Bacterias fototróficas púrpuras para la recuperación de recursos: bases fundamentales y estado actual de desarrollo

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Purple phototrophic bacteria for resource recovery: fundamental bases and current stage of development

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INTRODUCTION

Purple phototrophic bacteria (PPB) have re-emerged as mediators for resource recovery from wastewater. The photoheterotrophic metabolism of PPB allows them to use light as energy source and organics as source of carbon and electrons. This particular growth strategy enables the simultaneous assimilation of carbon and nutrients, which can be recovered as biomass at yields up to 1 g COD·g COD^{-1}. Median removal efficiencies of 76%, 53% and 58% for COD, N and P have recently been reported, depending on the wastewater COD:N:P ratio (Capson-Tojo et al., 2020).

As with any phototrophic process, the main limitation for implementing PPB technology is its high cost (Acién Fernández et al., 2019). Therefore, profits must be maximised (via optimised treatment performance and product value) and costs must be minimised. The latter entails that enriched cultures must be used (no sterilisation costs), and that sunlight must be the energy source (artificial light is prohibitive) (Capson-Tojo et al., 2020). Optimal reactor design is also paramount, providing efficient mixing, light distribution, and allowing load maximisation. Decades of research on microalgal reactor design can provide a starting point for PPB, but conclusions from algae cannot be directly extrapolated, as their underlying biochemical processes are completely different. The most common cultivation technologies for phototrophs are photobioreactors (PBRs) and open ponds (OPs) (Posten, 2009). While PBRs lead to improved performances and productivities, they are more expensive, which has made OPs the most common configuration for growing microalgae (IEA Bioenergy, 2017).

Here, we present the bases of PPB biochemistry, their fundamental differences *vs.* algae, and the implications that these have for the implementation of PPB processes. In addition, we also present results from a demonstration scale PPB PBR operated outdoors, treating industrial wastewater (the first of its kind). These results, together with recently published data, are used to validate our statements and to point research towards realising the full-scale implementation of PPB processes.

MATERIALS AND METHODS

The demonstration plant PBR was a flat plate reactor of 1 m^3, covered with UV-VIS absorbing foil (Hülsen et al., 2022b). The wastewater fed (from a poultry processing plant), the feeding regime, the growth strategy (suspended *vs.* attached) and the HRT were varied over the operational period (192 days; see Table 1). The concentrations of soluble and total COD, total phosphorus (TP) and total Kjeldahl nitrogen (TKN), NH_4^+-N, PO_4^{3-}-P, VFAs, and total and suspended solids were measured twice a week. The characteristics of the harvested biomass (via centrifugation) were also determined.

RESULTS AND DISCUSSION

Results from the demonstration scale photobioreactor

The PBR provided effective VFA, N and P removal despite the varying environmental conditions (*i.e.*, day-night cycles, peaks daily temperatures of 14-42 ºC, and daily irradiances of 3-32 $MJ·m^{-2}$), with average removal efficiencies of >90% (in most phases), 34-77%, and 21-45%, respectively (Figure 1). Photoheterotrophic assimilation of VFAs occurred at rates over 0.5 g $SCOD·L^{-1}·d^{-1}$ (peaks up to 1.0 g $SCOD·L^{-1}·d^{-1}$). The removal of N and P was limited by COD availability (*i.e.*, VFAs), as the wastewater had a COD:N:P ratio far from the PPB uptake ratio (Figure 1). Feeding prefermented wastewater only during daytime hours allowed to maximise biomass productivities and PPB relative abundances (up to 0.56), while maintaining treatment performance. Optimal retention times of 2.1-2.4 d (Table 1) lead to estimated biomass productivities up to 24 g $VS·m^{-2}·d^{-1}$ (conservative), at organic loading rates around 1.5 g $COD·L^{-1}·d^{-1}$. The produced biomass was harvested at 90±1% VS/TS, with a crude protein content of 58±14% and an amino acid profile suitable for animal feeding.

PPB fundamentals and implications for reactor design

The data shown above show that PPB can effectively generate a valuable product from wastewaters using outdoors PBRs. Nevertheless, while promising, the reported loads and productivities do not ensure a feasible economic process, even when using OPs. The given values are on the high-end for microalgal processes, suggesting process feasibility, but extrapolations and comparison from/with algae systems can be misleading. The main reasons for this are: (i) a more significant light attenuation in PPB cultures (Capson-Tojo et al., 2022) and (ii) the requirement of anoxic/anaerobic conditions. These differences have two main implications for reactor design: (i) light-path lengths need to be shorter in PPB systems (*e.g.* 5-10 cm *vs.* 30 cm deep OPs, (Capson-Tojo et al., 2022)), and (ii) O_2 diffusion must be minimised. The PBR used above had a thickness of 8 cm, showing that there was no effective light limitation. Results from OPs (100 L) reported light limitation at depths of 10-20 cm (Alloul et al., 2021) and at 15 cm (Sepúlveda-Muñoz et al., 2020). Light limitation at depths of 15 cm in OPs was confirmed by other studies, where longer retention times (4-11 d) than those usually applied were needed (García et al., 2019; López-Serna et al., 2019). Concerning anaerobic conditions, no oxygen was detected in the 8 cm demonstration PBR (despite being open at the top (Hülsen et al., 2022b)). A study using OPs recently suggested that this might be challenging in OPs, due to the larger liquid-gas exchange surface (Alloul et al., 2021). Minimisation of dissolved oxygen (DO) in 10-20 cm deep OPs by daytime mixing only increased the PPB proportions from 14 to 56%. The need to minimise DO concentrations in OPs by low mixing intensities and by small surface/volume ratios (S/Vs) to reduce O_2 diffusion (opposed to the high S/V needed for efficient light supply), will be a key challenge for designing PPB OPs. These challenges might tip the balance towards the utilisation of PPB PBRs (see Figure 2 for a comparison between PBRs and OPs).

CONCLUSIONS

Although dedicated research is needed using both PBRs and OPs under different conditions, the given results (and fundamental differences between PPB and microalgae) suggest that PBRs seem to be a more promising option for PPB systems. Data from the demonstration PBR show that the process in technically fasible, with promising loads, productivities and retention times in outdoor units. Nevertheless, dedicated economic-cost analyses are needed, as well as data from holistic demonstration-scale processes. Crucial aspects to be studied are the applicability and value of the harvested PPB biomass (including product biosafety assessment), as well as data on biomass post-treatment (*e.g.*, harvesting, drying, potential sterilisation, etc.), and PBR mixing.

FIGURES AND GRAPHICS

Table 1. Operational conditions of the photobioreactor during different periods (Hülsen et al., 2022b).

Parameter	Phase I	Phase II	Phase III	Phase IV	Phase V	Phase VI
Duration (d)	1-32	35-60	63-95	93-118	120-127	130-192
Substrate	FWW	FDAF	FDAF	FDAF	FDAF	FDAF
Feeding strategy	Continuous	Continuous	Continuous	Continuous	Daytime	Daytime
HRT (d)*	4.4-5.7	2	2	1	2.4	2.1
Growth strategy	Suspended	Attached	Suspended	Suspended	Suspended	Suspended
Average temperature inside reactor (ºC)**	26 (2.8)	25 (3.5)	25 (3.1)	24 (3.0)	18 (1.5)	16 (1.3)
Daily average irradiance ($MJ \cdot m^{-2}$)	26 (5.7)	20 (7.9)	19 (7.1)	18 (4.1)	15 (2.3)	13 (1.7)

FWW stands for fermented wastewater, FDAF for fermented dissolved air flotation effluent and HRT for hydraulic retention time.
* Note that when feeding only during daytime, the daytime (effective) HRT is half of the given value.
** These values correspond to the moments when the samples were taken (10-12 am).

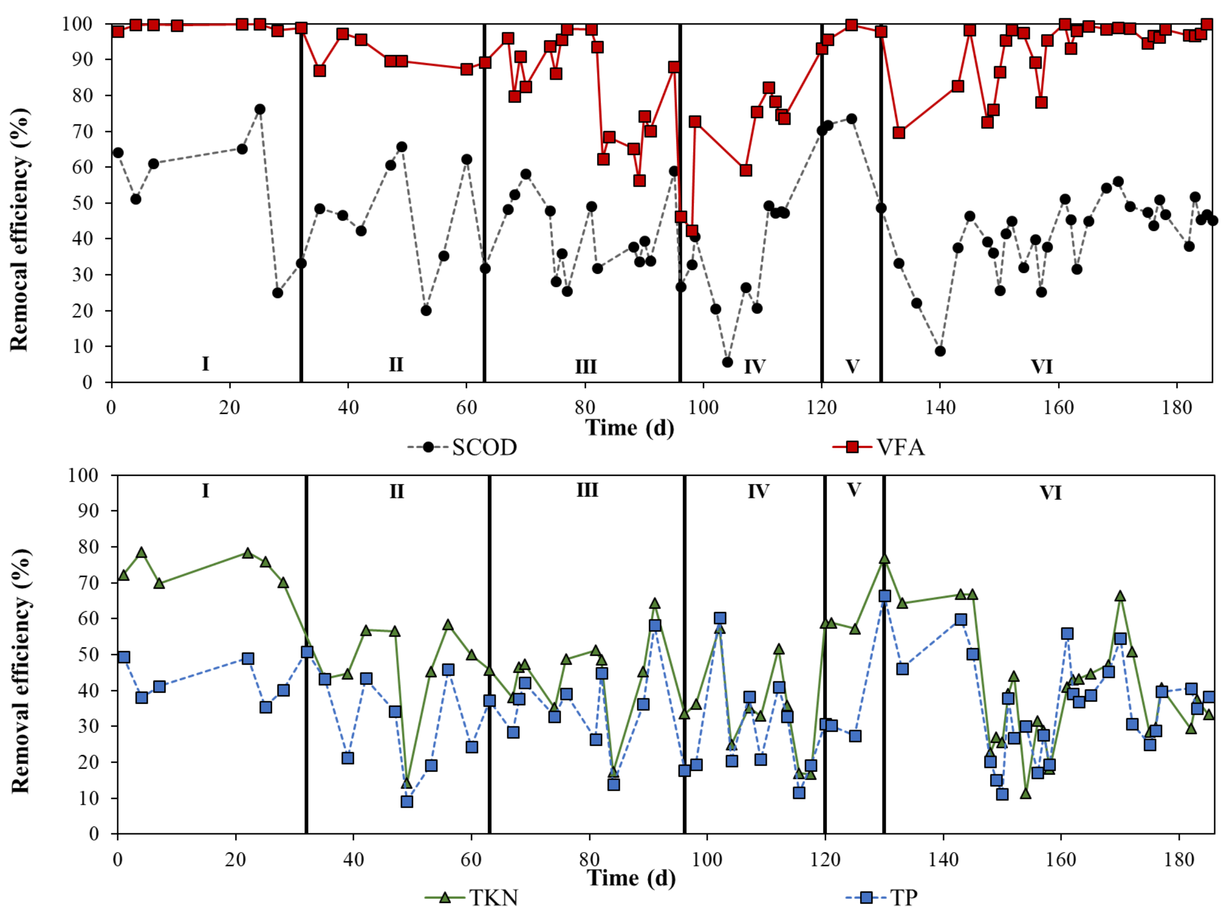

Figure 1. Removal efficiencies of (up) SCOD and VFAs and (down) total Kjeldahl nitrogen (TKN) and total phosphorus (TP) at the different operation periods. The roman numbers refer to the phases described in Table 1. Adapted from Hülsen et al. (2022b).

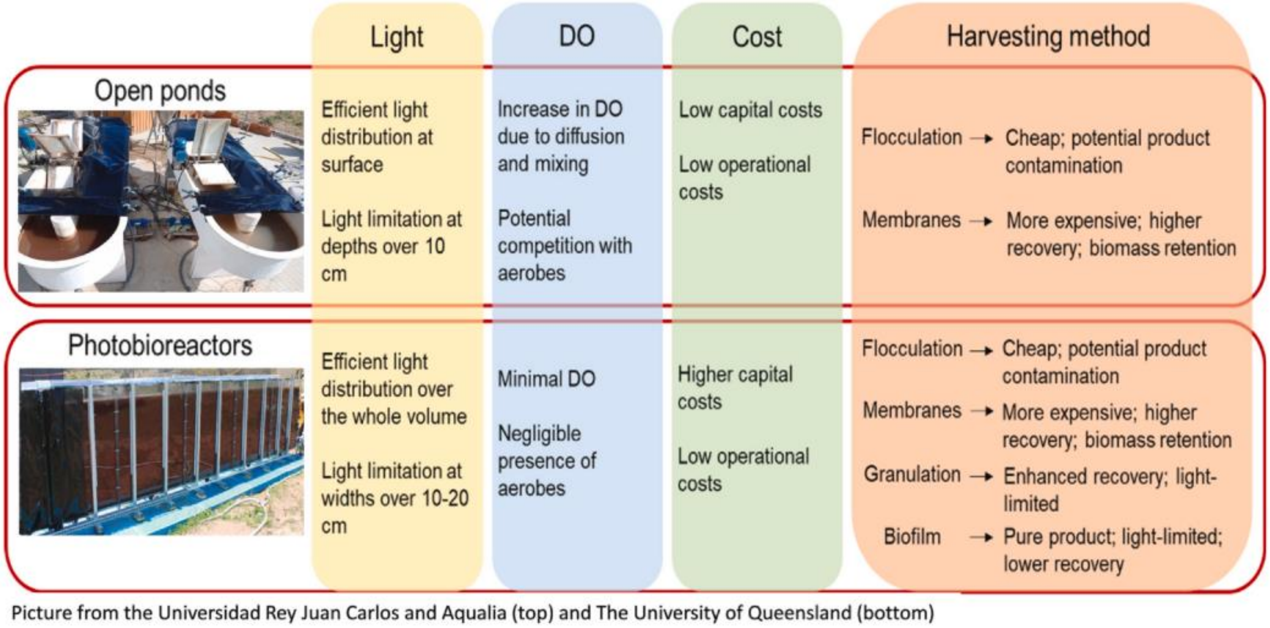

Figure 2. Qualitative comparison between open ponds (OPs) and photobioreactors (PBRs). Their main characteristics concerning light distribution, dissolved oxygen (DO) availability, cost, and applicable harvesting methods are discussed. Harvesting is not discussed in the text due to space limitations. Adapted from Hülsen et al. (2022a).

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