



Preparation of Ag/RGO nanocomposite as antibacterial waterborne paints additive

Fatemeh Farsinia *, Elaheh K. Goharshadi*, Navid Ramezani*, Masoomeh Mehraban*, Marjan Moghayedi*

١. F. Farsinia

Department of Chemistry, Faculty of Science, Ferdowsi University of Mashhad, Mashhad 9177948974, Iran

٢. Prof. E. K. Goharshadi

Department of Chemistry, Faculty of Science, Ferdowsi University of Mashhad, Mashhad 917794897, Iran

Abstract

Nanomaterials can be used as antimicrobial additives in different applications such as water-borne paints. The fabrication of antimicrobial paints is important because of the prevention of indoor biological colonization, paint bio-deterioration, and health problems in people and pets. The antipathogen paints have several applications in different areas such as health centers, elderly care centers, emergency rooms, and hospitals. In this work, Ag/RGO nanocomposite was synthesized as an antimicrobial waterborne paint additive. The antibacterial activity of a waterborne paint formulated with Ag/RGO nanocomposite was evaluated against *Escherichia coli* (ATCC25922) as a gram-negative bacterium using the microdilution method. The antibacterial tests showed that Ag/RGO nanocomposite has strong antibacterial activity.

Keywords: Water-borne paints, Antimicrobial, Microdilution method, Gram-negative bacterium.



1. Introduction

Paints are used for protective and decorative purposes. Their properties largely depend on their ingredients. They basically protect surfaces from corrosion, oxidation, environmental weathering, or other types of deterioration [1]. All paints contain the same primary constituents including a matrix or binder, pigments, extenders (which confer color and build), and an appropriate solvent [2]. The ingredients can act as a carbon source for several species of microorganisms. Binders provide the adhesion and cohesion, keep the pigments within the coating, and ensure that the paint remains attached to the substrate. Paints can be classified according to their kind of binder. The examples of organic binders include natural latexes or synthetic polymers like alkyd, acrylic, and poly (vinyl acetate) [1]. Pigments provide color and opacity to paints. Solvents dilute the paints and use in the preparation of paints. The solvents are removed after drying paints [3]. Solvents may be organic in nature or water. Other components besides the above ingredients may be used in the fabrication of paints which contribute of about 5% of total formulation of paint. These extra ingredients include emulsifiers (surfactants), defoamer, wetting agents, dispersants, and biocides. The growth of pathogens including bacteria, fungus, and viruses stains the paints and deteriorates the paint properties [1]. Pathogens damage the paint surface by different phenomena including discoloration, increase of porosity of the paint layer, reduction in physical resistance, and penetrating moisture to the surface.

The antimicrobial agents are chemical substances which inhibit or kill the growth of pathogens. Incorporation of antibacterial agents (biocides) in paints can inhibit the growth of microorganisms on their surfaces [4]. It has been proved that some nanomaterials (NMs) such as Ag nanoparticles (NPs), metal oxide NPs, and carbonaceous NMs have strong antimicrobial properties. Up to now, the antimicrobial activity of different NMs including Ag NPs [5], Cu NPs [6], TiO₂ NPs [7] and ZnO NPs [8] have been reported. In this study, we synthesized Ag/RGO nanocomposite as antimicrobial waterborne paint additive by assessing its antibacterial properties against *Escherichia coli* (ATCC25922) as gram-negative bacterium using the microdilution method.



2. Experimental Section

۲.۱. Microdilution method and MIC determination

In this work, Ag/RGO nanocomposite was prepared. *E. coli* was used to evaluate the antibacterial activity of Ag/RGO nanocomposite by using microdilution method. Before evaluation (assessment) of the samples, the *E. coli* solution with 1×10^6 CFU/mL was cultured in a tryptic soy broth. The suspensions of *E. coli* and Ag/RGO were introduced into 24-well culture plates and then were incubated at 37 °C in an incubator for 24 h. The optical density (OD) of the *E. coli* suspensions was measured at 625 nm. The antibacterial activity of the samples was calculated using the following equation:

$$\text{Antibacterial activity\%} = (OD_1 - OD_2) / (OD_1 - OD_3) \times 100 \quad (1)$$

where OD_1 , OD_2 , and OD_3 is the absorbance of the bacterial suspension without NPs, the bacterial suspension with NPs, and the culture medium, respectively.

2.2. Assessment of antibacterial properties of the prepared paints

In this work, the bactericidal activity of a waterborne paints formulated with Ag/RGO was assessed against *E. coli*. For this purpose, distilled water (50-55%), thickener (0.40–0.50%), defoamer (0.30–0.45%), titanium dioxide (10.00–15.00%), micronized calcium carbonate (20.00–25.00%), and acrylic resin (10.00–15.00%) were mixed to get an appropriate suspension by a high-speed disperser mixer. Then, Ag/RGO with different concentrations of as an additive was added to the above suspension under sonication (60 Hz) for 10 min.

۲.۳. Antimicrobial assessment of the waterborne paints

In a typical procedure, 15 ml of the bacterial suspension of *E. coli* was collected by centrifugation and washed three times using buffer phosphate (PBS). Then, the bacteria were resuspended and diluted to 1.0×10^6 CFU/mL in PBS. 100 μ l of diluted bacterial suspension was injected onto the



surfaces of the control and the painted plates (6 cm). Afterwards, the inoculated plates were incubated at 37 °C for 18 h. After incubation, the pristine and painted plates were rinsed and immersed in 5 mL of PBS and sonicated for 10 min to resuspended the bacterial cells. The retrieved cells were 10-fold diluted to a series of concentrations of bacterial suspensions for agar plating, and then the plates were incubated overnight for CFU counting. The log reduction of pathogens was calculated using the following equation:

$$\log(\text{reduction}) = \log(\text{cell counts of control}) - \log(\text{cell counts on the coated sample}) \quad (2)$$

۳. Results

Table 1 show the percent growth inhibition (GI%) of *E. coli* by Ag/RGO nanocomposite with different concentrations. As this table shows by increasing the concentration of the nanocomposite, the GI% value increases.

Table 1. The GI% of *E. coli* by Ag/RGO nanocomposite

Concentration (µg/mL)	GI%
230	89
280	94
330	97

Figure 1 shows the atomic force microscopy (AFM) images of bacteria before and after exposure to paint containing Ag/RGO. The bacteria cell in buffer phosphate without nanocomposite was used as a control. As shown in Figure 1a, 1b, *E. coli* cells have rodlike



shape with smooth membranes after 24 h in 37 °C of incubation. In contrast, the structure of *E. coli* was degraded in the paint containing 100 ppm Ag/RGO (Figure 1c, 1d). As Figure 1e, 1f reveals that when the concentration of the nanocomposite increases to 150 ppm, more degradations occur in *E. coli* structure with respect to that of 100 ppm.

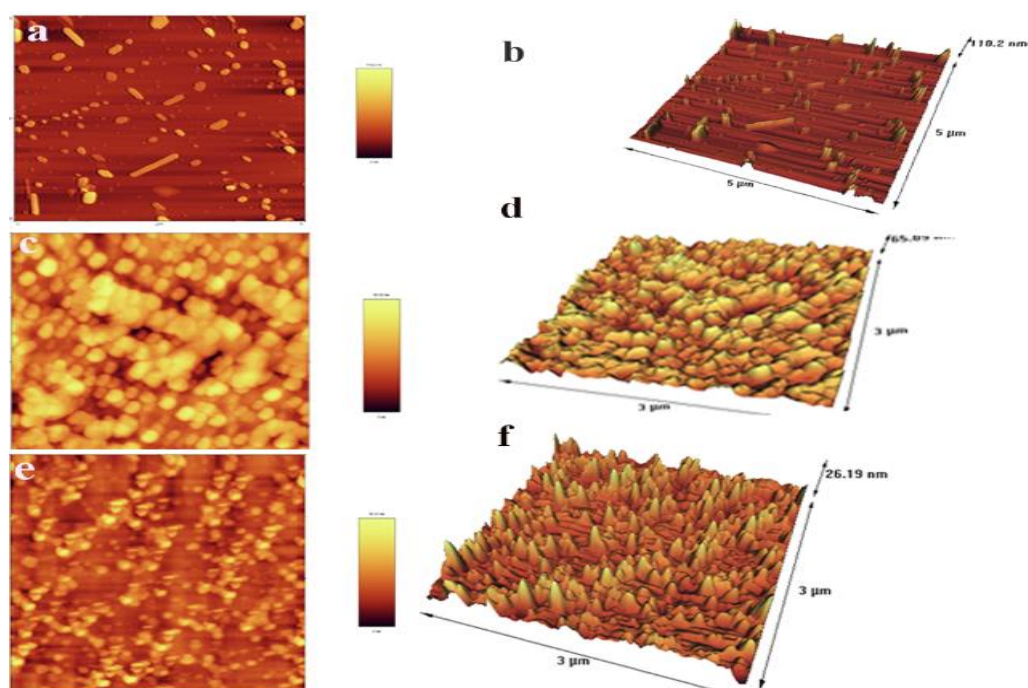


Figure 1. The 2D and 3D AFM images of *E. coli* cells. (a, b) control (c, d) *E. coli* incubation with Ag/RGO nanocomposite (100 ppm) after 24 h, and (e, f) *E. coli* after incubation with the Ag/RGO (150 ppm) nanocomposite after 24 h.

۳. References

[1] Maduka C M and Igwilo S O, Microorganisms survive in paints, Curr. Biotechnol; 2: 1-5, 2019.



- [2] Krishnamoorthy K, Jeyasubramanian K, Premanathan M, Graphene oxide nanopaint, Carbon; 72: 328-337, 2014.
- [3] Robertson J. Forensic Examination of Glass and Paint, 1st ed., Australian: Taylor & Francis Forensic Science Series; 2001.
- [4] Fernández Torneroa A C, García Blascoa M, Chiquirín Azquetab M, Fernández Acevedoc C, Salazar Castroc C and Ramos López S J, Antimicrobial ecological waterborne paint based on novel hybrid nanoparticles of zinc oxide partially coated with silver, Prog. Org. Coat; 121: 130-141, 2018.
- [5] Kumar A, Vemula P K, Ajayan P M and John G, Silver-nanoparticle-embedded antimicrobial paints based on vegetable oil, Nat. Mate; 7: 236-341, 2018.
- [6] Jiang G, Li G X, Che Y, Lv L, Liu F, Wang Y, Zhao C, Wang X, Antibacterial and anticorrosive properties of CuZnO@RGO waterborne polyurethane coating in circulating cooling water, Environ. Sci. Pollut. Res; 26: 9027-9040, 2019.
- [7] Hochmannova L and Vytrasova J, Photocatalytic and antimicrobial effects of interior paints, Prog. Org. Coat; 67: 1-5, 2010.
- [8] Saeed A M, Fattah M, Azzam A M, Synthesis of ZnO nanoparticles and studying its influence on the antimicrobial, anticorrosion and mechanical behavior of polyurethane composite for surface coating, Dyes Pigm; 121: 282-289, 2015.
- [9] Le Y, Hou P, Wang J and Chen J F, Controlled release active antimicrobial corrosion coatings with Ag/SiO₂ core-shell nanoparticles, Mater. Chem. Phys; 120: 351-355, 2010.