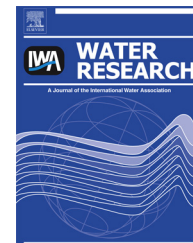


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# Investigating why recycling gravity harvested algae increases harvestability and productivity in high rate algal ponds

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## ABSTRACT

It has previously been shown that recycling gravity harvested algae promotes *Pediastrum boryanum* dominance and improves harvestability and biomass production in pilot-scale High Rate Algal Ponds (HRAPs) treating domestic wastewater. In order to confirm the reproducibility of these findings and investigate the mechanisms responsible, this study utilized twelve 20 L outdoor HRAP mesocosms operated with and without algal recycling. It then compared the recycling of separated solid and liquid components of the harvested biomass against un-separated biomass. The work confirmed that algal recycling promoted *P. boryanum* dominance, improved 1 h-settleability by >20% and increased biomass productivity by >25% compared with controls that had no recycling. With regard to the improved harvestability, of particular interest was that recycling the liquid fraction alone caused a similar improvement in settleability as recycling the solid fraction. This may be due to the presence of extracellular polymeric substances in the liquid fraction. While there are many possible mechanisms that could account for the increased productivity with algal recycling, all but two were systematically eliminated: (i) the mean cell residence time was extended thereby increasing the algal concentration and more fully utilizing the incident sunlight and, (ii) the relative proportions of algal growth stages (which have different specific growth rates) was changed, resulting in a net increase in the overall growth rate of the culture.

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## 1. Introduction

Algal biomass produced and harvested as a by-product of wastewater treatment in High Rate Algal Ponds (HRAPs) could be used as a substrate for biofuel conversion (e.g. biogas, bio-ethanol, bio-diesel, bio-crude oil) through various biological and chemical pathways (Sukias and Craggs, 2010; Vasudevan

and Fu, 2010; Campbell et al., 2011; Craggs et al., 2011). Both wastewater treatment and algal biofuel production require high production of algal biomass followed by rapid and cost-effective algal harvest from HRAP effluent. Therefore, improvements in algal biomass production and its subsequent harvest efficiency (i.e. 'harvestable algal biomass production') are essential to achieve both efficient wastewater treatment

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and economic algal biofuel production (Benemann, 2003; Chen and Yeh, 2005; van Harmelen and Oonk, 2006; Brennan and Owende, 2010; Pittman et al., 2011).

Researchers have proposed several practical options to improve algal biomass production and harvest efficiency in wastewater treatment HRAP including: (1) CO<sub>2</sub> addition to increase carbon availability for algal growth and mitigate pH inhibition (van Harmelen and Oonk, 2006; Heubeck et al., 2007; Park and Craggs, 2010); (2) promotion of natural aggregation of algae (i.e. bio-flocculation) to enhance simple gravity harvest (Benemann, 2008b; Schenk et al., 2008; Bhatnagar et al., 2010; Pittman et al., 2011) and; (3) promotion of the dominance of algal species with beneficial attributes such as efficient settleability (Weissman and Benemann, 1979; Tillett, 1988; Park et al., 2011c).

Algae commonly found in wastewater treatment HRAPs such as *Scenedesmus* sp., *Micractinium* sp., *Actinastrum* sp., *Pediastrum* sp., *Dictyosphaerium* sp., *Coelastrum* sp. can not only grow as large settleable colonies (colony diameter of 50–200 µm) but, in association with wastewater bacteria, can also form large aggregates (diameter: >500 µm) which enhances gravity sedimentation of algal biomass from the HRAP effluent (Benemann, 2008a; Craggs et al., 2011; Park et al., 2011c, 2011a). Therefore, promoting the dominance of readily-settleable colonial algae in wastewater treatment HRAP could be a simple and practical way to enhance the efficiency of algal biomass harvest.

Recycling harvested algal biomass has been previously shown to increase the dominance of readily-settleable algae in small-scale laboratory cultures (Benemann et al., 1977; Weissman and Benemann, 1979; Tillett, 1988). A previous one year pilot-scale HRAP study on domestic wastewater (Park et al., 2011b) indicated that recycling a portion of gravity harvested algae ('algal recycling') improved the dominance of the readily-settleable colonial algal species *Pediastrum boryanum* and resulted in improved algal settleability. This was the first study that showed the dominance of a single algal species (such as *P. boryanum*) can be controlled over similarly sized co-occurring algae in wastewater treatment HRAP by recycling harvested algae.

In this paper we attempted to replicate the pilot-scale findings in replicated mesocosm experiments conducted under different seasonal conditions. In addition to comparing species dominance and settleability, the research was extended to include biomass productivity (Experiments 1 and 2). To further investigate why algal recycling triggered these effects, a further experiment assessed the separated solid and liquid components of the recycled gravity harvested algae.

## 2. Materials and methods

In order to undertake multiple replicates, mesocosms were used (twelve including controls). The experiments were conducted next to two outdoor pilot-scale HRAPs that were also monitored, so that the validity of using the mesocosms to represent HRAP's could be verified.

### 2.1. Operation of pilot-scale HRAPs with algal recycling

Full details of the operation of the two pilot-scale HRAPs (surface area: 31.8 m<sup>2</sup>, depth: 0.30 m, volume: 8 m<sup>3</sup>, wastewater: primary sewage (0.5–1 m<sup>3</sup>/d)) one with algal recycling

(HRAP<sub>r</sub>) and one control without algal recycling (HRAP<sub>c</sub>) are described in (Park et al., 2011b).

### 2.2. HRAP mesocosm experiments

Twelve replicate mesocosms (plastic containers with a water depth of 0.3 m; filled volume of 18 L; surface area: 0.07 m<sup>2</sup>) were set-up and operated next to the two pilot-scale HRAPs located at the Ruakura Research Centre, Hamilton, New Zealand (37°47'S, 175°19'E). The containers were foil-wrapped to ensure that sunlight only entered through the mesocosm water surface. The experiments were sequentially conducted over three seasons (Experiment 1: autumn; Experiment 2: winter; Experiment 3: spring) with each experiment lasting 36–39 days. The mesocosm operational parameters including the pond water used (either HRAP<sub>r</sub> or HRAP<sub>c</sub>), initial algal dominance, algal recycling rate, and the hydraulic retention time (HRT) are summarized in Table 1 for each experiment. A schematic diagram for the mesocosm experimental set-up is given in Fig. 1.

In Experiments 1 and 2, six of the twelve mesocosms were initially filled with water from a pilot-scale HRAP<sub>r</sub> dominated by *Pediastrum boryanum* (a readily-settleable alga) and the other six with water from a pilot-scale HRAP<sub>c</sub> dominated by *Dictyosphaerium* sp. (a poorly-settleable alga). The HRAP waters used for the mesocosms were pre-filtered using 200 µm mesh to remove large invertebrates (e.g. *Daphnia* sp. or *Moina* sp.) to avoid potential algal grazing. Of the six, three were operated with algal recycling (M<sub>r</sub>) and three without recycling (as controls, M<sub>c</sub>). For example, the triplicate mesocosms denoted as M<sub>r</sub>(HRAP<sub>r</sub>) were initially filled with HRAP<sub>r</sub> water and operated with algal recycling.

Each day settled algal biomass was removed from the bottom of the algal settling cone (ASC<sub>r</sub>) (*P. boryanum* dominated algal biomass) and 2.5 ml was added to the mesocosms (M<sub>r</sub>), which was the same recycling rate as that used in the pilot-scale HRAP<sub>r</sub>. Ultimately, because we wished to determine the net algal biomass productivity, the mass of solids that was recycled was subtracted from the total yielded from the pond.

Recycling extended the mean cell residence time (MCRT) in the mesocosms (M<sub>r</sub>) and was calculated using Equation (1).

$$\text{MCRT} = \frac{VX}{Q_c X - Q_{re} X_h} \quad (1)$$

Where;

MCRT = Algal mean cell residence time (d)

V: HRAP volume (m<sup>3</sup>)

X: HRAP algal biomass concentration (VSS, g/m<sup>3</sup>)

Q<sub>c</sub>: Net evaporation compensated HRAP effluent flow rate (m<sup>3</sup>/d)

Q<sub>re</sub>: Algal biomass recycled per day (1 L/d)

X<sub>h</sub>: Harvested biomass concentration (VSS g/L)

The mesocosms were operated as semi-continuous cultures with the same HRT as the pilot-scale HRAPs (for the time of year) by daily replacement (at ~9 am) of a portion of the pond water with primary treated domestic wastewater. During Experiments 1 and 3 (conducted in the New Zealand autumn and spring respectively) a 6 d HRT was maintained by replacing 3 L of mesocosm water each day. During

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