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Treatment of acidic sulfate-containing wastewater using revolving algae biofilm reactors: Sulfur removal performance and microbial community characterization

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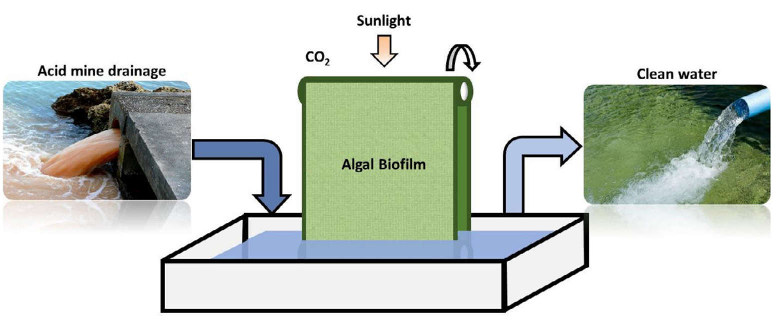
GRAPHICAL ABSTRACT



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ABSTRACT

Industries such as mining operations are facing challenges of treating sulfur-containing wastewater such as acid mine drainage (AMD) generated in their plant. The aim of this work is to evaluate the use of a revolving algal biofilm (RAB) reactor to treat AMD with low pH (3.5–4) and high sulfate content (1–4 g/L). The RAB reactors resulted in sulfate removal efficiency up to 46% and removal rate up to 0.56 g/L-day, much higher than those obtained in suspension algal culture. The high-throughput sequencing revealed that the RAB reactor contained diverse cyanobacteria, green algae, diatoms, and acid reducing bacteria that contribute the sulfate removal through various mechanisms. The RAB reactors also showed a superior performance of COD, ammonia and phosphorus removal. Collectively, the study demonstrated that RAB-based process is an effective method to remove sulfate in wastewater with small footprint and can be potentially installed in municipal or industrial wastewater treatment facilities.

1. Introduction

Sulfur is a contaminant commonly found in municipal and industrial effluents generated from various operations such as medication, tanning, mining, petrochemical, fermentation and food processing (Liu et al., 2012). The high sulfur-containing wastewater leads to severe environmental issues such as impoverishing aquatic flora and fauna, emissions of sulfur gases, subsidence and corrosion of foundations

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(Jarvis and Younger, 2000). Sulfate as the sulfur oxidation product is the most common sulfur compound in wastewater and is usually harmless to the environment. Under anaerobic environment, however, sulfate can be converted into sulfide by sulfate reducing bacteria (SRB). Compared to sulfate, sulfide is more toxic, corrosive and odorous, and more harmful to human health. The emission of sulfide-containing off-gases (e.g. H_2S) can lead to sulfur enrichment in a waterbody causing ecological and health hazards (Li et al., 2015). Because of the significant physiological and toxicological impacts on the environment, it is important to develop effective processes to remove sulfur contaminants from wastewater.

Sulfur in wastewater can be removed through physical, chemical and biological methods. The physical methods such as electrodialysis, ion exchange and membrane filtration require high energy input. Chemical methods, such as metal precipitation, need to use excessive chemicals and replace poisoned catalysts, and thus, cause liquid contamination and reactor corrosion (Lens et al., 1998). In the bacteria-based biological sulfur removal process, sulfate is reduced into sulfide and oxidized to elemental sulfur (Xu et al., 2014). This process can emit H_2S to the atmosphere as a result of sulfate reduction. It also requires strict anaerobic conditions which can be difficult to maintain. Considering these challenges, it is essential to develop a low-cost, simple and eco-friendly methods for sulfur removal.

In recent years, microalgae-based wastewater treatment is gaining increased attention due to its environmental friendliness and potential economic benefit compared to conventional wastewater treatment processes (Gross et al., 2015). Microalgae are capable of removing various pollutants such as oxygen consumption pollutants, nitrogen, and phosphorous and metals from wastewater, the biomass produced during the treatment process can be used as feedstock for fuels, feeds, and chemicals (Kesaano and Sims, 2014). When sulfur was the targeted pollutant, an algae-based sulfur removal process is also possible because algae need to absorb sulfur in the synthesis of amino acids cysteine and methionine (Mera et al., 2016).

Compared to the other nutrients such as nitrogen and phosphorous, however, sulfur removal from wastewater has been less studied. Among limited reports of algae-based sulfur removal, researchers have studied the municipal wastewater in which the sulfur concentrations were relatively low (~ 300 mg/L) and pH was neutral (Lv et al., 2017, Mera et al., 2016). Contrary to the municipal wastewater, effluents from industries such as medication, tanning, mining, and petrochemical operations contain a high sulfur level (> 2 g/L) (Galiana-Aleixandre et al., 2005). Among those industrial effluents, acid mine drainage (AMD) is a particular concern. In addition to high sulfur content, AMD also contains a diverse of metals (Orandi and Lewis, 2013) with an acidic pH ranging from 3.6–4.7 to 1.5 (Abinandan et al., 2018). Researches on AMD treatment have been mainly focusing on metal removal. For example, Orandi et al. (2012) demonstrated that an algal-microbial consortium in a rotating biological contactor was capable of removing heavy metals from AMD. Abinandan et al. (2018) reviewed the effect of microalgae and baceria interaction on the metal removal and concluded that the algae-bateria consortrium can remediate AMD. Das et al. (2009) reviewed the role of algae and fungi in the metal removal during the AMD treatment and its effects on sulfate reduction bacteria. In another study of sulfur-removal from AMD by algae-bacteria system, Sheoran and Bhandari (2005) reported that the main role of algae is to adsorb metals and nitrogen, leading to a rise of alkalinity and serving as the carbon source for sulfur reducing bacteria, which is the ultimate sulfur remover.

Our research laboratory has recently developed a revolving algal biofilm (RAB) reactor as an effective way growing microalgae (Gross et al., 2015; Gross and Wen, 2018). The RAB reactor relies on a vertically oriented materials for attached algal growth. The material travels through the water absorbing nutrients, then rotates out of the water to facilitate light exposure and CO_2/O_2 exchange. Compared to the conventional suspended growth systems, the RAB reactor allows for greater surface area exposure to sunlight in a much smaller footprint. The biomass productivity in the RAB reactor was 5–10 times higher than that of the open pond. Also the biomass can be harvested through scraping from the attachment material, which greatly reduced the cost compared to the centrifugation harvested processes (Gross et al., 2016). Recently, the RAB system has been successfully implemented in Metropolitan Water Reclamation District (MWRD) of Greater Chicago to remove nitrogen, phosphorus and metals from sludge thickening supernatant in MWRD facility (Kunetz et al., 2016, Zhao et al., 2018).

With prospective the success implementation of RAB reactor in municipal wastewater treatment, the aim of this study is to explore the utility of the RAB-based culture system for sulfur removal from AMD. Different from previous research on AMD treatment where metal removal was the focus (Orandi et al., 2012), this work focuses on a thorough evaluation of sulfur removal performance. In addition, a holistic view of the sulfur removal mechanisms was studied through identification and quantification of microbial consortium based on a high-throughput gene sequencing method.

2. Materials and methods

2.1. Microalgae culture

The microalgal seed culture was taken from a raceway pond (1000 L working volume) at the Algal Production Facility at Iowa State University in Boone, IA, USA. The pond was initially inoculated with *Chlorella vulgaris* (UTEX #265) and has been operated for four years. The pond culture has been maintained using Bold's Basal Medium (BBM) with half of the pond liquid being exchange with fresh medium every 7 days. Over the years, a stable algal community containing various green algae and cyanobacteria species has been established. The abundance of the mixed algal culture, particularly the original strain *C. vulgaris*, was determined based on illumina high-throughput sequencing as described in Section 2.6. This algal polyculture was used as inoculum for the bubble column and RAB reactors.

2.2. Synthesis wastewater composition

Synthetic wastewater mimicking acid mine drainage commonly found in the mining industry was used in this work. The basic recipe of the synthetic wastewater composed of (per L) 200 mg NH_4Cl, 50 mg KH_2PO_4, 66 mg $MgSO_4 \cdot 7H_2O$, 6 mg $CaCl_2$, 0.55 mg $FeSO_4 \cdot 7H_2O$, 2.86 mg H_3BO_3, 1.84 mg $MnCl_2 \cdot 4H_2O$, 0.22 mg $ZnSO_4 \cdot 7H_2O$, 0.39 mg $Na_2MoO_4 \cdot 2H_2O$, 0.08 mg $CuSO_4 \cdot 5H_2O$, 0.05 mg $Co(NO_3)_2 \cdot 6H_2O$ and 375 mg glucose. This receipt was adapted from the synthetic municipal wastewater reported previously (Lv et al., 2017). The addition of glucose was used to provide COD of the acid mining drainage. To mimic the high sulfur concentration in the acid mine drainage, sodium sulfate (Na_2SO_4) was added to the receipt at a concentration of 1 g/L, 2 g/L and 4 g/L sulfate, respectively. The pH of the wastewater was 3.5–4.0 adjusted by hydrochloric acid.

2.3. Bubble column cultures

Sulfur removal by microalgae was evaluated in suspension-based bubble column reactors in a batch culture mode. The bubble columns contained 1-L synthetic wastewater with different sulfate concentrations. To inoculate the bubble columns, the microalgae seed culture (with an inoculum ratio of 1:10, v/v) was first settled for 1–2 h, the settled slurry was then washed with DI water before being inoculated into the reactors. The bubble columns were placed at room temperature (25 °C) and aerated at a flow rate of 0.5 L/min throughout the culture. Fluorescent lights were used to provide 24-hr lighting at an intensity of 130 $\mu mol\ cm^{-2} s^{-1}$. During the culture, cell density was determined based on optical density at 680 nm (OD_{680}).