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Enhanced bioremediation of heavy metal from effluent by sulfate-reducing bacteria with copper–iron bimetallic particles support

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HIGHLIGHTS

- Cu/Fe bimetallic can enhance Cu^{2+} and Zn^{2+} removal and SRB resistance for metals.
- High level metal removal efficiency maintained at wide range pH in SRB–Cu/Fe system.
- The first time found that SRB–Cu/Fe preferable to SRB–Fe^0 for Cu^{2+} and Zn^{2+} removal.
- 97.9% Zn^{2+} and 97.2% Cu^{2+} reduction efficiency was obtained within the first 24 h.
- Heavy metals reduction in SRB–Cu/Fe system was feasible and effective.

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ABSTRACT

The purpose of this study was to investigate the potential of copper–iron bimetallic particles supported sulfate-reducing bacteria (SRB) in enhancing the reduction of Cu^{2+} and Zn^{2+} in effluent. The results showed that the copper–iron bimetallic particles can enhance Cu^{2+} and Zn^{2+} removal and the resistance of the sulfate-reducing bacteria towards metals toxicity, the inhibiting concentration of Cu^{2+} and Zn^{2+} for SRB was significantly increased (from 100 to 200 mg/L for Cu^{2+} and 300 to 400 mg/L for Zn^{2+}). The removal efficiencies of Cu^{2+} and Zn^{2+} (initial concentration 100 mg/L) were 98.17% and 99.67% in SRB–Cu/Fe system after 48 h, while only 29.83% Cu^{2+}, 90.88% Zn^{2+} and 63.81% Cu^{2+}, 72.63% Zn^{2+} were removed in the SRB and Cu/Fe system at the same condition.

1. Introduction

Heavy-metal pollution is a typical representation of environmental problem because of the toxic effects of metals, which can damage nerves, liver, bones and block functional groups of vital enzymes, and their accumulation throughout the food chain leads to serious ecological and health problems (Malik, 2004; Sanyal et al., 2005). These toxic metal ions generated from mining operations, metal-plating facilities, power generation facilities, electronic device manufacturing units, and tanneries and commonly existed in process waste streams (Liu et al., 2009), which has been increased dramatically during the last few decades in ambient environment. Thus, the removal of such toxic metal ions from effluent is a crucial issue.

Most heavy metal salts can dissolve in the water and form aqueous solutions and, as a consequence, cannot be separated by general physical separation methods (Hussein et al., 2004a,b). Physical–chemical methods such as chemical precipitation, chemical oxidation or reduction, electrodeposition and electrocoagulation, solvent exchange and membrane separation technologies have been widely used to remove heavy metal ions from industrial wastewater. But these processes may be ineffective or expensive, especially when the heavy metal ions are in solutions containing in the order of 1–100 mg dissolved heavy metal ions/L (Meunier et al., 2003; Volesky and Holan, 1995). Biological methods for the removal of heavy metal ions may provide an attractive alternative to physical–chemical methods. Many of them have been achieved successively, including removal of Cr (VI) in wastewater by fungi (Li et al., 2008), removal of chromium from aqueous solution using mixture of *Candida lipolytica* and dewatered sewage sludge (Ye et al., 2010), and removal sulfate and copper by a PVA-immobilized

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sulfate reducing bacterial (Hsu et al., 2010). Recently, sulfate-reducing bacteria (SRB) have been identified as the primary bacteria for the biological treatment of heavy metal in effluent. They are a diverse group of organisms capable of great metabolic diversity. They could oxidize organic matter or H_2 using sulfate as an electron acceptor to produce hydrogen sulfide and bicarbonate by the disassimilation as products. This biogenically produced sulfide can rapidly react with soluble heavy metal ions to form insoluble metal sulfides. These bacteria can also reduce metals directly by an enzymatic way (Goulhen et al., 2006; Pagnanelli et al., 2010; Sahinkaya et al., 2011; Sheng et al., 2011). Under anaerobic conditions, SRB play an important role in the treatment or remediation of heavy metals effluent. However, the technology had a few deficiencies; for instance, the time of reaction delay increased with the metal concentration increased. Furthermore, the processing time was longer than chemical or physical precipitation. In addition, their reducibility could be greatly affected by physicochemical parameters of the solution such as pH, ion strength and temperature. Based on Guha and Bhargava study (2005), Fe^0 had been applied in Cr-containing wastewater combined SRB, but the removal efficiency was limited when oxide layer produced and sulfide deposited on the Fe^0 surface. On the other hand, there has been a growing interest in the use of bimetallic replace of zero valent iron for treatment of contaminated groundwater. In previous research, bimetallic particles based on zero valent iron (the additional metals could be Pd, Ag, Ni, and Cu) could accelerate the degeneration rate of organic and inorganic contaminants (Cheng et al., 1997; Chun et al., 2010; Fennelly and Lynn Robert, 1998; Gui et al., 2000; Grittini et al., 1995; Hu et al., 2010; Kim and Carraway, 2000; Liou et al., 2005; Lin et al., 2004; Li and Klaubunde, 1998; Odziemkowski et al., 2000; Schrick et al., 2000). The same process may also work for Cu^{2+} and Zn^{2+} removal. Therefore, the copper–iron bimetallic particles have been brought in SRB system to enhance the stabilization and improve the removal efficiency of SRB.

In this work, batch experiments were performed at different metal ion concentration, pH and temperature to study the effects on the reductions of Cu^{2+} and Zn^{2+} by three systems including SRB–Cu/Fe, Fe/Cu and SRB, respectively. Additionally, the content of copper–iron bimetallic particles in this process was also investigated. Subsequently, the feasibility of Cu^{2+} and Zn^{2+} removal based on the SRB–Cu/Fe system was confirmed well. After that, the reductions of Cu^{2+} and Zn^{2+} with time by SRB–Cu/Fe process were analyzed, and the difference in reduction mechanisms between the Cu^{2+} and Zn^{2+} was completely investigated and discussed to evaluate the possibility of heavy metal remediation enhancement for subsequent studies and eventually practical application.

2. Methods

2.1. Preparation of SRB for reduction

The activated sludge contained SRB was collected at the sewage discharge canal bank of Qingshuitang industrial estate in Zhuzhou city. The bacteria were cultivated in closed infusion bottles using standard procedures reported in the literature (Postgate, 1984) at 37 °C. The pH value of medium was approximately equal to 6. High metal tolerance of SRB had been acquired by increased the concentrations of Cu^{2+} and Zn^{2+} in the medium orderly, which indicates the promising adaptation abilities for practical applications.

2.2. Preparation of copper–iron bimetallic particles

Copper–iron bimetallic particles were synthesized by mixing the solution of copper chloride with iron particles which were finer than 100 mesh. Initially, the copper metal stock solution was prepared by dissolving copper precursors in N_2-purged Milli-Q water at a concentration of 1000 mg/L as Cu. The desired amount of copper metal stock solution (2.5, 5.0, 7.5, and 10.0 mL of 1000 mg/L copper) was added by stirring to 10.0 g of iron particles. Therefore, the mass ratios of copper to iron were 2.5, 5.0, 7.5, and 10.0% (w/w), respectively. The mixture was filtered and washed four to five times with N_2-purged Milli-Q water, and then dried via a vacuum freeze-drying process for 24 h. Compared to the concentration of the stock solution (1000 mg/L), the residual copper concentration of each precursor solution was less than 10 mg/L, which can be negligible. Hence, the metathesis was complete in each experiment (Hu et al., 2010).

2.3. Analytical techniques

The concentrations of Cu^{2+} and Zn^{2+} were spectrophotometrically determined using sodium diethyldithiocarbamate and dithizone in UV-1100 spectrophotometer (Shanghai), respectively. The analytical wavelengths were set at 440 nm and 535 nm for detection Cu^{2+} and Zn^{2+}. The sulfate concentrations were determined using barium chloride by turbidimetry method. Each sample was read three times to get the average value. Each experiment was carried out in triplicate under identical condition to get the mean values.

2.4. Batch studies

All experiments were performed with 100 mL infusion bottles. In each bottle, 5% of SRB, 0.4 g of bimetallic particles and 100 mL synthetic solution (KH_2PO_4 500 mg/L, NH_4Cl 1000 mg/L, Na_2SO_4 500 mg/L, $CaCl_2$ 100 mg/L, $MgSO_4 \cdot 7H_2O$ 2000 mg/L, sodium lactate 4 mL/L, yeast extract 1000 mg/L) were added, leaving no headspace. Immediately, the vials were capped with rubber plug and sealing film for anaerobic conditions and then mixed at 100 rpm using a constant temperature shaker at ambient temperature (37 °C). In this case, the content of copper–iron bimetallic particles in this process was investigated and residual concentrations of Cu^{2+} and Zn^{2+} were measured after 48 h at various initial concentrations (50, 100, 200, 300, 400 and 500 mg/L). The solution pH was adjusted to the desired values including 1–10 by using HCl and NaOH to evaluate the effects of pH on adsorption of Cu^{2+} and Zn^{2+}, respectively. The temperatures were controlled at 16, 28, 37 and 45 °C to observe their influences on the removal of metal ions. Then, samples were determined periodically at 6, 12, 24, 36, 48 h and analyzed for the residual Cu^{2+} and Zn^{2+} concentrations. Meanwhile, the residual SO_4^{2-} concentration was tested at the same condition.

2.5. Scanning electron microscopy (SEM) analysis of copper–iron bimetallic particles and SRB

The scanning electron microscope (JEOL JSM-6360LV) was used to take pictures of copper–iron bimetallic particles and SRB at different states. Before taking pictures, the samples were dried via a vacuum freeze-drying process.

3. Results and discussion

3.1. Effect of copper loading

The effects of Cu loading on the removal efficiency of Cu^{2+} and Zn^{2+} using SRB–Cu/Fe system were examined with batch studies. The results were shown in Table. 1. It can be seen that the removal efficiencies of Cu^{2+} and Zn^{2+} were lower in the SRB–Fe^0 system without copper, and increased with the rise of copper loading. The Cu^{2+} removal efficiency increased significantly after the copper coating was applied. It may be that the insoluble film formed or