

## High Bactericidal Ability of Silver (Ag)-Coated TiO<sub>2</sub> Films via Doctor Blade Technique

Rungnapa Tongpool\*, Suppharat Inphonlek, Jidaporn Ninlapat,  
Preeyawis Na Ubol and Kongthip Setwong

National Metal and Materials Technology Centre (MTEC)  
114, Paholyothin Rd., Klong 1, Klong Luang, Pathumthani 12120, Thailand

\*Corresponding author. E-mail: [rungnapt@mtec.or.th](mailto:rungnapt@mtec.or.th)

### ABSTRACT

*Films and nanoparticles of P25, precipitated (ppt.) TiO<sub>2</sub>, 1.5%Ag-loaded TiO<sub>2</sub> and 1.5%Ag-loaded P25 were used as photocatalysts in bactericidal reactions. P25 powder was obtained commercially while nanoparticles of ppt. TiO<sub>2</sub>, Ag-TiO<sub>2</sub> and Ag-P25 were prepared by hydrolysis and co-precipitation reactions. Brookite crystal structure was found more in Ag-TiO<sub>2</sub> powder than in TiO<sub>2</sub> powder, indicating that Ag ions influenced the crystallization of TiO<sub>2</sub>. All the films were fabricated by doctor blade technique. The Ag-doped samples showed highest bactericidal activities. However after being washed, the doped powder became ineffective. Unlike the doped powder, the doped films still performed well after being washed, indicating that Ag was strongly adhered to the films. This work succeeded in preparing highly-effective bactericidal films via a simple doctor blade technique.*

**Keywords:** TiO<sub>2</sub>, bactericidal, Ag, film, dissolution

### INTRODUCTION

Disinfection is necessary in drinking and household water. It is an important function of swimming pool, floor tile, window and clothes. It has been known that, when exposed to UV light, TiO<sub>2</sub> generates holes which directly oxidise organic materials or react with water and oxygen, producing hydroxyl (OH) radicals which are highly reactive. These radicals damage the cell wall (Kuhn et al., 2003) and cell membrane of bacteria (Zhang et al., 2003). It causes abnormal cell division and physicochemical alteration of the cell membrane (Amezaga-Madrid et al., 2003]. Silver (Ag) and silver ions have been known to be able to kill germ. They could be reused, handled more easily and more economically if impregnated on substrates. Since TiO<sub>2</sub> is environmental friendly, Ag coated TiO<sub>2</sub> (Ag-TiO<sub>2</sub>) has been widely studied (Sokmen et al., 2001; Zhang et al., 2003). Several work reported that Ag-TiO<sub>2</sub> particles have high bactericidal abilities. However, we found that the dissolved Ag ions (Sokmen et al., 2001), not photocatalytic reaction, were the reason for bacteria reduction. The dissolution of Ag ions results in system contamination and life-time shortening of the photocatalysts. Various work incorporated Ag in TiO<sub>2</sub> films

via sol-gel process (Chang et al., 2006; Colmenares et al., 2006; Ge et al., 2006; Rengaraj and Li, 2006). This process was slow and complicated and the obtained films were expensive and had low surface area. Ag-TiO<sub>2</sub> films from sputtering or vapor deposition require sophisticated equipment. This work proposed high bactericidal ability Ag-TiO<sub>2</sub> films where Ag ions were strongly adhered, using a simple doctor blade technique.

## MATERIALS AND METHODS

TiO<sub>2</sub> nanoparticles (denoted as ppt. TiO<sub>2</sub>) was precipitated from hydrolysis reaction of tetraisopropyl orthotitanate (Ti(OiPr)<sub>4</sub>, Fluka) and calcined at 450°C for 1 h. The Ag-loaded TiO<sub>2</sub>, denoted as Ag-TiO<sub>2</sub>, was obtained from co-precipitation of AgNO<sub>3</sub> (BDH, AnalaR) solution and Ti(OiPr)<sub>4</sub> and then calcined at 450°C for 1 h. The Ag-loaded P25 (a commercial TiO<sub>2</sub>, J.J. Degussa), denoted as Ag-P25, was obtained from mixing P25 and AgNO<sub>3</sub> solution and then calcined at 450°C for 1 h. The molar ratios of Ag in TiO<sub>2</sub> and P25 were 1.5%.

The obtained powder of 1 g was mixed with 1 ml of glycerol (Aldrich Chemical Co., Inc., 99%) and then the slurry was cast on a cover glass, using doctor blade technique. The films were dried and calcined at 450°C for 30 min.

Structural properties of the samples were obtained by an X-ray diffractometer (XRD, JEOL JDX-3530). Particle sizes of the samples were investigated by transmission electron microscope (TEM, JEOL JEM 1220). Surface area of the samples was obtained by BET surface area analysis, using surface area analyzer (Quantachrome Instrument, Autosorb-1).

Before bactericidal study, the powder samples were washed 8 times with 5 ml of distilled water. Then 1 ml of *E. coli* (ATCC 25922) solution was mixed with 1 mg of the powder samples. The mixtures were irradiated with 365-nm UV light (UVP Inc., UVLS 26) for 1 min at the distance of 26 cm from the lamp. Bacteria solution without TiO<sub>2</sub> was also irradiated. Then they were diluted with 0.85% NaCl (Fluka, > 99.5%) and poured onto plate count agar (Oxoid, UK). The colonies were counted after incubation at 37°C for 24 h. Bactericidal ability was determined from % reduction of *E. coli* number as shown in the Equation (1);

$$\% \text{ Reduction} = (n_0 - n) / n_0 \times 100 \quad \dots\dots\dots(1)$$

where  $n_0$  is a concentration of *E. coli* solution without UV exposure and  $n$  is concentration of *E. coli* solution after UV exposure.

As the films can be easily detached from the cover glass, they were washed by immersing in 3 ml of water for 2 days. Then the films were sterilized, using UV light of 254 nm for 15 min. *E. coli* solution of 0.1 ml was dropped onto the film and the film was irradiated with 365-nm UV light for 1 min at the distance of 26 cm from the lamp. Then, the film was cultured in nutrient broth (Oxoid, UK) at 37°C for 24 h. The liquid-cultures of 1 ml was taken to a sterilized tube and mixed with 10 μl

of MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) solution (5 mg/ml), and incubated at 37°C for 4 h. Then 1 ml of 0.04 M HCl in isopropanol was added to all the tubes to dissolve the blue-violet crystals. The obtained solution of 200 µl was taken into a 96-well plate (Nunc, Wiesbaden, Germany) and the absorption at 570 nm was measured, using a microplate reader (Bio-Tek EIA reader, U.S.A). The absorption is proportional to the number of *E. coli*. The *E. coli* on a cover glass without UV exposure was denoted as an untreated sample. Bactericidal ability was determined from Equation (2);

$$\% \text{ Reduction} = (x_0 - x) / x_0 \times 100 \quad \dots\dots\dots(2)$$

where  $x_0$  is the absorption of the solution from untreated samples (no UV exposure) and  $x$  is the absorption of the solutions from the UV-exposed samples.

### RESULTS AND DISCUSSION

According to JCPDS-International Centre for Diffraction Data (2003), P25 and Ag-P25 nanoparticles contain anatase and a small amount of rutile phase, noted from the peak height at the 2-theta of 27.5° (rutile, Fig. 1). The peak height ratios of the 2-theta of 27.5° (rutile) and 25.4° (anatase) is 0.13 for both P25 and Ag-P25, indicating no change in the phase content. The ppt. TiO<sub>2</sub> and Ag-TiO<sub>2</sub> nanoparticles consist of anatase and a small amount of brookite crystal form, noted from the peak height at the 2-theta of 30.9°. The peak height ratios of the 2-theta of 30.9 (brookite) and 25.4 (anatase) is 0.08 and 0.18 for TiO<sub>2</sub> and Ag-TiO<sub>2</sub>, respectively, indicating an increase of the brookite content in Ag-TiO<sub>2</sub>. The phase change occurs probably because the doping process was carried out in a liquid phase where atomic arrangement is possible and the Ag ions might be responsible for this. Since Ag and Ag compound (AgO, Ag<sub>2</sub>O<sub>2</sub>, Ag<sub>2</sub>O) peaks superimpose on TiO<sub>2</sub> peaks (indicated by arrows), they are difficult to be distinguished. However, we found that the calcined doped samples became blackened when left in the room or after being washed. This occurs because the silver changed to silver oxide by oxidation and light (Kim et al., 2003).

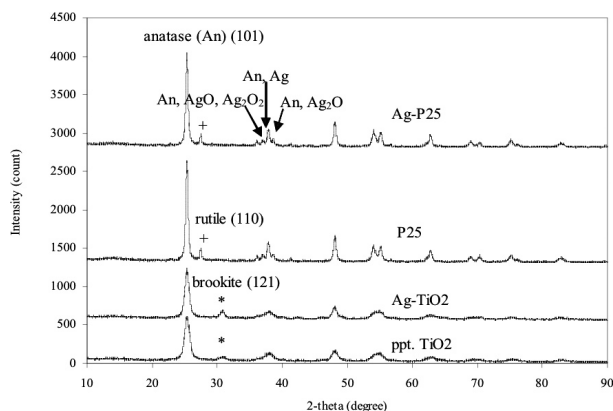
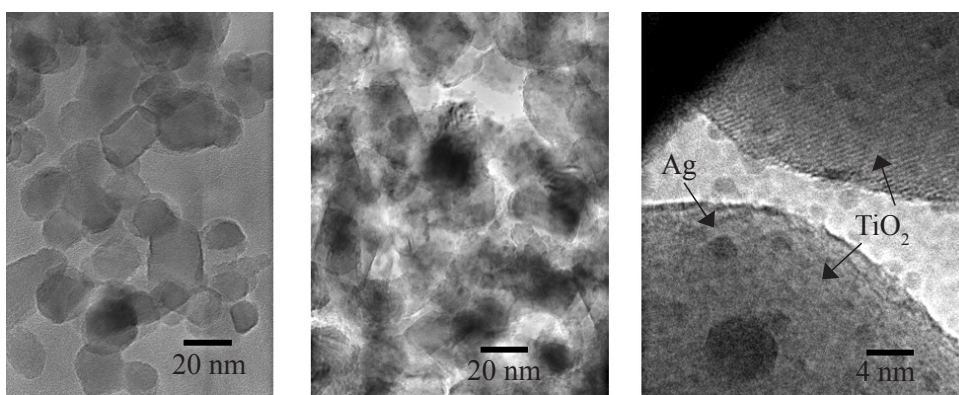


Figure 1. XRD patterns of the sample powders.

The synthesized ppt.  $\text{TiO}_2$  has much higher surface area than the commercial P25 (Table 1), correlating to the smaller particle sizes of the ppt.  $\text{TiO}_2$ . The surface areas of the doped powder were lower than those of the undoped powder, correlating to the larger sizes of the doped samples. Zhang et al (2003) also reported the reduction in surface area of the powder after being doped. Since ionic radii of Ag ( $\text{Ag}^+ = 1.26 \text{ \AA}$  and  $\text{Ag}^{+2} = 0.89 \text{ \AA}$ ) are much larger than that of  $\text{Ti}^{+4}$  ( $0.68 \text{ \AA}$ ), small spheres of Ag compound adhered on the surface of  $\text{TiO}_2$  particles (Fig. 2).

**Table 1.** surface area and particle size of nanoparticle samples.

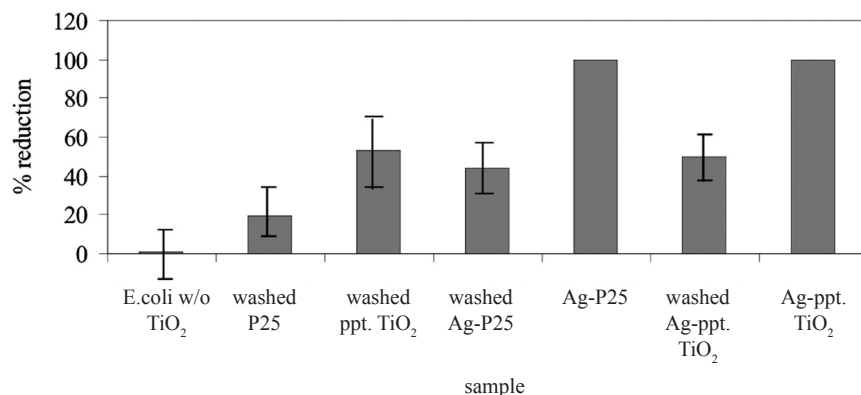
Sample	Surface area ( $\text{m}^2 \text{ g}^{-1}$ )	Particle size (nm)	size (nm)
ppt. $\text{TiO}_2$	126.07+1.10	9.4+2.5	9.43+2.51
1.5%Ag- $\text{TiO}_2$	79.07+0.57	11.3+2.5	
P25	56.07+0.77	23.3+3.9	18.70+8.27
1.5%Ag-P25	49.37+0.83	25.1+8.9	10.06+2.03



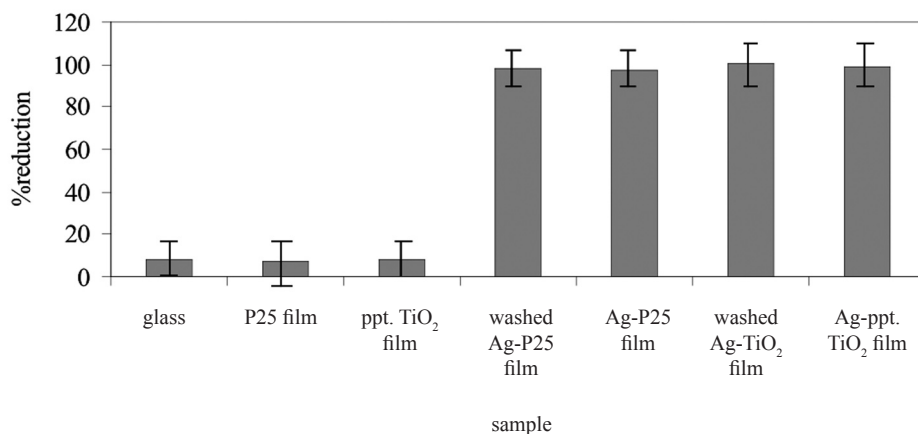
**Figure 2.** TEM images of (a) P25 (b) Ag-P25 powder and (c) Ag-P25 with high resolution.

Bactericidal activities of the samples are shown in Figs. 3 and 4. The ppt.  $\text{TiO}_2$  powder showed higher bacterial reduction than did P25 as a result of higher surface area. The effect of surface area is also demonstrated by the difference in % reduction obtained from the undoped powder which has high surface area and the undoped films which have low surface area. The former shows higher activity than the latter. The Ag-doped samples showed the highest bactericidal activities as % reduction of bacteria number was as high as 98-100%. The Ag-doped powder performed worse after being washed, implying that the Ag was dissolved out. Unlike the doped powder, the washed doped films perform well as the unwashed do, implying that the Ag was strongly adhered onto the films. Bactericidal activities of the doped films were much higher ( $> 11$  times) than the undoped films and the

cover glass, indicating that the photocatalytic activity of  $\text{TiO}_2$  was much enhanced by Ag.



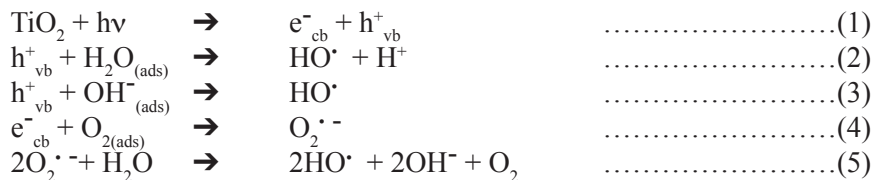
**Figure 3.** Reduction of *E. coli* on the powder samples.



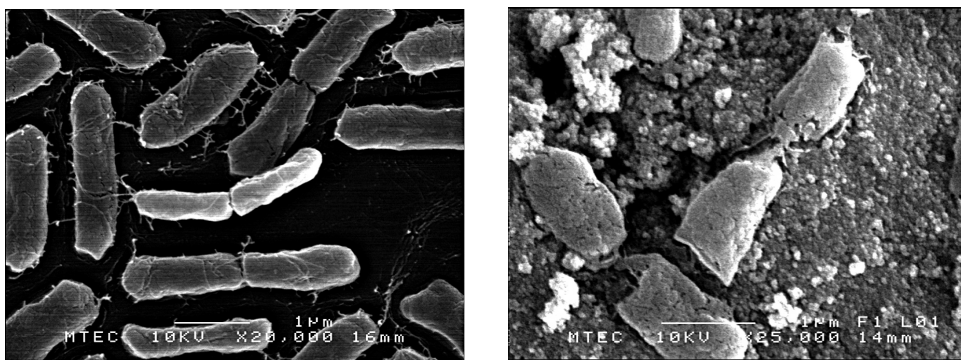
**Figure 4.** Reduction of *E. coli* on the film samples.

This is because of the fact that when exposed to UV, conduction band electrons and valence band holes in  $\text{TiO}_2$  are generated (Equation (1)) and recombined in a short time (Tachikawa et al., 2004). In the presence of Ag on  $\text{TiO}_2$  particle, photogenerated electrons from  $\text{TiO}_2$  were transferred to Ag clusters, as Ag has higher work function than  $\text{TiO}_2$ , inhibiting the electron-hole pair recombination (Zhang et al., 2003). As a result, the holes have longer life time to oxidize organic materials and to react with adsorbed water and hydroxyl groups, producing hydroxyl radicals (Equations (2) and (3)) (Ireland et al., 1993). Some of the photogenerated electrons can react with adsorb oxygen on  $\text{TiO}_2$  surface, generating superoxide ions (Equation (4)) which decompose to hydroxyl radicals (Equation (5)). Hydroxyl radicals are very reactive

and cause the death of the cells (Fig. 5).



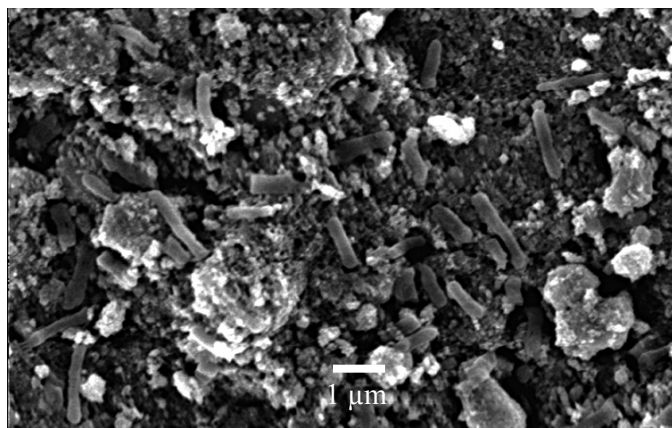
It is possible that the Ag itself (on the films) caused the cell damage as Zhang and Yu (2005) reported that  $\text{Ag}^+$  ions present during the photocatalysis can degrade adsorbed species on the catalyst. In addition to that, during the cell incubation, Ag might dissolve out in a small amount and cause the cell death. However, we found that the longer the illumination, the more the cell damage (Fig. 6). Bare in mind that *E. coli* cells were in contact with Ag-TiO<sub>2</sub> film surface all the time. This confirms that UV light influences the bactericidal ability of the samples.



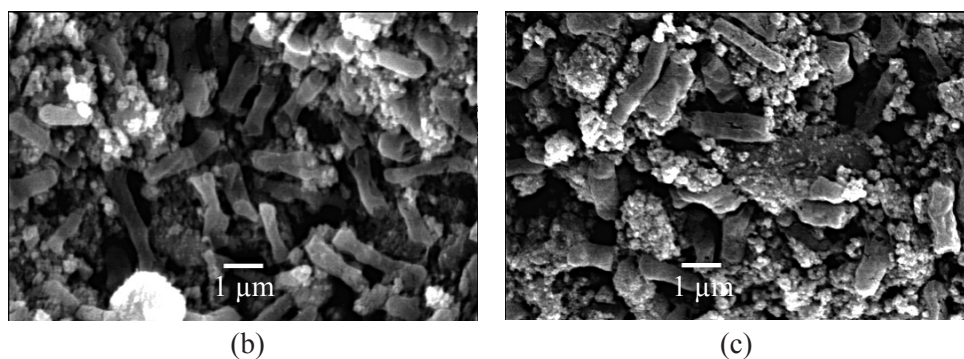
(a)

(b)

**Figure 5.** SEM images of *E. coli* on (a) cover glass without UV exposure and (b) Ag-TiO<sub>2</sub> film with UV exposure.



(a)



**Figure 6.** SEM images of *E. coli* on Ag-TiO<sub>2</sub> films (a) without UV exposure, (b) with 15-sec exposure and (c) with 30-sec exposure.

### CONCLUSION

It was found that there was more brookite structure in Ag-TiO<sub>2</sub> powder than in TiO<sub>2</sub> powder, indicating that Ag influenced crystallization of TiO<sub>2</sub>. Although the doped powder had lower surface area, they were more reactive than the undoped samples, indicating that Ag played an important role in bactericidal ability. There was Ag dissolution from the Ag-doped powder during the washing process, leading to a decrease in bactericidal ability. Unlike the doped powder, the doped films showed high reactivity after being washed, eleven times higher those without Ag. This work was a success in preparing effective doped films via doctor blade technique.

### ACKNOWLEDGEMENTS

This work was supported by National Synchrotron Research Centre, Thailand and National Metal and Materials Technology Centre, Thailand. We acknowledge Dr. P. Songsirithigul for his helpful discussion on XPS.

### REFERENCES

- Amezaga-Madrid, P., R. Silveyra-Morales, L. Cordoba-Fierro, G. V. Nevarez-Moorillon, M. Miki-Yoshida, E. Orrantia-Borunda, and F. J. Solis. 2003. TEM evidence of ultrastructural alteration on *Pseudomonas aeruginosa* by photocatalytic TiO<sub>2</sub> thin films. *Journal of Photochemistry and Photobiology B: Biology* 70: 45-50.
- Chang, C. C., C. K. Lin, C. C. Chan, C. S. Hsu, and C. Y. Chen. 2006. Photocatalytic properties of nanocrystalline TiO<sub>2</sub> thin film with Ag additions. *Thin Solid Films* 494: 274-278.
- Colmenares, J. C., M.A. Aramend'a, A. Marinas, J.M. Marinas, and F.J. Urbano. 2006. Synthesis, characterization and photocatalytic activity of different metal-doped titania systems. *Applied Catalysis A: General* 306: 120-127.

- Ge, L., X. Mingxia, and H. Fang. 2006. Photo-catalytic degradation of methyl orange and formaldehyde by Ag/InVO<sub>4</sub>-TiO<sub>2</sub> thin films under visible-light irradiation. *Journal of Molecular Catalysis A: Chemical* 258: 68-76.
- Ireland, J. C., P. Klostermann, E. W. Rice, and R. M. Clark. 1993. Inactivation of *Escherichia Coli* by titanium dioxide photocatalytic oxidation. *Applied and Environmental Microbiology* 59: 1668-1670.
- Kim, S. S., I. Park, B. C. Shin, and S. H. Lee. 2003. Antibacterial agents, and antibacterial and deodorizing solution comprising the same. US patent 20030190370.
- Kuhn, K. P., I. F. Chaberny, K. Massholder, M. Stickler, V. W. Benz, H. Sonntag, and L. Erdinger. 2003. Disinfection of surfaces by photocatalytic oxidation with titanium dioxide and UVA light. *Chemosphere* 53: 71-77.
- Rengaraj, S., and X. Z. Li. 2006. Enhanced photocatalytic activity of TiO<sub>2</sub> by doping with Ag for degradation of 2,4,6-trichlorophenol in aqueous suspension. *Journal of Molecular Catalysis A: Chemical* 243: 60-67.
- Sokmen, M., F. Candan, and Z. Sumer. 2001. Disinfection of *E. coli* by the Ag-TiO<sub>2</sub>/UV system: lipidperoxidation. *Journal of Photochemistry and Photobiology A: Chemistry* 143: 241-244.
- Tachikawa, T., S. Tojo, K. Kawai, M. Endo, M. Fujitsuka, T. Ohno, K. Nishijima, Z. Miyamoto, and T. Majima. 2004. Photocatalytic oxidation reactivity of holes in the sulfur- and carbon-doped TiO<sub>2</sub> powders studied by time-resolved diffuse reflectance spectroscopy. *Journal of Physical Chemistry B* 108: 19299-19306.
- Zhang, L., J. C. Yu, H. Y. Yip, Q. L., K. W. Kwong, A. Xu, and P. K. Wong. 2003. Ambient light reduction strategy to synthesize silver nanoparticles and silver-coated TiO<sub>2</sub> with enhanced photocatalytic and bactericidal activities. *Langmuir* 19: 10372-10380.
- Zhang, L., and J. C. Yu. 2005. A simple approach to reactivate silver-coated titanium dioxide photocatalyst. *Catalysis Communications* 6: 684-687.