ethno-medicinal herb (Fig. 1). The plant material for the present study was collected from the campus of K.M. Centre for Postgraduate Studies, Pondicherry, India. The leaf, stem and roots were collected from the healthy plants and washed thoroughly with distilled water, and finely cut into small pieces (Fig. 2,3 & 4A,B).

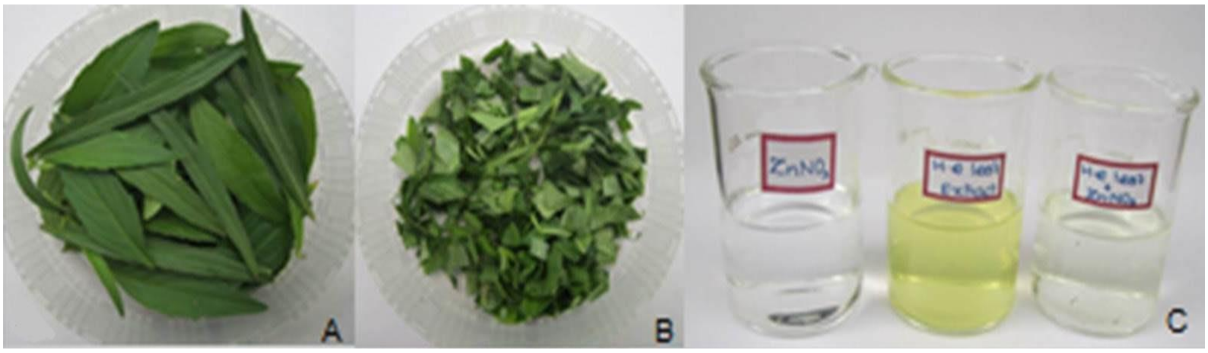


Figure 2. A, Mature leaves, **B,** Leaves in small pieces; **C,** Zinc Nitrate solution, aqueous extract of leaves and the mixture.

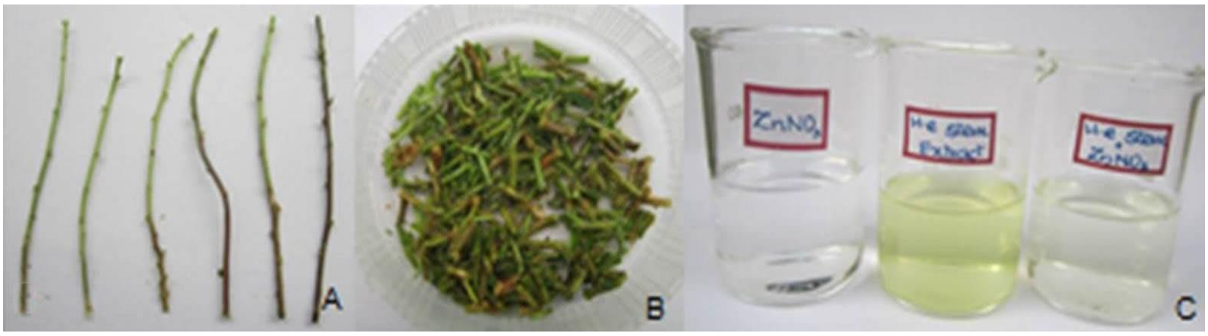


Figure 3. A–B, Stem cuttings; **C,** Zinc Nitrate solution, aqueous extract of stem and the mixture.

5 gm of chopped plant parts were boiled in a clean and sterilized conical flask of desired size with 50 ml of double distilled water for 5 min. to prepare broth solution. The extracts were filtered with Whatman filter paper No.1 after boiling and stored in refrigerator for further study.

Preparation of precursors and synthesis of Zinc oxide nanoparticles

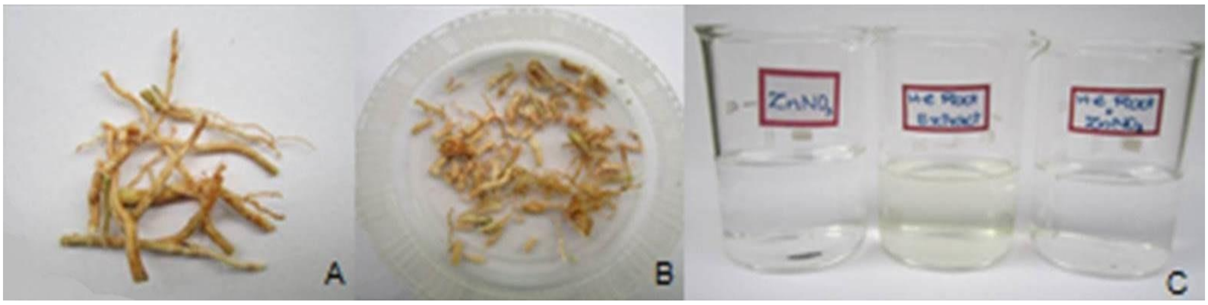


Figure 4. A–B, Root cuttings; **C,** Zinc Nitrate solution, aqueous extract of roots and the mixture.

Zinc Nitrate hexahydrate [$Zn(NO_3)_2.6H_2O$] (Merck, Mumbai, India) was used as precursor to synthesize ZnO nanoparticles using *H. enneaspermus.* 1 mM Zinc nitrate solution was prepared using Zinc Nitrate hexahydrate with double distilled water and stored in refrigerator at 4^{o}C for further use. Three boiling tubes were taken for the synthesis process; one containing 10 ml of 1 mM Zinc nitrate solution as control, the second tube containing 10 ml of broth solution from appropriate part of the plant to observe the color change and the third tube containing 9 ml of 1 mM Zinc nitrate solution and 1 ml of plant extracts as test solution.

Characterization of nanoparticles

The synthesized Zinc oxide nanoparticles using the plant extracts were centrifuged at 5000 rpm for 15 min to obtain the pellet which was used for further study. Supernatant was discarded and the pellet dissolved in deionized water. The synthesis of Zinc oxide nanoparticles were confirmed and characterized by using UV-Visible spectrophotometer (Model 2202, Systronics Ltd. India). The UV-Vis absorption spectra of the zinc colloids from various parts of the plants were confirmed by using wave length scan between 200 nm and 700 nm.

RESULTS AND DISCUSSION

The ethnobotanical herb *Hybanthus enneaspermus* has been reported to have anti-inflammatory, antitussive, antiplasmodial, anticonvulsant, anti-bacterial, anti-oxidant, antifungal, hypolipidemic and free radical scavenging activities (Boominathan *et al.* 2004, Sahoo *et al.* 2006, Satheesh & Kottai 2012, Patel *et al.* 2011, Arumugam *et al.* 2011). This plant is said to be rare because of its seasonal habitat and sporadic distribution (Prakash *et al.* 1999).

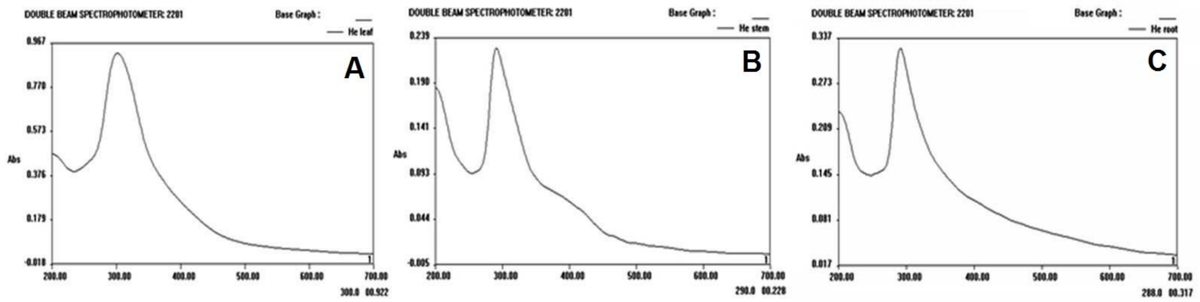
The bio-nano-synthesis of zinc oxide nanoparticles using leaf, stem and root extracts of *Hybanthus enneaspermus* was investigated in the present study. The color change was observed in the test solution of leaf and stem from colorless to pale yellow when 1 ml of broth solution was challenged with 9 ml of Zinc nitrate solution. The color was not changed in case of root extract without heating the mixture (Fig. 2,3 & 4C) even after two hours. Color change from colorless to pale yellow was observed when the test solution was heated in the oven at 60^o C for ten minutes, which was disappeared while cooling. The present observations were also confirmed by our previous study (Shekhawat *et al.* 2014a, 2014b).

Plants attributed the way to synthesize nanoparticles through green method because of their green chemistry principle. Sripriya *et al.* (2013) biosynthesized silver nanoparticles using *H. enneaspermus* and used these nanoparticles in antibacterial coatings and drug delivery applications. Zinc oxide nanoparticles are very special among the existing nanomaterials due to their organometallic properties. Now it is possible to prepare individual nano metal or oxide particles.

The whole plant body of *H. enneaspermus* has evident to possess aphrodisiac activities and it has significant role in maintaining maleness. The Siddha medicine explains it as rejuvenating herb and it is known to possess coumarin, which is responsible for the hypolipidemic activity (Senthil *et al.* 2013, Narayanaswamy *et al.* 2007). We strongly believe that the phytochemicals, like coumarin are working as reducing agent and responsible for the conversion of metallic oxide into nanoparticles.

The synthesized ZnO nanoparticles were characterized by UV-Vis spectrophotometeric analysis. The aqueous leaf extract showed absorption peak at 300 nm, stem extract at 290 nm and the root extract at 288 nm (Fig. 5). Jain *et al.* (2014) also observed same types of results and described the biological approach for the synthesis of Zinc oxide nanoparticles.

The absorption wavelength at about 300 nm of ZnO suggested the excitonic character of Zinc at room temperature. Vanheusden *et al.* (1996) described that the UV emission is attributed to the radiative recombination between the electrons in the conduction band and the holes in the valence band while working on ZnO phosphor powders.



Figures 5. A, Spectrophotometric absorbance peak of leaf reaction mixture; **B,** Spectrophotometric absorbance peak of root reaction mixture; **C,** Spectrophotometric absorbance peak of stem reaction mixture.

CONCLUSION

This is the first study to develop an efficient protocol for the biosynthesis of ZnO nanoparticles using *H. enneaspermus* to highlight eco-friendly approach for commercial application of Zinc nanoparticles in agriculture as nano-bio-fertilizers and in the field of medicine.

ACKNOWLEDGEMENTS

Authors are grateful to Department of Science, Technology and Environment, Govt. of Puducherry for providing financial support.

REFERENCES

Arumugam N, Sasikumar K, Malipeddi H & Sekar M (2011) Antifungal activity of *Hybanthus enneaspermus* on wet clothes. *International Journal of Research in Ayurveda & Pharmacy* 2(4): 1184–1185.

Bagabas A, Alshammari A, Aboud MFA & Kosslick H (2013) Room-temperature synthesis of zinc oxide nanoparticles in different media and their application in cyanide photodegradation. *Nanoscale Research Letters* 8(1): 516.

Boominathan R, Parimaladevi B, Mandal SC & Ghoshal SK (2004) Anti-inflamattory evaluation of *Ionidium suffruticosum* Ging. in rats. *Journal of Ethnopharmacology* 91: 367–370.

Dickson RM & Lyon LA (2000) Unidirectional plasmon propagation in metallic nanowires. *Journal of Physical Chemistry* 104: 6095–6098.

Fu L, Liu Z, Liu Y, Han B, Hu P, Cao L & Zhu D (2005) Beaded Cobalt oxide nanoparticles along carbon nanotubes: towards more highly integrated electronic devices. *Advanced Materials* 7: 217–221.

Gnanasangeetha D & Sarala TD (2013a) One pot synthesis of Zinc oxide nanoparticles via chemical and green method. *Research Journal of Material Science* 1(7): 1–8.

Gnanasangeetha D & Salara TD (2013b) Biogenic Production of Zinc Oxide Nanoparticles Using *Acalypha indica*. *Journal of Chemical Biological and Physical Sciences* Sec B 4(1): 238–246.

Gopal GV & Shah GL (1985) Some folk medicinal plants used for jaundice in Gujarat, India. *Journal of Research and Education in Indian Medicine* 4: 45.

Jain N, Bhargava A & Panwar J (2014) Enhanced photocatalytic degradation of methylene blue using biologically synthesized "protein-capped" ZnO nanoparticles. *Chemical and Engineering Journal* 243: 549–555.

Kheraro J & Bouquet A (1950) In-Plantes medicinales et toxiques de la cote d'lvoire-Haute-Volta. Vigot Freres, Paris, pp. 170.

Kirtikar KR & Basu BD (1975) Indian Medicinal Plants, *Vol. I, 2nd Edition*. International Book Distributors, Dehradun, India. pp. 212.

Malarkodi C & Annadurai G (2013) A novel biological approach on extracellular synthesis and characterization of semiconductor Zinc sulfide nanoparticles. *Applled Nanosciece* 3: 389–395.

Narayanaswamy VB, Manjunatha SM, Malini S & Annie Shirwaikar (2007) Preliminary aphrodisiac activity of *Hybanthus enneaspermus* in rats. *Pharmacologyonline* 1: 152–161.

Patel DK, Kumar R, Prasad SK, Sairam K & Hemalatha S (2011) Antidiabetic and *in vitro* antioxidant potential of *Hybanthus enneaspermus* (Linn.) F. Muell in streptozotocin-induced diabetic rats. *Asian Pacific Journal of Tropical Biomedicine* 1(4): 316–322.

Prakash E, Sha Vallikhan PS, Sairam Reddy P & Rao KR (1999) Regeneration of plants from seed-derived callus of *Hybanthus enneaspermus* (L) Muell., a rare ethnbotanical herb. *Journal of plant cell reports* 18(10): 873–878.

Pushpangadan P & Atal CK (1984) Ethno botanical investigations in Kerala, Some primitive tribal of Western Ghats and their herbal medicine. *Journal of Ethnomorpharmocology* 11: 59–77.

Rajendran R, Balakumar, Hasabo AM, Jayakumar S, Vaideki K & Rajesh EM (2010) Use of Zinc oxide nano particles for production of antimcrobial textiles. *International Journal of Engineering, Science and Technology* 2(1): 202–208.

Sahoo S, Kar DM, Mohapatra S, Rout SP & Dash SK (2006) Antibacterial activity of *Hybanthus enneaspermus* against selected urinary tract pathogens. *Indian Journal of Pharmaceutical Science* 68(5): 653–655.

Sangeetha G, Rajeshwari S & Venckatesh R (2011) Green synthesis of Zinc oxide nanoparticles by *Aloe barbadensis* miller leaf extract: Structure and optical properties. *Materials Research Bulletin* 46: 2560–2566.

Sarita D, Dash SK & Padhy SN (2004) Ethno botanical information from Orissa State, India. A Review. *Journal Human Ecology* 14(3): 227.

Satheesh SK & Kottai MA (2012) Comparative evaluation of flavone from *Mucuna pruriens* and coumarin from *Ionidium suffruticosum* for hypolipidemic activity in rats fed with high Fat diet. *Lipids Health Dis* 11: 126.

Senthil KB, Vijaya KJ & Selvaraj R (2013) Aphrodisiac activity of *Cycas circinalis* L. and *Ionidium suffruticosum*. Ging on male Wister albino rats. *Asian Journal of Pharmaceutical and Clinical Research* 6(3): 215–217.

Shearer AEH, Paik JS, Hoover JG, Haynie SL & Kelley MJ (2000) Potential of an antibacterial ultraviolet-irradiated nylon film. *Biotech and Bioengineering* 67: 141–146.

Shekhawat MS & Manokari M (2014a) Biogenesis of Zinc oxide Nanoparticles using *Morinda pubescens* J.E. Smith Extracts and their Characterization. *International Journal of Bio Engineering and Technology* 5(1): 1–6.

Shekhawat MS, Ravindran CP & Manokari M (2014b) Biosynthesis of Zinc oxide nanoparticles from *Passiflora foetida* L. Extracts and their Characterization. *International Journal of Green and Herbal Chemistry* 3(2): 518–523.

Sripriya J, Anandhakumar S, Achiraman S, Antony JJ, Siva D & Raichur AM (2013) Laser receptive polyelectrolyte thin films doped with biosynthesized silver nanoparticles for antibacterial coatings and drug delivery applications. *International Journal of Pharmacy* 457(1): 206–13.

Udayan PS & Indira B (2009) *Medicinal plants of Aryavaidyasala herbs garden*. Aryavaidyasala, Kottakkal publications, Kerala, India. pp. 199.

Vanheusden WL, Warren C, Seager H, Tallant DR, Voigt JA & Gnade BE (1996) Mechanisms behind green photoluminescence in ZnO phosphor powders. *Journal of Applied Physics* 79: 7983–7990.

Zhang L, Jiang Y, Ding Y, Daskalakis N, Jeuken L, Povey M, Alex J, O'Neill & York DW (2010) Mechanistic investigation into antibacterial behaviour of suspensions of ZnO nanoparticles against *E. coli*. *Journal of Nanoparticle Research* 12: 1625–1636.

Zhao L, Lu PF, Yu ZY, Guo XT, Shen Y, Ye H, Yuan GF & Zhang L (2010) The electronic and magnetic properties of (Mn, N)-codoped ZnO from first principles. *Journal of Applied Physics* 108: 113924–113930.