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Using a mechanistic model to further develop purple phototrophic bacteria systems for resource recovery

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Introduction

Processes based on purple phototrophic bacteria (PPB) are a promising option for resource recovery from wastewater. The photoheterotrophic metabolism of PPB (using light as energy source) allows them to simultaneously remove organics and nutrients (*e.g.*, N and P), recovering them as value-added products (*e.g.*, biomass (to be used as single-cell protein source or as fertiliser), or polyhydroxyalkanoates) at high biomass yields (up to 1 g $COD_{biomass}$·g $COD_{removed}^{-1}$) (Capson-Tojo et al., 2020).

The single mechanistic model available to represent nutrient recovery by PPB is the Photo-anaerobic model (PAnM) (Puyol et al., 2017). The PAnM is accurate to represent laboratory-scale conditions working in a controlled environment, but it cannot predict the behaviour of outdoor PPB reactors treating complex wastewater streams. Consequently, the PAnM has been recently extended to account for: changing environmental conditions, light attenuation, the complex nature of waste streams, microbial synergies and competitions, inihibitory processes, and the diverse metabolic capabilities of PPB (Capson-Tojo et al., 2022b). All these factors are essential for the accurate simulation of potential full-scale PPB-based processes for resource recovery.

After calibration and validation with data from a flat-plate demonstration-scale photobioreactor (PBR), the resulting extended model (ePAnM) has been used here to estimate optimal operational and design parameters, and to evaluate scenarios that could foster the implementation of PPB-based systems (*e.g.*, intermittent night aeration). With a clear lack of experimental data from outdoors PBRs, these results might be tremendously useful for the future implementation of PPB reactors.

Methodological Approach

The model was calibrated and validated using data from a 10 m demonstration-scale PPB PBR, with a volume of 1 m^3 (the first of its kind). A period of 33 days, working at varying operating/environmental conditions, was selected (see Hülsen et al. (2022b) and Capson-Tojo et al. (2022) for the plant description and the calibration procedure).

The resulting model was used to simulate scenarios to determine optimal operational and desing parameters (*e.g.*, hydraulic retention time (HRT) and reactor width).

Simulations assumed an ideal influent with constant composition, with excess of nutrients, and with volatile fatty acids (VFAs) concentrations in the influent of 500-3,000 mg COD·L^{-1}. Simulation time was 30 days, assuming a reactor equivalent to the aforementioned PBR. Anaerobic, continuously illuminated conditions were assumed (near infrared intensities of 5-200 W·m^{-2}), with continuous feeding (pH controlled at 7) and temperatures of 10-45 ºC. Tests at different HRTs assumed a width of 0.08 m and those at varying widths assumed an HRT of 0.5 days. MATLAB (R2015a) was used.

A scenario that could improve the system performance by allowing continuous-outdoors operation was also simulated. This approach (proposed in Hülsen et al. (2022b)) combines anaerobic-photosynthetic growth during daytime and aerobic-heterotrophic growth during night hours. HRTs of 1 and 2 days were tested, under the same illumination profile used for model validation (real day-night variations). The reactor characteristics were those used in the simulations at different HRTs (influent of 1,500 g COD·L^{-1}). At night, dissolved oxygen (DO) levels were 0-5.7 mg DO·L^{-1}.

Results and Discussion

Figure 1 shows the total VFA removal efficiencies (used as performance crtiteria) from the simulations at different HRTs and reactor widths.

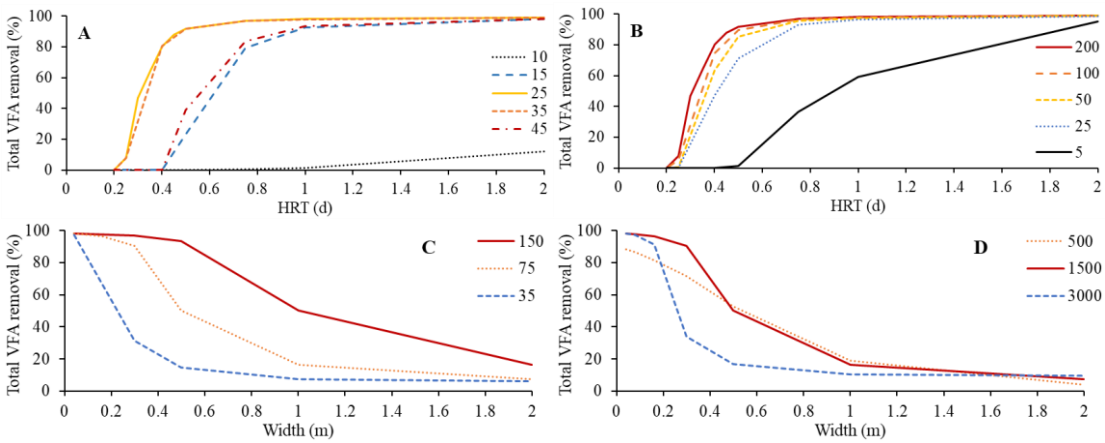

Figure 1 Simulation results showing the total VFA removal efficiencies in a flat plate PBR at different (A) HRTs and temperatures (ºC), (B) HRTs and light intensities (W·m^{-2}), (C) reactor widths and light intensities (W·m^{-2}), and (D) reactor widths and influent concentrations (mg COD·L^{-1}).

The results show that, at optimal temperatures and light intensities (*e.g.*, 25-35 ºC and >25 W·m^{-2}) HRTs above 0.5 d must be kept if PPB biomass washout is to be avoided (Figures 1A-B), thus keeping an efficient removal performance. For outdoors systems, this implies that a semicontinuous reactor fed only during daytime will have to run at a minimum HRT of 1 d. This is in agreement with previous limits presented in the literature (Hülsen et al., 2022b) and with values calculated using common PPB growth rates (Capson-Tojo et al., 2020). Non-optimal temperatures (10, 45 ºC) resulted in longer required HRTs, as they resulted in slower kinetics. Reducing the light intensities did not jeopardise the process performance until very low values (*i.e.*, below 25 W·m^{-2}). This is obviously also related to the reactor width, which affects light attenuation

through the reactor. Simulations at different reactor widths show that, at common average light intensities and influent concentrations (*e.g.*, 75 $W \cdot m^{-2}$ and 3,000 mg $COD \cdot L^{-1}$), the performance falls drastically at values over 15-30 cm (assuming 2-sided illumination). This value is also in agreement with numbers from the literature, where it was proven that light attenuation through the reactor is mostly caused by light absorption by pigmented biomass, and that this effect is more pronounced in PPB than in microalgal cultures (Capson-Tojo et al., 2022a). Lower incident intensities obviously resulted in shorter optimal widths, and higher influent concentrations lead to more biomass accumulation, favouring light attenuation and reducing the allowable widths.

The simulations results from the day-anaerobic/night-aerobic operation at different HRTs (1 and 2 d) are presented in Figure 2, where the predicted concentrations of VFAs, nutrients, and PPB biomass under different aeration regimes are provided.

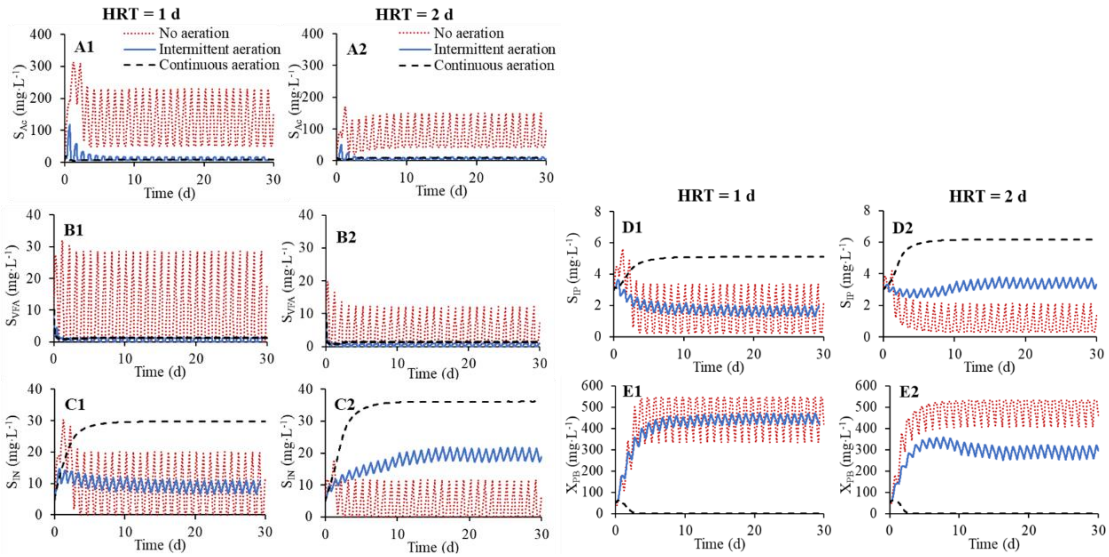

Figure 2 Simulation results showing the concentrations of (A) acetate (S_{AC}), (B) other VFA (S_{VFA}), (C) inorganic nitrogen (S_{IN}), (D) inorganic phosphorus (S_{IP}), and (E) PPB biomass (X_{PB}) in an outdoors flat plate PBR working at an HRT of (A1-E1) 1 day, and of (A2-E2) 2 days. Three aeration strategies are compared: no aeration, intermittent aeration during night-time, and continuous aeration. The reactor was continuously fed with an influent containing 600 mg $COD \cdot L^{-1}$ of total VFAs. A natural illumination profile was assumed, with an incident NIR fraction in the reactor of 100%.

While continuous aeration resulted in PPB outcompetition (and lower nutrient removal due to dominance by heterotrophs) and no aeration resulted in no removal of organics and nutrients during night-time (lack of light), intermittent aeration merged the best of both options. It provided total VFA removal continuously, while keeping an average nutrient removal similar to that achieved in non-aerated reactors (particularly at an HRT of 1 days, thanks to the wash-out of aerobic grazers and PPB aerobic growth).

Conclusion

The ePAnM was used to determine crucial optimal operational and design parameters in PPB flat plate PBRs, concluding that: (i) a minimum HRT of 0.5 d must be kept if PPB biomass washout is to be avoided and, (ii) reactor widths below 15-30 cm should

be used (implying 7.5-15 cm depths for ponds). These values (still unknown in the literature) might be starting points for coming techno-economic analyses, which will determine the feasibility of PPB-based systems for resource recovery. The proposed daily-anaerobic night-anaerobic approach might be an option to improve the system performance, allowing continuous operation, but slightly sacrificing biomass yields due to lower aerobic biomass yields. Further experimental validation is needed.

Opinion

As a technology still in its infancy, there is a current debate around which cultivation strategy (*e.g.*, open ponds or PBRs) should be used in PPB systems. Defenders of using ponds base their reasoning mostly on results from lab-scale systems and on cost-analysis using data from microalgae reactors (Alloul et al., 2021, 2019; Puyol et al., 2019). Although this approach has merits (and obviously we could be wrong), we think that PBRs might be the right approach to pursue (if not the only feasible one). Fundamental differences between PPB and microalgae (reviewed in Hülsen et al. (2022)) impose that open ponds for PPB need to be too shallow (<15 cm), and thus too expensive. Using results from the presented simulations, we give numbers to the maximum depth that ponds could have, further confirming their limitations. Although dedicated economic analyses should be performed using experimental data and the results presented here, PBRs appear as the sole potentially feasible alternative.

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