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Research Article

SYNTHESIS OF ZINC OXIDE NANOPARTICLES UTILIZING LEAVES OF *TARAXACUM OFFICINALE RADIX:* AN ADAPTABLE TOOL AGAINST BACTERIAL, FUNGAL STRAINS AND A METHODOLOGY TOWARDS ECOLOGICAL REMEDIATION AS A PHOTOCATALYST

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ABSTRACT

The aim of this research work is to report a novel, facile green route method concentrate of leaves extract of *Taraxacum officinale radix* as a diminishing and covering generation of NPs was approved by the Yellowish shade of the colloidal suspension, which on additional UV-Vis spectrophotometer focused at 372 nm. Further morphological investigations using TEM, EDX, SEM uncovered that the incorporated particles were consistently round (21.47nm), monodisperse, basically made out of Zinc and Oxygen (3 keV,88.2(wt%) Antifungal Activity of zinc oxide nanoparticles (ZnO NPs) were examined against *Candida albicans* and *Aspergillus niger* by influencing cell capacities, which caused deformation in fungal hyphae. ZnO NP acted as an excellent catalyst (2.88 eV band gap energy) and mediated the reduction of toxic dyes. Further, promising antibacterial activity was depicted by ZnO NPs against two gram positive and three gram negative pathogens *viz*, *Staphylococcus aureus, Klebsiella pneumoniae*, *Streptococcus pneumoniae*, *Escherichia coli* and *Pseudomonas aeruginosa* respectively. Overall the experiments suggested that green NPs can be used effectively in agricultural and food safety application as well as it can be address for future medical concern. These nanoparticles went about as effective electron transfers thereby leading to enhanced catalytic reduction/degradation of model toxic and pollutant dyes.

Keywords: *Taraxacum Officinale radix*, ZnO-NPs, Antibacterial, Anti Fungal activity, Dye deprivation.

1. INTRODUCTION

Recent era have seen a period where causalities because of microbial organism based diseases have occurred. In spite the fact that the enormous undertakings are being made but this serious situation is yet to be resolved. Another basic issue is the exhaustion of the environment. The consistent release of dyes in the water bodies has drastically diminished the quality of water. These colors are primarily mutagenic and cancer-causing in nature and lead to serious geno and cytotoxicity [1-3]. Nanoparticle treated heterogeneous photodegradation has appeared as the powerful methods of water purification as it is capable of changing natural toxins in to products like CO_2 and H_2O. The new creating discipline, nano-technology, animated the creation of metal NPs particularly ZnO NPs described by low harmful impacts to human and high bactericidal potential. ZnO NPs might be utilized as an option in contrast to anti-infective drugs showing better impact on multidrug safe bacteria. The presences of protein covers on nanoparticles uphold both adjustment and authoritative to bacterial cell [4-7]. The method of activity of ZnO antibacterial potential is examined based on bacterial cell penetrability, cell breath just as infiltration inside the bacterial cell causing harm through responding with DNA and protein (phosphate and sulfate containing compounds) [8]. Apart from this, Fungal infections influence different parts of the body and can be difficult to treat.

Aspergillus contamination causes a scope of arranged sicknesses especially in lung according to host immunity. *Candida* contaminations can be shallow or intrusive. Shallow infections routinely impact the skin or mucous films. A comprehensive response for this graving issue has been offered by "Green Method" reasonable, eco-accommodating, One pot synthesis, cheap and liberated from chemicals [9, 10].

The class *Taraxacum* is from the family Asteraceae, subfamily Cichorioideae. The weed has been known for its accommodating properties and has been utilized for the treatment of various contaminations, For instance,

dyspepsia, heartburn, spleen and liver battles, hepatitis and anorexia. Explicit thought has been given to diuretic, choleretic, against coagulatory and prebiotic impacts [11-13].

In recent years, ZnO has been used because of its exceptional optical and electrical properties and potential applications including sensors, photodiodes, sunlight based cells, memory gadgets, UV-light diodes, piezo-electric transducers and photocatalysts. ZnO has been most often times utilized for degradation of many organic pollutants. In small concentration ZnO nanoparticles emphatically restrains the activity of pathogenic organisms. On literature basis, it is known that ZnO demonstrates significant growth inhibition of a broad spectrum of bacteria [14]. Preliminary studies shown that the antibacterial movement of ZnO NPs may be identified with the arrangement of free radicals on the outside of the NPs, and the harm to the lipids in bacterial cell film which thusly lead to the breakdown of bacterial cell layer [15, 16].

2. MATERIAL AND METHODS

Leaves of *Taraxacum officinale radix* were collected from Tehri Garhwal, Uttarakhand. Zinc nitrate was acquired from Sigma Aldrich and NaOH and methanol were procured from Fischer Scientific. Double distilled water was used for the experiment. The fungal strains used in the study were *Candida albicans* (ATCC 10231) and *Aspergillus niger* (ATCC 16404).These strains were obtained from National Chemical Laboratory (NCL), Pune, Maharashtra, India. Clinical isolate *E.coli* were secured from V.C.S.G. Gov. Institute of Medical Science and Research, Srikot, Srinagar Garhwal, Uttarakhand. *Staphylococcus aureus* (MTCC-1144), *Klebsiella pneumoniae* (MTCC-4030), *Streptococcus pneumoniae* (MTCC-655) and *Pseudomonas aeruginosa* (MTCC-2474) were collected from Institute of Microbial Technology (IMTECH) Chandigarh, India.

2.1. Plant extraction prepration

Fresh leaves were gathered and washed several times with water to eliminate the dust particles residue particles and afterward air dried to eliminate the humidity. The concentrate utilized for the reduction of zinc ions (Zn^{2+}) to zinc nanoparticles (Zn°) was set up by placing 50g of washed dried fine leaves in 250mL glass beaker alongside 100 mL of sterile distilled water. The mixture was then boiled for 35 minutes at 45°C. The extract was cooled to room temperature and filtered using filter paper. The concentrate was put away in a fridge to be utilized for extra trials.

2.2. Green synthesis of zinc oxide nanoparticles

A progression of ZnO NPs were initially synthesized at room temperature by utilizing diverse precursor (Zinc Nitrate hexahydrate, Zinc acetate dehydrate) and distinctive bases (Sodium and Potasium hydroxide). To standardize optimal conditions for the preparation of 25 ml of the plant, extract is heated at 50°C for 15 min under magnetic stirring and then 50ml solution of the $Zn(NO_3)_2.6H_2O$ in double distilled water was added drop insightful to the plant separate. The mixture was heated under continuous stirring for 20 min and few drops of NaOH were added for maintaining the pH of solution. After complete disintegration of the combination, the supernatant was disposed off and centrifuged at 3000 rpm for 15 min and afterward washed with de-ionised water to dispose of the contaminations [17]. Resulted material was squashed in a mortar-pestle to improve nature for depiction.

2.3. Characterization techniques

This analysis was proceeded via an X-Ray diffractometer (PANalytical, X*PERT PRO) applying monochromatic radiation of CuKα with 1.54 Å wavelength. XRD was used to analyze the crystalline nature of nanoparticles and for providing information about unit cell dimensions. XRD also determined the crystalline size of nanoparticles using Debye Scherrer's equation. Scanning Electron Microscope (CARI ZEISS, MA15/EVO18) was used to determine the morphology of the green synthesized zinc oxide nanoparticles. A drop of nanosuspension (prepared in ethanol) was placed on the carbon-coated copper grid (Tecnai TF20 equipped with FEG source) at an accelerating voltage of 200 kV, subsequently, the grid was permitted to air dry for 5 min. Post-air-drying, the grid was analyzed and respective microphotographs were acquired ±70 degrees inclined PC controlled stage and is furnished with a 4K x 4K Eagle CCD Camera with a 4-port readout and 15µm pixel size. EPU programming has been utilized to additional investigation the data. The natural organization of the as-arranged ZnO NP was interpreted utilizing QUANTAX EDS.

2.4. Antibacterial assay

The agar well diffusion is a semi-quantitative test to decide the antibacterial action. NPs were evaluated against two gram positive *Staphylococcus aureus*, *Streptococcus pneumoniae* and three gram negative pathogens *viz*, *klebsiella pneumoniae*, *Escherichia coli* and

Pseudomonas aeruginosa separately by utilizing the agar well diffusion method [18]. A suspension of zinc nanoparticles of varying concentrations of 50µg/ml, 100µg/ml and 150µg/ml in 0.5%DMSO was utilized for screening the antimicrobial movement of nanoparticles against the test microorganisms. DMSO was utilized as a control and erythromycin has been utilized as a standard antibiotics. Test organisms were scattered over the surface of agar plates. A small amount of sample gently pushed over the center of nutrient agar plate inoculated with bacterial cells from intimate contact of the sample. Plates were incubated at 37°C for 24 h. The antibacterial activity of ZnO nanoparticles were appeared by the broadness of the zone of restriction made in and around the example.

2.5. Antifungal activity determination

The growth medium utilized for growth of fungal pathogens, Sabouraud's dextrose agar/broth was used. The fungal pathogens (10^5 Cfu/ml) were inoculated in Sabouraud's dextrose stock for 48-72 h followed with pouring in Sabouraud's dextrose agar medium at 28°C for 48 h. The antifungal movement was constrained by well dispersal method with slight modifications [19]. The fungal cultures were pre suspended in sterilized Sabouraud's dextrose broth till solidification. The specific sized diameter wells were punched into the agar and each of the wells were filled with test samples (1mg/ml) prepared in DMSO. The diluent, DMSO was utilized as adverse control. For antifungal activity, Fluconazole (1mg/ml) was used. The fungal cultures were kept for incubation at 28°C.The wells having antifungal especially was considered as an exceptional antifungal trained professional.

2.6. Dye degradation

To check the reactant activity, different parameters such as catalyst concentration, initial concentration of dye, initial pH and temperature of the dye solution were investigated. To make colloidal suspension of ZnO -NPs with each dye (Acridine orange, Methylene orange, Rose bengal), 1 mg dye in 100 ml distilled water was constantly stirred. Then 5 mg ZnO-NPs added in 25 ml of dye solution and kept under instantaneous exposure to sunlight. Known volume (5ml) was withdrawn at different time spans such as 0, 60, 120, 180, 240, 300 minutes and measured using a spectrophotometer at 200-650 nm to assess the rate of degradation. The % degradation was calculated using the equation [20]

The aliquot was taken out at different time intervals. The absorption spectra of the dye solutions were recorded and the rate of degradation was observed seen at various time intervals in terms of change of intensity at λ_{max} i.e 464nm (MO), 450-465(AO)and 450-650 nm (RB).

3. RESULTS AND DISCUSSION

3.1. XRD analysis

XRD analysis was determined from Start Position [°2Th] 20.0150 to end Position [°2Th] 69.9950. Sharp peaks showed the highly crystalline nature with crystallite size of 25.8nm. The detected peaks corresponded to the hexagonal phase ZnO nanoparticles are found in the lattice planes (h,k,l) of (1,0,0),(0,0,2),(1,0,1),(1,0,2), (1,1,0), (1,0,3) ,(2,0,0),(2,0,1),(1,0,4) in the Pos. [°2Th.] value of 32.10, 32.18, 34.71, 34.8, 36.55, 36.64, 56.89,57.05 with relative intensity(%)66.19, 33.10,76.27, 38.13, 100, 50,37.37,18.68 respectively.

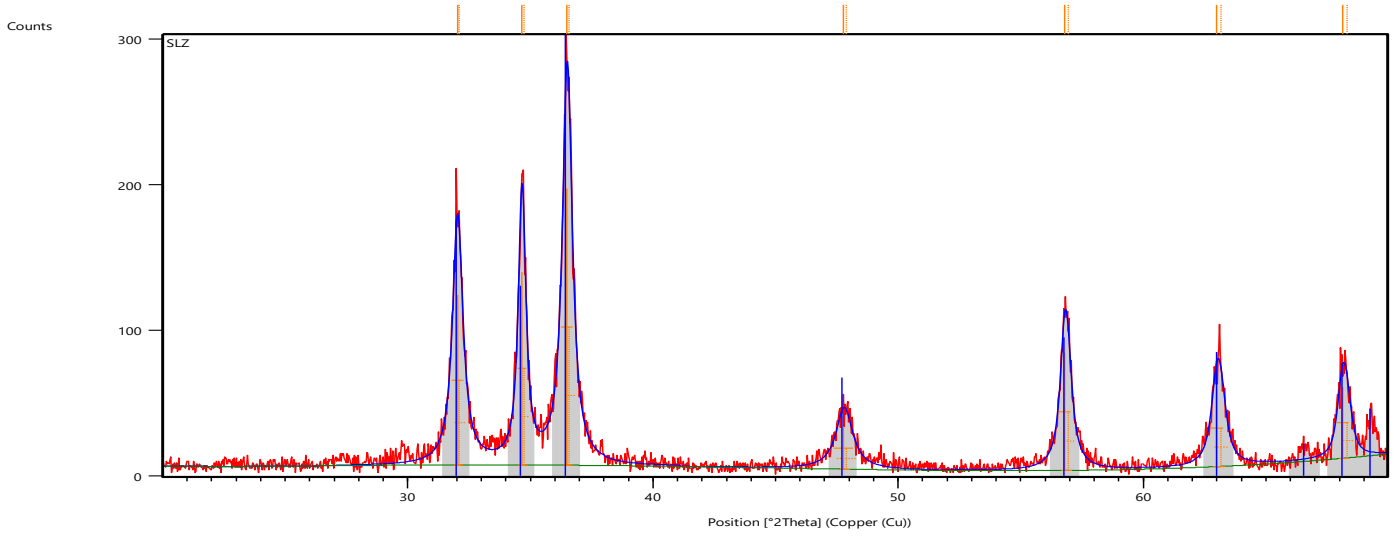

Fig. 1: XRD pattern of ZnO-NPs of the leaf extract of *Taraxacum Officinale radix*

3.2. SEM and EDX analysis

SEM picture is utilized to know the morphology of the ZnO nanoparticles. SEM pictures of ZnO nanoparticles under various amplifications are shown in fig. 2(a) & 2(b). It is seen that the majority of the nanoparticles are spherical in shape with agglomeration. The boundary of the single nanoparticles can be clearly visible in SEM pictures.

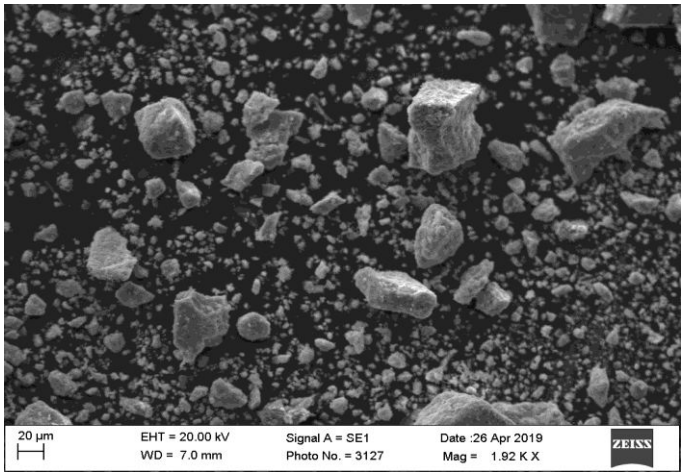

Fig. 2(a)

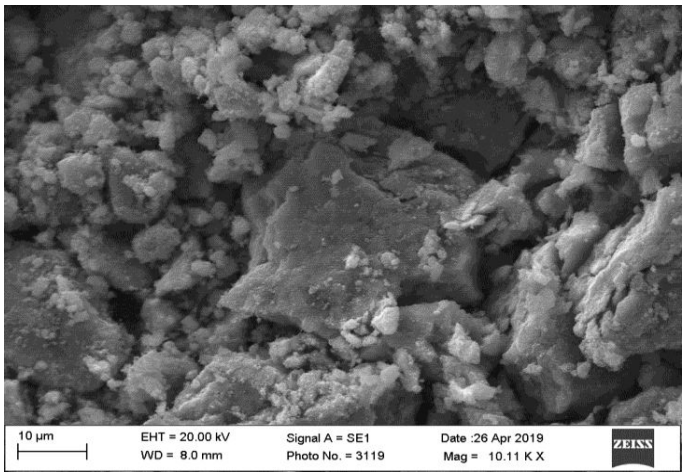

Fig. 2(b)

Fig. 2(a) & Fig. 2(b): SEM pattern of ZnO-NPs of the leaf extract of *Taraxacum officinale radix* at various magnifications

Further Analysis of the ZnO nanoparticles by EDX spectrum in fig.3 confirmed the signal characteristic of zinc and oxygen. Presented peaks are assigned for Zn and O proves the composition of ZnO Nanoparticles.

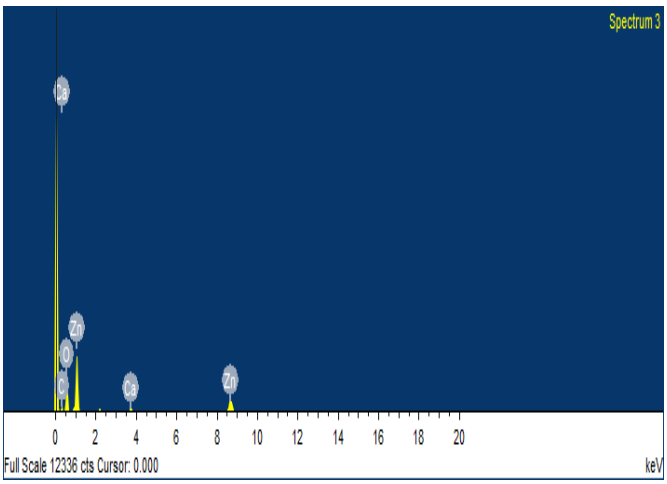

Fig. 3: EDX spectrum of the leaf extract of *Taraxacum officinale radix*

3.3. TEM image of the leaf extract of *taraxacum officinale radix*

Transmission Electron Microscope image of ZnO-NPs is displayed in Fig. 4(a) & (b) which clearly demonstrates the agglomerated particles are spherical in shape having size 21.47 nm for extract of *Taraxacum officinale radix*.

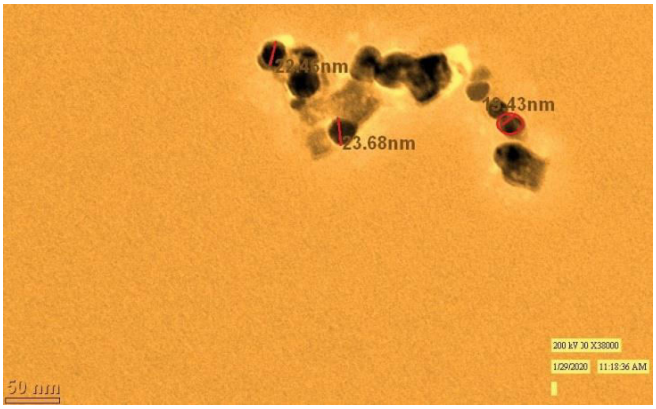

Fig. 4(a)

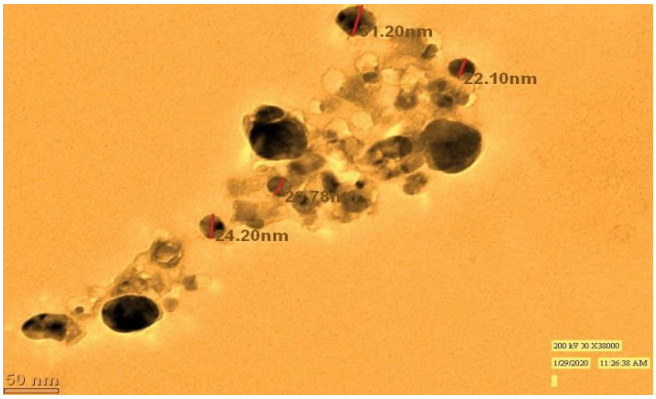

Fig. 4(b)

Fig. 4(a) & Fig. 4(b): TEM pattern of ZnO-NPs of the leaf extract of *Taraxacum officinale radix* at various magnifications

3.4. Antimicrobial assay

Synthesized ZnO-NPs has better antibacterial activity than available marketed drug and can be used as an alternative (Table 1). Holds result at various concentration against different pathogens.

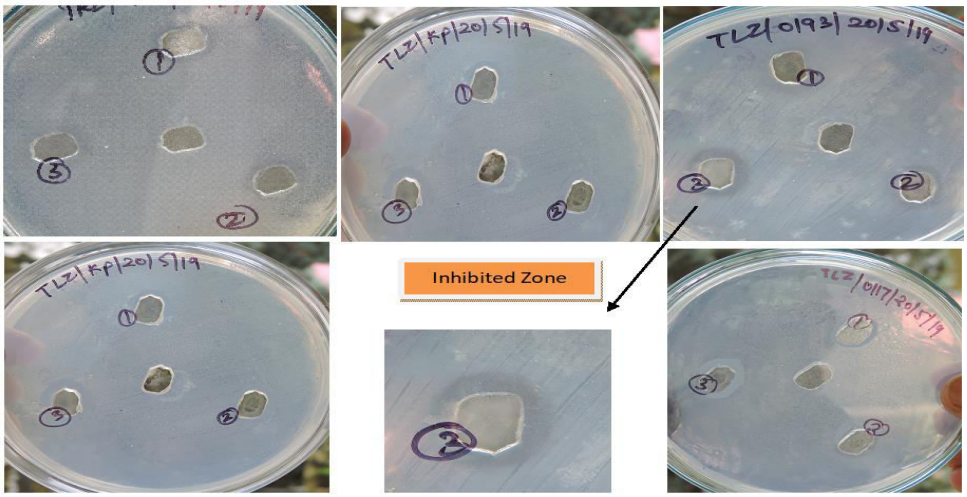

Fig. 5: Inhibition Zone

Table 1: Antimicrobial assay of ZnO-NPs of the leaf extract of *Taraxacum Officinale radix*

S.N.	Pathogen	Control	50 µL	100 µL	150µL
1.	*k. pneumonia*	-	14mm	14mm	16 mm
2.	*S. pneumonia*	-	15mm	16mm	18mm
3.	*E. coli*	-	13mm	15 mm	16 mm
4.	*P. aeruginosa*	-	-	-	-
5.	*S. aureus*	-	12mm	16 mm	17 mm

Cork diameter- 8mm

3.5. Antifungal activity determination via well diffusion method

The antifungal activity was investigated by well diffusion method with slight changes [21]. The fungal cultures were pre-suspended in disinfected Sabouraud's dextrose stock till solidification.

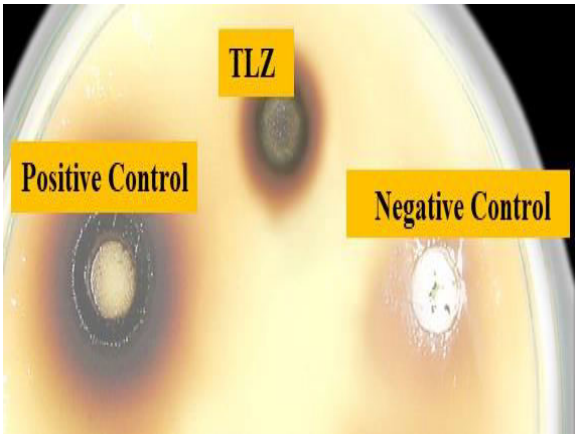

Fig. 6(a): Antifungal activity of test samples against *Aspergillus niger*

Fig. 6(b): Antifungal activity of test samples against *Candida albicans*

The specific sized diameter wells were punched into the agar and each of the wells were filled with test samples (1 mg/ml) prepared in DMSO. The diluent, DMSO was utilized as negative control. For antifungal action, Fluconazole (1 mg/ml) was utilized. The fungal cultures were kept for incubation at 28°C. The wells having antifungal very much was considered as a powerful antifungal specialist.

Table 2: Antifungal efficacy of ZnO-NPs of the leaf extract of *Taraxacum Officinale radix*

Test samples/Controls	Diameter of Zone of inhibition (mm)	
	Test organisms	
	Aspergillus niger	*Candida albicans*
TLZ	22.0± 0.045	25.0± 0.027
Positive Control Fluconazole (1 mg/ml)	36.0± 0.034	32.0± 0.038
DMSO	NA	NA

3.6. Dye degradation

Distinctive interaction boundaries like photocatalyst doze, initial dye concentration, pH and temperature were studied in order to get the maximum color corruption. Absorbance intensity was found to be diminished constantly with increment in time interval and maximum degradation of MO was found to get degraded to the extent of 77.45%, AO to 67.9 % and RB to 76.13% at pH 7.0. The photo degradation of dye was determined by normalized change in concentration and degradation efficiency. For pure AO arrangement, degradation efficiency was 14.5% which after 4 hours, went to 36.25%. On mixing ZnO-NPs, it was noticed 27% following an hour, which turned 67.9% after 5 hrs. The standardized fixation change of unadulterated MO, was 3.6% after an hour which turned 32% after 5 hours on direct openness to daylight. On mixing ZnO-NPs, the efficiency was settled 7.5% after an hour which on the steady presentation of sun based light after 300 minutes got 77.44%. For unadulterated RB game plan debasement proficiency was 14.5% which following 4 hours went to 36.25%. On mixing ZnO-NPs, it was noticed 27% following an hour, which turned 76.6% after 4 hrs. It may be due to the reason that catalyst used is a semiconductor which upon solar irradiation generates electron-hole pairs. The generation of electron-hole pairs is responsible for the photo-degradation of dyes. Further the photodegradation was discovered to be dye concentration dependent.

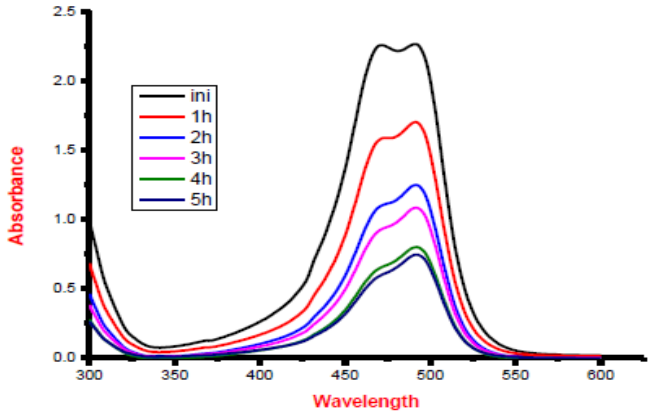

Fig. 7(a): AO degradation at different intervals

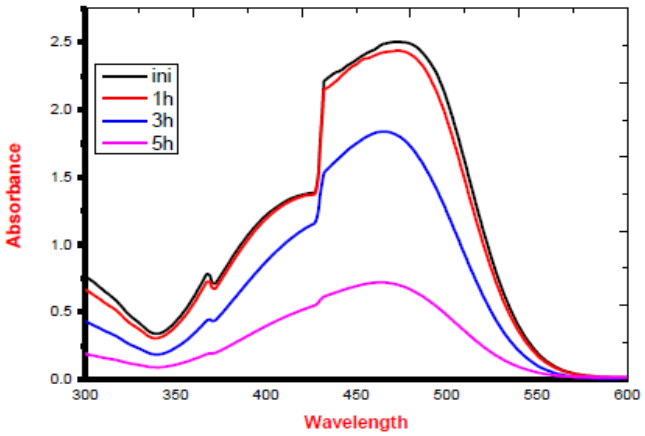

Fig. 7(b): MO degradation at different intervals

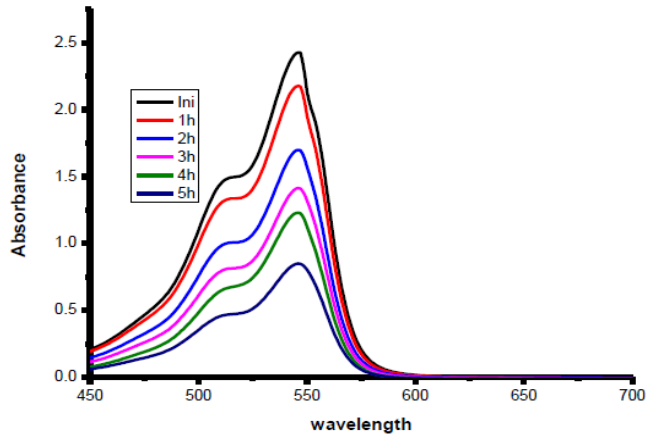

Fig. 7(c): RB degradation at different intervals

4. CONCLUSION

The exploratory outcome suggests the upgrade of an expedient, cheap, biodegradable and secured approach of ZnO-NPs for battling against bacterial contaminations and environmental pollution. The novel fabrication was accomplished with an idea to check antibacterial drug resistance and the congesting threat of environment toxicology utilizing plant with confirmed preferences. Hexagonal phase ZnO-NPs using leaves of *Taraxacum officinale radix* at 60°C formed with in size restricted about 21.47nm is evaluated by XRD, SEM and TEM. The assessment of photocatalaytic movement might be considered the photocatalyst NP as

a heterogeneous photocatalysis to handle water contamination and reduce environmental pollution. This biosynthesized ZnO-NPs has portrayed particularly well antibacterial action as well as antifungal property. Thus, on the premises of these realities and discoveries, it tends to be said that the current framework includes a more extensive scope of uses, which can be only utilized in various field. ZnO nanoparticles could be appropriate sanitizer specialists and utilized as dynamic element for dermatological applications.

Conflict of interest

None declared

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