Bioresource Technology 153 (2014) 383–387

Contents lists available at ScienceDirect

Bioresource Technology

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

Short Communication

Algae-facilitated chemical phosphorus removal during high-density *Chlorella emersonii* cultivation in a membrane bioreactor

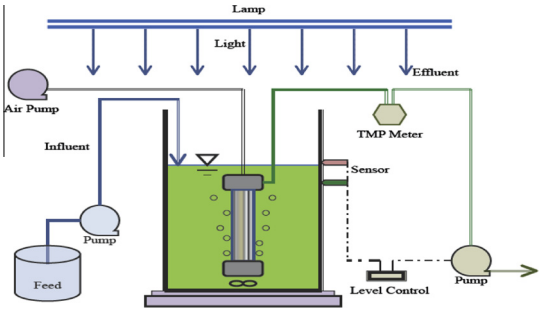
Meng Xu [a], Matthew Bernards [b], Zhiqiang Hu [a,*]

[a] Department of Civil and Environmental Engineering, University of Missouri, Columbia, MO 65211, USA
[b] Department of Chemical Engineering, University of Missouri, Columbia, MO 65211, USA

HIGHLIGHTS

- An algae-based membrane bioreactor was evaluated for wastewater treatment and nutrient removal.
- The extracellular P accounted for >90% of the total phosphorus in the algae biomass.
- Algae-induced P precipitation is key to P removal in high-density algae cultivation.
- The P enriched algal biosolids had excellent settling properties.
- Low tendency for membrane fouling due to the low algal EPS production.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 23 October 2013
Received in revised form 3 December 2013
Accepted 7 December 2013
Available online 14 December 2013

Keywords:
Algae
MBR
Phosphorus removal
Chemical precipitation
Membrane fouling

ABSTRACT

An algae-based membrane bioreactor (A-MBR) was evaluated for high-density algae cultivation and phosphorus (P) removal. The A-MBR was seeded with *Chlorella emersonii* and operated at a hydraulic retention time of 1 day with minimal biomass wastage for about 150 days. The algae concentration increased from initially 385 mg/L (or 315 mg biomass COD/L) to a final of 4840 mg/L (or 1664 mg COD/L), yielding an average solids (algae biomass + minerals) production rate of 32.5 g m^{-3} d^{-1} or 6.2 g m^{-2} d^{-1}. The A-MBR was able to remove 66 ± 9% of the total P from the water while the algal biomass had an average of 7.5 ± 0.2% extracellular P and 0.4% of intracellular P. The results suggest that algae-induced phosphate precipitation by algae is key to P removal and high-density algae cultivation produces P-rich algal biomass with excellent settling properties.

1. Introduction

Owing to the potential for simultaneous nutrient removal and renewable energy production, algae-based photobioreactor techniques are being revived for wastewater treatment and nutrient removal (Bordel et al., 2009; Ruiz-Martinez et al., 2012). Algal assimilation of N and P plays an important role in nutrient removal from wastewater. Algae also have the ability to uptake more P than required for their growth known as "luxury uptake", which occurs especially under P limitation (Riegman et al., 2000). Furthermore, the induced flocculation of microalgae can facilitate chemical phosphate precipitation. Therefore, P can be removed by algae through a combination of adsorption and algae-induced chemical precipitation (Sanudo-Wilhelmy et al., 2004). It is desirable to recover the phosphorus from water/wastewater through algae-induced P precipitation due to the zero chemical addition and possible P reuse from algae biomass as a valuable product.

Traditional algae systems for water and wastewater treatment face many challenges due to the slow growth of algae, poor settling properties, low algal biomass concentrations, and difficulty in algae harvesting. With continued reductions in membrane cost,

* Corresponding author.
E-mail address: huzh@missouri.edu (Z. Hu).

0960-8524/$ - see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.biortech.2013.12.026

membrane bioreactor (MBR) technologies may be a solution for high-density algae cultivation, because of the MBR's excellent solid/liquid separation performance. By using the submerged membrane filtration the starting period can be shortened, as substrates are replenished without diluting the algae. However, key issues such as the phosphorus removal mechanism, membrane fouling, and MBR performance at controlled SRT has not been fully resolved. The objectives of this study were therefore (1) to determine the P removal efficiency and explore the P removal mechanisms by algae, and (2) to assess the membrane fouling trend and biomass settling properties and evaluate whether high-density algae cultivation facilitates algae separation and harvesting.

2. Methods

2.1. Algae seeding of the A-MBR

A green alga *Chlorella* was selected because of its high photosynthetic efficiency, fast growth (Liang et al., 2009), and its efficient organic and nutrient removal characteristics (Mayo and Noike, 1994). The algal species (ATCC® 13482) with mixotrophic growth characteristics was initially erroneously labeled as *Chlorella vulgaris*, but it was determined to be *Chlorella emersonii* by gene sequencing analysis (data not shown). It was cultured under continuous light (85 μmol m^{-2} s^{-1}) at 25 ± 1 °C on a solid agar plate and later in a liquid medium before seeded in the A-MBR.

2.2. A-MBR setup and operation

The A-MBR had an effective volume of 7.2 L with dimensions of 0.195 m × 0.195 m × 0.19 m (length, width, height) (see Supporting Information, Fig. S1). From the start-up period onward, the algae bioreactor was operated at a hydraulic retention time (HRT) of 1 d with little biomass wastage except for 280 mL of mixed liquor (corresponding to a solids retention time or SRT of 180 d). Detailed information about the setup of the MBR is available elsewhere (Liang and Hu, 2012). Briefly, a ZeeWeed hollow fiber membrane module (GE Water and Process Technologies, Trevose, PA) made of polyvinylidene fluoride (PVDF) with a nominal pore size of 0.1 μm, a total effective filtration area of 0.047 m^2, and a designed membrane flux of 15.0 L m^2 h^{-1} was directly submerged in the bioreactor. A periplastic pump operated intermittently after manually setting the permeate flux/effluent flow rate to two times the influent flow rate. The algal mixed liquor volume was maintained almost constantly (with less than 5% volume change) by upper and lower water level sensors (Cole-Palmer, Vernon Hills, Illinois) in the A-MBR by operating the effluent pump. A digital pressure gauge (Cole-Palmer) was installed to measure the daily change in the transmembrane pressure (TMP) as an indication of membrane fouling. Aeration was supplied to the built-in orifices at the bottom of the membrane module at a constant flow rate of 8 L/min for membrane fouling control and algae mixing. Fluorescent lamps were used as the light source, and fixed 12 h dark/12 h light cycle was applied on a daily basis with a light intensity of 80 ± 5 μmol m^{-2} s^{-1} at the surface of algal mixed liquor during light exposure. The A-MBR was operated as a continuous stirred tank reactor (CSTR) and it was fed with a synthetic wastewater (see detailed composition in Table S1).

2.3. A-MBR performance monitoring

The water quality parameters including TP, NO_3^- − N, and chemical oxygen demand (COD), and the sludge settling properties in the A-MBRs were measured in duplicate according to the standard methods (APHA et al., 2012). The algal biomass concentration was measured in both dry weight/mixed liquid suspended solids (MLSS) and COD units. A standard plate count method was applied to determine the abundance of bacteria (in colony forming units or CFU) in the open algae growth system even though a transparent plastic plate was placed on top of the A-MBR.

Specific oxygen production rate (SOPR, defined as gram of oxygen production per gram algal biomass COD per unit of time) was used to determine algal photosynthetic activity. The detailed procedure of SOPR measurements can be found elsewhere (Choi et al., 2010). There is a direct relationship between the SOPR and the specific algal growth rate (μ) as shown in equation below:

$$106\ CO_2 + 81\ H_2O + 16\ NO_3^- + PO_4^{3-} + 19\ H^+ \xrightarrow{\text{light}} C_{106}H_{181}O_{45}N_{16}P + 150\ O_2$$

Under the assumption that the empirical formula for algae is $C_{106}H_{181}O_{45}N_{16}P$, which indicates 1.56 g COD/g algae, the following calculations display the relationship between SOPR and μ assuming nitrate as the nitrogen source:

$$\mu = \frac{1\ \text{mol}\ C_{106}H_{181}O_{45}N_{16}P}{150\ \text{mol}\ O_2} \times \frac{2428\ \text{g}\ C_{106}H_{181}O_{45}N_{16}P / \text{mol}\ C_{106}H_{181}O_{45}N_{16}P}{32\ \text{g}\ O_2 / \text{mol}\ O_2} \times \frac{1.56\ \text{g biomass COD}}{\text{g}\ C_{106}H_{181}O_{45}N_{16}P} \times \left(\frac{\text{g}\ O_2}{\text{g biomass COD t}}\right)$$

μ = 0.79 (SOPR)

2.4. Intracellular and extracellular phosphorus contents in algae

Intracellular and extracellular phosphorus contents in *chlorella* were differentiated by the oxalic acid rinse method followed by the total P measurement (APHA et al., 2012). Although the analysis of intracellular and extracellular P is more operationally-based, the oxalic acid rinse method is widely used to differentiate between the intracellular and surface-bounded phosphate and trace metals (Sanudo-Wilhelmy et al., 2004). Detailed information about the intracellular and extracellular phosphorus measurement is available elsewhere (Sanudo-Wilhelmy et al., 2004).

2.5. Membrane fouling monitoring and control

The TMP of the submerged membrane module was closely monitored while the permeate flux in the A-MBR was maintained relatively constant at 12.6 ± 0.5 mL/min throughout the study period. To better understand the membrane fouling behavior in the A-MBR, a resistance-in-series (RIS) model (Eq. (1)) was applied to determine the source of resistance based on the relationship between the permeate flux and TMP (Eq. (2)):

$$R_t = R_m + R_c + R_f \quad (1)$$

$$J = \frac{\Delta P}{\mu \cdot R_t} \quad (2)$$

where J is the permeate flux, μ is the viscosity of permeate, ΔP is the TMP, R_t is the total hydraulic filtration resistance, R_f is the fouling resistance caused by irreversible adsorption and pore plugging, R_c is the cake resistance caused by cake layer formation, and R_m is the intrinsic membrane resistance. Extracellular polymeric substances (EPS) have been identified as a major fouling factor in MBR operations (Le-Clech et al., 2006). The polysaccharide content was determined by the phenol–sulfuric acid method with glucose as a standard (Dubois et al., 1956). The protein content was quantified by a modified micro pyrogallol red method using a total protein assay kit (Sigma–Aldrich).