GREEN APPROACH FOR FABRICATION OF ZnO NANO PARTICLES FROM EICHHORNIA CRASSIPES AND ITS ANTI-OXIDANT, ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY

Abstract

Aquatic weed plant, Eichhornia crassipes is utilized for this synthesis. The nanoparticles were prepared using the Zinc nitrate and plant extract of the Eichhornia crassipes. White coloured paste procured and then desiccated and used for further studies. The characterization of ZnO carried out using FESEM, XRD, EDX, and FTIR. FESEM detects the presence of ZnO nanoparticle's surface. XRD is applied to predict the size of the nanoparticles was estimated to be 54 nm. EDX results denote the composition of Zinc and Oxygen. Fourier Transform infra red spectra analysis interprets absorption frequency of all functional of synthesized nanoparticle. The thermal activity checked by TGA/DTG. The biological properties of ZnO Nps such as antioxidant, antimicrobial were examined.

Keywords: XRD, EDX, TGA/DTG, FESEM, Eicchornia crassipes, ZnONps

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I. INTRODUCTION

Nanotechnology involves various applications of biotechnology [1, 2]. The utilization of metal oxide nanoparticles for antimicrobial, antibacterial, antifungal, anticorrosion activities against various microorganisms is more focused in recent years[3]. The biological activity of nanoparticles hinges on their dimension. The magnitude is very important because they can enter bacterial plasma membrane. Metal oxide nanoparticles give rise to hydrogen peroxide in the cell. This property of metal oxide nanoparticles utilized for lesion rehab [4]. Furthermore, metal oxide nanoparticles used as an antimicrobial agent because of their security, strength and usage against a extensive range of infectious agent bacteria [5]. Phyto chemical compositions of plants generate antioxidant property to the nanoparticles [6].

Nano ZnO particle can be used in various fields due to its special physical, chemical properties and its applications [7,8]. In the manner of many literately studies, plants extract has been suggested as beneficial proxy to other method of synthesis [9].

The extensive growth of this weed plant creates enormous problems like eutrophication, depletion of oxygen etc. A proper method of disposal is yet to be found. Green synthesis of nanoparticles using Eichhornia crassipes is gaining importance nowadays which may be a better alternative for its disposal by a constructive mean. Green synthesis of ZnO nanoparticles from the aqueous extract of Eichhornia crassipes leaves using NaOH, its characterization and its antibacterial, antifungal, anti-oxidant activity were reported in this work.

II. EXPERIMENTAL

1. **Materials:** Zinc Nitrate hexahydrate, sodium hydroxide,ethanol used in this work was purchased from Merck and sigma-Alrich chemicals respectively.

2. **Preparation of Eichhornia Crassipes Leaf Extract:** Eichhornia crassipes leaves collected from kuruchi lakes at Coimbatore, Tamilnadu, India. It was washed with distilled water thoroughly. The plant boiled in distilled water at 50^0C for 30 minutes and then filtered with using whatmann 40 filterpaper. It was cooled and stored in refrigerator for further studies.

3. **Synthesis of ZnONps from Eichhornia Crassipes Leaf-Extract:** Zinc Nitrate used as precursor. 1M Zinc nitrate solution prepared and heated for an hour. It is mixed with plant extract under constant stirring for one hour.Sodium hydroxide was added in the solution for maintain pH range and then stirred for 30 minutes. White precipitate obtained which was filtered and dried.

4. **Characterization of Synthesized Eichhornia Crassipes Leaf- ZnONps:** Crystallinity and crystal phases were identified with X-Ray Diffraction [Perkin Elmer] spectrometer. Functional groups in Eichhornia crassipses and the presence of Zinc oxide analysis was done with FTIR [Shimadzu] Spectrometer. Elemental Analysis was done with the Energy Dispersive X-Ray [RONTEC'S EDX system] spectrometer. Particle morphology was analyzed by Scanning Electron Microscope [Model JSM6390LV]. The thermal analysis of ZnO nanoparticles was done by TGA/DTG.

III. RESULT AND DISCUSSION

1. **X-ray Diffraction Spectroscopy:** XRD results of ZnNPs are illustrated in figure 1. The XRD spectra of ZnNPS depicted the sharp peak at 35^0,38^0 and confirm single monoclinic structure. There are no any other peaks shows no impurities present in the synthesized ZnO nano particles. The magnitude of the nano particle is determined by XRD using Debye Scherer equation. The Size of the ZnO NPs is 24-54nm[10].

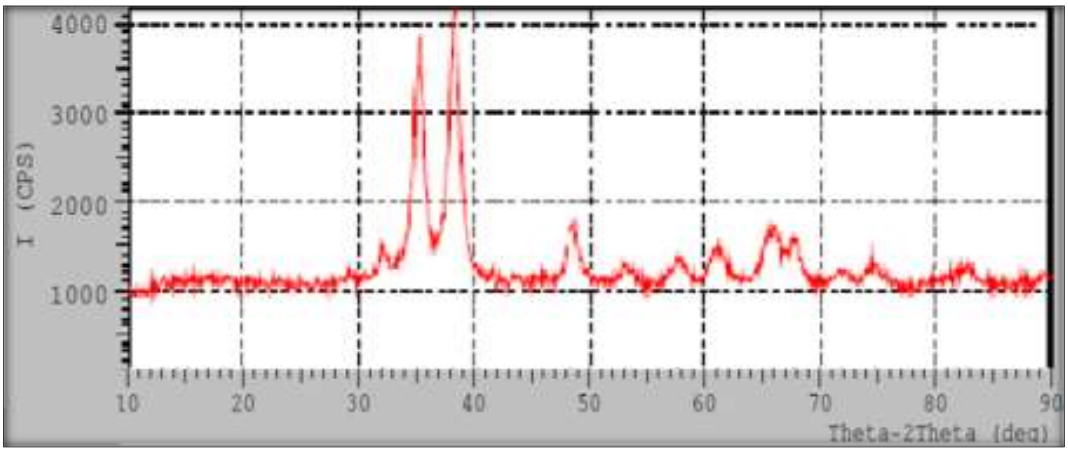

Figure 1: XRD of ZnNPS

2. **FTIR Spectroscopy:** The FTIR spectrum of ZnNPs was depicted in figure3. The peak at 3440.39 cm^{-1}, 2067.32 $cm^{-1}$are confirmed the-OH, N-H stretching. The adsorption band at 794 cm^{-1}, 876.48 cm^{-1} depicted the presence of -CH bending vibration frequency. The frequency band at 1118 cm^{-1} showed the C-O stretching. The frequency range at 633 cm^{-1} confirmed the stretching frequency of ZnONps [11,12].

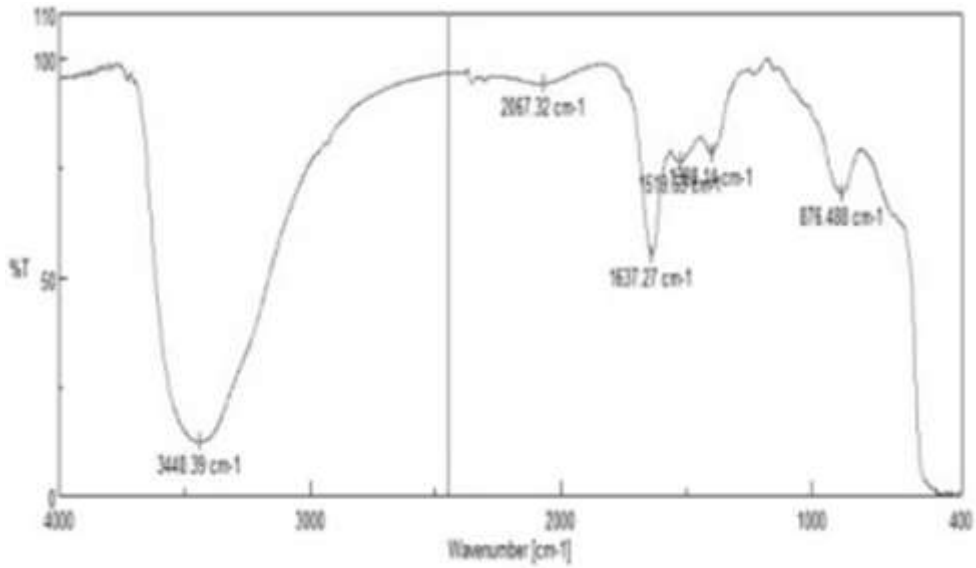

Figure 2: FTIR of ZnNPS

3. **FESEM and EDX Analysis:** The ZnO NPs nanoparticles surface was analyzed by FESEM and EDAX analysis with elemental mapping. The FESEM images of ZnO NPs are denoted in the figure 4. ZnO NPs were formed with uniform structures [13]. This property occurred due to polarization, Coulomb force of ZnO NP [14]. In EDAX studies Zn (45.96%), O (52.36%), were the elements present in the ZnO NPS. Figure 3 represents different elemental distribution maps in the ZnO nanoparticles. Figure 4(b) shows the elemental distribution maps, using of ZnO NPS[15].Elements anlaysis of ZnO nanoparticles given in the table1.

Table 1: Elemental Analysis of Zno Nps

Element	Percentage of weight	Percentage of Atomic
CK	15.85	37.82
OK	18.67	33.44
Cuk	3.11	1.40
ZnK	12.36	27.33

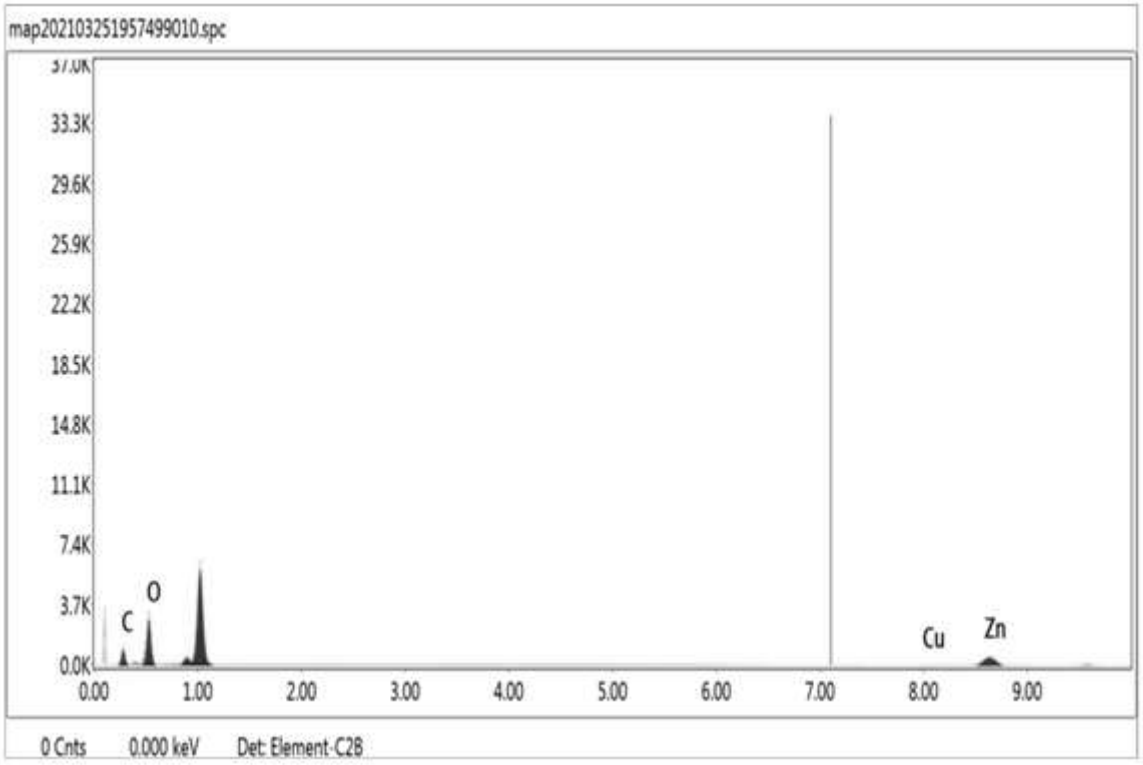

Figure 3: EDX- Elemental Analysis of ZnO Nps

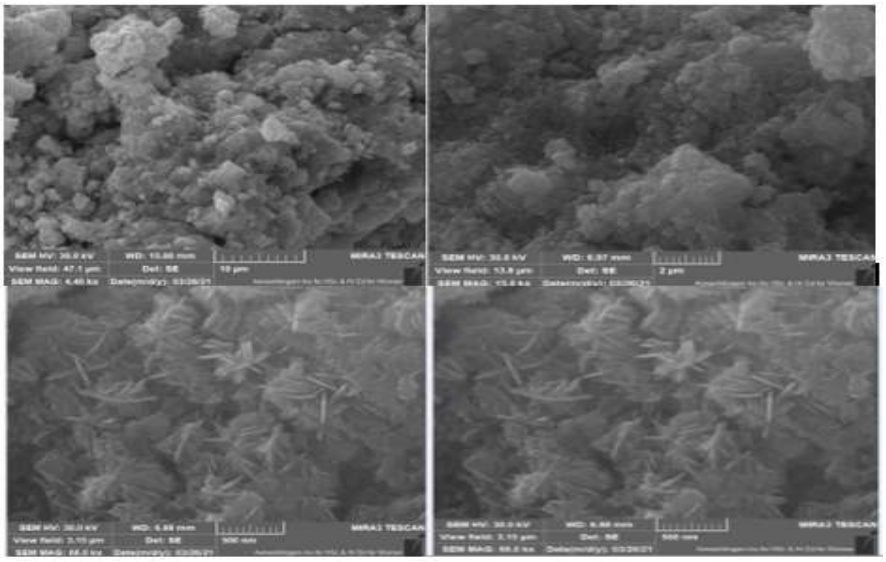

Figure 4: FESEM images of ZnONP

4. **TGA /DTG Analysis:** TGA and DTG have been widely used to quantitatively characterize the dehydration and desolvation of solvates and hydrates at constant temperature. The thermal analysis result of ZnO nanoparticles shown in the figure. At 30 to 120 °C, the weight loss that had been noted, it has been related to the vaporization of water molecules. The range of 121 to 300 °C another weight loss, reported due to associated with the burning of extract with chemicals [16]. The temperature range of 300 to 800°C one more weight loss has retained, due to the formation of ZnO .The mass change occurred via -1.35% at temperature .30-120^{0}C,-18.24% at temperature 30-800^{0}C. -6.66% at temperature121-300^{0}C.-10.23% weight loss occurred at temperature 301-800 ^{0}C.The least amount of mass changes occurred. So ZnO nanoparticles thermally stable.

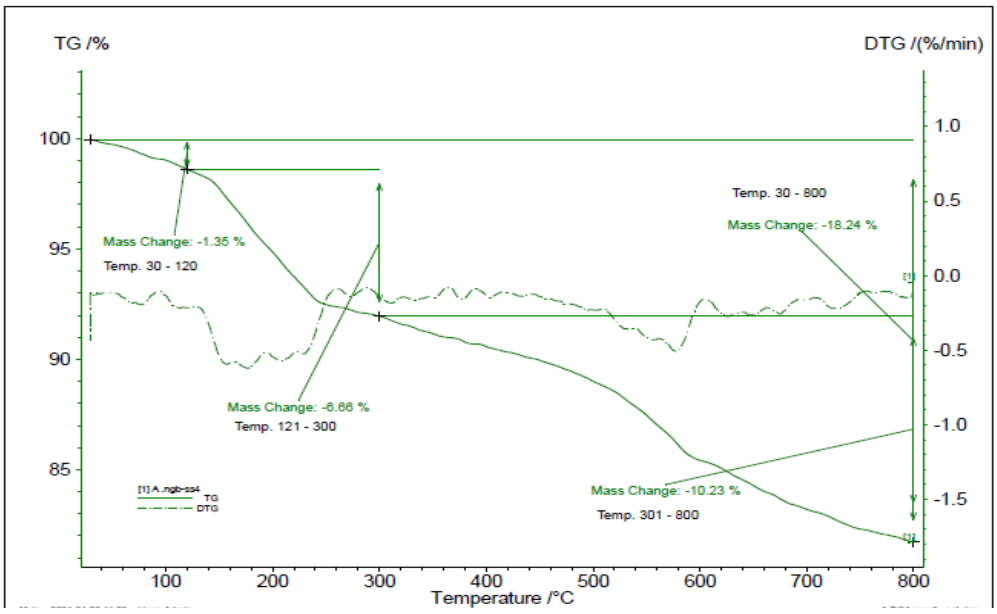

Figure 5: TGA/DTG of ZnO NPS

IV. ANTI OXIDANT ACTIVITY

1. **Free Radical Scavenging Activity:** DPPH assay is used to find out antioxidant property. The free radical scavenging activity was calculated as follows

   $$\text{Scavenging Activity\%} = \frac{\text{control} - \text{Test}}{\text{control}} \times 100$$

2. **ABTS Scavenging Activity:** ABTS assay is used for these studies. The test solution showed good scavenging activity shown in table 2.Increasing concentration of the solution increasing antioxidant activity. The scavenging activity is calculated by using the following formula

   $$\text{Scavenging Activity\%} = \frac{\text{control} - \text{Test}}{\text{control}} \times 100$$

Table 2: Antioxidant Activity

Volume (ml)	Concentration (µg)	DPPH Scavenging Activity (%)	ABTS Scavenging Activity (%)
0.1	250	27.2	95.2
0.2	500	46.5	96.3
0.3	750	75.1	98.4

The results clearly indicate the potential of ZnO nanoparticles [17]. Various phytochemicals such as polyphenols and, flavonoids are already reported in the aqueous extracts of leafs. These chemicals provide antioxidant property to nano particles.

3. **Analysis of Minimum Inhibitory Concentration (MIC):** Antimicrobial susceptibility testing was carried out by MIC, agar well diffusion method. For this antibacterial activity,the test sample was diluted to get series of concentrations from 50mg/ml to 100mg/ml in sterile nutrient broth. The microorganism suspension of 50µl was added to the broth dilutions.For fungal activity, the microorganism suspension of 50µl was added to the broth dilutions The test samples were allowed in Sabouraud dextrose broth and 50 µl of Aspergillus niger spore suspension was also added.

4. **Agar- Well Diffusion Method:** Escherichia coli, and Staphylococcus aureus were used to predict antibacterial activity. ZnO NPs had shown a good antibacterial activity against E.Coli. The volume of test samples were taken as 20,40,60µl. The antibacterial activity due to the large surface area of the ZnO nanoparticles.The anti-bacterial activity,concentration of ZnO NPs given in the table4. Another reason for the anti-bacterial potential could be due to the active oxygen species produced by metal oxide particles [18, 19].

Table 3: Mic Method

Concentration of ZnO NPS mg/µl	
Bacteria	
Staphylococcus aureus	**E.coli**
50	100

Table 4: Zone Of Inhibition Efficiency

Volume of samples (µl)	**Zone of inhibition (mm)**	
	Staphylococcus aureus	**E.coli**
20	-	-
40	-	1
60	-	3

\+ : No bacterial growth occurred
\- : bacterial growth occurred

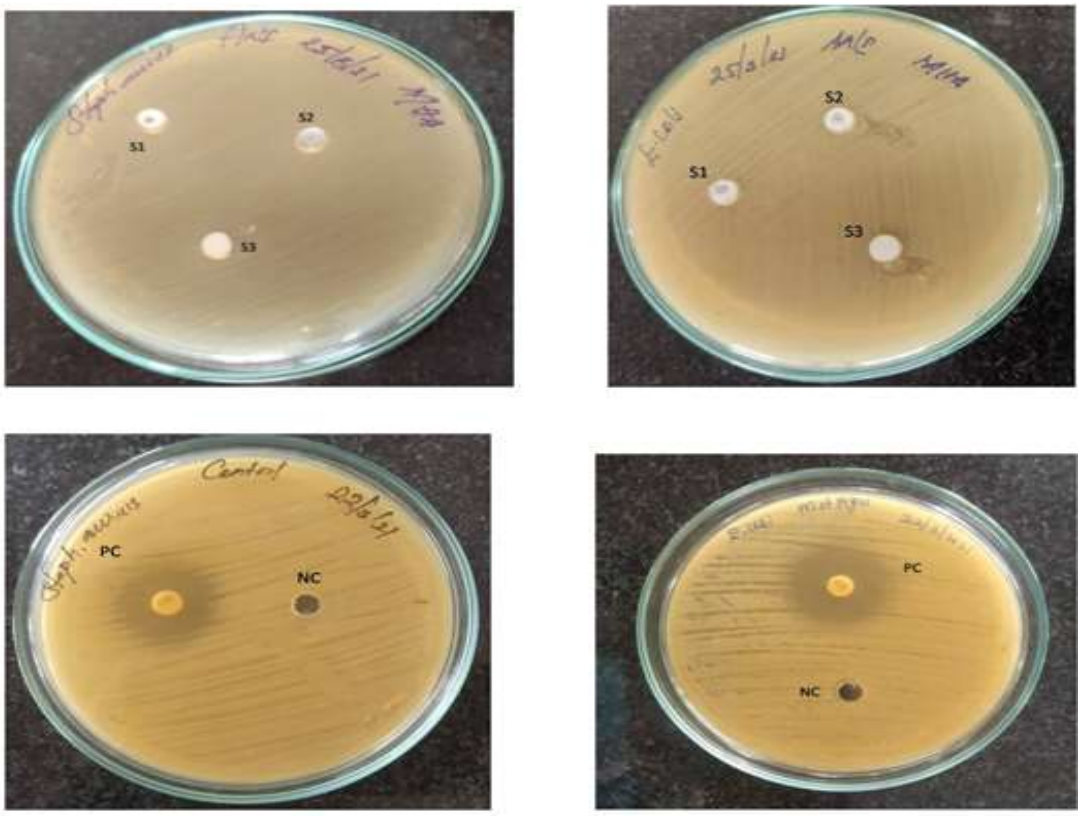

Figure 6: Zone inhibition of Staphylococcus aureus,E.coli

5. **Anti-Fungal Activity:** Lactophenol cotton stain is used for these studies. Antifungual studies tested against Aspergillus niger fungi. Positive sign denotes Spore inhibition caused by test sample [20].The inhibition efficiency was shown in the table5.

Table 5: Spore Inhibition Efficiency

(Concentration of ZnO NPS mg/l) against Fungi	**Sample Name**	**Spore inhibition of Aspergillus niger**
Aspergillus niger	Control (Without test sample)	-
100	Test sample	+

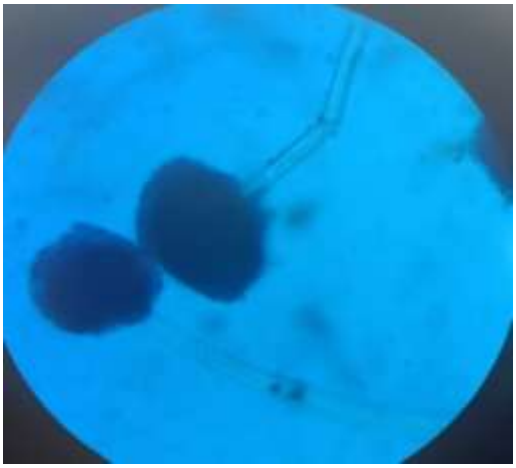

Figure 7: Control (Without test sample)

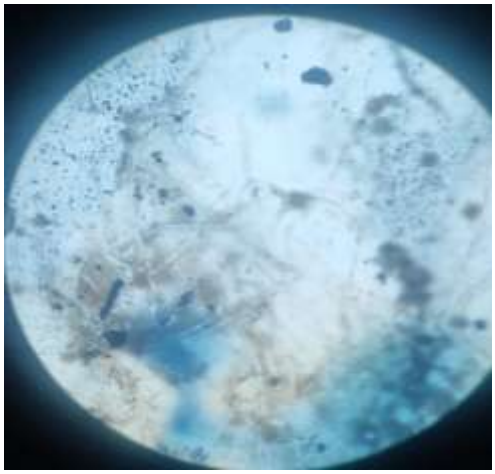

Figure 8: Test sample

This activity reveals the destructions of microbial plasma membrane due to the discharged zinc ions from ZnO NPs[21,22].

V. CONCLUSION

In the present study on synthesis of zinc oxide nanoparticles using leaf extract of Eichhorrnia crappises. XRD was used to calculate the dimension of the nanoparticles as 54 nm. EDX results show the composition of Zinc is 45.96% and Oxygen is 52.36%, respectively. Fourier Transform Infra Red (FTIR) spectroscopic analysis depicts absorption peak of Zinc Oxide bonding at 633 cm^{-1}.The thermal activity checked by TGA/DTG. FESEM images analysed the surface morphology nanoparticles. ZnO nano particle showed good anti oxidant activity, good antibacterial against E.coli bacteria. The antifungal potential, these nanoparticles showed good activity against tested fungal.

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