Bioresource Technology 245 (2017) 10–17

Contents lists available at ScienceDirect

Bioresource Technology

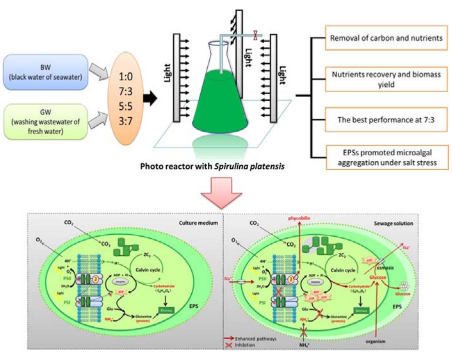
journal homepage: www.elsevier.com/locate/biortech

Nutrients removal and recovery from saline wastewater by *Spirulina platensis*

Weizhi Zhou*, Yating Li, Yizhan Gao, Haixia Zhao

School of Environmental Science and Engineering, Shandong University, Jinan, Shandong 250100, China

GRAPHICAL ABSTRACT



ARTICLE INFO

Keywords:
Saline wastewater
Spirulina platensis
Nutrients removal
Nutrients recovery
Valuable biomass

ABSTRACT

As an important alternative to alleviate the pressure of fresh water shortage, seawater application is facing a great challenge on the wastewater treatment due to the salinity brought from seawater. *Spirulina platensis* originated from salty lake was used to treat mixed synthetic toilet flushing wastewater of seawater with washing wastewater of freshwater. It was showed that 79.96% of TN (to 15.69 mg/L), 93.35% of TP (to 1.03 mg/L) and 90.02% of COD_{Cr} (to 90.24 mg/L) were removed with 0.76 g/L of biomass production in the optimal ratio 7:3 of the above mixed synthetic wastewater. The performance was better than that of current strategy of seawater toilet flushing treatment. With the evaluation of nutrients uptake, biomass composition and microalgal aggregation, a model of nutrients recovery and metabolism of *Spirulina platensis* in saline wastewater treatment was proposed. It is provided a promising strategy for saline wastewater treatment with valuable biomass yield.

1. Introduction

Water scarcity has become a leading issue in many countries since rapid urbanization and increasing environmental pollution (WHO, 2014). Toilet flushing with high quality water accounts for 20–35% of typical household freshwater in most cities. Compare to other alternative water resources, seawater flushing in coastal cities is more optimal for: seawater accounts for 97.5% of all water resources on the planet (Shannon et al., 2008), and it requires considerably less treatment, consumes less energy, resulting in a lower operating cost (Wang et al., 2015b).

In Hong Kong, a dual water supply system providing freshwater for potable uses and seawater for toilet flushing has covered 80% of the population, and fulfills about 20% of the city's total water demand since 1950s. However, the salts that is about 0.9% originating from the mixed seawater toilet flushing does not affect BOD and nitrogen removal efficiency significantly (Leung et al., 2012), but it will lead to more than 50% inhibition of the anaerobic metabolism of phosphate accumulating organisms (PAOs) and practically fully inhibit the aerobic metabolism when exposure to chloride concentrations as low as 10 g NaCl L^{-1} (1%

* Corresponding author.
E-mail address: wzzhou@sdu.edu.cn (W. Zhou).

http://dx.doi.org/10.1016/j.biortech.2017.08.160
Received 22 June 2017; Received in revised form 22 August 2017; Accepted 27 August 2017
Available online 31 August 2017

salinity) for hours (Tang et al., 2007; Welles et al., 2015). Thus current license effluent standard of seawater toilet flushing sewage was without regard to P effluent, which results in the treated sewage with excess P discharging into the seas with causing eutrophication of continental shelves and gulf areas. Therefore, alternative salt tolerant biotreatment should be utilized in the high strength specific wastewater treatment with considering nutrients recovery and resource. In addition, low carbon/nitrogen of seawater toilet flushing sewage (about 5–7) is another challenge which leads to insufficient carbon sources for phosphorus removal or additional cost by dosing carbon sources (Wang et al., 2015a). As photosynthetic organism, microalgae-systems have been gradually used in wastewater treatment and especially in advanced wastewater treatment without additional carbon sources (Noüe et al., 1992) (e.g. high rate algal ponds (HRAP)) for their excellent economical value (Maity et al., 2014) and environmental potential (Ge and Champagne, 2015; Ferreira et al., 2012).

Compare to other salt tolerant organisms, *Spirulina platensis* possesses superior performance in nitrogen recovery (Chang et al., 2013), high removal efficiency of CO_2 which is 10 times than that of terrestrial plants (Skjånes et al., 2007) as well as high contents of proteins (70%), polysaccharides, essential fatty acids and vitamins (Madkour et al., 2012; Pulz and Gross, 2004). Depending on microbiological characteristic of physiological and metabolic, several earlier investigations of cultivated *S. platensis* have been separately elucidated the effects of salinity stress, mainly on the function of photosystem I and II by inhibiting the electron transport (Lu and Vonshak, 2002), the biomass composition change of protein, carbohydrate and photopigment (Vonshak et al., 1996), and the relationship of photosynthetic and respiratory electron transport (Berry et al., 2003) in specific autotrophic medium or natural bulks (Kebede, 1997a). With respect to wastewater treatment, digested sago factory wastewater (Phang et al., 2000), municipal wastewater combined Zarrouk medium (Park et al., 2013) and wastewater of monosodium factory mixed with Zarrouk medium (Jiang et al., 2015) were used for *S. platensis* cultivation with carbon and nutrients removal from wastewater. To our knowledge, however, *Spirulina platensis* used in treatment of nutrients wastewater with high salinity, particularly in the seawater application remediation, have yet been reported but of great significance.

In this study, we investigated the performance of effluents indexes and biomass yield of *S. platensis* in the treatment of saline wastewater with different collected ratios (express as different blending ratios in batch tests) of toilet flushing wastewater of seawater (referred to as black water (BW)) with washing wastewater of freshwater (referred to as grey water (GW)). Based on the systematical statistic analysis, the optimal collected strategy and key limited factors of seawater toilet flushing sewage treatment by *S. platensis* was identified and characterized. Further, the mechanism of nutrients recovery, biomass composition and the aggregation abilities for biomass harvest in response to the function of extracellular polymeric substances (EPSs) were explained on the basis of the physiological and metabolic characteristics of *S. platensis*.

2. Materials and methods

2.1. Microalgae strain

Spirulina platensis was obtained from Freshwater Algae Bank of Chinese Academy of Sciences (CAS), cultivated in Zarrouk medium (ZM: $NaHCO_3$ 16.8 g/L, $NaNO_3$ 2.5 g/L, NaCl 1.0 g/L, K_2HPO_4 0.5 g/L, K_2SO_4 1.0 g/L, $MgSO_4{\cdot}7H_2O$ 0.2 g/L, $CaCl_2$ 0.031 g/L, Na_2EDTA 0.08 g/L, $FeSO_4{\cdot}7H_2O$ 0.01 g/L) at room temperature (25.0 ± 0.5 °C) under continuous irradiation. To maintain the purity of algae strain, the collected algal suspension was filtered through 0.45 μm cellulose acetate membrane every week and resuspended in fresh ZM for four cycles. The ZM used for cultivation was prepared with de-ionized (DI) water, sterilized at 121 °C for 20 min. Bulk of algal suspension was taken from ZM and centrifuged at 10,000 rpm for 10 min with discarding supernatant, the remaining pellet was washed by DI water twice. The initial biomass monitored as optical density at 560 nm (OD_{560}) was equal 0.2 for batch tests.

Table 1
Characteristics of composite wastewater used in this study.

Blending ratio (BW:GW)	COD (mg O_2/L)	NH_4^+-N (mg/L)	TN (mg/L)	TP (mg/L)	Salinity (%)	pH
1:0	1200.00	140.00	180.00	20.00	3.20	8.00
7:3	900.00	90.00	130.00	15.00	2.24	7.79
5:5	700.00	70.00	95.00	12.00	1.63	7.65
3:7	500.00	40.00	60.00	10.00	0.93	7.51

2.2. Experimental wastewater

Wastewaters were prepared by mixing synthetic seawater-originated black water and freshwater-originated grey water together with blending ratios 1:0, 7:3, 5:5 and 3:7 according the parameters of the report by Todt (Todt et al., 2015). Black water (BW) here we referred to toilet flushing sewage using seawater (COD 1200 mg/L, NH_4^+-N 140 mg/L, TN 180 mg/L, TP 20 mg/L, salinity 3.20% and pH 8.00); Grey water (GW) parameters were based on house hold washing sewage (COD 200 mg/L, NH_4^+-N 1.6 mg/L, TN 7.4 mg/L, TP 5 mg/L, salinity 0.08% and pH 7.20). The characteristics of composite wastewater were listed in Table 1.

Actual municipal wastewater, which composition was approximately 600 mg/L chemical oxygen demand (COD), 45 mg/L total nitrogen (TN) and 5 mg/L total phosphorous (TP), was obtained from a sedimentation tank from Everbright Water (Jinan) Company Limited. The parameters of actual wastewater were adjusted to the optional blending ratio of BW:GW according to Table 1.

2.3. Batch test setup

Batch tests were carried out in 500 mL erlenmeyer flasks filled with 250 mL sterilized synthetic wastewater with different blending ratio of BW:GW and actual municipal wastewater adjusted to the optimal ratio was used to investigate the performance of *Spirulina platensis* in the actual wastewater. The flasks were placed in an incubator which was operated at 25 °C with a 14/10 h light/dark cycle illuminated at the intensity of 90 μmol m^{-2} s^{-1}. A certain volume of each samples was taken periodically after light cycle and the samples were divided into supernatant and sediment through centrifugation for 10 min at 10,000 rpm for determination of water quality and microbial biomass respectively. All of the experiments were carried out in triplicate.

In order to study the mechanism of nutrients recovery and biomass composition, five different salinity levels (0.93%, 1.49%, 2.07%, 2.49% and 3.04%) in batch tests were conducted and kept other parameters the same (COD 400 mg/L, NH_4^+-N 40 mg/L, TN 42 mg/L, TP 10 mg/L and pH 7.70). The operation conditions and sampling were the same as above batch tests.

Salinity and pH were measured with corresponding meters (ATAGO, ES-421, Japan and Sartorious, PB-10, Germany). Total phosphorus (TP), chemical oxygen demand (COD), ammonium nitrogen (NH_4^+-N) and total nitrogen (TN) were measured by the standard method (APHA, 1998). COD was pretreated by $AgNO_3$ to remove Cl^- (Shi et al., 2012). Liquid samples were filtered through a 0.22 μm filter to remove microalgal cells and other particles prior to analysis.

2.4. Extraction and determination of biomass composition

Biomass production monitored as optical density at 560 nm (OD_{560}) was measured using a spectrophotometer (Metash, UV – 5500,