



Effects of dissolved oxygen concentration on photosynthetic bacteria wastewater treatment: Pollutants removal, cell growth and pigments production



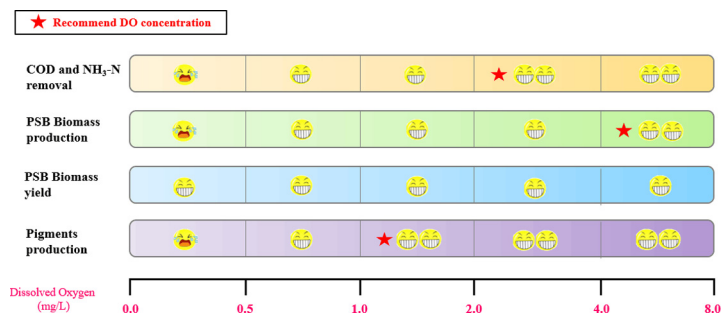
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HIGHLIGHTS

- This paper for the first time studied DO effects on PSB wastewater treatment.
- DO of 2–4 mg/L was recommended for pollutants removal.
- PSB biomass reached 1645 mg/L when DO concentration was 4–8 mg/L.
- DO > 1.0 mg/L was preferred for pigments production.

GRAPHICAL ABSTRACT



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ABSTRACT

Dissolved oxygen (DO) is an important parameter in photosynthetic bacteria (PSB) wastewater treatment. This study set different DO levels and detected the pollutants removal, PSB growth and pigments production. Results showed that DO significantly influenced the performances of PSB wastewater treatment process. The highest COD (93%) and NH₃-N removal (83%) was achieved under DO of 4–8 mg/L, but DO of 2–4 mg/L was recommended considering the aeration cost. PSB biomass reached 1645 mg/L under DO of 4–8 mg/L with satisfying co-enzyme Q10 content. The biomass yield was relatively stable at all DO levels. For bacteriochlorophyll and carotenoids, DO > 1 mg/L could satisfy their production. On the other hand, DO < 0.5 mg/L led to the highest dehydrogenase activity. According to the different purposes, the optimal treatment time was different. The most pigments production occurred at 24 h; biomass reached peak at 48 h; and the optimal time for pollutants removal was 72 h.

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1. Introduction

Photosynthetic bacteria (PSB) wastewater treatment can realize pollution reduction and resource recovery simultaneously. This method solved the problem of residual sludge in traditional wastewater methods, so it was studied by many researchers (Kobayashi, 1978; Grady et al., 1999; Sasaki et al., 2005). PSB treat many wastewaters effectively, such as soybean wastewater, sac-

charides wastewater, pharmaceutical wastewater, domestic wastewater (Lu et al., 2011; Hülsen et al., 2016; Prachanurak et al., 2014). What's more, PSB biomass can realize resource recovery. PSB biomass can be used in livestock breeding, aquaculture and soil additives because it contains 40–60% protein. The market price of PSB biomass is 2–16 RMB/L. PSB biomass also contain high value substances like coenzyme Q10 (CoQ10), polyhydroxyalkanoates, 5-aminolevulinic acid, and carotenoids (Petritir and Suling, 1998; Muñoz and Guieysse, 2006). CoQ10 is a kind of fat soluble quinone compounds present in nature that plays an important role in the heart function, and is widely used in food, cosmetics, and

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dietary supplements. The price of CoQ10 can reach 3500 RMB/g. Bacteriochlorophyll and carotenoid are both important pigments with different roles (Connelly et al., 1997). Carotenoids can be used as food and fat colorants since they can protect the human visual system and skin tissue to resist adverse environment. Human body cannot synthesize carotenoids, thus must obtain from outside. However, carotenoids are low in many plants, and are difficult to synthesize by chemical methods, thus microbial synthesis of carotenoids has attracted many attentions.

In the process of PSB wastewater treatment, the most important factor is the light-oxygen condition (Siefert et al., 1978; Izu et al., 2001; Prachanurak et al., 2014). There are three typical light-oxygen conditions, namely light-anaerobic, dark-aerobic and natural light micro-aerobic. The wastewater treatment efficiency and PSB biomass were evidently different under those conditions (Prasertsan et al., 1993; Azad et al., 2004; Lu et al., 2011). More in-depth studies of light condition on PSB have been reported. The light recipe could influence biomass and COD removal in the wastewater treatment, including light intensity, light source and photoperiod (Zhou et al., 2014, 2015a,b). Kaewsuk et al. (2010) reported that the factors relating to light energy must be incorporated into engineering design in PSB for dairy wastewater treatment. Suwan et al. (2014) also reported that organic removals in the photo-bioreactor lighted with infrared Light Emitting Diodes (91–95%) was higher than those in photo-bioreactor with tungsten lamps with filter (79–83%).

The other aspect, oxygen, is noted as a more important factor comparing to light for organics reduction. PSB carried out oxidative phosphorylation under aerobic condition, in which pollutants were sufficiently utilized with high mineralization degree (Lu et al., 2011). The oxygen condition is an important factor in other biological wastewater treatment methods. Yadav et al. (2014) reported the higher DO level achieved higher degradation efficiency. Liao et al. (2011) reported that low DO led to poorer bio-flocculating ability and less bound extracellular polymeric substances content of the activated sludge. So it was speculated that the oxygen concentration influences the PSB wastewater treatment significantly, however there is no in-depth study on this subject, all available reports involved the change of light condition simultaneously. In addition, DO level is of important economical consideration since aeration counted for almost 1/3 of operational cost in wastewater treatment plants (Fletcher et al., 2007; Aymerich et al., 2015; Stadler and Love, 2016). So it is meaningful to study the effects of DO level on PSB wastewater treatment for both scientific and practical reasons.

The DO concentration set in this study was referred to the practical biological wastewater treatment. In traditional activated sludge methods, anoxic condition is DO < 0.5 mg/L, micro-aerobic condition is DO of 0.5–1 mg/L, aerobic condition is DO of 2–4 mg/L, and strong aeration condition is DO 4–8 mg/L. These different DO conditions have decisive effects on pollutant removal and microbial cell synthesis. So this research studied PSB treatment performance under these DO conditions. The specific set of DO concentration was <0.5, 0.5–1, 1–2, 2–4, and 4–8 mg/L. The PSB wastewater treatment performances under these DO levels were examined, included COD removal, NH₃-N removal, PSB cell growth, pigments production, and dehydrogenase activity.

2. Material and methods

2.1. Microorganism and culture conditions

PSB strain was *Rhodospseudomonas* (Zhou et al., 2014), which was isolated from a local pond, and the Gene Bank accession number is CP001151.1. PSB strain was incubated in a thermostat shaker

(120 r/min, 26–30 °C) with improved RCVBN medium (70/30, v/v) for 48 h before use (Lu et al., 2013).

2.2. Wastewater

Artificial sugar wastewater was prepared with saccharose (5 g/L), malic acid (3 g/L), sodium bicarbonate (1 g/L), ammonium sulfate (2 g/L), potassium dihydrogen phosphate (0.4 g/L), and magnesium sulfate heptahydrate (0.2 g/L). The wastewater had a COD and NH₃-N concentration of 6650 mg/L and 290 mg/L respectively, and the initial pH was 7.0. Sugar wastewater is a highly concentrated and non-hazardous wastewater that can provide nutrients for PSB growth.

2.3. Experimental set-up

The experiments were conducted in batch photo bioreactors on a laboratory scale using 1000 ml borosilicate Pyrex bottles. Wastewater and PSB (80%/20%, v/v) were added to the reactors. The dissolved oxygen (DO) concentration of <0.5, 0.5–1, 1–2, 2–4, 4–8 mg/L were set using agitators, DO meters and automatic control. The temperature was maintained at 26–30 °C in a thermostatic shaker (Yiheng Q-III, China). The light condition was natural light (no extra light).

2.4. Analysis methods

Samples collected from reactors were centrifuged at 9056g for 10 min. The supernatant was used to test the COD and NH₃-N; the collected PSB were used to measure the biomass, carotenoid, bacteriochlorophyll, and dehydrogenase activity. The COD, NH₃-N, biomass and dehydrogenase activity were tested using APHA standard methods (Eaton et al., 2005). The pH was measured using a pH tester (Mettler Toledo FE20, Swiss), and the DO was measured using a dissolved oxygen meter (YSI 550A, USA). The pigments were measured using a spectrophotometer (TU1900, Puxi, China), and the carotenoid and bacteriochlorophyll concentrations were calculated following Zhou et al. (2014).

2.5. Statistical analysis

Three parallel experiments were conducted to ensure the accuracy of data. All reported values were the average. Tukey's test was adopted to analyze the significance of the data.

3. Results and discussion

3.1. Pollutants removal under different DO levels

COD removal and NH₃-N removal under different DO concentrations were tested. As Fig. 1 shows, higher DO concentration improved COD and NH₃-N removal. COD removal under DO 4–8 mg/L reached 93%, which was 3.5 times of the value under DO < 0.5 mg/L. This phenomenon was consistent with other studies that reported higher COD removal under dark-aerobic than natural light-micro oxygen and light-anaerobic conditions (Lorrungruang et al., 2006; Sirianuntapiboon and Srikul, 2006; Lu et al., 2011). The NH₃-N removal under aeration was higher compare to anaerobic condition (Prachanurak et al., 2014). Tao et al. (2013) also reported that the NH₃-N removal of livestock breeding wastewater by PSB under anaerobic condition was 29.79% while the removal under aerobic condition was 91.74%. In this study, DO of 4–8 mg/L was the best for pollutants removal. But in general DO > 2 mg/L was good enough for COD and NH₃-N removal. The aeration cost was much lower for DO of 2–4 mg/L comparing to DO of

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