Synthesis, Structural and Toxicity Characters of Nano-sized Titanium Dioxide/Magnetite Nanoparticles

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Abstract. Titanium dioxide/magnetite nanocomposites become important materials due to their sophisticated potential applications regarding their significant roles in medical science. In this work, we reported the preparation of the titanium dioxide/magnetite nanocomposites prepared using a sol-gel method. The natural sand was explored as an important raw material to prepare the sample. Based on the structural characterizations, the sample size in nanometric ranged of 7.8 nm with amorphous and crystalline phases for titanium dioxide and magnetite, respectively. The vibrational characters of the samples were overlapped originating from Fe-O and Ti-O on the wavenumber ranged between 500-700 cm\textsuperscript{-1}. Microscopy investigation showed that the sample agglomerated in a 3-dimensional fractal dimension as a spherical shape with the size of 34.8 nm. Interestingly, the samples had the optical band gap value of 2.1 eV and presented an excellent character with a low toxicity.

Keywords: Titanium dioxide, magnetite, structure, toxicity, natural sand.

1. Introduction

In the last years, titanium dioxide has been extensively explored one of them related to its performance as an active photocatalyst [1]. In physics, titanium dioxide is one of the materials with good optical and electronic properties [2]. Besides that, titanium dioxide is also stable in biology system, thus it has a great potential in the application of cancer therapy [3]. However, titanium dioxide has a limitation in terms of being relatively complex to be separated from its reaction medium. Therefore, there needs a development of preparation method in order titanium dioxide can be effortlessly separated from the reaction medium, particularly for medical application means.

The previous research showed that the development of titanium dioxide composites with magnetic nanomaterial especially magnetite contributes a vast opportunity to separate titanium dioxide with ease from its reaction medium under external magnetic field [4]. Furthermore, the utilization of magnetite nanomaterial has also been widely developed related to its superparamagnetic properties. Generally,
titanium dioxide/magnetite composites in the nanometer order also create a new application in diverse fields, one of them is in the medical field. The nanocomposites have a massive prospective for the application of drug delivery system [3].

To be applicable in the drug delivery system, one of the pivotal properties from the material used is to have a low toxicity. In this context, the importance of toxicity testing related to liver organ because it is an organ which has an imperative role in the detoxification of toxic process. If one of the materials used in the process of treatment categorized as a drug delivery agent, then the sample needs to have a proper toxicity rate. The latest study showed that toxicity from magnetite and titanium dioxide in the dosage under 100 mg/kg BB does not cause liver damage [5,6]. Unfortunately, the study merely analyzed separately each magnetite and titanium dioxide with commercial material as the main material in producing the samples. Therefore, fundamental studies related to the characteristics of the toxicity of titanium dioxide/ magnetite nanocomposites based on local natural materials that have not been previously reported become urgent. Certainly, other characteristics such as structure, optical properties, and functional groups are also important to be studied before the development of medical applications is carried out.

In relation to material synthesis, several previous studies showed that various methods have been invented to synthesize the titanium dioxide/magnetite nanocomposites such as hydrolysis method [7], solvothermal [8], precipitation [9], and sol-gel [4]. In this research, we opted to select a sol-gel method since this method has advantages in a material synthesis at low temperature, does not need a high cost, and the particle generated can obtain a good chemical composition homogeneity [4]. Interestingly, in this research, we utilized natural iron sand in the synthesis of titanium dioxide/magnetite nanocomposites to reduce the preparation cost.

2. Methods

The synthesis process of titanium dioxide/magnetite nanocomposites was performed in two stages namely magnetite synthesis process using a coprecipitation method which proceeded with a synthesis process of titanium dioxide/magnetite nanocomposites using a sol-gel method. In order the particle distribution generated is homogeneity with the nanometer size, polyethylene glycol was also used in this research (PEG-4000) as template media. In the mixing process of magnetite dan titanium dioxide, magnetite dissolution process was conducted using ethanol followed by the titration process of titanium dioxide solution. Furthermore, the washing and drying processes were done to gain the titanium dioxide/magnetite in the form of powder.

The obtained sample was characterized using XRD, SEM, FTIR, UV-Vis, and SAXS to study its structural characteristics and optical properties. Meanwhile, the toxicity testing was performed to find out its toxicity rate on the test animal (mice) categorized into 4 groups. The samples of titanium dioxide/magnetite were dissolved in 7.5 ml of aqua pro-injection and CMC-Na 0.05% of aqua pro-injection solvent and stirred until homogeneous. Each sample of 0.5 ml of mice was inducted using a feeding tube. The level of concentration given to each group was different, namely, in the first group were given a sample with a concentration of 20 mg/kg body weight, the second group was 40 mg/kg body weight, the third group was 60 mg/kg body weight, and the fourth group was used as a control. After the induction process, an observation for mice was conducted for 7 days. On the seventh day, an autopsy was performed and blood samples were taken from each mouse to conduct SGPT and SGOT testing.

3. Results and Discussion

Figure 1 showed an XRD pattern of titanium dioxide/magnetite nanocomposites with the radiation source of Cu-Kα (λ = 1.54 Å). Based on the analysis result of XRD data in Figure 1, peaks emerged on the 2-theta angle ranged 30.3°, 35.7°, 43.2°, 53.9°, 57.3°, 63.0°, and 74.9°. The diffraction peaks were appropriate with the magnetite crystallography data with the number ICSD 30860, where the sample has a cubical phase with the space group of F d 3 m and lattice constant of $a = b = c = 8.344$ Å. However, the diffraction peaks from the titanium dioxide phase which was expected to emerge on 2-theta angle
ranged 25.2°, 38.1°, 47.1°, 54.8°, and 62.8° was invisible. This X-ray diffraction pattern showed a similar pattern with the results of previous research [10]. Generally, it was due to titanium dioxide having an amorphous property as reported in the previous research [8,11]. This phenomenon can be predicted physically because the heating temperature used in the synthesis process is still relatively low, i.e. at a temperature of 100 °C. The low synthesis process is chosen so that the sample still retains the superparamagnetic properties of magnetite particles. If the sample is synthesized at high temperatures, the superparamagnetic character of the magnetite particles will be damaged and even saturation magnetization will also be reduced dramatically. In relation to this, the previous research also revealed that the heating temperature in the synthesis process also has an essential contribution to the subsiding magnetic property of a material [8].

Figure 1. XRD pattern of titanium dioxide/magnetite nanocomposites

To ensure that the synthesized titanium dioxide/magnetite composite has a nanometer size, Scherrer equation used to calculate of sample particle size as formulated in the Equation (1) was also performed.

\[ D = \frac{k\lambda}{B \cos \theta} \]  

where \( \lambda \) is the wavelength of Cu-Ka, \( \beta \) showed the peak width at half maximum and \( \theta \) was the diffraction angle [10,12]. The value of \( \beta \) is obtained from the results of fitting X-ray diffraction using the Gaussian function by taking the highest diffraction peak. The results of data analysis showed that the sample had a crystal size of 7.8 nm. The crystal size obtained from this analysis tends to be smaller than the magnetite crystal size in general. Thus, PEG-4000 as a template in this study has an important role in controlling the size of nanocomposite titanium dioxide/magnetite particles. The previous study showed that the distribution of PEG is not only effective at controlling a particle size but also in controlling the particle distribution and morphology [13].

FTIR characterization shown in Figure 2 was conducted to find out the functional group and the bonds exist in the titanium dioxide/magnetite nanocomposite. This characterization was performed with an infrared spectroscopy with the wavenumber ranged between 400-4000 cm\(^{-1}\). Based on Figure 2, the atom Fe-O vibration emerged on the wavenumber of 422 cm\(^{-1}\). Interestingly, on the wavenumber of
500-700 cm\(^{-1}\), there was 2 bond vibrations on titanium dioxide/magnetite nanocomposite [9] which was the overlapping between the Fe-O and Ti-O vibration. This can be figured from the peak dilation on the range of the wavenumber \([9,14–22]\). Meanwhile, on the wavenumber of 1630 and 3386 cm\(^{-1}\), there was H-O-H vibration and on the wavenumbers of 1419 and 2876 cm\(^{-1}\) there was C-H atom strain vibration. The previous research also showed that H-O-H vibration appeared at the wavenumber of 1636 cm\(^{-1}\) [23]. Therefore, these data strengthen the analysis results of the previous diffraction that in the titanium dioxide phase there was an amorphous phase undetectable from the X-ray diffraction peaks.

![FTIR spectrum of titanium dioxide/magnetite nanocomposite](image)

**Figure 2.** FTIR spectrum of titanium dioxide/magnetite nanocomposite

![SEM image of titanium dioxide/magnetite nanocomposite](image)

**Figure 3.** SEM image of titanium dioxide/magnetite nanocomposite
The morphological characteristics of titanium dioxide/magnetite nanocomposite can be investigated observed through SEM photography with 100,000 times magnification shown in Figure 3. SEM results show that agglomeration of titanium dioxide/magnetite nanocomposites occurs. Gaussian fitting analysis to obtain the average particle size of the sample is shown in Figure 4. Based on the data analysis, the sample has a particle size of 34.82 nm. Interestingly, this shows different results from the results of the size data analysis obtained from XRD data. Thus, further data analysis that can cover the particle size of the sample from the smallest to the largest size in the fractal structure needs was carried out through the SAXS test as shown in Figure 7. Furthermore, in this research UV-Vis characterization was also carried out to determine the optical properties from titanium dioxide/magnetite nanocomposites such as absorbance and band gap. Figure 5 shows the absorbance for the wavelength of 200–800 nm.
In Figure 5, it can be seen that the absorbance of the sample emerged at the wavelength of 512, 283, 259, and 227 nm, with the magnetite absorbance rate ranged 400–600 nm, while the titanium dioxide absorbance ranged between 200–400 nm. The amount of band gap energy value can be measured using a Tauc plot method, where the measurement was conducted by pulling a linear line on the graphic of $hv$ and $(h\alpha v)^n$ relation until cutting the $hv$ axis. The equation used is shown in the Equation (2).

$$ (h\alpha v)^n = A(hv - Eg) $$  \hspace{1cm} (2)

where $h$ is Planck constant, $\nu$ is frequency, $E_g$ is band gap, and $A$ is proportional constant. The $n$ value showed the transition type with $n = \frac{1}{2}$ for direct transition and $n = 2$ and $\frac{3}{2}$ for the indirect transition. The results of data analysis showed that the band gap energy value of titanium dioxide/magnetite nanocomposites was 2.078 eV.

Further analysis that was important to be performed in this research was SAXS testing to find out the detail information of sample nanostructure as presented in Figure 7. The horizontal axis in the image shows scattering vector (q), while the vertical axis shows scattering intensity (I). Statistically, the experimental data that is shown by the star symbol is well-matched as shown by the blue line. The mathematical model used in SAXS data fittings is lognormal distribution and mass fractal. The results of data analysis showed that the sample had a building block size of about 6.0 nm which had similar results to the results of the XRD analysis. The building block forms secondary particles with a size of about 40.0 nm which has a size similar to the results of the SEM test data analysis. Generally, this structure has a similar result with the previous research results on the nanoparticle magnetite sample [24]. Furthermore, the titanium dioxide/magnetite nanocomposites building block of synthesis results in this research formed a greater aggregation in the 3-dimensional fractal.
In order to observe the toxicity properties of titanium dioxide/magnetite nanocomposites, toxicity testing was conducted using SGPT and SGOT testing to find out the enzyme level in the body. The SGPT enzyme level is usually found in the liver, while SGOT enzyme level not only can be found in the liver, but also in the heart and muscles. The test results of SGPT and SGOT enzyme level for titanium dioxide/magnetite nanocomposites samples are presented in Table 1 and 2.

| Dosage     | SGPT Enzyme Level (mU/ml) | Average (mU/ml) |
|------------|----------------------------|-----------------|
|            | Animal 1 | Animal 2 |                  |
| 20 mg/kgBB | 93       | 96       | 94.5             |
| 40 mg/kgBB | 125      | 130      | 127.5            |
| 60 mg/kgBB | 110      | 273      | 191.5            |

| Dosage     | SGOT Enzyme Level (mU/ml) | Average (mU/ml) |
|------------|----------------------------|-----------------|
|            | Animal 1 | Animal 2 |                  |
| 20 mg/kgBB | 194      | 246      | 220.0            |
| 40 mg/kgBB | 252      | 245      | 248.5            |
| 60 mg/kgBB | 255      | 275      | 265.0            |

Based on the results of toxicity testing in Table 1, it can be seen that the results of SGPT results showed there is an increase of enzyme correlated with the increased of dosage given to the test animal. Whereas the SGPT value of control animals is equal to 243 mU/ml. When compared with the test results, it can be concluded that the dose given to these test animals is still normal and acceptable to the body. The result in Table 2 showed a similar result as in Table 1 where the average of SGOT enzyme increases along with the increase of the dosage given to the tested animals. However, the result is still in the normal level since the value is still under the SGOT value which is 504 mU/ml.
Generally, the SGPT and SGOT enzyme level, on the mice given a treatment, is relatively distinct. It is because the enzyme level depends on the health of the liver organ on each mouse. The result of previous research conducted by Hajshafiei et al. [25] showed that magnetite with doses below 200 mg/kgBW did not have a significant effect on the SGPT and SGOT enzymes, so it could be said to be non-toxic. The same results can also be seen from the results of research conducted by Jia et al. [26] which showed that a magnetite with the dosage under 100 mg/kgBB did not have a significant effect on the results of SGPT and SGOT; instead on the dosage between 150–200 mg/kgBB, the SGPT and SGOT value increased.

Titanium dioxide/magnetite nanocomposites given to the tested animals with the lowest to the highest dosage did not cause any death after 7-day treatment. Eventually, the LD50 value is unknown. LD50 is a maximum dosage which can cause 50% of the tested animal population die. The dosage of 60 mg/kgBB was the highest dosage used in this research, thus this dosage is considered as quasi-LD50 because it does not cause death in tested animals. The results obtained from this study cannot be separated from the factors that affect such as the size, shape, and solubility of the sample. Particle sizes below 100 nm have a high surface area, high reactivity and can easily pass through the cell membrane [27], thus the smaller the particle size, the higher toxic properties [28]. Besides that, concentration, exposal duration, and the experimental model used also caused various results [24]. Thus, the sample of the analysis results in this result is potential to be further developed as a material candidate in medical application.

4. Conclusion
The titanium dioxide/magnetite composites have been synthesized using a sol-gel method. The prepared titanium dioxide/magnetite nanocomposites created a spinel cubic structure with lattice parameter of $a = b = c = 8.344$ Å with the crystal size of 7.8 nm originated from magnetite structure. Meanwhile, from the sample morphology, it can be seen that there is an aggregation with the sample grain size of 8–15 nm. Furthermore, the results of SPGT and SGOT testing showed that titanium dioxide/magnetite composites have a low toxicity.

References
[1] K. Tedsree, N. Temnuch, N. Sitrilai, and S. Pinitsoontorn, “Ag modified Fe$_3$O$_4$@TiO$_2$ magnetic core-shell nanocomposites for photocatalytic degradation of methylene blue,” Mater. Today Proc., vol. 4, no. 5, pp. 6576–6584, 2017.
[2] S. Valencia, J. M. Marin, and G. Restrepo, “Study of the Bandgap of Synthesized Titanium Dioxide Nanoparticles Using the Sol-Gel Method and a Hydrothermal Treatment,” Open Mater. Sci. J., vol. 4, no. 1, pp. 9–14, Feb. 2010.
[3] S. Shen et al., “Core–shell structured Fe$_3$O$_4$@TiO$_2$-doxorubicin nanoparticles for targeted chemosonodynamic therapy of cancer,” Int. J. Pharm., vol. 486, no. 1–2, pp. 380–388, May 2015.
[4] S. Khashan, S. Dagher, N. Tit, A. Alazzam, and I. Obaidat, “Novel method for synthesis of Fe$_3$O$_4$@TiO$_2$ core/shell nanoparticles,” Surf. Coat. Technol., vol. 322, pp. 92–98, Aug. 2017.
[5] K. Parivar, F. Malekvand Fard, M. Bayat, S. M. Alavian, and M. Motavaf, “Evaluation of Iron Oxide Nanoparticles Toxicity on Liver Cells of BALB/c Rats,” Iran. Red Crescent Med. J., vol. 18, no. 1, Jan. 2016.
[6] M. Shakeel, F. Jabeen, S. Shabbir, M. S. Asghar, M. S. Khan, and A. S. Chaudhry, “Toxicity of Nano-Titanium Dioxide (TiO$_2$-NP) Through Various Routes of Exposure: a Review,” Biol. Trace Elem. Res., vol. 172, no. 1, pp. 1–36, Jul. 2016.
[7] W.-J. Chen, P.-J. Tsai, and Y.-C. Chen, “Functional Fe$_3$O$_4$/TiO$_2$ Core/Shell Magnetic Nanoparticles as Photokilling Agents for Pathogenic Bacteria,” Small, vol. 4, no. 4, pp. 485–491, Mar. 2008.
[8] H. Niu et al., “Visible-Light Active and Magnetically Recyclable Nanocomposites for the Degradation of Organic Dye,” Materials, vol. 7, no. 5, pp. 4034–4044, May 2014.
[9] Q. He, Z. Zhang, J. Xiong, Y. Xiong, and H. Xiao, “A novel biomaterial — Fe$_3$O$_4$:TiO$_2$ core-shell nano particle with magnetic performance and high visible light photocatalytic activity,” *Opt. Mater.*, vol. 31, no. 2, pp. 380–384, Oct. 2008.

[10] A. Listanti, A. Taufiq, A. Hidayat, and S. Sunaryono, “Investigasi Struktur dan Energi Band Gap Partikel Nano TiO$_2$ Hasil Sintesis Menggunakan Metode Sol-Gel,” *JPSE J. Phys. Sci. Eng.*, vol. 3, no. 1, pp. 8–15, 2018.

[11] Z. Li, Y. Zhu, J. Wang, Q. Guo, and J. Li, “Size-controlled synthesis of dispersed equiaxed amorphous TiO$_2$ nanoparticles,” *Ceram. Int.*, vol. 41, no. 7, pp. 9057–9062, Aug. 2015.

[12] S. Bhukal, T. Namygal, S. Mor, S. Bansal, and S. Singhal, “Structural, electrical, optical and magnetic properties of chromium substituted Co–Zn nanoferrites Co$_{0.5}$Zn$_{0.4}$CrFe$_{2-x}$O$_4$ (0⩽x⩽1.0) prepared via sol–gel auto-combustion method,” *J. Mol. Struct.*, vol. 1012, pp. 162–167, Mar. 2012.

[13] B. Dutta *et al.*, “PEG mediated shape-selective synthesis of cubic Fe$_3$O$_4$ nanoparticles for cancer therapeutics,” *J. Alloys Compd.*, Dec. 2017.

[14] Y. Wei, B. Han, X. Hu, Y. Lin, X. Wang, and X. Deng, “Synthesis of Fe$_3$O$_4$ Nanoparticles and their Magnetic Properties,” *2011 Chin. Mater. Conf.*, vol. 27, no. Supplement C, pp. 632–637, Jan. 2012.

[15] A. Atta, G. El-Mahdy, H. Al-Lohedan, and S. Al-Hussain, “Synthesis of Environmentally Friendly Highly Dispersed Magnetite Nanoparticles Based on Rosin Cationic Surfactants as Thin Film Coatings of Steel,” *Int. J. Mol. Sci.*, vol. 15, no. 4, pp. 6974–6989, Apr. 2014.

[16] W. Li *et al.*, “Synthesis and characterization of magnetically recyclable Ag nanoparticles immobilized on Fe$_3$O$_4$@C nanospheres with catalytic activity,” *Appl. Surf. Sci.*, vol. 335, pp. 23–28, Apr. 2015.

[17] S. Maria Dhiyva, S. M. Sathiya, G. Manivannan, and M. A. Jothi Rajan, “A Comparative Study on the Biopolymer Functionalized Iron Oxide Nanocomposite for Antimicrobial Activity,” *Int. Conf. Mater. Res. Appl. ICMRA -2016 11–13th March 2016 Dep. Phys. CMR Tech. Campus Hyderabad Telangana State India*, vol. 3, no. 10, Part B, pp. 3866–3871, Jan. 2016.

[18] N. Bachan, A. Asha, W. Jothi Jeyarani, D. Arun Kumar, and J. M. Shyla, “A Comparative Investigation on the Structural, Optical and Electrical Properties of SiO$_2$:Fe$_3$O$_4$ Core–Shell Nanostructures with Their Single Components,” *Acta Metall. Sin. Engl. Lett.*, vol. 28, no. 11, pp. 1317–1325, Nov. 2015.

[19] B. Gaihre, S. Aryal, M. S. Khil, and H. Y. Kim, “Encapsulation of Fe$_3$O$_4$ in gelatin nanoparticles: Effect of different parameters on size and stability of the colloidal dispersion,” *J. Microencapsul.*, vol. 25, no. 1, pp. 21–30, Jan. 2008.

[20] R. A. Ismail, G. M. Sulaiman, S. A. Abdulrahman, and T. R. Marzoog, “Antibacterical activity of magnetic iron oxide nanoparticles synthesized by laser ablation in liquid,” *Mater. Sci. Eng. C*, vol. 53, pp. 286–297, Aug. 2015.

[21] S. Khashan *et al.*, “Photo-thermal characteristics of water-based Fe$_3$O$_4$@TiO$_2$ nanofluid for solar-thermal applications,” *Mater. Res. Express*, vol. 4, no. 5, p. 055701, May 2017.

[22] V. Panwar, P. Kumar, A. Bansal, S. S. Ray, and S. L. Jain, “PEGylated magnetic nanoparticles (PEG@Fe$_3$O$_4$) as cost effective alternative for oxidative cyanation of tertiary amines via C H activation,” *Appl. Catal. Gen.*, vol. 498, pp. 25–31, Jun. 2015.

[23] J H Wei and C J Leng and X Z Zhang and W H Li and Z Y Liu and J Shi, “Synthesis and magnetorheological effect of Fe$_3$O$_4$:TiO$_2$ nanocomposite,” *J. Phys. Conf. Ser.*, vol. 149, no. 1, p. 012083, 2009.

[24] A. Taufiq *et al.*, “Studies on Nanostructure and Magnetic Behaviors of Mn-Doped Black Iron Oxide Magnetic Fluids Synthesized from Iron Sand,” *Nano*, vol. 12, no. 09, p. 1750110, Aug. 2017.

[25] P. Hajishafiee, S. Fatahian, and K. Shahanipoor, “In Vivo Toxicity Assessment of Bovine Serum Albumin and Dimercaptosuccinic Acid Coated Fe$_3$O$_4$ Nanoparticles,” *Iran. J. Biotechnol.*, vol. 12, no. 2, Jun. 2014.
[26] X. Jia, S. Wang, L. Zhou, and L. Sun, “The Potential Liver, Brain, and Embryo Toxicity of Titanium Dioxide Nanoparticles on Mice,” *Nanoscale Res. Lett.*, vol. 12, no. 1, Dec. 2017.

[27] L. Sadeghi, F. Tanwir, and V. Yousefi Babadi, “In vitro toxicity of iron oxide nanoparticle: Oxidative damages on Hep G2 cells,” *Exp. Toxicol. Pathol.*, vol. 67, no. 2, pp. 197–203, Feb. 2015.

[28] M. Kumari et al., “Repeated oral dose toxicity of iron oxide nanoparticles: biochemical and histopathological alterations in different tissues of rats,” *J. Nanosci. Nanotechnol.*, vol. 12, no. 3, pp. 2149–2159, 2012.

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