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Synthesis and Characterization of Nano Zinc Oxide by using Red Cabbage (Brassica Oleraceae) Extract

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Abstract - **In the present work, we describe a low-cost and simple procedure for biosynthesis of Zinc oxide nanoparticles (ZnO NPs) using different volumes from red Cabbage extract. Zinc acetate hydrate [$Zn(OAc)_2.2H_2O$] solution was used as a precursor to synthesize Zinc oxide nanoparticles. The prepared ZnO nanoparticles were characterized using scanning electron microscopy (SEM), Elemental Dispersion Analysis of X-ray (EDAX), Fourier transform infrared spectroscopy (FTIR), X-Ray diffraction (XRD) and Brunauer–Emmet-Teller (BET) surface area. The suitable volume of plant extract can effectively regulate the morphology, particle size and distribution of ZnO, and form more mesopores. FT-IR spectra revealed the functional groups and the presence of protein as the reducing/capping agents for surrounding the ZnO nanoparticles. The average crystallite size of the as prepared ZnO samples (calculated by Scherrer formula) is about 35 nm, 28 nm, 23 nm, and 16 nm for samples obtained in the presence of 5, 10, 15 and 20 ml of the aqueous extracts, respectively. The EDAX analysis, confirmed the presence of metallic zinc oxide in biosynthesized ZnO NPs.**

Keywords: Biosynthesis, ZnO nanoparticles, Red Cabbage, XRD, FTIR, SEM, EDX, BET.

I. INTRODUCTION

In modern research era of any branch of science, nanotechnology has found enormous interest. This technology is a term encompassing the science, engineering, and applications of submicron materials– involves the harnessing of the unique physical, chemical, and biological properties of Nano scale substances (Nanoparticles, NPs) in fundamentally new and useful ways[1,2,3].Recently, metal nanoparticles have attracted much attention due to their high surface-to-volume ratio, surface energy, and spatial confinement and reduced imperfections. Also, metal nanoparticles have unique physical, chemical, electrical, mechanical, magnetic, thermal, dielectric, optical and biological properties [4, 5, 6].

Biosynthesis of nanoparticles is a bottom-up approach which employs biological sources or their components for the synthesis of nanoparticles. Among the biological entities employed, plant mediated synthesis is very common where the bioactive phytochemicals derived from plants are utilized for the production of nanoparticles. Recently, plant extract mediated synthesis of nanoparticles has become one of the popular alternatives over the conventional methods. Biosynthesis or green synthesis approaches has been gaining attention as they are cost effective, novel and usage of toxic chemicals and harsh conditions for reduction and stabilization are avoided. Various metal and metal oxide nanoparticles have been successfully synthesized using biological sources [7-10]. ZnO nanoparticles (ZnO NPs) has been gaining much attention for its semi conducting properties, high catalytic and photochemical activity, unique antifungal, antibacterial, wound healing ,UV- filtering, and anti- inflammatory properties owing to its large surface area to volume ratio[11,12].ZnO NPs are an ideal for biological applications as they are environment friendly, non-toxic, bio safety and biocompatible. Use of plants as a green source for nanoparticles synthesis is most commonly practiced as plants are the hub of a wide range of phytochemicals which can act as reducing and stabilizing agent for nanoparticles synthesis. Different phytochemicals like flavonoids, ketones, aldehydes, amine, amide, organic acid are responsible for the reduction of Zinc ion to nanoparticles form and different proteins aid in the stabilization of synthesized nanoparticles [13].

Previously, many studies have been reported for the green synthesis of Zinc Oxide nanoparticles using various plants extract such as Origanummajorana [14], Polygonumchinense [15], Isodonrugosus [16], Passifloracaerulea [17], MoringaOleifera [18], Conyzacanadensis [19], red tomato fruit (Lycopersiconesculentum M.) [6], and Euphorbia Petiolata[20].

Cabbage (Brassica oleracea L. var. capitata) is one of the most important vegetables grown worldwide. It belongs to the family Cruciferae, which includes broccoli, cauliflower, and kale. The different cultivated types of cabbage show great variation in respect of size, shape and color of leaves as well

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as the texture of the head [21]. Nieuwhof (1969) categorized the different forms of cultivated cabbage into white cabbage, red cabbage and savoy cabbage [22]. Species of Brassica are good sources of phytochemicals and water-soluble and - insoluble vitamins [23].In addition to vitamins, a large number of minor non nutritious phytochemicals - that is, phenolic acids, flavonoids, flavones, iso-flavones, anthocyanins, catechins, and iso-catechins - are present in cabbage, which reduce the risk of oxidative damage (antioxidant activity) caused by free radicals [24]. Due to its antioxidant, anti-inflammatory and antibacterial properties, cabbage has widespread use in traditional medicine. The colors of anthocyanins extracted from red cabbage vary from red at low pH to blue and green at high pH [25]. Thus, this broad color change makes it attractive for application as natural pH indicators. Some studies have been conducted to use of anthocyanin as a capping agent to fabricate ZnO nanoparticles [26,27]. The reports published on the green synthesis of ZnO NPs using red cabbage vegetable are very few [28] with fragmented knowledgebase. In the present study, cost-effective green synthesis of ZnO NPs using red cabbage aqueous extract as a capping agent and their characterization using various techniques are reported.

II. METHODS

2.1 Preparation of Plant Extracts

The natural solution was extracted from vegetable sample (red Cabbage) through a very rigorous process which employing the following procedure as previous literature [29]. Fresh red Cabbage was washed with water and left to dried. 10 gm. From red Cabbage partly were mixed with 100 ml ethanol acidified with 0.01% acetic acid(as solvent) and macerated in a warring blender at full speed for 5 min. Solid residues were filtrated out to obtain clear solutions. The residue on the filter paper was washed rapidly with the extracting solvent until collecting of about 300 ml from extracted solution**.** The resulting extract (Plant Extract) was concentrated in a rotary vacuum evaporator until 50 ml at >40^{o}C and stored in the absence of light for further experimental use.

2.2 Synthesis and Purification of Zinc Oxide Nanoparticles

ZNPs were synthesized by the Pyrolytic method reported previously by Sabura*et al.*, [30] with some modifications. In order to find the optimal method of synthesizing the nanoparticles, we needed to do several trial and error testing. ZnO nanoparticles prepared with different volumes (5, 10, 15, 20, 25 and 30mL) of plant extracts were named S5, S10, S15, S20, S25 and S30 sequentially. In order to synthesize the ZnO nanoparticles, 25 mL of zinc acetate, $Zn(OAc)_2$. $2H_2O$ (0.01M) solution was mixed with different volumes of plant extract. The reaction mixture was stirred for 2 h. using a magnet stirrer at room temperature. Then 25ml of NaOH(0.02M) solution was added drop by drop under continuous stirring in order to adjust pH solution to 12. The mixture was stirred for 1 extra hour until a solid product with a light yellow color was obtained which confirmed the synthesis of ZnO NPs. The precipitate was purified by several re-dispersions in de ionized water and Then the precipitate was collected by centrifugation at 16 000 rpm for 10 min. The final product was a white color powder material which was pyrolysed at 300ºC for 3 hours.

III. Experimental Set-up

The synthesized samples were characterized for their structure by x-ray diffractometer (XRD; Philips, PW1800) using Cu-Kα radiation (λ = 0.15418 nm, 40 kV, 30mA). The average crystallite size of the as-prepared ZnO samples was calculated from the diffraction peaks using the Scherrer formula: [31]

$$D_c = K * \lambda / \beta_{1/2} \cos\theta,$$

Where D_c is the crystallite diameter, K is the Scherrer constant (0.89), λ is the x-ray wavelength (0.15418 nm), $\beta_{1/2}$is the full width at half maximum intensity (FWHM) of the diffraction peak, and θ is the diffraction angle of the peak. Fourier transforms infrared (FT-IR) spectrophotometer was used to test the functional groups in the molecules by Avatar 360 FTIR spectrograph. The morphology and particle size of the-as prepared ZnO samples were characterized by scanning electron microscopy (SEM) using a Philips XLS30 model. Elemental com positions of the synthesized nanoparticle were characterized using Elemental Dispersion Analysis of X-ray (EDAX). The Brunauer–Emmet-Teller (BET) surface area (m^2g^{-1}) of the synthesized ZnO was detected using N_2 adsorption–desorption measurements (Micro meritics Gemini 2375 Surface Area Analyzer, USA).

IV. RESULT AND DISCUSSION

4.1 X-ray Diffraction

The XRD patterns of four ZnO powders were determined and nearly similar results were obtained. Here, four samples (S5, S10, S15, and S20) were selected as an example to reveal the effect of plant extract volume on the XRD patterns of synthesis ZnO NP by varying the ratio of plant extract to zinc acetate (v/v) (Fig. 1).

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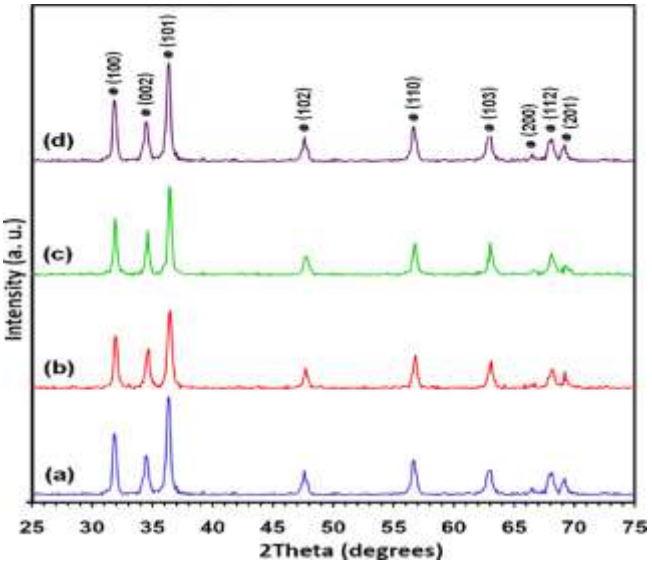

Figure 1: XRD patterns of ZNPs prepared by varying the ratio of plant extract to zinc acetate (v/v), (a) 5ml. plant extract / 25ml Zinc acetate (S5), (b) 10ml. plant extract / 25ml Zinc acetate (S10), (c) 15ml. plant extract /25ml Zinc acetate (S15) and (d) 20ml. plant extract / 25ml Zinc acetate (S20)

There are no noticeable changes in the crystallographic patterns and intensity ratios among peaks. However, clear sharpening and attenuation of peaks can be observed with increasing of plant extract volume (red Cabbage Extract). The sharp and narrow peaks also illustrate that ZnO particles enjoy high crystallinity and purity. The sharp diffraction peaks were observed at 2θ values 31.46, 34.29, 36.33, 47.51, 56.50, 62.84, 66.70, 67.79and 68.55 degrees. These peaks are indexed as (100), (002), (101), (102), (110), (103), (200), (112), and (201) diffraction lattice planes respectively which confirm the hexagonal wurtzite structure for the synthesized nanoparticles (JCPDS card 36–1451). In general, the increase in the intensity of diffraction peaks is attributed to the increase in the crystallinity of the obtained powder [32, 33]. It is also found that the ratio of the intensity of (100) peak to that of (102) peak of ZnO powder tends to increase as volume of plant extract increases. The average crystallite size of the as prepared ZnO samples (calculated by Scherrer formula) is about 35 , 28 nm, 23 nm, and 16 nm for samples obtained in the presence of 5, 10, 15 and 20 ml of the aqueous extracts, respectively. These results reveal that all theas-prepared ZnO samples are nano-size and their crystallite size was decreased by increasing the volume of plant extract. The particle size of the synthesized ZNPs was in close agreement with the previous findings [34, 35].

4.2 FTIR Analysis

FTIR analysis was performed to find the functional groups on aqueous extract of plants and identify their role in the synthesis of ZnO-NPs. In another meaning, this study was carried out in order to ascertain the purity and nature of the nanoparticles and also the presence of phytochemicals in the extract. The phytochemicals such as alcohols, phenols, amines, carboxylic acids and so on can interact with the zinc surface and aid in the stabilization of ZNPs. FT-IR studies of red cabbage aqueous extract and the synthesized ZNPs are given in figure 2.

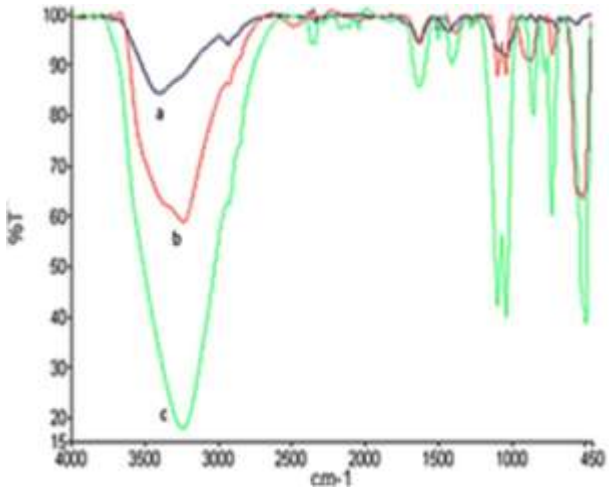

Figure 2: FT-IR spectra of ZNPs prepared by varying the ratio of plant extract to zinc acetate (v/v), (a) red Cabbage, aqueous extract; (b)5ml. plant extract / 25ml Zinc acetate (S5), and (c) 20ml. plant extract / 25ml Zinc acetate (S20)

This figure shows the broad absorption peak in the range of 3000–3500 cm−1 is present in both the figures which can be attributed to the characteristic absorption of hydroxyl groups (O–H) [36, 37]. The band at 2865, 2300, 1595, 1490, 1020, 850 and 680 cm-1 of *red cabbage* extract shifted to 2856, 2337, 1624, 1420, 1060, 900, 750 and 490 cm−1. The peaks at 2922.59 and 2855cm^{-1} show the C-H Stretching vibration. Further, the two weak absorption peaks near at 2900-2750 cm-1 indicates the presence of aromatic aldehydes [38]. The broad and intense band at 3237-3500 cm-1 is owing to OH stretching. The Zn–O frequencies observed for the synthesized ZNPs are in accordance with literature values between 450-540 cm^{-1}[39,40]. The peaks at 1034 and 2924 cm-1 are due to the presence of C–O and C–H vibration modes of starch, which acts as a capping agent. These soluble elements could have acted as both reduction and stabilizing agents preventing the aggregation of NPs in solution, extracellular biological synthesis of zinc oxide NPs [37,39].

4.3 Scanning Electron Microscopy and EDAX

The shape, structure and size of the synthesized ZNPs were determined by the SEM analyses (Figure 3).

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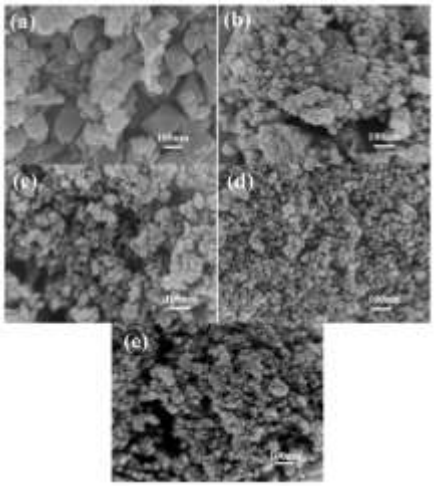

Figure 3: Scanning electron microscopy (SEM) images of ZnO nanoparticles samples synthesized with different volume of plant extract to zinc acetate (v/v). (a) 5ml. plant extract / 25ml Zinc acetate (S5), (b) 10ml. plant extract / 25ml Zinc acetate (S10), (c) 15ml. plant extract /25ml Zinc acetate (S15), (d) 20ml. plant extract / 25ml Zinc acetate (S20) and (e) 25ml. plant extract / 25ml Zinc acetate (S25)

The micrographs of ZNPs proved that they had nano-sized range, spherical shape and uniform distribution. The SEM results illustrated that using different volumes of plant extract affected the size and shape of the nanoparticles. With increasing volume of plant extract (Figure 3b–d), the morphology ZnO nanoparticles becomes smaller and more uniform. S25 (Figure 3e) shows more regularly spherical shape and the minimal size of 10–20 nm. When the volume of plant extract is lower (S10 and S15), the aggregation behavior of ZnO nanoparticles is improved. On the contrary, the structures of ZnO nanoparticles become more compact when the volume increases. As shown in Figure 3c, S15 looks looser and exhibits irregularly spherical particles with the size of about 20–30 nm and the existence of abundant mesopores. (Fig. 4) shows the EDAX analysis, confirmed the presence of metallic zinc oxide in biosynthesized ZnO NPs.

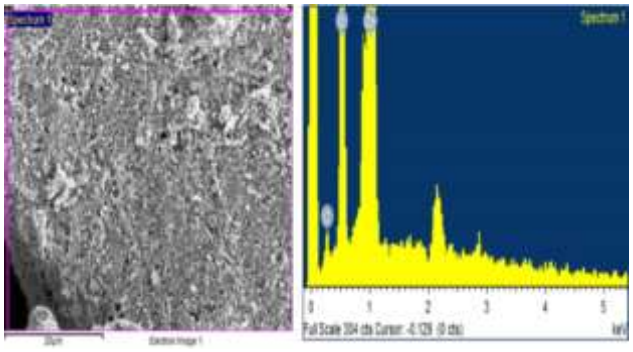

Figure 4: EDAX Spectrum of ZnO NPs synthesized by 25ml. plant extract / 25ml Zinc acetate (S25)

The composition obtained from EDAX analysis was Zinc (75.38%), Oxygen (22.34%), and Carbon (2.28%). The presence of carbon in trace amount indicates the involvement of plant phytochemical groups in reduction and capping of the synthesized ZnO NPs [41].

4.4 Surface Area Values for Synthesized ZNPs

The BET surface area and pore size distributions of as-prepared ZnO particles synthesized at different concentrations of plant extract were investigated by N_2 adsorption-desorption measurement. As shown in Figure 5, according to the International Union of Pure and Applied Chemistry (IUPAC)classification of adsorption and desorption isotherms, all five isotherm profiles can be classed as type IV with a hysteresis loop in the relative pressure range of 0.55–0.75, which indicate that the samples have the adsorption properties of porous materials [42,32].

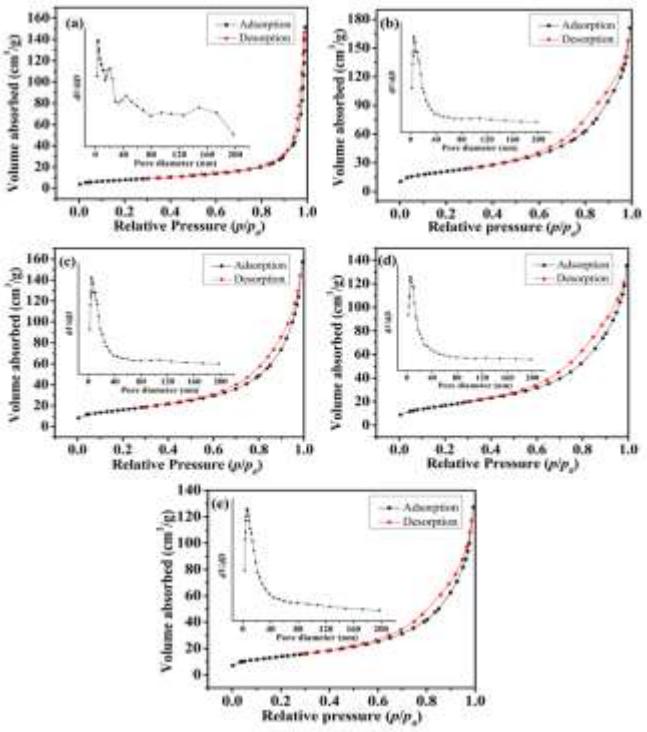

Figure 5: Nitrogen adsorption-desorption isotherms and the pore size distribution (insert) of ZnO samples. (a) S5 (b) S10, (c) S15, (d) S20, (e) S25

For S10, S15, S20 and S25 samples, the hysteresis loop shifts to lower relative pressure, indicating smaller pores size. The Barrett-Joyner-Halenda (BJH) pore size distribution curves demonstrate the existence of more abundant mesopores and relatively homogeneous pore distribution for the ZnO samples synthesized using different volumes of plant extract. Table 1 exhibits the results of the measured BET specific surface area and the BJH pore size distribution of the samples.

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TABLE I
BET surface area and pore volume of as-prepared ZnO nanoparticles

Samples No	Surface Area, S_{BET} ($m^2 g^{-1}$)	Average Pore Diameter, (AP nm)	Total Pore Volume V_{tot} ($cm^3 g^{-1}$)
S5(5ml. plant extract / 25ml Zinc acetate)	28.64	32.24	0.231
S10(10ml. plant extract / 25ml Zinc acetate)	75.70	13.56	0.257
S15(15ml. plant extract / 25ml Zinc acetate)	58.79	15.91	0.234
S20(20ml. plant extract / 25ml Zinc acetate)	62.64	12.89	0.202
S25(25ml. plant extract / 25ml Zinc acetate)	49.61	15.31	0.190

For S10, it is suggested that the largest specific area of 75.7 $m^2 g^{-1}$ comes from the more micropores and smaller mesopores formed by the assembly of finer ZnO nanoparticles. S25 has the smallest specific surface area of 49.6 $m^2 g^{-1}$, due to the close packing of ZnO nanoparticles. S15 has higher pore diameter of 15.9 nm, higher specific area and larger pore volume resulting from the abundant and larger mesopores (Figure 5c inset). These results are consistent with that of SEM.

V. CONCLUSION

Among other conventional techniques to manufacture nano materials, a greener route stands up as promising fields that provides a non-toxic, eco-friendly, and a practical approach to fabricate new materials. Here, we present a greener synthesis of ZnO NPs using red Cabbage plant extract. The phytochemicals present in plant extract played an important role in the formation and stabilizing ZnO nanoparticles. The size and morphology of ZnO nanoparticles were optimized at different volumes of plant extract i.e. 20 ml and 25 ml.

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