



Comparison between jet and paddlewheel mixing for the cultivation of microalgae in anaerobic digestate of piggery effluent (ADPE)

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ABSTRACT

There is great need to improve turbulent mixing of microalgae cultures grown in turbid wastewater to ensure efficient use of light and nutrients for higher biomass productivity and nutrient removal rates. In this outdoor study, we compared the turbulent mixing and nutrient removal efficiency of conventional paddlewheel driven raceway ponds (PWP) with customized jet-nozzle raceway pond (JNP) on microalgae grown in undiluted anaerobic digestate of piggery effluent (ADPE). Overall, the concentration of microalgae consisting mainly of Cyanobacteria and *Chlorella* sp. trended higher in the JNP than the PWP with notable absence of diatoms in JNP. The average percentage of ammonium removal rates were found to be significantly higher in the JNP ($36.8\% \pm 3.93$) than the PWP ($23.5\% \pm 4.42$). The measured amount of turbulent kinetic energy (TKE), as an indicator of algal movement at eight distinct locations of both ponds, trended higher in the JNP than the PWP, suggesting improved mixing performance with higher shear stress on cultures in the JNP. Based on the higher ammonium removal rates and turbulence mixing, JNP was found to be more efficient for the cultivation of microalgae in ADPE than PWP.

1. Introduction

Wastewater arising from animal farming facilities such as piggeries is typically characterized by elevated nutrient content (i.e. ammonia nitrogen and phosphorus) which can result in the eutrophication of water streams when directly exposed to the environment [1]. The use of microalgae for the bioremediation of undiluted wastewaters such as anaerobic digestate of piggery effluent (ADPE) is of great interest due to the inherent ability of microalgae in recycling organic and inorganic nutrients for their growth [2,3].

Microalgae represent simple photoautotrophs which are unicellular in size and can be classified to various different phyla (major taxonomic groups) [4]. The cultivation of microalgae in ADPE holds enormous benefits as it combines the treatment of a waste stream together with biomass production which can be converted into multiple commodities such as animal feed, biofertilisers, bioenergy and nutraceuticals [5,6]. The use of ADPE as a growth medium also represents an innovative solution in reducing the high overall cost commonly associated with the commercial cultivation of microalgae [5].

Owing to their larger capacity and lower capital cost, open

cultivation systems of microalgae such as raceway ponds are mainly used for the treatment of wastewater [7]. Achieving optimum algal growth and nutrient removal rates are directly influenced by various physical, operational and biotic factors and the understanding of these factors are essential to optimize the integration of microalgae cultivation with wastewater treatment [8]. For example, for maximum light penetration and good mixing, raceway ponds are typically operated between 20 cm to 60 cm depth with an average water velocity of 30 cm/s generated by one or more paddlewheels [9,10].

Low power requirements and high productivity are by far the most important factors in commercial microalgae cultivation and wastewater treatment [11]. When cultivated in undiluted ADPE, a combination of the effluent turbidity and the depth of the algal pond significantly reduce the availability of light for algal cells, negatively affecting their growth and productivity [3]. Thus, there is great need for innovative options to improve mixing at a lower power requirement to maximise the availability of light for algal cells for nutrient removal when cultivated in ADPE.

Turbulent mixing is a critical factor for achieving high productivity in all types of algal cultivation systems because of its role in the

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distribution of light and nutrients, diffusion of gases and maintenance of uniform temperature throughout the culture [12]. Turbulence by paddle wheels, aeration and pumping can result in different sensitivities or complete inhibition of different algal species. The relative sensitivities of algal species ranges from green algae < blue-green algae < diatoms < dinoflagellates with dinoflagellates being most sensitive to turbulence [13,14].

In open cultivation systems, such as raceway ponds, there is substantial room for improvement in terms of mixing technology in order to prevent algae sedimentation and to improve the availability and utilization of light by algal cells [15]. Efficient turbulence mixing of algal culture is seen to favourably improve input energy requirement, costs and particularly productivity in open cultivation systems [7,15]. Eight-blade paddle wheel designs represent an attractive option in terms of efficiency, weight and construction costs [10]. Alternatively, water jets have been previously tested, mainly in small systems and have been found to have a high power requirement [16]. Recently, a jet-type circulation system for algae ponds has been patented and is currently being tested in large raceway algal ponds in Australia [17].

Despite being commonly used, only limited information is available on the quantification of mixing in paddle wheel and jet driven raceway ponds. Such quantification of turbulent flow will certainly aid in the understanding of vertical mixing at various locations of the raceway pond and the different sensitivities and restriction brought forward to algal growth by turbulence for efficient of nutrient from wastewaters.

The main objective of this study is to characterize the fluid flow of both paddle wheel and jet driven cultures of microalgae grown in undiluted ADPE using an Acoustic Doppler Velocimeter at eight different locations and quantify the level of turbulence created by both these mixing mechanisms with an emphasis on increasing vertical mixing. To enhance our understanding of mixing rates for efficient use of light and nutrients for higher biomass productivity and nutrient removal rates in ADPE. We also compared the effects of both these mixing regimes on the ammonium removal rates of the microalgal cultures present to assess the effect of mixing on efficiency of ammonium removal rates and the energy required.

2. Materials and methods

2.1. Microalgae culture

The consortium of microalgae used in this study consists of a mixed population of Cyanobacteria, *Chlorella* sp., and pennate diatoms isolated and established previously from undiluted ADPE. The microalgae were grown and operated as batch cultures for a period of four months with periodical harvesting and renewal of cultures with fresh ADPE whenever they reached a predetermined cellular concentration.

2.2. Anaerobic digestion of piggery effluent (ADPE) and growth media

The ADPE used as culture medium in this study (Table 1) was

Table 1
Chemical composition of untreated and undiluted ADPE used for the growth of the microalgae [2].

Parameter	Value
Ammonia ($\text{mg L}^{-1} \text{NH}_4^+\text{-N}$)	960–1000
Total phosphate ($\text{mg L}^{-1} \text{PO}_4\text{-P}$)	25.0–26.5
Nitrite ($\mu\text{g L}^{-1} \text{NO}_2\text{-N}$)	8.0–8.5
Magnesium ($\text{Mg L}^{-1} \text{mg}$)	165–175
Potassium ($\text{mg L}^{-1} \text{K}$)	530–545
Total iron ($\text{mg L}^{-1} \text{Fe}$)	8.5–9.5
Nitrate ($\text{mg L}^{-1} \text{NO}_3\text{-N}$)	14.0–14.5
Chemical oxygen demand, COD (mg L^{-1})	1200–1350
Total nitrogen ($\text{mg L}^{-1} \text{N}$)	1050–1101

obtained from a covered anaerobic digestion pond located at Medina Research Station in Kwinana, Western Australia ($32^\circ 13' 16''\text{S}$, $115^\circ 48' 30''\text{E}$). The ADPE has been previously partially characterized by its high nutrient content (e.g. ammonium and phosphorus) [18].

2.3. Experimental setup and cultivation conditions

Outdoor studies were carried out between 21 September 2015 to 11 January 2016 (Austral Summer) using two 1 m^2 fiberglass raceway ponds at the Algae R&D Centre of Murdoch University (31.575S , 115.51E). The first raceway pond was mixed using a conventional 4 blade paddle wheel and was known as the paddle wheel pond (PWP) while the other pond was mixed using as jet nozzle (Fig. 1) based on the design of Parsheh et al. [17] and fabricated on a 3D printer (Makerbot 2) from polylactic acid (PLA). This pond was termed as the jet nozzle pond (JNP). Both raceway ponds were inoculated with the same concentration of microalgae stock culture and were operated at a working volume of 160 L and at a liquid velocity of 30 cm/s which was determined using the tracer method with 1 M HCl [19,20]. Daily evaporative losses during the experiment period were compensated using freshwater up to the initial working depth before sampling. Weather records illustrating solar irradiance and air temperature for the period of the experiment was obtained from Murdoch University Weather Station (<http://wwwmet.murdoch.edu.au>). Sampling of the ponds was done at 10 am on alternative days. Samples collected were used for cell counts, photosynthesis assessment and measurement of ammonium nitrogen concentration in the media.

2.4. Analytical methods

Cell concentration of samples over time was measured using an improved Neubauer counting chamber, while organic biomass (AFDW, mg L^{-1}) was assayed according to the methods of Moheimani et al. [21] by filtering 5 ml of culture through pre-combusted and pre-weighed GF-C microfiber filters. Filters were first dried at 90°C for 7 h and were subsequently combusted at 450°C for 6 h in a furnace to obtain dry and ash-free dry weights. Temperature, dissolved oxygen (DO) and pH of both ponds were monitored in situ using individual YSI 6-Series multi parameter Sondes. Ammonium measurement was carried out using a photometer (Spectroquant Move 100, kit models).

The maximum quantum yield value in light (F_q'/F_m') and the maximum quantum yield of dark adapted samples (F_v/F_m) were measured using an AquaPen-C portable fluorometer (Photon Systems Instruments, Czech Republic). F_q'/F_m' values represented the immediate photo-physiological status of samples collected from the ponds while F_v/F_m values represented the recovery potential of photosynthesis after samples were dark adapted for 30 min. The estimated maximum quantum yield of PSII photochemistry (F_q'/F_m' and F_v/F_m) is regularly used as an indicator of plant stress [22].

A micro Acoustic Doppler Velocimetry (MicroADV by SonTek, San Diego, CA, USA) was used to measure the three velocity components of the flow field in both the PWP and JNP as installed at eight locations in each pond shown in Fig. 1. The instrument is based on the physical principle of the Doppler Effect with a 10 Hz sampling frequency for 10 min in each location. The ADVs were located at distances 7 cm above the bed both in the paddle and jet experiments as mentioned in Fig. 1. It is noteworthy to mention that the physical dimensions of the instruments determined the distance above the bed such that the sensors did not touch the flume bottom during the experiments. Data from the near-bed ADVs (at $z = 7 \text{ cm}$ height above the bed) is used for the analysis in this study.

Spectral analysis was conducted based on spectral energy cascade theory [23] for the three-dimensional inertial subrange spectrum:

$$E(k) = C_k \varepsilon^{2/3} k^{-5/3},$$

where $E(k)$ is the energy spectrum based on the wave number; C_k is the

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