Synthesis of $Ag/ZnTiO_3$ and Ag/Zn_2TiO_4 by sol-gel method and their antibacterial properties

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ABSTRACT

Antibacterial materials have been studying in many researches. In this study, sol-gel method was applied to synthesis of antibacterial materials from initial components Ag, TiO_2 and ZnO. Ethylenediaminetetraacetic acid (EDTA) was used to produce the medium complexion compounds. $Ag/ZnTiO_3$ powder was formed when the ratio of Ag/EDTA/(ZnO,TiO_2) is 0.1:2:1 while Ag/Zn_2TiO_4 powder was formed when this ratio is 0.1:6:1. The X-ray Diffraction (XRD) results were obtained from these compounds. Then both compounds were tested the antibacterial property with Staphylococcus aureus (S.aureus) in the concentrations of 20mg/ml in 1– 4 hrs.

Keywords: *$Ag/ZnTiO_3$, Ag/Zn_2TiO_4, antibacterial materials.*

1. INTRODUCTION

Nowadays, nanotechnology is a rapidly growing technology to develop new methods in several research fields. These developed materials for reducing bacterial activities which synthesized from metal oxides such as titanium and zinc oxides are also investigated in many researches [1]. The antibacterial activity of nano titanium oxide was proved from the previous researchers that its activity is very highly effective when it was activated by the ultraviolet light with below 380nm wavelengths [2]. While the activity of nano zinc oxide can be also highly effective without the sunlight, but unluckily, it decreases over time [3]…From the above analysis, each kind of these nano oxides meet still some weak points so the combination of them can be made new materials which can be used with or without the sunlight. Therefore, the new materials were created with the more outstanding properties by combining TiO_2 and ZnO together [4, 5]…

The antibacterial materials $ZnTiO_3$, and Zn_2TiO_4 with the nano sizes were made by sol-gel method through medium complex compounds. By changing the reactants ratio,

$ZnTiO_3$ or Zn_2TiO_4 can be formed. Most of zinc metatitanate $ZnTiO_3$ are made with the ratio of $Zn(NO_3)_2$: $TiCl_4$: EDTA is 1:1:1. For the ratio of 2:1:6, Zn_2TiO_4 is more dominant than $ZnTiO_3$.

Meanwhile, antibacterial activities of $ZnTiO_3$ and Zn_2TiO_4 exhibited very weak antibacterial activities against S. aureus bacteria. Silver element was chosen to modify and enhance their antibacterial. It is well known that silver is considered one of the most common antibacterial elements and exhibits potent antibacterial properties with low toxicity for humans and animals by comparison with other heavy metals [7,8]. Ag is effective for both Gram-negative and Gram-positive bacteria, whereas the efficacy of conventional antibiotics varies with the species of bacteria[8]. Many researchers have recently reported that nano Ag demonstrate excellent antibacterial activity [9,10, 11, 12]. Despite of its excellent antiactivity, its price is very expensive so a small amount of it was used

S. aureus is one of the leading causes of infections acquired in the community and after surgery or hospital. It is present in the nose of about 30% of healthy adults and on the skin of about 20%. S. aureus infections range from mild to life threatening. The bacteria tend to infect the skin often causing abscesses. However, the bacteria can travel through the bloodstream and infect almost any site in the body, particularly heart valves and bones. It is the most popular bacteria so many researches are focusing to do research on it.

Nowadays, the environmental pollution in Vietnam is very serious, moreover the booming of population here also makes the good condition for viruses or bacteria grow and develop very fast, a lot of diseases concerning the respiratory, intestine, skin and other cancers keep occurring. So it is nessary to make a new kind of glazed tile which has the highly effective antibacterial properties with or without the sunlight.

In this study, nano powders $Ag/ZnTiO_3$ and Ag/Zn_2TiO_4 was used at rather high concentration because these powders will be covered on the glazed tile to make the antibacterial glazed tile and this kind of material can be used in a long time.

2. EXPERIMENTAL

2.1. Materials

Zinc nitrate hexahydrate ($Zn(NO_3)_2.6H_2O$), silver nitrate ($AgNO_3$) and tetra-n-butyl othotitanate $(C_4H_9O)_4Ti$ were used as the precursors. Ethylenediaminetetraacetic acid – EDTA ($C_{10}H_{16}N_2O_8$) was used to produce the medium complex compounds and ethylene glycol ($C_2H_4(OH)_2$) was used as a dispersing agent. In the process, ammonium hydroxide (NH_4OH) was used to adjust pH of the solution. Tetra-n-butyl othotitanate was obtained from Merck – German, other chemicals were obtained from China through Hoa Nam Company–Vietnam.

The antibacterial property was checked with *Staphylococcus aureus (S.aureus)* (ATCC 29523) which were provided by University of Medicine and Pharmacy - Ho Chi Minh City, Vietnam.

2.2. Synthesis of $Ag/ZnTiO_3$ and Ag/Zn_2TiO_4

$Zn(NO_3)_2$ and EDTA were dissolved in distilled water at 80^0C under stirring. Continuously, NH_4OH was dripped gradually until the final pH of solution was 4.5±0.5.

$(C_4H_9O)_4Ti$ was dissolved in ethylene glycol at 80^0C with stirring and then this solution was poured into the above solution.

$AgNO_3$ was dissolved in ethylene glycol separately with $(C_4H_9O)_4Ti$, then it was poured into this mixtures. After that the solution will make an dark gray gel. This gel was dried at 150^0C for 5 hours. Finally, the ash was calcined at 650^0C in air for 3 hours and was ground for 5 hours by mill ball to obtain the final samples.

2.3. Synthesis of $ZnTiO_3$ and Zn_2TiO_4

The synthesis processes are the same to the previous, but here $AgNO_3$ was not used.

2.4. Characterization

Thermal decomposition of the dried gel samples was analyzed by thermo gravimetric analyzer (TGA), where the sample was heated from room temperature to 900^0C with a heating rate of 10^0C/min in air.

The calcined samples were characterized by X-ray diffraction (XRD, Japan D8–Advance) Powder X-ray diffraction (XRD) patterns of the materials were recorded on a Japan D8 – Advance diffractometer operated at 40kV voltage and 40 mA current and calibrated with a standard silicon sample, using Ni-filtered Cu K□(□=0.154184 nm) radiation. Structural characteristics of as-prepared powders were observed using field emission transmission electron microscopy (FE-SEM).

2.5. Antibacterial test

The antibacterial properties were tested by two methods (Colony Count Method and Minimum Inhibitory concentration (MIC) for $ZnTiO_3$ and Zn_2TiO_4 without the sunlight. The antibacterial activity of the prepared samples was investigated using *S.aureus* (ATCC 29523) with an initial cell density of approximately 10^5 colony forming units (CFU) per ml.

Colony Count Method was used to determine the number of surviving bacterial colonies for a defined period of time (1hr, 2hrs, 4hrs…). The agar plate with a certain concentration of the antibacterial material was inoculated with a standardised inoculum of the bacteria and incubated under standardised conditions. The removal efficiency, was calculated as:

$$\eta = \frac{N_1 - N_2}{N_1} x100\%$$

where η is the percentage of bacterial reduction, N_1 is the number of surviving bacterial colonies from the control sample, and N_2 is the number of surviving colonies from test sample.

The Minimum Inhibitory Concentration Assay is a technique used to determine the lowest concentration of a antibacterial material that inhibits the visible growth of the bacterium under defined test conditions after overnight incubation.

3. RESULTS AND DISCUSSION

3.1. Thermal analysis

TGA/DTA analysis was used to determine the calcination temperature. Fig. 1 shows TGA/DTA curves of dried gel powder which heated in air at the ratio of 10^0C/min. The result shows that the weight loss of dried gel takes place at three distinctly separable levels. During the first level, a small exothermic peak appeared at about 76.79^0C as shown in DTA curve, which indicated the loss of water and ammonium hydroxide in the composite sol (6.092% weight loss). Decomposition of the organic components and EDTA is showed in the second level. The total weight loss in this level is about 26.9%. The exothermic peak at 351^{0}C seems to be corresponded to the decomposition of organic components. A dramatically peak at 643.76^{0}C with the weight loss about 49.75% between 580 and 700^{0}C because of the forming of $ZnTiO_3$. In the temperature range of about 650 to 900^{0}C there is no significant change in the weight of sample.

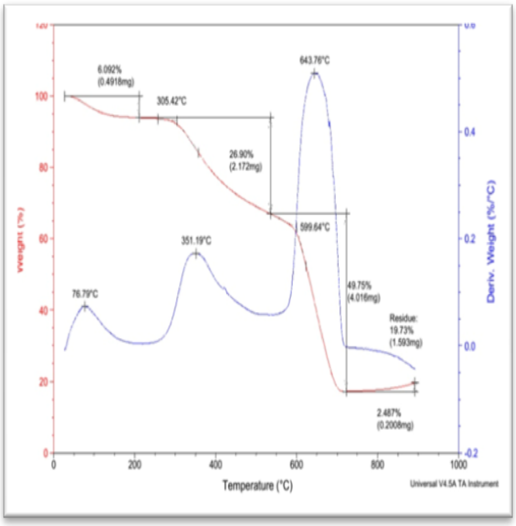

Figure 1. DTA-TGA analysis of $ZnTiO_3$ powders derived from $ZnTiO_3$ precursor solution.

3.2. X-ray Diffraction

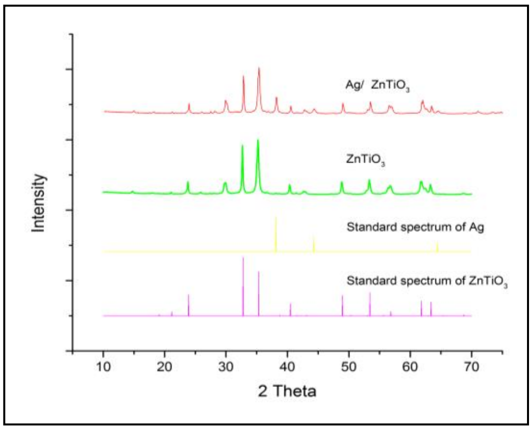

Figure 2. Synthesis $ZnTiO_3$ and Ag/$ZnTiO_3$

The results in Fig.2 showed that, the sample with the ratio $Zn(NO_3)_2$:$(C_4H_9O)_4Ti$:EDTA of 1:1:1 is mainly contained zinc metatitanate $ZnTiO_3$. With the same experiment, $AgNO_3$ was added with the ratio $AgNO_3$:$(C_4H_9O)_4Ti$ of 0.1:1 is a trace of Ag was made with most of zinc metatitanate $ZnTiO_3$.

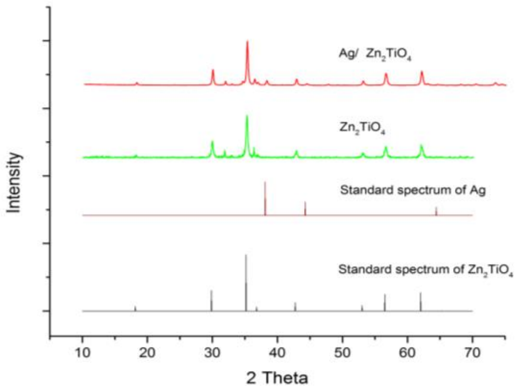

Figure 3. Synthesis Zn_2TiO_4 and Ag/Zn_2TiO

In Fig.3, the sample with the ratio $Zn(NO_3)2$:$(C_4H_9O)_4Ti$:EDTA of 2:1:6 is Zn_2TiO_4 is more dominant than $ZnTiO_3$.

As above, $AgNO_3$ was added with the ratio $AgNO_3$:$(C_4H_9O)_4Ti$ of 0.1:1 is a trace of Ag was made with most of zinc metatitanate Zn_2TiO_4.

3.3. Comparing antibacterial property of the materials with other

- ***The effect of Ag/Zn_2TiO_3 and $ZnTiO_3$ on removal of S.aureus in nutrient medium***

The experiments were done at initial 3.3×10^5CFU (colony forming unit)/ml and 20 mg/ml antibacterial materials. *S.aureus* was cultured in nutrient solution medium. It is easy to find out the antibacterial activity of sample with Ag was much higher than the sample without Ag. Ag/$ZnTiO_3$ could kill all bacteria in 4 hours as showed in Fig.4.

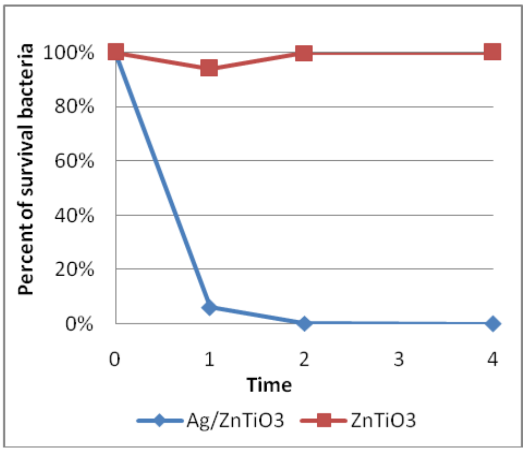

Figure 4. Antibacterial activity of $ZnTiO_3$ and Ag/$ZnTiO_3$ with *S. aureus*.

- ***The effect of Ag/Zn_2TiO_4 and Zn_2TiO_4 on removal of S.aureus in nutrient medium***

The effect of Ag/Zn_2TiO_4 and Zn_2TiO_4 on removal of S.aureus are presented in Fig. 5. The experiments were done at initial 7.7×10^4 CFU/ml and 20mg/ml antibacterial materials and the environment is the same to the previous experiment. In fig.5, it proved that the sample with a small amount Ag had much higher antibacterial activity than that the sample without Ag. All of the bacteria could be killed within 2 hours.

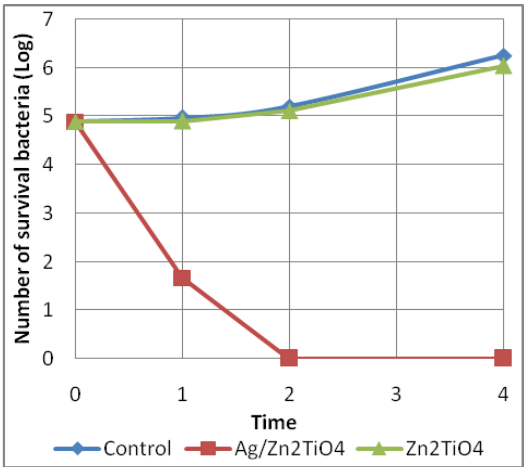

Figure 5. Antibacterial materials Zn_2TiO_4 and Ag/Zn_2TiO_4 with against *S. aureus*.

The effect of Ag/$ZnTiO_3$ and Ag/Zn_2TiO_4 on removal of S.aureus by MIC method are showed in table below:

Table 1: Effect of Ag/$ZnTiO_3$ and Ag/Zn_2TiO_4 on removal of *S.aureus*

Type	MIC (mg/ml)
$ZnTiO_3$	25 mg/ml
Ag/$ZnTiO_3$	2.5 mg/ml
Ag/Zn_2TiO_4	0.78 mg/ml

From above results, it is easy to conclude that the antibacterial activity of Ag/Zn_2TiO_4 is much higher than Ag/$ZnTiO_3$.

4. CONCLUSIONS

Nanosized $ZnTiO_3$, Ag/$ZnTiO_3$, Zn_2TiO_4, Ag/Zn_2TiO_4 and ZnO powders were synthesized by sol-gel method through medium complex compounds. The results showed that nano powders have high crystalinity and $ZnTiO_3$ was obtained when the ratio $Zn(NO_3)_2$:$(C_4H_9O)_4Ti$:EDTA of 1:1:1, and the ratio $Zn(NO_3)_2$:$(C_4H_9O)_4Ti$:EDTA of 2:1:6. The antibacterial activities were tested with *S.aureus* which was cultured in nutrient solution medium to control their development, antibacterial activities of $ZnTiO_3$và Zn_2TiO_4 exhibite very weak antibacterial activity against *S. aureus* bacteria. However, a small amount of $AgNO_3$ (with the ratio $AgNO_3$:$(C_4H_9O)_4Ti$ of 0.1:1) was added to make nano Ag/$ZnTiO_3$ or Ag/Zn_2TiO_4, their antibacterial activies were significantly increased.

Tổng hợp Ag/$ZnTiO_3$ và Ag/Zn_2TiO_4 bằng phương pháp sol-gel và các tính chất kháng khuẩn của nó

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TÓM TẮT

Các vật liệu kháng khuẩn đã đang được nghiên cứu từ nhiều nhà nghiên cứu. Trong nghiên cứu này, phương pháp sol-gel được áp dụng để tổng hợp các vật liệu kháng khuẩn từ các tác chất ban đầu như Ag, TiO_2 và ZnO. Ethylenediaminetetraacetic acid (EDTA) được sử dụng để tạo thành các hợp chất phức chất trung gian. Bột Ag/$ZnTiO_3$ đã được tạo thành khi tỷ lệ Ag/EDTA/(ZnO,TiO2) là 0.1:2:1 trong khi bột Ag/Zn_2TiO_4 được tạo thành khi tỷ lệ này là 0.1:6:1. Các kết quả nhiễu xạ tia X (XRD) thu được từ những hợp chất này. Kế tiếp các hợp chất này đã được kiểm tra tính chất kháng khuẩn với Staphylococcus aureus (S.aureus) với nồng độ 20mg/ml trong 1– 4 giờ.

Từ khóa: *Ag/$ZnTiO_3$, Ag/Zn_2TiO_4, antibacterial materials.*

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