ISSN(Online): 2319-8753
ISSN (Print): 2347-6710

International Journal of Innovative Research in Science, Engineering and Technology

(An ISO 3297: 2007 Certified Organization)

Vol. 5, Issue 8, August 2016

The effect of Nano-Multi-Functional Photocatalyst on the Bacteria Motion

Mohammadreza Elahifard

Engineering Department, Ardakan University, Iran

Abstract: The synthesized antibacterial nano-composite which includes TiO_2, hydroxyapatite, Ag, and AgBr as photocatalyst, is nontoxic and shows bacteria-static properties in the dark media. In this work the rotational motion of bacteria in the dark at a fixed position in the presence of the nano-composite is reported for the first time. The same bacteria-static effect of synthesized nano-composite on all types of bacteria (Gram-positives and Gram-negatives) in the dark is another result that introduced the new antibacterial mechanism. Transmission Electron Microscopy (TEM) and optical microscopic observation in the dark indicated that the nano-composite, only traps the bacteria by chemical bonding and the rotational motion begins when the bacteria becomes heavier due to adhering of catalyst particles to its surface which causes it to have circular motion in a fixed position.

Keywords: Rotational Motion, Antibacterial Mechanism, Bacteria-Static.

I. INTRODUCTION

The various interactions between the chemical, biological and environmental agents and bacteria are very important for a better understanding of its mechanism. Bactericides have been extensively investigated for the killing of bacteria with various antibacterial mechanisms [1,2]. The main drawbacks are that most bactericides only affect a specific type of bacteria [3] and furthermore, many of them have not been applied due to their toxic characterization and environmental degradation [4,5]. Antibacterial effect on modified TiO_2 [6-8] has been investigated as a result of photocatalytic properties. The mechanism of the antibacterial photocatalyst has been extensively studied and has been shown that reactive oxygen species are responsible for photokilling mechanism [9-12]. In an earlier work, [13] we introduced the antibacterial composite with two mechanisms that can affect on various bacteria in different conditions. This composite affects on the *E.coli* as a photocatalyst and bacteria-static agent in the visible and dark respectively. In this communication the effect of nano-composite on the motion of bacterium as one of the biological activity was considered.

Bacillus subtilis, and *Escherichia coli* that are a Gram-positive bacterium and Gram-negative bacteria, respectively, were used as the photo typical micro-swimmer in this study.

II. MATERIALS AND METHODS

Apatite-coated $Ag/AgBr/TiO_2$, was prepared as a sample of synthesized antibacterial group, through deposition of hydroxyapatite, as adsorption bioceramic, and AgBr, as photosensitive material. The preparation procedure has been explained elsewhere [5].

To measure the catalytic activity under dark media, 24 mg of photocatalyst was added to bacterial suspension. The Escherichia coli (ATCC, 8739) and Bacillus subtilis (ATCC, 6633) were prepared in 1×10^7 colony-forming units (CFU/ml) bacteria cell concentration. The reaction mixture was stirred with a magnetic stirrer to prevent the precipitation of the photocatalysts, and in certain time intervals, 2 ml of the reaction mixture was diluted with 0.9% saline. Then 1 ml of diluted solution was incubated at 37°C for 24 h on sybeen caseion digest agar, and the colonies were counted.

ISSN(Online): 2319-8753
ISSN (Print): 2347-6710

International Journal of Innovative Research in Science, Engineering and Technology

(An ISO 3297: 2007 Certified Organization)

Vol. 5, Issue 8, August 2016

For the TEM analysis the sample were prepared according to the standard procedure [10]. A photo camera coupled with optical microscope was used to follow the motion of bacteria.

III. RESULT AND DISCUSSION

The effect of apatite-coated $Ag/AgBr/TiO_2$ on growth of *E.coli* and B.sub were evaluated in the dark. Results indicated that after 3h nano-composites can inhibit the growth of two types of the bacteria (Figure 1).

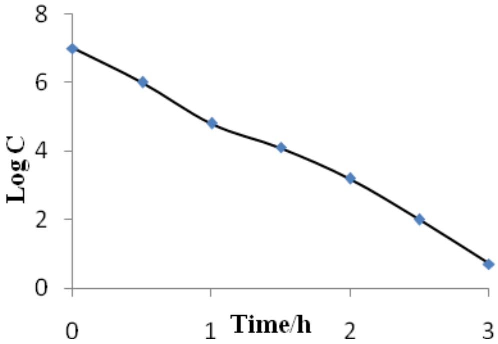


Figure 1: Average of the *E.coli* and B.sub inactivation (1×10^7, 30 mL) in aqueous dispersions containing 24 mg of nano-catalysts apatite (5%) $Ag/AgBr/TiO_2$ (P-25) under dark condition.

The multi composite cannot be toxic due to the fact that the deposition method to make the composite included deposition of hydroxyapatite on $Ag/AgBr/\ TiO_2$ which is not toxic as well [10, 13], The Transmission Electron Microscopy (TEM) investigation showed that the catalyst nanoparticles adhere to the outer membrane of the cell with possible hydrogen and covalent bonding (with dehydration). This linkage between outer membrane of bacteria, peptidoglycan and lipopolysacarid from Gram-positive and Gram-negative respectively, and phosphate group of apatite were formed. As shown in TEM image of *E.coli* after photocatalytic test (Scheme1) the outer membrane of the cell was not damaged because the colored interior of the cell did not become white [10].

The motion of bacteria in certain time intervals was followed as complimentary investigation which demonstrated that the motion of bacteria was still continued after 3 h. this serves as the other reason for the bacteria-static effect of nano-composite. However, this motion is circular which is different from natural motion of bacteria (running and tumbling) (Figure 2). Thus, the apatite-coated $Ag/AgBr/TiO_2$ does not kill the bacteria but only traps and inhibit them from the growth in the dark.

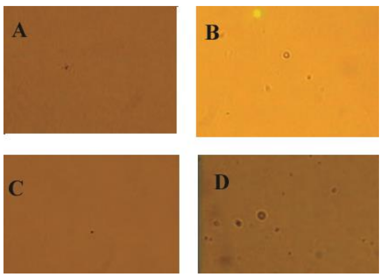


Figure 2: The microscopic image of B.sub in (a) first time (in absent of photocatalyst), (b) final time (after 3h in presence of photocatalyst), and *E.coli* in (c) first time, and (d) final time.

ISSN(Online): 2319-8753
ISSN (Print): 2347-6710

International Journal of Innovative Research in Science, Engineering and Technology

(An ISO 3297: 2007 Certified Organization)

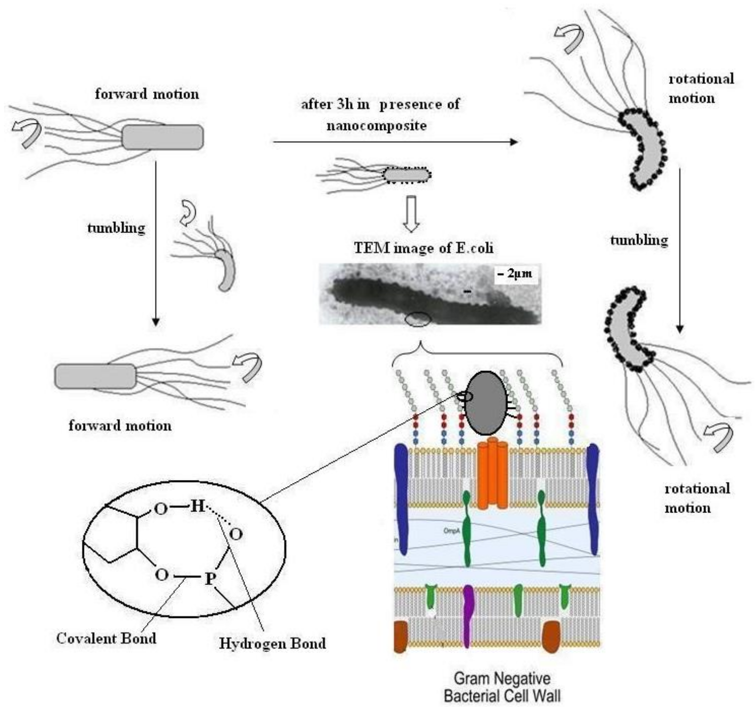
Vol. 5, Issue 8, August 2016

Bacteria swim in a random walk by the action of rotary motors embedded in the cell wall. If all the motors of bacteria rotate counter-clock wise the bacteria cell swims forward, when one or more motors rotate clock wise, the bacteria reorient rapidly and swim in the opposite direction [14].

Berg and Turner [15] reported that *E.coli* does not have straight runs near solid surfaces but they observed a clockwise circle, when viewed from above which is due to the flagellar bundle rotates counter-clock wise.

This rotational motion is different from circular trajectory near solid boundaries and glass surface which can be summarize as follows; (a) New motion observed in a fixed position, (b) both clockwise and counter-clockwise circular motion were observed due to tumbling force, and (c) the speed of the rotational motion is very slow (approximately 1Hz) compared to the circular trajectory which rotates at speeds of 10 Hz (see supporting information).

This motion is due to the asymmetrically adhering of nano-composites on the bacterium surface. Therefore, we proposed the bacterium becomes heavier unequally due to this adherence, and thus, the rotary motors cannot power the bacteria forward and consequently, the bacterium is forced to a rotational motion in a fixed position (Scheme 1).



Scheme 1: Various kind of *E.coli* motion.

Supplementary Information

Movie 1 and movie 2: These movies show the initial (without catalyst) and final (after 3 hours in presence of catalyst) motion of *E.Coli* in dark operation condition.

Movie 3 and movie 4: These movies show the initial (without catalyst) and final (after 3 hours in presence of catalyst) motion of B. Sub in dark operation condition.

IJIRSET

ISSN(Online): 2319-8753
ISSN (Print): 2347-6710

International Journal of Innovative Research in Science, Engineering and Technology

(An ISO 3297: 2007 Certified Organization)

Vol. 5, Issue 8, August 2016

The samples were diluted in movie 1, 3 to follow the motion of one bacterium. No dilution was performed in sample of movie 2, 4 to emphasise that all of bacteria motion was changed. This thorough investigation demonstrated that synthesized composite has inactivation effect on bacteria in the dark or traps them in a circular motion in vivo. The proposed mechanism introduces the new kind of antibacterial composites that affect on all type of bacteria similar to each other.

REFERENCES

[1] Dantas G, Sommer MOA, Oluwasegun RD, Church GM, “Bacteria Subsisting on Antibiotics. Science” 2008; Vol 320 pp: 100-103.

[2] Villain-Guillot P, Gualtieri M, Bastide L, Roquet F, Martinez J, et al. “Structure−Activity Relationships of Phenyl-Furanyl-Rhodanines as Inhibitors of RNA Polymerase with Antibacterial Activity on Biofilms”, J Med Chem Vol 50 pp. 4195-204, 2007

[3] Martínez JL, “Antibiotics and Antibiotic Resistance Genes in Natural Environments”, Science Vol 321 pp. 365-367, 2008

[4] Adams A, Freeman WA, Holland A, Hossack D, Inglis J, et al. “Sulphasomizole (5-p-Aminobenzenesulphonamido-3-Methylisothiazole): A New Antibacterial Sulphonamide” Nature Vol 186 pp. 221-222, 1960.

[5] Michael R, Brown W, Richards RME, “Effect of Ethylenediamine Tetraacetate on the Resistance of Pseudomonas aeruginosa to Antibacterial Agents”, Nature Vol 207 pp.1391-1393, 1965

[6] Asahi R, Morikawa T, Ohwaki T, Aoki K, Taga Y, “Visible-Light Photocatalysis in Nitrogen-Doped Titanium Oxides”, Science Vol 293 pp. 269-271, 2001

[7] Ryu J, Kim KY, Hahn BD, Choi JJ, Yoon WH, et al. “Photocatalytic nanocomposite thin films of TiO2-β-calcium phosphate by aerosol-deposition”, Catal Commun Vol 10 pp.596-599, 2009

[8] Vamathevan V, Amal R, Beydoun D, Low G, McEvoy S, “Photocatalytic oxidation of organics in water using pure and silver-modified titanium dioxide particles”, J Photochem Photobiol A, Vol 148 pp. 233-245, 2002

[9] Fu G, Vary PS, Lin CT. “Anatase TiO2 Nanocomposites for Antimicrobial Coatings”, J Phys Chem B, Vol 109 pp. 8889-8898, 2005

[10] Hu C, Lan Y, Qu J, Hu X, Wang A. Ag/AgBr/TiO2 Visible Light Photocatalyst for Destruction of Azodyes and Bacteria. J Phys Chem B. Vol 110 pp. 4066-4072, 2006.

[11] Jacoby WA, Maness PC, Wolfrum EJ, Blake DM, Fennell JA, “Mineralization of Bacterial Cell Mass on a Photocatalytic Surface in Air” Environ Sci Technol Vol 32, pp. 2650-2653, 1998

[12] Ma N, Fan X, Quan X, Zhang Y, “Ag–TiO2/HAP/Al2O3 bioceramic composite membrane: Fabrication, characterization and bactericidal activity” J Membr Sci Vol, 336, pp. 109-117, 2009

[13] Elahifard MR, Rahimnejad S, Haghighi S, Gholami MR, “Apatite-coated Ag/AgBr/TiO2 visible light photocatalyst for destruction of bacteria”, J Am Chem Soc. Vol 129, pp. 9552-9553, 2007

[14] Lauga E, Diluzio WR, Whitesides GM, Stone HA, “Swimming in Circles: Motion of Bacteria near Solid Boundaries” Biophys J 90: pp. 400-412, 2006

[15] Berg HC, Turner L, “Chemotaxis of bacteria in glass capillary arrays. Escherichia coli, motility, microchannel plate, and light scattering” Biophys J, Vol 58, pp. 919-930, 1990.