Colloids and Surfaces B: Biointerfaces 152 (2017) 49–57

Contents lists available at ScienceDirect

Colloids and Surfaces B: Biointerfaces

journal homepage: www.elsevier.com/locate/colsurfb

Enhanced visible-light-driven photocatalytic bacteria disinfection by g-C_3N_4-AgBr

Jun Deng, Jialiang Liang, Mian Li, Meiping Tong*

The Key Laboratory of Water and Sediment Sciences, Ministry of Education, College of Environmental Sciences and Engineering, Peking University, Beijing 100871, PR China

ARTICLE INFO

Article history:
Received 3 November 2016
Received in revised form
27 December 2016
Accepted 3 January 2017
Available online 4 January 2017

Keywords:
g-C_3N_4-AgBr
Bacteria disinfection
Photocatalysis
Active species
Visible light irradiation

ABSTRACT

g-C_3N_4-AgBr was synthesized by depositing AgBr nanoparticles onto g-C_3N_4. Scanning electron microscopy (SEM), Transmission electron microscope (TEM), X-ray diffraction (XRD), X-ray photoelectron spectroscopy (XPS), UV–vis diffuse reflectance spectra (DRS) and Photoluminescence (PL) spectra were employed to characterize the as-synthesized photocatalysts. The disinfection activities towards representative Gram-negative strain *E. coli* and Gram-positive strain *S. aureus* were examined under visible light irradiation. Complete inactivation of 3×10^6 CFU/mL viable cell density was reached in 60 min for *E. coli* and 150 min for *S. aureus*, respectively. Ag^+ released from the photocatalysts did not contribute to the photocatalytic disinfection process. Direct contact of g-C_3N_4-AgBr composites and bacterial cells, as well as the presence of O_2 was indispensable for the cell inactivation. Photo-generated holes, surface bounded •OH, and indirect generation of intracellular active species played important roles in disinfection process of g-C_3N_4-AgBr under visible light irradiation. The disruption of outside structure of cells as well as inner cell injury led to the inactivation. High pH condition led to increasing the cell disinfection due to the generation of surface bounded •OH.

1. Introduction

Microbial contamination of drinking water has become great threats to human health [1,2]. Although chlorination, ozonation and ultraviolet (UV) irradiation have been shown to be effective in water disinfection [3–5], these techniques yet were found to have some drawbacks, such as the generation of carcinogenic disinfection byproducts (DPBs) and the regrowth of harmful bacteria [6,7]. Therefore, it is urgent to develop innovative water purification technologies. Photocatalytic materials especially the visible light driven (VLD) photocatalysts have been demonstrated to have great potentials to disinfect microbes in water. Great efforts thus have been recently devoted to fabricate different types of VLD photocatalysts [8–15]. For instance, CNT-doped TiO_2 [10,11], graphene oxide deposited TiO_2 [12], Ag/AgX deposited carbon nanotubes (CNTs) [13], Ag/Ti decorated BiOI [14,15] have been synthesized and found to inactivate microbe efficiently with visible light irradiation.

With a medium band gap originating from the tri-s-triazine units connected with planar amino groups, graphitic carbon nitride (g-C_3N_4) was found to exhibit efficient visible light utilization and electric conductivity [16]. As it can also be easily prepared via one-step polymerization of nitrogen-rich precursors such as cyanamide, dicyandiamide, melamine and urea [17], g-C_3N_4 has recently attracted tremendous attention in VLD disinfection field [18–21]. However, due to the small surface area, low visible-light utilization efficiency as well as high recombination of photo-generated electron (e^-)-hole (h^+) pairs, pure g-C_3N_4 exhibited relatively weak disinfection activity under visible light irradiation [22,23]. Different techniques thus have been employed to improve the disinfection efficiency of g-C_3N_4. For example, to increase the surface areas, mesoporous g-C_3N_4 was synthesized by using silica as templates [20]. It was found that with the increase of surface areas, mesoporous g-C_3N_4 showed improved disinfection efficiency [20]. Coupling g-C_3N_4 with cyclooctasulfur (S_8) [21] and TiO_2 [24] have also been demonstrated to significantly enhance the photocatalytic disinfection activity due to the more efficient separation of photo-generated e^--h^+ pairs.

As a fascinating photosensitizer containing strong antibacterial capacity, silver bromide (AgBr) has been widely employed to inactivate microbe [25–27]. Previous studies showed that coupling AgBr

* Corresponding author.
E-mail address: tongmeiping@pku.edu.cn (M. Tong).

http://dx.doi.org/10.1016/j.colsurfb.2017.01.003
0927-7765/

with semiconductors could depress the recombination of charge carriers and thus increase the photocatalytic efficiencies [17,25,28]. Moreover, combination of AgBr with other materials would also enhance the stability of AgBr, which was important for the disinfection with light irradiation [29]. Therefore, decorating g-C_3N_4 with AgBr is expected to increase the stability of composites and also enhance antibacterial activity.

Therefore, the objective of this study is to fabricate g-C_3N_4-AgBr and investigate their bactericidal performance as well as disinfection mechanisms. g-C_3N_4-AgBr was synthesized through decorating AgBr nanoparticles on the surface of g-C_3N_4 via a deposition method. The bactericidal effects of the as-synthesized photocatalysts towards representative Gram-negative strain *Escherichia coli* (*E. coli*) and Gram-positive strain *Staphylococcus aureus* (*S. aureus*) under visible light irradiation were investigated. Mechanisms involved in the VLD disinfection processes as well as the effect of pH were also verified.

2. Materials and methods

2.1. Materials

$AgNO_3$, KBr, NaOH, HCl, NaCl, $FeCl_2{\cdot}4H_2O$, sodium oxalate ($Na_2C_2O_4$), potassium dichromate, ammonium molybdate, bacto-yeast extract, melamine, tryptone, potassium chromate, L-histidine, nitroblue tetrazolium (NBT) and terephthalic acid (TA) were purchased from Sinopharm Corporation Ltd. (Shanghai, China). KI, 4-hydroxy-2,2,6,6-tetramet-hylpiperidinyloxy (TEMPOL) and isopropanol (IPA) were purchased from Sigma-Aldich Chemical Co. (St. Louis, MO). All the chemicals were analytical grade and used without further purification.

2.2. Synthesis and characterization of g-C_3N_4-AgBr

g-C_3N_4 was synthesized by a facile thermal polymerization method [19]. In detail, melamine was placed in an alumina crucible with a cover to form a semi-closed system. It was then heated at 550 °C for 4 h with a ramp rate of about 5 °C/min. The obtained yellow product was bulk g-C_3N_4 powder, which was washed with acid water to neutral. Subsequently, bulk g-C_3N_4 was heated at 550 °C for another 3 h to form g-C_3N_4.

g-C_3N_4-AgBr was synthesized by adsorption–deposition method. In a typical experiment, 0.92 g g-C_3N_4 and 1.7 g $AgNO_3$ were dispersed in 40 mL deionized water and then sonicated it for 30 min. Subsequently, the mixture was stirred for 12 h at room temperature (25 °C) under dark condition. To minimize the formation of AgBr in the bulk solution, the suspension was filtered and washed with deionized water for three times. Then, the solid was dispersed in 40 mL solution with 1.190 g KBr and sonicated for 15 min. After that, the mixture was vigorously stirred for 2 h and the product was collected by filtration, which was washed with deionized water and ethanol for at least five times, respectively. Finally, g-C_3N_4-AgBr was obtained after being dried at 60 °C for 4 h. Composites with different molar ratios of g-C_3N_4 to AgBr (denoted as g-C_3N_4-xAgBr, x = 0.5, 1, 2, 4) were also prepared according to the same procedure with different dosages of $AgNO_3$ and KBr. Pure AgBr without adding g-C_3N_4 was also prepared via precipitation method.

Different techniques including powder X-ray diffraction (XRD), X-ray photoelectron spectroscopy (XPS), scanning electron microscopy (SEM), and transmission electron microscope (TEM) were employed to characterize the as-synthesized photocatalysts. Powder XRD was performed by DMAX-2400 (Rigaku, Japan) with Cu Kα (λ = 0.154 nm) irradiation at 40 kV and 100 mA. XPS pattern was carried out on an Axis Ultra (Kratos Analytical Ltd.) imaging photoelectron spectrometer using a monochromatized Al Kα anode with an internal standard of C1s peak at 284.8 eV. SEM and TEM of the photocatalysts were determined by FEI Nova Nano scanning electron microscopic 430 and Hitachi H9000 transmission electron microscope, respectively. The UV–vis diffuse reflectance spectra (DRS) were recorded for the dry pressed disk samples by a PerkinElmer Lambda 650S spectrophotometer, and $BaSO_4$ was used as a reference. Photoluminescence (PL) spectra were also utilized to characterize the optical properties of the prepared material. Zeta-sizer Nano ZS90 (Malvern Instruments, U.K.) was employed to determine the zeta potentials of synthesized material.

2.3. Bacteria preparation

E. coli ATCC15597 and *S. aureus* ATCC6538 were used in this study as model Gram-negative and Gram-positive cells, respectively. Both *E. coli* and *S. aureus* were cultivated in 100 mL of Luria Broth growth medium, consisting of 10 g/L tryptone, 5 g/L bacto-yeast extract, and 10 g/L NaCl. After inoculation, the growth medium was placed into a thermostatic incubator at 37 °C with a shake of 200 rpm (16 h for *E. coli* and 24 h for *S. aureus*). Then centrifugation (4000 × *g* for 8 min at 4 °C) was used to separate the cells from the growth medium. After that, the bacteria were washed three times to remove the residual growth medium with sterilized physiological saline (0.9% of NaCl at pH 7.0). Bacterial stock solutions were then obtained after the sedimentary bacteria cells were re-suspended in appropriate sterilized physiological saline. The cell density in the bacterial stock solutions was about 3×10^8 colony forming unit (CFU) per mL.

2.4. Disinfection experiments

A Xenon arc lamp (300 W, Osram Instruments, USA) was used as light source in the photocatalytic disinfection experiments and a 100 mL double wall beaker was chosen as the reactor. A UV cut-off filter (kP400 nm) was used to filtrate the UV light irradiation and a solar power meter (TM-207, Tenmars Electronics Co.) was employed to measure the visible light intensity in the center of the reaction suspension. The light intensity was 64 ± 1 mW/cm^2. The schematic diagram of the photocatalytic experimental set-up was provided in Fig. S1 and the irradiance spectrum of the arc lamp was given in Fig. S2. During the experiments, the suspension of bacterial cells and photocatalysts were put in a double wall beaker, and a circulating cooling water bath was employed at outer layer to remove the heat from the light and ensure a constant temperature at 25.0 °C. The light irradiation passed through the cutoff filter and then focused on the suspension solution.

All glass apparatuses utilized were sterilized by an autoclave at 121 °C for 20 min prior to the experiments. In a typical disinfection experiment, 5 mg g-C_3N_4-AgBr was added into 49.5 mL sterilized deionized water, and 0.5 mL of the bacterial stock solution was added after complete mixture. The initial cell concentration was about 3×10^6 CFU/mL. The cell suspension was stirred with a magnetic stirrer throughout the duration of experiment. 0.5 mL of the water samples were collected at different time intervals and then serially diluted with sterilized deionized water to yield the viable cell density by employing the method of plate count. Details of viable cell determination could be found in the supplementary information. To make comparison, disinfection experiments were also conducted under dark condition. Experiments without g-C_3N_4-AgBr under visible light irradiation were also conducted as blank control. Three parallel experiments were conducted for each set of experiments.

To clarify the effects of active species, different scavengers were introduced into the disinfection system to remove the corresponding active species. Specifically, IPA was employed to remove