DEGRADATION OF METHYLENE BLUE DYE IN AQUEOUS SOLUTION USING GREEN SYNTHESIZED NANO-SIZED ZnO PARTICLES

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Abstract

This research deals with degradation of methylene blue (MB) dye in aqueous solution using green synthesized nano-sized ZnO particles. The aim of this research is to prepare, characterize and study the degradation efficacy of green synthesized nano-sized ZnO particles. Leaves of Ma-yo-gyi (*Calotropis gigantea* L.) were collected from Kathitkan village in Aung Lan Township, Magway Region, Myanmar. Nano-sized ZnO particles were synthesized from zinc nitrate and zinc acetate sources using aqueous leaves extract of Ma-yo-gyi as a reducing agent via green synthesis. The green synthesized nano-sized ZnO particles were characterized by TG-DTA, XRD, SEM and TEM techniques. The effects of calcination temperature on the preparation and average crystallite size of particles were also studied. The green synthesized nano-sized ZnO particles have hexagonal phase with average crystallite size of (24-34) nm. Supported SEM and TEM images by the degradation efficacy of green synthesized nano-sized ZnO particles on MB nethylene blue(MB) dye were studied via various parameters such as effect of pH (3, 4, 5, 6, 7 and 8), effect of contact time (0, 30, 60, 90, 120, 150 and 180 min) and effect of dosage of nano-sized ZnO particles (0.02, 0.03, 0.04 and 0.05 g). Dye removal activity was highest at pH 7 using both of green synthesized nano-sized ZnO particles. From the contact time experiment, at the end of the reaction (after 180 min), MB dye removal efficacy was upto 80-85 % of its initial value. Degradation percent of MB dye solution increased gradually with an increase in dosage of nano-sized ZnO particles.

Keywords : Nano-sized ZnO, Ma-yo-gyi, methylene bue dye, degradation

Introduction

The green synthesis of metal oxide nanoparticles using biological material as the reducing and stabilizing agents has attracted a lot of attention and consideration in the field of pharmaceuticals and biomedical sectors as compared to the toxic chemical and physical methods due to the usage of ecofriendly, non-toxic and safe reagents during the green synthesis process (Tamanna *et al*., 2015). Furthermore, synthesis in plants tends to be faster than microorganisms, is more cost-effective and is relatively easy to scale up for the production of large quantities of nanoparticles (Shah *et al*., 2015). Zinc oxide nanoparticles have received considerable attention due to their unique antibacterial, antifungal, and UV filtering properties, high catalytic and photochemical activity (Shabnan *et al*., 2019). Photocatalytic activity of nanoparticles offers a promising method for wastewater treatment and ZnO nanoparticles behave as wastewater purifiers (Jawad *et al*., 2018). Methylene blue is one of the most common organic pollutants discharged from the industries directly or indirectly into water sources causing water pollution. ZnO exhibits very good photochemical reactivity and efficiently degrades toxic water pollutants released from textile and dying industries by utilizing natural source of energy, sunlight. This is due to the presence of many active sites and fabrication of hydroxyl radicals on ZnO surface (Tamanna *et al*., 2015). When the ZnO nanoparticles are irradiated with UV light, valence band electrons are excited to the conduction band, which leaves holes behind. Then the generated holes create hydroxyl radicals by oxidizing H_2O and OH^- and the excited electrons are captured by oxygen in the air. The resulting anionic radicals are highly reactive and degrade the organic dyes into carbon dioxide and water (Isik *et al.*, 2019). The objective of this study was to study the synthesis, characterization and the effectiveness of green synthesized zinc oxide

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nanoparticles as catalyst for the removal of Methylene Blue (MB), a cationic dye from aqueous solutions. Laboratory batch studies were conducted to estimate the dye degradation efficacy using the nano-sized ZnO particles via the effect of dosage, effect of pH and effect of contact time on the degradation of MB dye.

Materials and Methods

Sample Collection and Scientific Classification of Ma-yo-gyi

The leaves of Ma-yo-gyi were collected from Kathitkan village, Aunglan Township in Magway Region, Myanmar. The botanical name was identified in Department of Botany, Pyay University. *Calotrophis gigantea* L. is also known as Crown flower plant, Maddar, Rui. It is a large shrub growing to 4 m tall. Largely found in Cambodia, Indonesia, Malaysia, Philippines, Thailand, Sri Lanka, India, China and Pakistan. Botanical name of Ma-yo-gyi is *Calotropis gigantea* L(Chandrabhan *et al.*, 2011). Figure 1 shows the plant of Ma-yo-gyi.

Figure 1 *Plant of Ma-yo-gyi*

Preparation of Nano-sized ZnO Particles by Green Synthesis

(a) Preparation from zinc nitrate as precursor

Ma-yo-gyi leaves extract was prepared by placing 10 g of cleaned and dried crushed leaves in 250 mL glass beaker along with 100 mL of distilled water. The mixture was then boiled for 20 min until the color of aqueous solution changed from watery to brown-yellow. Then, the mixture was cooled to room temperature and filtered with Whatman No. 1 filter paper.

For the synthesis of nano-sized ZnO particles, 50 mL of aqueous leaves extract of Ma-yo-gyi was taken and boiled to 60-80 ºC using a magnetic stirrer heater. Zinc nitrate (5 g) was added to the boiled solution and maintained the temperature about 80 ºC. This mixture was continued boiling until it reduced to a deep yellow coloured paste. This paste was then collected in a ceramic crucible and calcined in muffle furnace at 200, 300, 400, 500 and 600 ºC for 2 h. A white coloured powder was obtained and this was carefully collected and stored in air-tight container.

(b) Preparation from zinc acetate as precursor

The nano-sized ZnO particles were also green synthesized from zinc acetate. Briefly, zinc acetate (2 M) was prepared in 50 mL of deionized water under constant stirring conditions. After complete dissolution of the mixture, 1 mL of 25 % aqueous leaves extract of Ma-yo-gyi and 50 mL of 2 M NaOH were added to the prepared solution of zinc acetate. The mixture was stirred continuously for 2 h on magnetic stirrer resulting in white precipitate. The precipitate was filtered and washed repeatedly with distilled water and then, followed by ethanol in order to remove the impurities. Finally, a white-grey powder ZnO was obtained after overnight drying of the purified precipitate at 60º C in oven. It was calcined in muffle furnace at 500 ºC for 2 h and white powder ZnO was obtained.

Characterization of the Green Synthesized Nano-sized ZnO Particles

The green synthesized nano-sized ZnO particles from two metal sources were characterized by TG-DTA, XRD, SEM and TEM techniques.

Investigation on Degradation of Methylene Blue Dye by using the Green Synthesized Nano-sized ZnO Particles

The wavelength of maximum absorption (λ_{max}) should be necessarily determined prior to the quantization of a substance by UV-visible spectrophotometry (Dod, 1967). UV-vis spectrophotometer (UV-vis 240, Shimadzu) was used throughout the degradation experiments. The wavelength of maximum absorption (λ_{max}) of MB solutions was recorded at wavelength range of 500-700 nm and standard calibration curve was constructed at λ_{max} 664 nm using 0.2-1.0 $\times$ 10^{-5} M MB solution at pH 5. Degradation experiments were conducted via effect of dosage, effect of pH, effect of contact time. A 0.04 g of prepared nano-sized ZnO particles was separately added into 250 mL capacity of six clean and dry beakers each containing 50 mL of 2.97 x 10^{-5} M methylene blue solution at pH of 3, 4, 5, 6, 7 and 8 by adjusting with 0.01 M HCl and NaOH solutions. The mixtures were stirred for 120 min and sampling out 10 mL and centrifuged immediately the sampling mixture at 100 rpm for 20 min to obtain the clear blue solution. The absorbance of each solution was measured at wavelength 664 nm using UV-Vis spectrophotometer. In the effect of contact time experiment, after the pH was adjusted at 7 and the solution was taken out 10 mL in every 15 min interval. In effect of dosage experiment, 0.02, 0.03, 0.04 and 0.05 g of prepared nano-sized ZnO particles was separately added into beakers each containing 50 mL of 2.97 x 10^{-5} M methylene blue solution at pH 7. The absorbance of each solution was recorded according to above procedure.

Results and Discussion

Characterization of the Green Synthesized Nano-sized ZnO Particles by TG-DTA Analysis

Thermal stability of green synthesized nano-sized ZnO particles from nitrate salt was investigated by TG-DTA technique (Figure 2). TGA data of the prepared nano-sized ZnO particles before calcination showed the total weight loss 52.98 % in the temperature range of 38.59-601.76 °C. This is attributed to the evaporation of trapped water in the crystal, decomposition of organic residue and zinc nitrate to zinc oxide in the preparation of green synthesized zinc oxide. The DTA curve indicated the two endothermic peaks at 139.56 °C and 198.98 °C corresponding to the loss of trapped water, decomposition of organic matter in sample. The small exothermic peak at 369.97 °C was related with decomposition of organic residue. TG-DTA data indicated that the green synthesized nano-sized ZnO particles were found to be thermally stable in the temperature range of 460-500 °C.

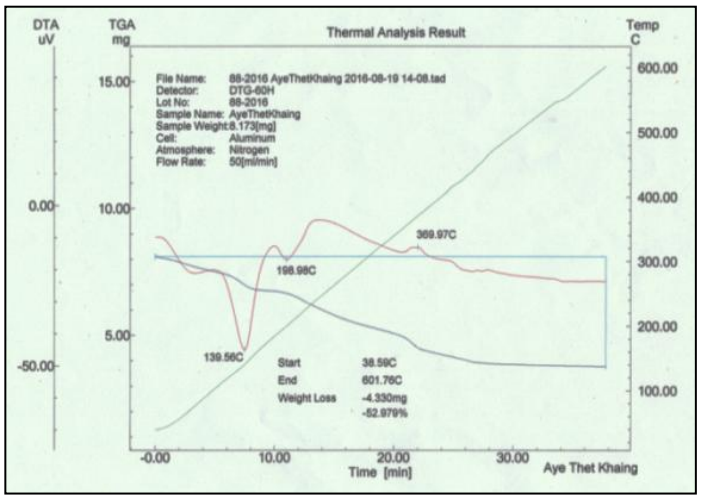

Figure 2 TG-DTA thermogram of the green synthesized ZnO from zinc nitrate

Characterization of the Green Synthesized Nano-sized ZnO Particles by XRD

Figures 3 (a) - (e) show the XRD diffractograms of the calcined ZnO nanoparticles synthesized from nitrate source at different calcination temperatures of 200, 300, 400, 500 and 600 °C respectively. **Figure. 3** (a) shows the XRD diffractogram of the green synthesized ZnO nanoparticles obtained at calcination temperature of 200 °C which is matched with the PDF file of 80-0074 of ZnO. It can be seen not only ZnO peak but also other phases were present due to incomplete formation of ZnO. Figure 3 (b) shows the XRD diffractogram of the calcined ZnO nanoparticles at calcination temperature of 300 °C. There are nine typical diffraction peaks, (100), (002), (101), (102), (110), (103), (200), (112) and (201) which may be assigned to the characteristic peaks of hexagonal ZnO crystal and are matched with library card number 80-0074 of ZnO. Some of the $Zn(NO_3)_2$ were transformed to ZnO, but other phases still exist. Figure 3 (c) shows the XRD diffractogram of the calcined ZnO nanoparticles at calcination temperature of 400°C. There are nine typical diffraction peaks, (100), (002), (101), (102), (110), (103), (200), (112) and (201) which may be assigned to the characteristic peaks of hexagonal ZnO crystal and are matched with PDF library card number 80-0074 of ZnO. Most of the $Zn(NO_3)_2$ are transformed to ZnO, but a few other phases still exist. This may be due to the presence of some impurities in the sample preparation.

The XRD diffractogram of the calcined ZnO nanoparticles at 500 °C is shown in Figure 3 (d). There are nine typical diffraction peaks, which may be assigned to the characteristic peaks of hexagonal ZnO crystal and are matched with the PDF library card number 89-1397 of ZnO. The ZnO crystals were developed at this temperature, it can be said that $Zn(NO_3)_2$ were completely transformed to ZnO. So that, transformation of $Zn(NO_3)_2$ to ZnO between temperature 400-500 °C and it is also consistent with TG-DTA data. At calcination temperature of 600°C, the XRD diffractogram of the calcined ZnO nanoparticles is shown in **Figure 3** (e) with nine typical diffraction peaks, which may be assigned to the characteristic peaks of hexagonal ZnO crystal and are matched with the PDF library card number 89-1397 of ZnO. From the study on effect of calcination on preparation of green synthesized ZnO nanoparticles, diffraction angles (2θ) of hkl planes were increased with increased in calcination temperature from 200 to 500 °C and slightly decreased at the calcination temperature 600°C. As the diffraction angle (2θ) increase, crystallite sizes of ZnO nanoparticles also increased with increased in calcination temperature. When calcination temperature reached 600 °C, the diffraction angle slightly decreased and average crystallite size also decreased. This may be due to agglomeration slightly occurring due to the presence of phytochemicals that can stabilize the nanoparticles formed.

The XRD diffractogram of nano-sized ZnO particles from zinc acetate source after calcined at 500 °C is shown in **Figure 4**. There are nine typical diffraction peaks, (100), (002), (101), (102), (110), (103), (200), (112) and (201) which may be assigned to the characteristic peaks of hexagonal ZnO crystal and are matched with library card number 89-1397 of ZnO. High purity and crystallinity of nano-sized ZnO particles was obtained at this temperature.

Characterization of Green Synthesized Nano-sized ZnO Particles by SEM and TEM

The morphologies of green synthesized nano-sized ZnO particles from zinc nitrate source, ZnO (I) and zinc acetate source, ZnO (II) after calcined at 500 ºC for 2 h were investigated by SEM and TEM techniques. **Figures 5**(a and b) show the SEM images of green synthesized nano-sized ZnO particles from zinc nitrate and zinc acetate sources, ZnO (I) and ZnO (II), after calcined at 500 ºC for 2 h, respectively. According to SEM micrographs of green synthesized nano-sized ZnO particles, the green synthesized nano-sized ZnO (I) have porous nature where those of ZnO (II) have aggregate and dense particles.

Figures 6 (a) and (b) show the TEM images of green synthesized ZnO particles from zinc nitrate source, ZnO (I) and zinc acetate source, ZnO (I) and ZnO (II) after calcined at 500 ºC for 2 h. According to TEM images, the green synthesized nano-sized ZnO (I) has within the nano scale 21.70-37.29 nm and ZnO (II) has 31.25-35.71 nm. The size distribution of green synthesized of ZnO particles from TEM images were closed to XRD data.

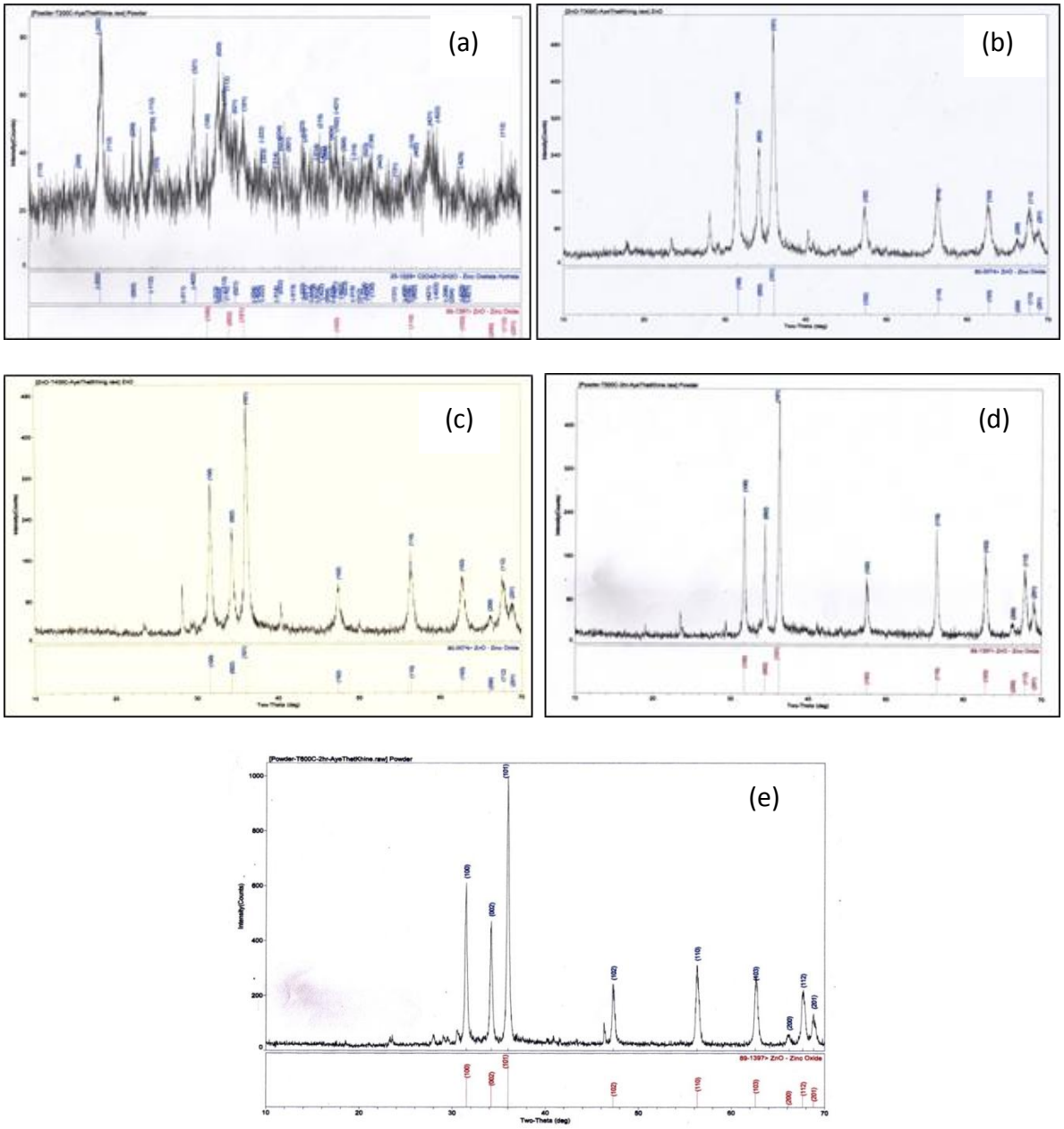

Figure 3 X-ray diffractograms of the green synthesized nano-sized ZnO particles from zinc nitrate source after calcination at (a) 200 ºC (b) 300 ºC (c) 400 ºC (d) 500 ºC (e) 600 ºC

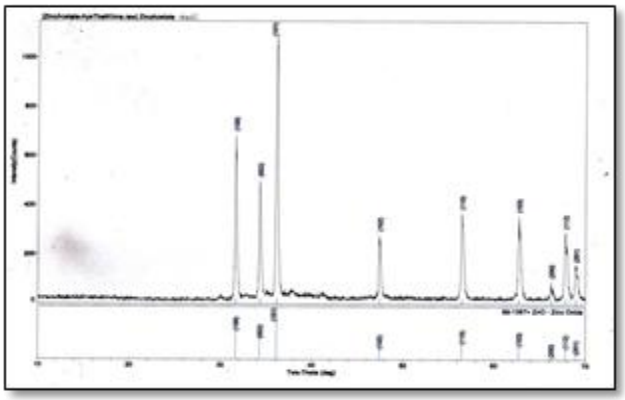

Figure 4 X-ray diffractogram of the green synthesized nano-sized ZnO particles from zinc acetate source after calcination at 500 ºC

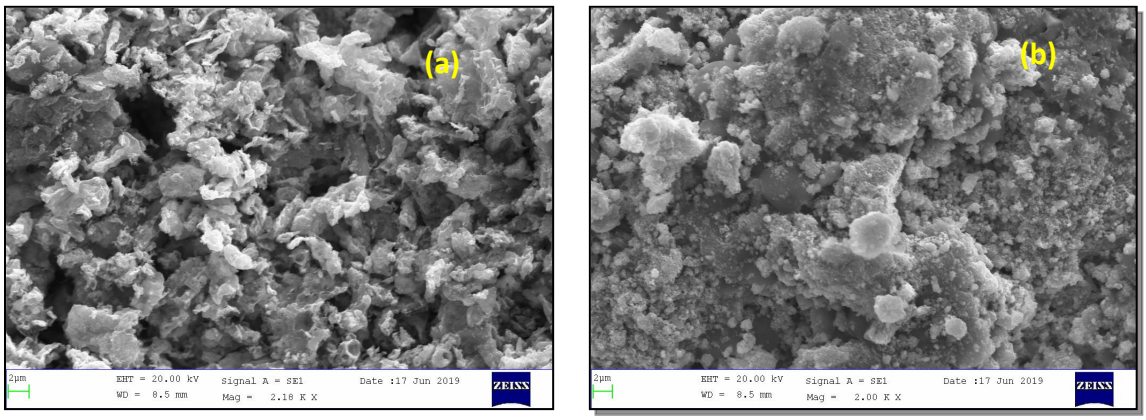

Figure 5 SEM images of the green synthesized nano-sized ZnO particles (a) ZnO (I) (b) ZnO (II) after calcined at 500 ºC

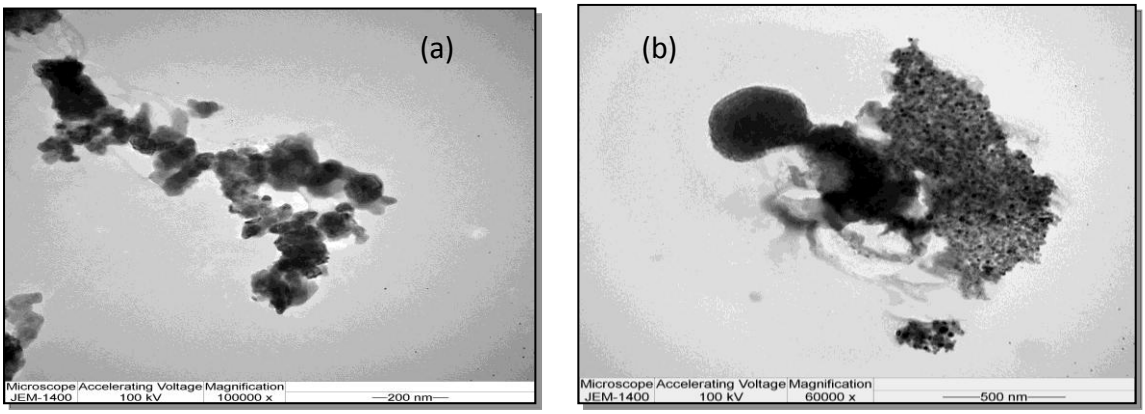

Figure 6 TEM images of the green synthesized nano-sized ZnO particles after calcined at 500 ºC (a) ZnO (I) and (b) ZnO (II)

Average crystallite sizes of the green synthesized nano-sized ZnO particles

The average crystallite sizes of green synthesized nano-sized ZnO particles were calculated from XRD data by using Debye-Scherrer equation. After calcined at 400 °C, amorphous nature disappeared and crystalline nature of ZnO appeared but its crystal structure has some impurities peaks. When the calcination temperature was higher than 500 °C, the XRD patterns showed the strong diffraction peaks of ZnO. Moreover, the characteristic peaks of ZnO become sharper and stronger when the calcination temperatures changed from 300 to 400, 500 and 600 °C, indicating that ZnO were getting better nanocrystalline size. The size data revealed

that the crystallite size increased with the increase in the final calcination temperature. Their difference in crystallization and crystallite size could be mainly attributed to the relative calcination temperature. The average crystallite sizes of green synthesized nano-sized ZnO particles from nitrate source at different calcinations temperature were calculated to be 24.82 nm at 300 °C, 25.99 nm at 400 °C, 31.26 nm at 500 °C and 30.31 nm at 600 °C, respectively. It was found that average crystallite size increase with increase in calcination temperature up to 500 °C but decreased in calcination temperature of 600 °C. It may be due to agglomeration in the calcination process. However, the average crystallite sizes of green synthesized nano-sized ZnO particles from acetate source after calcinations at 500 °C the average crystallite sizes were found to be 34.21 nm. The data are shown in Table 1. In order to obtain smaller ZnO particles with well-developed crystal structures, calcination temperature of 500 °C was chosen for the study the application of nano-sized ZnO particles.

Table 1 Average Crystallite Size of the Green Synthesized Nano-sized ZnO Particles at Different Calcination Temperatures

Metal Source	Calcination Temp. (ºC)	Average Crystallite Size (nm)
Zinc nitrate	300	24.82 (17.30-33.33)
	400	25.99 (22.10-30.37)
	500	31.26 (24.65-37.81)
	600	30.31 (24.79-35.13)
Zinc acetate	500	34.21 (30.47-38.96)

Degradation Efficacy of Green Synthesized Nano-sized ZnO (I) and ZnO (II) on Methylene Blue Dye

In this work, the absorption spectra of MB dye (0.2-1.0 × 10^{-5} M at pH 5) were recorded in the wavelength range of 500-700 nm. It was observed that the wavelength of maximum absorption was 664 nm. Standard calibration curve for methylene blue dye at various concentrations (1.0 ×10^{-5}, 0.8 × 10^{-5}, 0.6 × 10^{-5}, 0.4 × 10^{-5} and 0.2 × 10^{-5} M) was constructed. Removal efficacy was calculated by the equation, A_0-A/A_0 x 100, where A_0 is the absorbance of MB solution without nano-sized ZnO particles and A is the absorbance of the MB solution in reaction mixture with nano-sized ZnO particles after irradiation for time t. The degradation efficacy of green synthesized nano-sized ZnO particles was performed and assessed by evaluating the degradation of MB dye as model contaminant under daylight. The degradation efficacy was studied by employing the green synthesized nano-sized ZnO (I) and ZnO (II) in order to study the degradation of aqueous solution of methylene blue dye via effect of contact time, effect of pH and effect of dosages. Figure 7 shows UV-visible absorption spectra of MB dye showing degradation efficacy at different time intervals (0, 30, 60, 90, 120, 150, 180 min up to overnight) using green synthesized nano-sized ZnO particles from nitrate salt, ZnO (I) under day light without controlling any parameters. It has been observed that the characteristic absorption peak of MB at 664 nm diminishes sharply indicated that green synthesized nano-sized ZnO particles act as photocatalyst for degradation of MB dye.

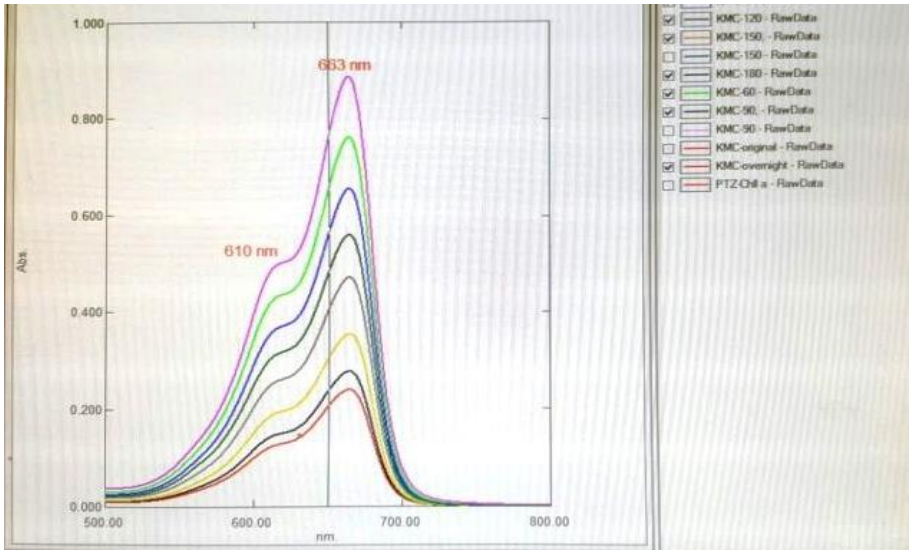

Figure 7 UV-visible absorption spectra of methylene blue dye showing removal efficacy of green synthesized nano-sized ZnO particles from nitrate source, ZnO (I) at different time intervals

Effect of pH

Effect of pH on removal of MB dye under daylight was studied via 50 mL of 2.97×10^{-5} M MB dye solution using of 0.04 g green synthesized nano-sized ZnO particles at various pH after 2 h contact time. Removal efficacy was found to be 21.08 %, 21.31 %, 25.25 %, 31.79 %, 80.38 % and 73.73 % of MB dye by using green synthesized nano-sized ZnO particles synthesized from nitrate source, ZnO (I) and 35.12 %, 40.30 %, 43.40 %, 65.78 %, 68.21 % and 32.98 % of MB dye by using nano-sized ZnO particles synthesized from acetate source, ZnO (II) at various pH of 3, 4, 5, 6, 7 and 8, respectively shown in Table 2 and Figure 8(a). Dye removal activity was highest at pH 7 using both of nanoparticles.

Effect of contact time

Effect of contact time on degradation of 2.97×10^{-5} M/100 mL MB dye solutions was studied at different time interval (0, 30, 60, 90, 120, 150, 180 min) using 0.1 g of green synthesized nano-sized ZnO particles at pH 7. The degradation efficiency of nano-sized ZnO particles as percent degradation was plotted as a function of time shown in Table 3 and Figure 8(b). The percent degradation of MB under daylight were 60.32 %, 72.77 %, 74.86 %, 84.49 %, 84.67 % and 84.89 % using green synthesized nano-sized ZnO particles synthesized from nitrate source, ZnO (I) and 70.07 %, 71.48 %, 73.90 %, 79.48 %, 79.93 % and 80.44 % using green synthesized nano-sized ZnO particles synthesized from acetate source, ZnO (II) at 0, 30, 60, 90, 120, 150 and 180 min contact time, respectively. Interestingly, methylene blue was degraded to 60 % of its initial value within first 30 min of exposure and degradation was increased up to 120 min, after that few degradation of dye was observed (Table 3). At the end of the reaction after 180 min, MB dye removal efficacy was up to 85 % of its initial value. This is in good match with the observed decolourization of MB in first 30 min (Tamanna *et al.,* 2015).

Effect of dosage of nano-sized ZnO particles

Effect of dosage on removal of MB dye under daylight was studied via 2.97 $x10^{-5}$ M/50 mL MB dye solution using various dosage of green synthesized nano-sized ZnO particles at contact time 2 h. Different amounts of nano-sized ZnO particles were used (0.02 to 0.05 g) and the results are given in Table 4 and Figure 8(c). Removal percent of methylene blue dye solution increased gradually with an increase in dosage of nano-sized ZnO particles. The removal percent of methylene blue dye using green synthesized nano-sized ZnO particles at dosages of 0.02, 0.03, 0.04 and 0.05 g were found to be (50.78 %, 59.47 %, 70.41 %, 79.65 %) using nano-sized ZnO (I) and (53.95 %, 66.85 %, 71.53 %, 82.13 %) using nano-sized ZnO (II), respectively. This study provides proof of concept for nano-sized ZnO particles, ZnO (I) and ZnO (II) to be used as efficient degrading agent in environmental remediation applications.

Table 2 Percent Removal of MB Dye at Various pH using Nano-sized ZnO particles

No.	pH	Absorbance at 664 nm		Percent Removal (%)	
		ZnO (I)	ZnO (II)	ZnO (I)	ZnO (II)
1	3	1.400	1.151	21.08	35.12
2	4	1.396	1.059	21.31	40.30
3	5	1.326	1.004	25.25	43.40
4	6	1.210	0.607	31.79	65.78
5	7	0.348	0.564	80.38	68.21
6	8	0.466	1.189	73.73	32.98

Table.3 Percent Removal of MB Dye at Various Contact Time

No.	Contact time (min)	Absorbance at 664 nm		Percent Removal (%)	
		ZnO (I)	ZnO (II)	ZnO (I)	ZnO (II)
1	0	1.774	1.774	-	-
2	30	0.704	0.531	60.32	70.07
3	60	0.483	0.506	72.77	71.48
4	90	0.446	0.463	74.86	73.90
5	120	0.275	0.364	84.49	79.48
6	150	0.272	0.356	84.67	79.93
7	180	0.268	0.347	84.89	80.44

Table.4 Percent Removal of MB Dye with Various Dosages of Nano-sized ZnO particles

No.	Dosage (g)	Absorbance at 664 nm		Percent Removal (%)	
		ZnO NPs (I)	ZnO NPs (II)	ZnO NPs (I)	ZnO NPs (II)
1	0.02	0.873	0.817	50.78	53.95
2	0.03	0.719	0.588	59.47	66.85
3	0.04	0.525	0.505	70.41	71.53
4	0.05	0.361	0.317	79.65	82.13

Where, ZnO NPs (I) and ZnO NPs (II) are nano-sized ZnO particles green synthesized from zinc nitrate source and zinc acetate source after calcined at 500 ºC for 2 h

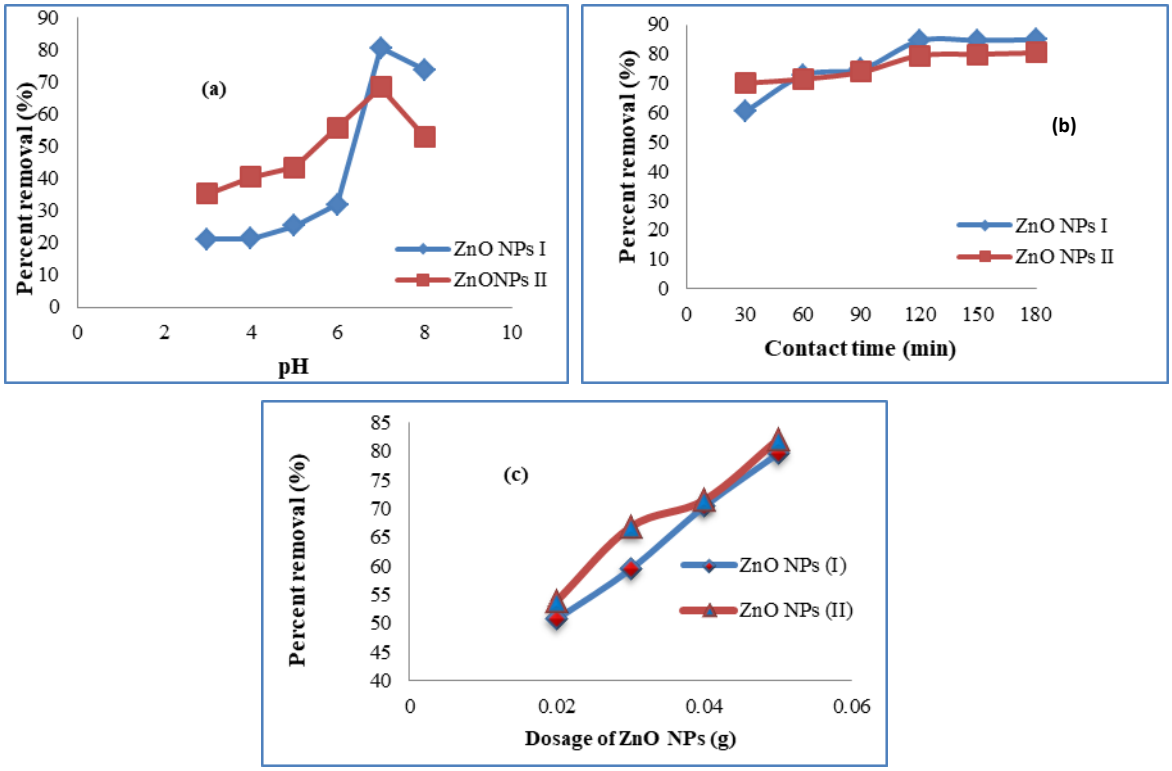

Figure 8 Changes of percent removal of MB dye (a) at various pH using nano-sized ZnO particles (b) at various contact time (c) with various dosage of nano-sized ZnO particles

Conclusion

The present research reported eco-friendly and inexpensive approach for the green synthesis of nano-sized ZnO particles from zinc nitrate and zinc acetate sources using aqueous leaves extracts of Ma-yo-gyi, which act as an effective reducing and stabilizing agent. TG-DTA data of green synthesized zinc oxide from zinc nitrate source revealed the decomposition of $Zn(NO_3)_2$ to ZnO start at 369.9 ºC and completely formed zinc oxide between 400-500 ºC. XRD data confirmed complete formation of pure zinc oxide at 500 ºC with hexagonal phase. The green synthesized nano-sized ZnO particles from two sources have average crystallite size in the range of 24-34 nm with hexagonal structure. SEM and TEM images of the green synthesized zinc oxide from zinc nitrate source and zinc acetate source confirmed the XRD data. It was found that the green synthesized nano-sized ZnO particles form zinc nitrate source have nanoporous and those from zinc acetate source have aggregate particles. From the study on the effect of calcination temperature on average crystallite size of green synthesized nano-sized ZnO particles from zinc nitrate source, average crystallite size increased with increased in calcination temperature up to 500 ºC and at 600 ºC, small decreased in size due to agglomeration of particles in the preparation step. From this finding, calcination temperature was chosen at 500 ºC for green synthesized nano-sized ZnO particles from zinc acetate source. The degradation efficacy of green synthesized nano-sized ZnO particles prepared from two metal sources on degradation of methylene blue dye provided proof of concept for green synthesized nano-sized ZnO particles to be used as efficient degrading agent for environmental remediation application.

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