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## Effect of Bimetallic Zero Valent Iron Nanoparticles Ag/NZVI on Bacterial Growth

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**Abstract:** In this study, a 13-day operation was conducted in order to investigate the effect of Ag-Fe nanoparticles on bacterial growth. To do so, three samples (S1, S2, S3) made of freshly domestic wastewater were treated under anaerobic conditions. S1 was considered as the control batch while 50mg/L of zero valent iron nanoparticles (NZVI) and Ag-Fe bimetallic nanoparticles were added to S2 and S3, respectively. Results showed that the addition of NZVI was effective in activating the bacterial growth. However, Ag-Fe nanoparticles inhibited the bacterial growth. A comparison of the chemical oxygen demand COD of the three samples confirmed the obtained results.

Keywords: Ag-Fe nanoparticles; zero valent iron; bacterial growth; COD.

#### 1. INTRODUCTION

Recently, population growth and modernization have extensively raised the global demand for clean energy and water [1,2]. Around the world, 1.3 billion people lack access to electricity and 783 million people lacking access to clean water [3]. The electrical energy generation through Microbial Fuel Cells (MFCs) using microorganisms is a renewable and sustainable approach. It creates truly an efficient technology for power production and wastewater treatment. The most important part of MFC is the microbes.

Using Nano zero-valent Iron NZVI technique was successfully applied in degrading the chemical pollutants and cleaning wastewater. However, the use of NZVI for enhancing the bacterial growth is still not confirmed yet.

The reactivity of NZVI can be substantially improved by impregnating with a second metal, typically Ni, Ag, or Cu to form so-called bimetallic nanoparticles [4].

A considerable number of investigations have evaluated the effect of NZVI on bacterial growth. In [5], results showed a negative impact of NZVI on Bacillus cereus growth capability, consistent with the entrance of cells in an early sporulation stage, observed by TEM. In [6], the impact exerted by NZVI nanoparticles on bacteria population highlights that toxicity should be dose and species dependent. The objective of this research is to investigate the feasibility of using bi-metallic iron nanoparticles NIZVI/Ag to wastewater to promote the bacterial growth.

#### 2. MATERIALS & METHODOLOGY 2.1 Preparation of NZVI and Ag/NZVI nanomaterials

Zero valent iron nanoparticles were prepared by the chemical reduction of 0.093 M ferric chloride hexahydrate (FeCl<sub>3</sub>.6H<sub>2</sub>O, 99.0%, Junsei Chemical Co., Japan) by 0.58 M sodium borohydride (NaBH<sub>4</sub>, 98.0%, Sigma–Aldrich Inc., USA) within anaerobic conditions provided by continues nitrogen purging [7,8]. NZVI precipitates were formed by the 400 RPM vigorous mixing during the whole synthesis time at  $30 \pm 0.5$  °C. Vacuum filtration was used for the particles separating after washing with deionized water (DIW). The Ag/NZVI bimetallic particles used in this study were prepared by

adding silver nitrate AgNO<sub>3</sub> to the FeCl3·6H2O solution (5% Ag) before dropping the reducing reagent of NaBH<sub>4</sub>.

#### 2.2 Wastewater source

The wastewater samples used in this study were obtained from Mikasagawa domestic wastewater purification center located in the city of Fukuoka, Japan. The wastewater was saved under  $4^{0}$ C in order to inhibit the bacterial all along the period of experiments.

## 2.3 Batch experiments

Batch experiments were conducted using three bottles S1, S2, and S3 filled with 100 mL of wastewater. The samples were saved at 37 °C under anaerobic conditions for 13 days. S1 was the control batch without any additives. NZVI and NZVI/Ag nanoparticles were added to S2 and S3 respectively, with a concentration of 50mg/L.

### 2.4 Measuring Microbial growth

Plate count method was adopted for bacterial growth counting. To do so, 0.5 ml of the saved sample is diluted five times. Then, 1 ml of each dilution is placed in the center of a petri dish. 15 ml of melted agar was poured and well mixed. The plate was saved for 24 hours at 37 °C. Each growth colony is carefully counted and represented a colony forming unit CFU.

# RESULTS & DISCUSSION Effect of NZVI and Ag/NZVI on Bacterial growth



Fig. 1. Effect of NZVI and Ag/NZVI particles on bacterial growth.

For a 13-day operation, cell colonies were counted, and the number of cells were plotted over time as it is presented in figure1.

Results show that adding iron nanoparticles to the medium had a positive effect on bacterial growth. The maximum number of cells was 3.6 E+11 colonies /g.Vs for the S2 while 2.6 E+11 colonies /g.Vs for S1.

Therefore, using NZVI particles activates the bacterial growth, thus the digestion of organic matter is enhanced. However, Ag/NZVI had a toxic effect as it inhibited the growth of bacterial colonies. The maximum number of cells was just 1.4 E+11 colonies /g.Vs.

3.2 Impact of NZVI and Ag/NZVI particles on COD



Fig. 2. Impact of NZVI and Ag/NZVI particles on COD.

The chemical oxygen demand was calculated in order to check the effect of adding nanoparticles on the bacterial activation thus, on the organic matter degradation.

At 0 hour, the COD concentration was 792 mg/l. After 13 days of operation, the COD of the three samples S1, S2 and S3 were 529, 354, 331 mg/L, respectively. Therefore, using NZVI particles enabled the bacterial growth and activation during the period of experiments. However, bacterial communities could be active only since the COD decreased (during the operating days), but their growth was limited as it was presented in figure1.

### 4. CONCLUSIONS

In this study, the microbial growth was subjected to the treatments of NZVI particles and Ag/NZVI bimetallic nanoparticles with concentration of 50 mg/L for 13 days. When treated directly with NZVI particles under anaerobic condition, bacterial growth capability was improved. However, no growth was observed for bacterial communities when they were treated with Ag/NZVI.

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