Graphitic Carbon Nitride as A Promising Antimicrobial Photocatalyst

Hongchen Shen¹, Danmeng shuai¹*

¹ Department of Civil and Environmental Engineering, The George Washington University
* Corresponding author, E-mail: danmengshuai@gwu.edu

Background & Introduction

• Drinking water remains a potential source of epidemic outbreaks, and each year hundreds of thousands deaths worldwide are resulted from pathogen contamination of drinking water;
• Bacteria, protozoa, helminths and virus are main waterborne pathogens (Figure 1);
• Photocatalysts harvest photons the energy of which is equal to or greater than the band gap to produce excited electrons and electron byproducts (DBPs), which are commonly present in current chemical disinfection strategies and are harmful to human health as well as environment;
• In addition, the raw materials for the production of g-C₃N₄ photocatalysts are the earth abundant, inexpensive C- and N-containing precursors;
• A series of reactive oxygen species (ROS), such as hydroxyl radicals, superoxide anion radicals, and hydrogen peroxide are generated during photocatalysis process;
• Waterborne pathogens will be oxidized and inactivated by these ROS;

Methods-Material Preparation

• In this study, g-C₃N₄ powder was first prepared via thermal polycondensation of melamine, cyanuric acid, and barbituric acid (Figure 3.a);
• g-C₃N₄ coupons were fabricated from the powder via a hydraulic press (Figure 3.b);

Methods-Bacterial Strains

• Staphylococcus epidermidis (S. epidermidis) is a Gram-positive bacterium and it is a pathogen which causes hospital acquired infections. Escherichia coli (E. coli) is a Gram-negative bacterium and it is commonly found in the lower intestine of warm-blooded organisms (endotherms).
• Staphylococcus epidermidis (S. epidermidis) and Escherichia coli (E. coli) were selected as target microorganisms in this study;

Methods-Disinfection

• S. epidermidis and Escherichia coli were cultured in Luria-Bertani broth (LB) or tryptic soy broth (TSB) at 37 °C with mixing (120 rpm), respectively;
• Both strains were harvested during their late-exponential phase by centrifugation and diluted in a phosphate-buffered saline (PBS) buffer to prepare bacterial suspension (OD₆₀₀ = 0.5);
• 25 ml of bacterial suspension was mixed with 0.001 g of g-C₃N₄ powder in a sterile glass beaker. The beaker was placed under a white light emitting diode (LED) lamp (7 W) for bacterial inactivation;
• The distance between the surface of the bacterial suspension and the LED lamp was maintained at 15 cm. Bacterial suspension samples were withdrawn from the beaker with pipette every half an hour. The samples were duplicated, diluted in series with the PBS buffer, and plate counting was conducted to determine bacterial viability;

Methods-Biofilm

• S. epidermidis bacterial suspension was prepared as described in disinfection experiment;
• g-C₃N₄ coupons were placed into a sterile six-well plate, and completely submerged by the bacterial suspension (2 ml for each coupon in each well);
• The system was first incubated at 37 °C for 24 h without light and mixing and then mixed to ensure effective bacterial attachment on coupon surface;
• Next, the coupons were transferred to a new sterile six-well plate, and 2 ml of 10 fold diluted TSB was added to submerge the coupons. The system was incubated at 37 °C with a mixing rate of 80 rpm under LED irradiation and in the dark (control experiment). The experimental setup is shown in figure 4;

Objective

The objective of our study is first to develop a new g-C₃N₄ photocatalytic material for antimicrobial applications. The inactivation of planktonic bacteria and biofilms is investigated under indoor lighting. The study will shed light on the development of reactive antimicrobial materials for pathogen transmission control and public health protection.

Result

• Confoocal microscope images of biofilms developed on g-C₃N₄ coupons in the dark condition and under LED irradiation are illustrated in Figure 6. The green spots in the figure illustrates live cells, while the red ones represent dead cells;
• Confocal microscopy images of biofilms developed on g-C₃N₄ coupons. S. epidermidis developed a dense and live biofilm with a thickness of 40-80 μm in three days in the dark condition, and the number of dead cells was limited;
• While very limited cells, dead or live, was observed on the coupon surface under white LED irradiation;

Figure 2. Mechanism of a photocatalyst-TiO₂

Figure 3. g-C₃N₄ powder and coupons

Figure 4. Experimental setup for biofilm development

Figure 5. Disinfection experiment result for E.coli

Figure 6. S. epidermidis biofilms on g-C₃N₄ coupons

Figure 7. S. epidermidis biofilms analyzed by OCT

Figure 8. A biofilm eliminated after white LED irradiation