Edward Barry

Distributed parameter modelling of phototrophic bioreactor systems
List of corrections as agreed upon during examination

Examiners
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The changes highlighted in the oral examination supersede those in the individual examiners' reports, with the exception of the minor corrections at the end of Prof. Leslie's report.

1 Page 5: Some details for reference [39] are missing (volume and page numbers).

I have updated the bibliography entry for Krujatz et al., 2015 to include the volume (112) and page numbers (2439-2449).

2 Page 5: The reference by Minkevich et al., 2004 cites data collected at 860 nm. This contradicts the statement on the lack of specific and relevant wavelength data for cultivation of Rb capsulatus. Candidate should clarify if this data is relevant or not for the current study.

This reference shows that there is a higher photosynthetic efficiency for each photon entering the system. This means that the efficiency changes for different wavelengths, but does not quantify the resulting growth rate or limiting parameters on wavelength or intensity.

The most interesting study done in that respect was with the series of three papers by Zhou et al. (2014, 2015b,a). There they look at photoperiod, light intensity, and wavelength however clarifications sought from the corresponding author showed these studies could not be used with confidence in my thesis. They used a photometric basis, not a radiometric basis for all wavelengths and intensities, and when asked, could not explain which particular wavelengths were used. By knowing the exact band of wavelengths, one could back-calculate to find a radiometric basis in the experiments.

3 Page 7: Equation 1.1 S_C is not defined in list of symbols. Nomenclature and symbols for all equations should be checked before final submission

I have included the following paragraph to explain the meaning of the symbols in this equation, and added those symbols to the list of symbols at the start of the thesis.

The second term on the left side of Eq. 1.1 represents convection of the particles with the velocity of the fluid phase(s), coupling the flow and concentration fields. This coupling is one-way since the solids concentration does not influence the flow field. The first term on the right hand side of the equation represents diffusion processes, and is normally included for numerical stability

with the diffusion coefficient Γ set arbitrarily low. The second term on the right hand side of the equation (S_C) represents all sources and sinks of the scalar quantity C.

4 Page 8: Equation 1.2. Subscripts a, D and p not defined in list of symbols

*I have added the following in order to clarify the meaning of the subscripts **a**, **D** and **p**.*

For Eulerian-Lagrangian references, a particle (P) with velocity $\mathbf{U}_P$ is tracked through the fluid field ($\mathbf{U}$), and is subjected to drag force ($\mathbf{F}_D$), gravity and particle/bulk density differences $\left(g\frac{\rho_P-\rho}{\rho_P}\right)$, and any additional forces ($\mathbf{F}_a$). Eq. 1.2 describes this behaviour.

5 Page 8: The use of Reference [57] does not support the statement as this work does not compare Eulerian and Lagrangian approaches for particle transport in enclosed air spaces. Please revise.

This was a referencing error. I have now pointed this claim to Zhang and Chen (2007).

6 Page 8: There is a section of text (paragraph) that is missing between page 8 and 9. Logic of the two paragraphs does not flow.

I have added the following into that section, and removed the mentions of the various CFD packages as they don't add much to the analysis.

To summarise, the most common and important physical phenomena to consider when modelling photobioreactors are:

- hydrodynamics, mixing and mass transfer
- radiative transfer
- physiology and biokinetics
- biofilms
- methods for coupling all physical phenomena

Microbial physiology and growth kinetics can be simulated well in lumped parameter systems, where spatial variations are considered negligible, however when coupling occurs with the remaining physical phenomena, spatial variations should be taken into account. Computational fluid dynamics is a discipline allowing users to account for these changes in space, however these simulations should be accompanied by proper error and convergence analysis.

7 Reference [75] is missing the year of publication. (should be 2013). Also, as the results from reference [75] conclude that Type 2 models best approach for out door but type 3 may be better for indoor systems. Features of these models are reiterated in the introduction to section 1.3 which is confusing.

I used IEEE referencing style for this thesis which puts the publication year at the end of the reference. Reference 75 Béchet et al. (2013) is now reference 76 due to adding in Zhang and Chen (2007) above.

I have removed the references to the type I, II and III models and have maintained what I wrote about the scattering effects, and range of biochemical models for algal systems.

8 Page 10. There is some confusion on the use of symbols. In Equation 1.4, β is listed in text as extinction coefficient – It is assumed that this is comparable to the attenuation coefficient as described in the symbols list. Please use consistent nomenclature.

I have now written some extra information on the Beer Lambert law (Eq 1.4) where I explain what β is. I have also clarified that β is the sum of κ and σ and what their units are [m^{-1}]. I have also changed the extinction coefficient to β from ϵ elsewhere in the document so that some consistency is maintained.

I have changed all instances of "attenuation" to "extinction" in the document. They have the same meaning, but using them interchangeably is confusing for the reader.

This approach lumps the absorption (κ_λ) and out-scattering (σ_λ) coefficients into one extinction coefficient (β_λ) by addition. The spectral absorption, out-scattering and extinction coefficients all of the units of m^{-1}.

9 Page 12: Difficult to follow the logic of paragraphs 2,3 and 4. The sudden introduction of "solution scheduling" is confusing. This could be improved by beginning paragraph 4 with the sentence: "Examples of where spectral simulations were carried out using the FVDOM or DOM...."

The first few sentences did appear to be out of place. I have moved them to the end of that paragraph as they have a concluding role rather than introducing anything.

10 Page 16: Equation 1.6 has no units. Please provide units for all equations.

For each term in the equation, I have included the units and the description of what each variable does physically.

11 Objective 1: please explain, more clearly, the selection of the anaerobic model and the Monod kinetics

I have added this to research objective 1:

The model presents a generic representation of anoxygenic phototrophic growth for a stoichiometric balance. The kinetics were inspired by the IWA-family models and are appropriate in this case as it captures substrate-cell interactions in ways that Michaelis-Menten kinetics can not. Other kinetic models proposed for photosynthetic organisms such as the Gompertz or Droop models were not selected as they do not reflect anoxygenic phototrophic growth modes better than Monod kinetics (the former accounting for a lag phase at the beginning of experimental runs, and the latter being a better descriptor of photosynthetic growth (growth on inorganic carbon, nutrients and water) accounting for excesses of nutrients. The model considered a closed anaerobic system where the mass balance could be closed on organic compounds (chemical oxygen demand) without defining a new electron acceptor.

12 Chapter 3: please plot velocity distributions predicted for both configurations, and add a paragraph to discuss about the effects of configurations/geometries on the results; acknowledge any shortcomings of the work in this regard

I have added the following paragraph to the chapter in the section entitled "Limitations of the model (section 3.3.4). The corresponding figure is included below the paragraph here.

Fig. 1 shows the velocity fields for the FPR (a) and the CR (b). The important characteristics of these velocity fields are that there is a greater maximal velocity in the case of the CR, and that leads to a fairly uniform velocity profile in the lower half of the reactor. However, there is a distinctly different profile above the impeller where the velocity magnitude decreases by roughly an order of magnitude. The profile in the CR differs from that of the FPR where there is a lower maximal velocity magnitude, but the distribution is more uniform in the sense that it is within the same order of magnitude throughout the reactor with only a small proportion of the reactor with low-velocity zones. It is difficult to conclude that the different velocity profiles of each reactor contribute significantly to different biochemical behaviour within the reactors compared with radiative field, and short term particle radiation dynamics. This uncertainty can serve as a base for future work in decoupling these factors for phototrophic growth.

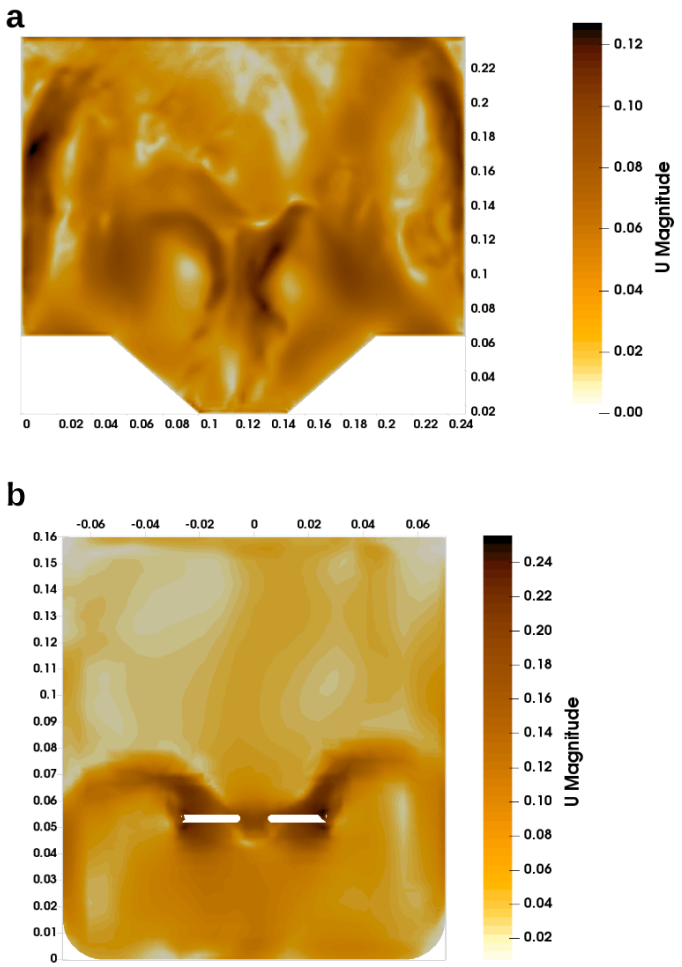

Figure 1: Liquid velocity field variations between the two reactors. The velocity fields are within an order of magnitude of each other, but there is a more uniform distribution in the FPR due to the geometry and mechanical mixing.

13 In Equation 3.12, D should be taken out of the Laplacian operator; Diffusion plays an important role in biofilm modelling, the effect of the biofilm matrix (e.g. EPS) and cell mobility on the D value should be discussed

I have taken the diffusion coefficient outside of the Laplacian in both Chapters 3 (Eq 3.12) and 4 (Eq 4.3 and 4.6). I have started a discussion on the role of the diffusivity coefficient and its differential nature in more realistic modelling scenarios. I added the following passage to the general species transport equation in section 4.2.3 as it is a relevant discussion for biofilms

It is important to note that the diffusion coefficient has been placed outside of the Laplacian operator. This means that there is no spatial variation of the apparent diffusivity through the biofilm. There are cases where the diffusion coefficient can vary in both time and space Vourc'h et al. (2018). These changes in diffusion coefficient can be indirectly due to the excretion of extracellular polymeric substances (EPS) over time. It is thought that the deposited EPS triggers cellular changes affecting cell motility Vourc'h et al. (2018). Spatial variation of the diffusion coefficient in biofilms has also been reported Sankaran et al. (2019). Operating conditions such as void fraction, biofilm tortuosity, and internal EPS can affect the molecular diffusivity of sub-

stances at different depths. The whole structure serves as a sort of molecular sieve Sankaran et al. (2019).

14 In Chapter 3, Newtonian flow was assumed, which is reasonable for this work. However, the assumption should be stated more clearly, and conditions for such an assumption to be valid/invalid should be discussed

I have included this paragraph on the Newtonian fluid assumption in section 3.2.2.

In wastewater treatment, sludge does not generally behave like a Newtonian fluid. This assumption can hold for low concentrations of biomass (around 3 gTSS/L) Hong et al. (2018). Temperature also has an impact on the viscosity of sewage sludge with lower temperatures correlating to a departure from Newtonian behaviour Hong et al. (2018). The Newtonian fluid assumption would therefore be voided under different modelling applications of purple bacteria such as biomass thickening applications, or low temperature wastewater treatment Hülsen et al. (2016).

15 Chapter 3, Figure 3.4: explain how the plot was generated; explain why the frequencies are different; explain the physical reason(s) for the differences; also, the take-home messages for readers should be made clearer

I have included this as a discussion on the frequencies:

A question arising from the dynamic radiative field is whether the PPB biomass is sufficiently fast to respond to these changes. If the biomass response is much faster than the change in intensity, the growth kinetics can be treated as a continuous function (with dependence on intensity). If biomass response kinetics are also on the order of seconds, lower-level expressions would be required (which also consider photo-enzyme kinetics). The response time for *Rhodobacter sphaeroides* was determined in the literature to be in the order of 10^{-11} seconds Slouf et al. (2012); a time constant much smaller than the periods seen in Fig. 2. Therefore, this justifies the use of a continuous function with respect to radiation for both these cases, and in scaled-up systems, since they will generally have longer time constants due to the larger dimension with respect to velocity. This also justifies the approach used in this study to generate results, with biomass growth coupled to the radiative field, and phototrophic organisms self-shading as they grow.

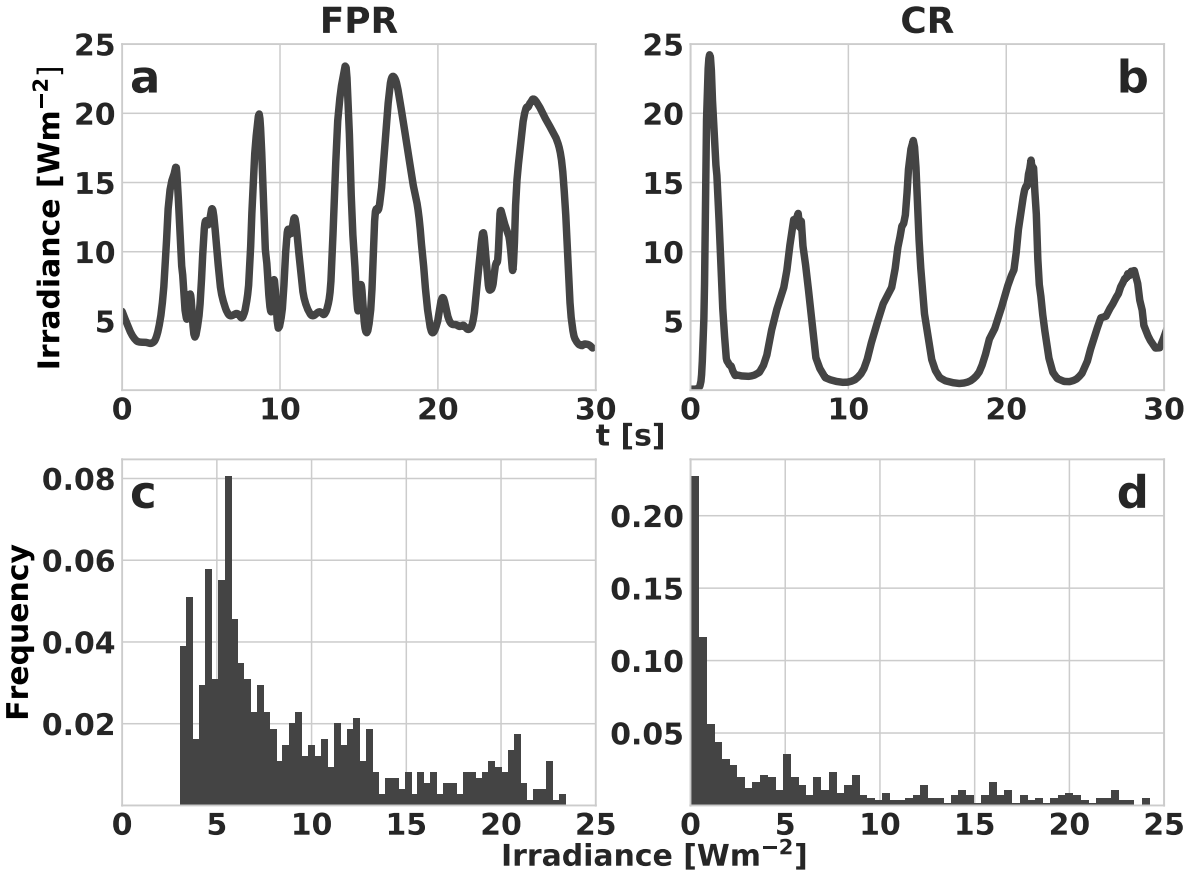

Figure 2: Particle radiation dynamics for an 850 nm band of irradiance. The top row (a, b) shows the transient radiative field experienced by a PPB particle for the flat plate and cylindrical reactors respectively. The same field is represented for both reactors (c, d) as a histogram on the bottom row. Both reactors have an incident irradiance of 30 Wm^{-2}

16 Chapter 3: add "mesh independence test"

I have added some information about the mesh independence test in section 3.2.5 where I stated that both cases had 260 000 cells as the optimal result/computational power.

17 Fig 4.18: Please comment on the oscillations on the curves, explain the reason(s) – is it a numerical issue? Discuss what could happen after 10 days.

The following was added to Section 4.5.3: Volume averaged irradiance and growth rate over time.

For the simulation conditions where the radiant energy was deployed from above the domain, there was a lag in specific growth rate for the sparsely initialised biofilm (case 5) compared with the uniformly initialised biofilm (case 3). This can be explained by the fact that the phototrophic biofilm was initially closer to the radiative field when compared with case 5. The biofilm in case 3 experienced 2 days of growth before reaching a steady state of growth of 2.5 kgCOD m^{-3} d^{-1}.

The biofilm in case 5 experienced 8 days of growth before reacting a steady state. Both cases had similar volume averaged irradiance throughout the simulations, with the sharper initial rate in decline for case 3 (the uniformly irradiated biofilm) being due to the greater volume of biomass initially in the system. The oscillations visible in this figure do not detract from the general trend of growth characteristics, but are mostly due to both numerical and physical diffusion. More simulations would be required to decouple physical phenomena from potential numerical issues. To extend the simulations beyond 10 days, a larger domain would be required. As there are no external forces mechanically removing biofilm from the structure, the biofilm would expand to fill the entire simulation domain. This highlights a need to explain biofilm relationships with external flow fields for longer term steady-state simulations.

18 Fix up any other spelling mistakes throughout the thesis

I have fixed up spelling and grammatical errors to the best of my ability throughout the thesis.

References

Béchet, Q., A. Shilton, and B. Guieysse
2013. Modeling the effects of light and temperature on algae growth: State of the art and critical assessment for productivity prediction during outdoor cultivation. *Biotechnology Advances*, 31(8):1648–1663. ISBN: 0734-9750.

Hong, E., A. M. Yeneneh, T. K. Sen, H. M. Ang, and A. Kayaalp
2018. A comprehensive review on rheological studies of sludge from various sections of municipal wastewater treatment plants for enhancement of process performance. *Advances in Colloid and Interface Science*, 257:19–30.

Hülsen, T., E. M. Barry, Y. Lu, D. Puyol, and D. J. Batstone
2016. Low temperature treatment of domestic wastewater by purple phototrophic bacteria: Performance, activity, and community. *Water Research*, 100:537–545.

Krujatz, F., R. Illing, T. Krautwer, J. Liao, K. Heibig, K. Goy, J. Opitz, G. Cuniberti, T. Bley, and J. Weber
2015. Light-field-characterization in a continuous hydrogen-producing photobioreactor by optical simulation and computational fluid dynamics. *Biotechnology and Bioengineering*, 112(12):2439–2449.

Minkevich, I. G., T. V. Laurinavichene, and a. a. Tsygankov
2004. Theoretical and experimental quantum efficiencies of the growth of anoxygenic phototrophic bacteria. *Process Biochemistry*, 39(8):939–949.

Sankaran, J., N. J. H. J. Tan, K. P. But, Y. Cohen, S. A. Rice, and T. Wohland
2019. Single microcolony diffusion analysis in Pseudomonas aeruginosa biofilms. *npj Biofilms and Microbiomes*, 5(1):1–10.

Slouf, V., P. Chabera, J. D. Olsen, E. C. Martin, P. Qian, C. N. Hunter, and T. Polivka
2012. Photoprotection in a purple phototrophic bacterium mediated by oxygen-dependent alteration of carotenoid excited-state properties. *Proceedings of the National Academy of Sciences*, 109(22):8570–8575. bibtex*[pmid=22586075].

Vourc'h, T., H. Peerhossaini, J. Léopoldès, A. Méjean, F. Chauvat, and C. Cassier-Chauvat
2018. Slowdown of surface diffusion during early stages of bacterial colonization. *Physical Review E*, 97(3):032407.

Zhang, Z. and Q. Chen
2007. Comparison of the Eulerian and Lagrangian methods for predicting particle transport in enclosed spaces. *Atmospheric Environment*, 41(25):5236–5248. ISBN: 1352-2310.

Zhou, Q., P. Zhang, and G. Zhang
2014. Biomass and carotenoid production in photosynthetic bacteria wastewater treatment: Effects of light intensity. *Bioresource Technology*, 171:330–335.

Zhou, Q., P. Zhang, and G. Zhang
2015a. Biomass and pigments production in photosynthetic bacteria wastewater treatment: Effects of light sources. *Bioresource Technology*, 179:505–509.

Zhou, Q., P. Zhang, G. Zhang, and M. Peng
2015b. Biomass and pigments production in photosynthetic bacteria wastewater treatment: Effects of photoperiod. *Bioresource Technology*, 179:505–509.