Technical University of Liberec

Faculty of Mechatronics and Interdisciplinary Engineering Studies

Resumé of Ph.D. Thesis

Photobioreactors for cultivation of microalgae under strong irradiances: Modelling, simulation and design

by

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Contents

Notation

Abbreviations

Abbreviation	Meaning
BC(s)	Boundary condition(s)
BLSSI	Bilinear system with single input variable
CFD	Computational fluid dynamics
CV	Control volume
IC(s)	Initial condition(s)
L – D cycles	Light/Dark cycles
ODE, PDE	Ordinary/partial differential equation
OS	Operator splitting
PBR	Photobioreactor
PFD	Photon flux density
PSF	Photosynthetic factory
PSU	Photosynthetic unit
UHCD	Ultrahigh cell densities

Physical quantities

Quantity	Unit	Meaning
c_x	kg–cell m^{-3}	biomass concentration (or cell density)
c_{xt}	kg–cell m^{-3}	biomass concentration at time t
f_{ij}	-	relative flowrate between adjacent compartments (from i to j)
h	s (or m)	discretisation parameter
I	μE m^{-2}s^{-1}	irradiance or PFD
I_0	μE m^{-2}s^{-1}	incident irradiance
k	s^{-1}	rate constant (or a general constant with a corresponding unit)
Me	s^{-1}	Maintenance term in PSF model
t	s	time - independent variable
T	s	total period of the L – D cycle
V	m^3	total PBR (culture) volume
$\dot{V}$	m^3 s^{-1}	total flowrate
V_i	m^3	volume of compartment i
$\vec{u}$	m s^{-1}	fluid velocity
$\vec{u}_m$	m s^{-1}	molar velocity of the mixture
$\bar{u}$	m s^{-1}	mean axial velocity
x_1, x_2, x_3	-	states (both labels and molar fractions) of the PSF model

Greek letters

Quantity	Unit	Meaning
α	μE^{-1} m^2	rate constant of PSF model
β	μE^{-1} m^2	rate constant of PSF model
γ	s^{-1}	rate constant of PSF model
δ	s^{-1}	rate constant of PSF model
κ	1	yield of photosynthesis production of PSF model
μ	s^{-1}	specific growth rate
ν	s^{-1}	frequency of light/dark cycles
ϕ	-	ratio between the light period and the total period of the light/dark cycles
ϕ_a	-	ratio between time interval corresponding to I_a and total period of generalized L-D cycles
ϕ_b	-	ratio between time interval corresponding to I_b and total period of generalized L-D cycles

1 State-of-the-art of photobioreactor modelling

1.1 Introduction

My thesis deals with modelling and design of algal bioreactors operating under strong irradiances. It covers the most relevant physical, biological and mathematical aspects of the modelled phenomena, allowing the growth simulation of microalgal mass culture in a general photobioreactor (further PBR) without being constrained by a special geometry or structure.

Nevertheless, an important aspect of this thesis is its nexus to the novel photobioreactor with Fresnel lenses, designed in Třeboň and built in Nové Hrady, Czech Republic, in the past years (see Fig. 1). The tag of 'strong irradiance' appears in the thesis title not only due to the close relation between my thesis and the photobioreactor with Fresnel lenses, alowing several fold greater incident irradiance than the ambient value is, but also for the other reasons: Firstly, the strong incident irradiance makes possible economically advantageous *ultra-high cell density cultures* (see Subsection 1.3.6 of my thesis); Second, the strong irradiance imposes the necessity of the common study of photolimitation and photoinhibition processes, and finally third, according to the optimal principle of Pontryagin, the regular solution of an optimisation problem with restrictions resides in the switching between the extreme values, and consequently, the so-called light/dark cycles gains new insight in this context.

In the following pages, I mention the state-of-the-art of PBR modelling. A special attention is paid to the so-called dynamic model of photosynthetic factory (PSF), which forms the cornerstone of my thesis, nevertheless the method of extending the PSF model to the whole PBR (i.e. the multicompartment approach) is generally valid independently on the form of basic growth model with lumped parameters. Afterward, in the next sections of this *Resumé of Ph.D. thesis* the objectives of my thesis are enumerated, the methods used are described, and finally, the original results and conclusions are briefly presented.

Figure 1: Tubular photobioreactor with Fresnel lenses. In Institute of Physical Biology, Nové Hrady, Czech Republic.

1.2 Motivation – looking for an optimal PBR design

Several challenging and sometimes not entirely clear statements of the following authors were my motivation when I started to work on the project of design of an algal photobioreactor with Fresnel lenses:

- Ogbonna and Tanaka (1995)[38]: It is thus utmost necessity to develop a general method for quantitative evaluation of the light conditions inside PBR. For a given cell and process, the optimum light condition must be determined and the engineering priority should be to maintain the optimum light condition both during the design and scale-up of PBR.

- Richmond (2000)[53]: Intermittent illumination is without exception mandatory for effective utilization of strong light...

- Janssen *et al.* (2002)[19]: ...The scalability of the PBR described has ranged from modest to poor.

- Wu X., Merchuk J.C. (2002)[64]: ...Since the light history of the cells is controlled by fluid dynamics, a description of the fluid dynamics in the bubble column bioreactor is required. An optimal column diameter could be defined for a given set of operation conditions.

- Hu-Ping Luo *et al.* (2003)[26]: ...Although substantial work exists on the culturing of phototrophs, both flow patterns and their effects on the overall performance of PBRs remain unclear.

Hence, it became obvious that first of all I needed to describe and decouple the three coupled phenomena: (i) the biochemical process, (ii) the flow field of algae in suspension, and (iii) the radiant light energy transfer and distribution. Only after that, the problem of optimisation of photobioreactor design parameters and operating conditions could be resolved.

1.3 Introduction to the state-of-the-art of PBR modelling

First of all, I will restrict the term *PBR Modelling* to the activity which consists of the construction of a system of algebraico-differential equations governing the problem of algal growth in a general PBR. The PBR structure and dimensions enter as the boundary conditions for the system of PDE or ODE. In this sense, neither the curve fitting e.g. the fitting of relation *biomass concentration* vers. *time* for the experimental data measured in a determinated PBR, nor the theoretical derivation of a scale-up criterion (see e.g. [32], [33] and [34], where the scale-up of tubular PBR is studied) is the subject of our interest. Nevertheless, I do not state that this kind of 'modelling' is useless.

Although the purpose of mathematical modelling is very clear and very attractive: an accurate model with predictive capacity would be the marvelous tool for the PBR design, for the optimisation of the PBR operating conditions, and for the PBR control and state (e.g. biomass concentration) estimation in real time, only few attempts to model microalgal growth in PBR are reported. Excluding the kinetic models, which are unsuitables for their limited capacity to model the dynamic behaviour of the system, I chosed for my purposes the so-called three-state model of photosynthetic factory (PSF), proposed by Eilers and Peeters (1988) [11]. The same model of PSF was employed in the works of Prof. Merchuk and co-workers (see e.g. [63], [64], [65], [66]). The direct application of PSF model in PBR modelling resides in the possibility of algal cell trajectories (or pathlines) description. Knowing the trajectories of individual algal cells, the *irradiance history* (i.e. the time course of irradiance perceived by cell) could be determined. Consequently, the cell growth has to obey the relations (8, 9, 10).

There is a research team, which follows this way (see [26], [27]). The main difficulty is the determination of irradiance histories for billions of cells in relation to the hydrodynamical parameters. In [26], this problem is solved by the CARPT (computer-automated radioactive particle tracking) technique. The effects of the biomass concentration, reactor geometry, and the aeration rate on the irradiance patterns are discussed. The authors of paper [26] state that their results demonstrate that the CARPT technique is promising for PBR analysis. Nevertheless, the CARPT technique is very interesting device, it supposes that the experimental reactor or its physical model in laboratory scale is already built. I mean that the 'philosophy' behind this approach is always the *scale-up* methodology of PBR design.

An other attempt to receive the time course of irradiance for the equations of PSF model was presented in [66]. The cells movement is supposed to be perfectly organised, and the temporal irradiance patterns are obtained by coupling the cells trajectories and the irradiance distribution.

An other way to couple the algal cell trajectory with the irradiance distribution in PBR, is by using a CFD code for trajectory (or pathline) calculation. In [9], which is the work of recently graduated student of Czech Technical University in Prague, Faculty of Mechanical Engineering, a tubular PBR is divided into the light and dark volume, then the commercial CFD code FLUENT provides the coordinates of several hundreds of particles while passing through the system, and thereafter a special program makes statistical description of time interval spent by algal cells in each zone without interruption.

The results in stochastic form of irradiance history, adapted from CFD code runs, could be employed as the stochastic input variable into the deterministic model of PSF. Although this problem can be readily solved, in some moment in past, I rejected this proceeding, and I started to prefer the so-called *multicompartment/CFD* approach. While the *multicompartment/CFD* approach harmonically combines with the main result of my thesis,[1] the Lagrangian approach is

[1] I.e.: The resulting specific growth rate μ in certain volume in the culture is approaching

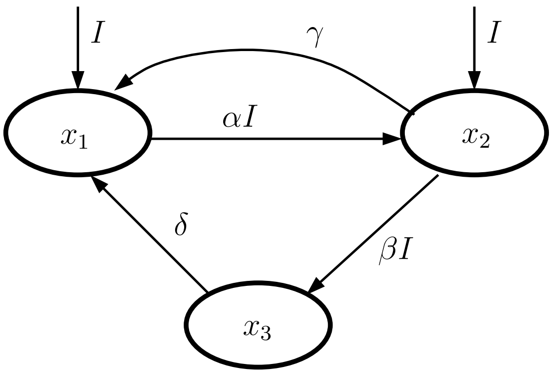

Figure 2: States and transition rates of a photosynthetic factory (PSF) in Eilers and Peeters model. The three states of a PSF are: x_1 resing state, x_2 activated state, x_3 inhibited state.

not so appropriate in point of view of PBR design. Against, while the separation between the description of the irradiance profile and algal suspension flow field (there are only flow rates among compartments which count) is relatively easy, the decoupling of *irradiance history* and fluid pattern is not obvious. Thus, for the determination of the influence of geometric parameters on *irradiance history*[2] the repeated runs (e.g. in an optimisation procedure) of CFD code have to be performed.

Resuming, in the next, only the *multicompartment/CFD* approach, which shows the synergic effect linking the property of a well mixed unit (or compartment) with the more simple description of the irradiance field by the compartment average irradiance, will be polished.[3]

1.4 Three-state model of photosynthetic factory

Eilers and Peeters presented in 1988 [11] an improvement of a model developed earlier by Crill (1977) [5]. It also had strong connection to the proposal by Kok (1965) [23].

The authors of the original paper [11] works with the probabilities that a PSF is in one of the states x_1, x_2 or x_3 (represented by the variables P_1, P_2 and P_3). The PSF can only be in one of these states, so:

(while the extent of mixing in the corresponding compartment is growing) to the limiting value, which only depends on average irradiance in the compartment.

[2]The proper term *irradiance history* is not defined unambiguously, and even more: how to define the frequently used term *light regime* or *Light/Dark cycles*, when the light distribution is continuous?

[3]Nevertheless, for the identification of the relative flow rate in the case study, the value of mean residence time which spent an algal cell in the compartment will be needed. Thereupon, this value can be calculated from residence time distribution based on above mentioned method applying Lagrangian formulation.

$$P_1 + P_2 + P_3 = 1 \tag{1}$$

From the possible transitions folows (see Fig. 2):

$$\begin{bmatrix} \dot{P}_1 \\ \dot{P}_2 \\ \dot{P}_3 \end{bmatrix} = \begin{bmatrix} 0 & \gamma & \delta \\ 0 & -\gamma & 0 \\ 0 & 0 & -\delta \end{bmatrix} \begin{bmatrix} P_1 \\ P_2 \\ P_3 \end{bmatrix} + I \begin{bmatrix} -\alpha & 0 & 0 \\ \alpha & -\beta & 0 \\ 0 & \beta & 0 \end{bmatrix} \begin{bmatrix} P_1 \\ P_2 \\ P_3 \end{bmatrix} \tag{2}$$

For given values of the parameters and irradiance I, the equation system (2) is a system of linear (*if the time course of I is given*) differential equations with constant (*if the photoacclimation does not count*)[4] coefficients, that can be solved explicitly be classical means.

The steady-state solution of the equation system (1) and (2) could be calculated from the algebraic equations resulting from the system (2) after vanishing the derivatives. The result is:

$$\bar{P}_1 = \frac{\delta\ (\gamma + \beta I)}{\alpha\beta I^2 + \delta\ (\alpha + \beta) I + \gamma\delta} \tag{3}$$

$$\bar{P}_2 = \frac{\delta\ \alpha I}{\alpha\beta I^2 + \delta\ (\alpha + \beta) I + \gamma\delta} \tag{4}$$

$$\bar{P}_3 = \frac{\alpha I\ \beta I}{\alpha\beta I^2 + \delta\ (\alpha + \beta) I + \gamma\delta} \tag{5}$$

The rate of photosynthetic production (μ) is proportional (there is a proportional constant κ) to the number of PSFs (N) and the number of transition from x_2 to x_1:

$$\mu = \kappa\gamma\bar{P}_2\ N = \frac{\kappa\ \gamma\delta\ \alpha I\ N}{\alpha\beta I^2 + \delta\ (\alpha + \beta) I + \gamma\delta} \tag{6}$$

Equation (6) gives the relation between irradiance and production (growth) rate in the steady state. Only a short moment is needed to realise, that the steady state growth kinetics is of Haldane type or *Substrate Inhibition Kinetic*. Only three of the six parameters ($\kappa, N, \alpha, \beta, \gamma, \delta$) could be estimated from the steady-state production curve ($P - I$ curve). To estimate the other parameters we need a dynamic measurements. The authors Wu and Merchuk in [63] used the measurement of the chlorophyl fluorescence parameters for the estimation of the remaining parameters for the red marine microalga *Porphyridium* sp. (see Fig. 3).

[4]Having an on-line measurement technique to the model parameter estimation, the time dependency of the constants of PSF model can be naturally included.

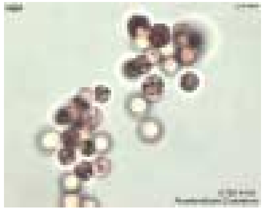

Figure 3: Red marine microalga *Porphyridium* sp. Reprinted from website http://www.bio.utexas.edu/.

1.5 Further improvements of three-state model of PSF

In the second and last paper (until now) of Eilers and Peeters on PSF [12], the authors slightly changed the interpretation of concept of PSF and the dynamic behaviour of the model were examined. The probabilities of transitions (the variables P_1, P_2 and P_3) were changed to the fractions of the total number of PSFs (represented by the variables x_1, x_2 and x_3) that are in the respective states 1, 2 and 3. The number of PSFs for calculating an overall production rate (see Eq. 6) was comprehended into the constant κ (see Eq. 10). Hence, an equivalent system of ordinary differential equations (ODE) were obtained (see Eq. 8).

Among others, in their last paper Eilers and Peeters satisfactorilly explained some experimental observation (differences between short and long incubations, the effect of intermittent illumination, the influence of prior illumination and hysteresis-effects). After that, many authors employed their concepts.[5] However, in the photobioreactor modelling only Wu and Merchuk (see [63], [64], [65]) and recently Hu-Ping Luo and Muthanna H. Al-Dahhan in [27] and myself, employed the same model.

Wu and Merchuk in [63] introduced to the PSF concept some innovation, which will be shown in the next sub-subsection. The novelty of my approach consists in the employment of the PSF model for the succesfull theoretical explanation of flashing light experiments, and subsequently (having good reason for doing this), the PSF model were implemented in the distributed parameter model of algal growth in PBR.

1.6 Wu and Merchuk's improvements of three-state model of PSF

In 2001 Wu and Merchuk published the paper entitled *A model integrating fluid dynamics in photosynthesis and photoinhibition processes*, see [63]. This model, schematically presented by the same Fig. 2, had an additional modification, which allows the negative production rate (see the equation 10).

Wu and Merchuk explain the concept of the photosynthetic factory in [63] as follows: The PSF is defined as the sum of the light trapping system, reaction centers and associated apparatus, which are activated by a given amount of light energy to produce a certain amount of photo-product. The three possible states

[5]There are 75 citations in the citation database Web of Science, until now, June 2005.

are assumed for the PSF: the resting state (open) x_1, the activated (closed) state x_2, and the inhibited state x_3. The process of culture growth can be summarized as the result of four sub-processes (each probably encompassing a number of steps) occuring simultaneously:

1. Photon capture starting the chain of biochemical reaction and leading to biomass synthesis. In terms of PSF, it would be indicated by $x_1 \rightarrow x_2$, and occur only during the light period.

2. Initiation of the chain of dark reactions, $x_2 \rightarrow x_1$, the light is not need to proceed.

3. Reversible loss of photon trap activity due to high PFD (photoinhibition), which in terms of the PSF model is indicated as $x_2 \rightarrow x_3$, and occur only during the light period.

4. Photon trap recovery, that can be indicated as $x_3 \rightarrow x_1$, the light is not need to proceed.

The observed growth rate is the result of the integration of all the processes described above over the history of the culture. As mentioned above, the original model of Eilers and Peeters did not respect the facts observed in the experiments of Wu and Merchuk that biomass decreases at weak light. To take this fact into consideration, a term that represents the maintenance process[6] was added. Wu and Merchuk accepted the proposition of Lee and Pirt in [24] and the maintenance process, which involves thousands of enzymatic reactions, was modelled by a negative constant (see Eq. 10). This gross assumption is usually acceptable if the biological material is in the growth phase, and no secondary metabolite is being produced. However, if a growth-associate secondary metabolite is produced, the model can still be valid.

According to the description above, the equations can be written as follows, expressing the dynamic process of the transition of PSFs within the three states, and the production rate of biomass.[7]

$$\begin{bmatrix} \dot{x}_1 \\ \dot{x}_2 \\ \dot{x}_3 \end{bmatrix} = \begin{bmatrix} -\alpha I & \gamma & \delta \\ \alpha I & -\gamma - \beta I & 0 \\ 0 & \beta I & -\delta \end{bmatrix} \begin{bmatrix} x_1 \\ x_2 \\ x_3 \end{bmatrix} \tag{7}$$

The system matrix in (Eq. 7) can be also splitted into two parts, the first constant, and the second part depending on irradiance. This form will be preferred in the next. The reasons are two: (i) the splitting form has the form of a general

[6]The energy for internal metabolism is needed!

[7]Note that the labels of respective states (i.e. $x_1,\ x_2,\ x_3$) are 'quantified' by the authors of [63], i.e. are used as a scalar quantity corresponding to the respective ratio of concentration of open, closed or inhibited state to the overal cell concentration.

bilinear system, (ii) that of more simple manipulation in case of application of PSF model in PBR modelling, when different compartments with different values of irradiances are treated together.

$$\begin{bmatrix} \dot{x}_1 \\ \dot{x}_2 \\ \dot{x}_3 \end{bmatrix} = \begin{bmatrix} 0 & \gamma & \delta \\ 0 & -\gamma & 0 \\ 0 & 0 & -\delta \end{bmatrix} \begin{bmatrix} x_1 \\ x_2 \\ x_3 \end{bmatrix} + I \begin{bmatrix} -\alpha & 0 & 0 \\ \alpha & -\beta & 0 \\ 0 & \beta & 0 \end{bmatrix} \begin{bmatrix} x_1 \\ x_2 \\ x_3 \end{bmatrix} \tag{8}$$

$$x_1 + x_2 + x_3 = 1 \tag{9}$$

$$\mu = \kappa\gamma x_2 - Me \tag{10}$$

where α, β, γ and δ are rate constants of PSF model (see Fig. 2), κ is the photosynthesis yield and Me is the maintenance term considering endogenous metabolism.

Now I will add the last remark of this subsection concerning the attractivity of the PSF model: Is worth to note that the term $\kappa\gamma$ is of the order 10^{-5} [s^{-1}], while the states of the PSF model reach the values between 0 and 1 [-]. By this way, the transition[8] from time scale of light/dark cycling and photosynthesis time constants in a basic level (time micro-scale) to time scale of biomass growth (time macro-scale) is effectuated without loss of accuracy. This is the main reason why to use the *state description* instead of the 'classical' description by means of biomass concentration or cell density. Further I will underline this verity, explaining why the growth simulation via c_x description could neither account with the transport by convection, nor by dispersion. The other interesting property of the solution of ordinary differential eqution system (8) for the model parameters corresponding to red marine microalga *Porphyridium* sp., and for some 'reasonable level' of irradiance (e.g. I_{opt}= 250 μE m^{-2} s^{-1}), is that the eigenvalues of the matrix A of the system of linear ODE (i.e. time constants of the process) are also different of several orders. This fact classifies the system as **stiff**.

[8] More impressive expression could be: *the scale jump*.

2 Objectives of the thesis

Research and Development in the field of algal biotechnology and photobioreactor design has definitelly **multidisciplinary** character. First of all, while describing and modelling the biological process, we deal with the biological system. In my case, the **biological system** is microalgal mass culture. The biological system comunicates with its environment, which has some physical and chemical properties. Thus, the description of these physical and chemical properties must be done. Only two most relevant physical phenomena for microalgal growth, i.e. the **radiant light energy transfer and distribution**, and **fluid pattern** of algal cells in dilute suspension, are involved. Neither the cell damage due to the hydrodynamic (shear) stress, nor the mass transfer phenomena (nutrient and CO_2 supply, photosynthetic O_2 accumulation) are taken into account. The other discipline – **mathematical modelling** – covers the mathematical formulation of the problem, numerical methods and their implementations.

From this brief summary of disciplines and tasks involved in photobioreactor modelling, the objectives of my thesis are derived:

1. Unification and elucidation of the terminology, methods and theoretical and experimantal approaches, providing from different disciplines involved in PBR modelling and design. The particular aim of this item was to *decouple the principal phenomena influencing algal growth in PBR*.

2. Formulation and development of *Lumped parameter model of microalgal growth*. The particular aim of this item was to explain the right role of the light regime parameters, i.e. (i) the frequency of light/dark cycles and (ii) the ratio between average and incident irradiance, by means of mathematical analysis. The consequent particular aim was to reconcile and explain some different results of two papers about the quantitative description of the effects of intermittent light on microalgal growth (see: [60], [36]).

3. Formulation and development of *Distributed parameter model of microalgal growth*. The special aims of this item were the PBR spatial discretisation, according to the non-uniform irradiance distribution across the PBR and the non-uniform flow regime, and the choice of such a model arrangement that the multi-scale problem of the algal cell growth actually could involve all the phenomena in different time and spatial scales.

4. *Case study - Simulations of microalgal growth in a PBR.* In this point, a simple case study to test the model properties was thought, aiming to elucidate the methodology of the optimisation of PBR operating conditions and design parameters, based on mathematical modelling and numerical simulations.

3 Methodology used

The theory of photosynthetic microorganisms growth modelling has long been regarded as a well-defined discipline in algal biotechnology, consisting of the adequate coupling between photosynthesis and irradiance, resulting in the light response curve (so-called P – I *curve*) which represents the microbial kinetics. However, several phenomena, e.g. flashing light enhancement, can not be explain by a simple kinetic relation. The main difficulty in considering the role of light/dark cycles induced by algal suspension flow in PBR consists in different time scales of both processes. While the characteristic time of algal growth is in order of hours, the transport of cells from light to dark zone and viceversa occurs generally in seconds.

Nevertheless, a simple dynamic model of photosynthetic factory (PSF), proposed by Eilers and Peeters (1988) [11], has proved to be an effective means to model all relevant phenomena. Firstly, the lumped parameter model of microalgal growth was derived in the form of: $\mathrm{d}x/\mathrm{d}t = Ax + uBx + Cu$, i.e. the bilinear system, which is linear in state x (state vector x has three components representing three states of a photosynthetic unit), linear in control variable u (single scalar input u represents the irradiance in the culture), but not jointly linear in both. Thereafter, the solution of boundary condition (Dirichlet) problem was resolved. Inspite of the fact that the rate constants among the states of the PSF model, which characterise the growth and the photoinhibition, are different of several orders (the system is stiff), the results of microalgal growth obtained by numerical simulations are in good agreement with the experimental work of Nedbal *et al.* (1996) [36]. In addition to the numerical results obtained for a specific microorganism, a general conclusion is presented. This result is based on the Lipschitzean dependence of trajectories of bilinear systems on control (see the work of Dr. Čelikovský (1988) [8]). The application of special control signal, which consists of periodic switching between two constant values, leads to the elucidation of the right role of so-called light-regime parameters, i.e. (i) the frequency of light/dark cycles and (ii) the ratio between average and incident irradiances.

Only an apparent contradiction appears when the Terry's concept of *'Photosynthetic Efficiency Enhancement'* is regarded (see Terry (1986) [60]). In fact, the Terry's concept has to be corrected (for details see Chapter 4 of my thesis).

Because of the non-uniform irradiance distribution across the PBR, the whole system, i.e. PBR, has to be treated as a distributed parameter model. The so-called multicompartment/CFD approach provides a natural framework for its spatial discretisation and governing equations derivation. More specifically, the reactor is divided into a limited number of compartments, and for each compartment an ordinary differential equation based on balance equation is derived. The biological process dynamics, i.e. the source term in the mass balance equation, is described by PSF model, and the flow rates across the compartment borders can be delivered by a CFD code. Consequently, the spatial averaging of the so-called closed state of PSF model is made every time step (i.e. in time micro-scale), and then the time averaging of PSF closed states is performed. The resulting growth in time macro-scale is proportional, according to the PSF model, to the spatial and time averaged value of PSF closed states.

Finally, two case studies are presented. In the first case study, the optimal operating conditions for the flat panel bioreactor are derived analytically. Afterwards, the optimisation of a design parameter, based on numerical simulations is carried out. In the second case study, the model of microalgal growth in photobioreactor with Fresnel lenses is developed and prepared for the further examination.

By this way, putting together mathematical modelling, numerical simulation and optimal design, the main goal of this work: i.e. *the proposal of a 'novel' methodology for microalgal bioreactor design* is accomplished.

4 New results

In this section the most important results of my thesis are presented after brief introduction to the bilinear systems.

4.1 Bilinear systems with single input

Let us consider the following control system which we shall call the bilinear system with single input (BLSSI):

$$\dot{x} = Ax + (Bx + c)u, \quad x(t_0) = x_0, \tag{11}$$

where A, B are $(n \times n)$-dimensional constant matrices and c is vector in R^n space. Scalar control u is assumed to be a measurable function on every finite time interval $[t_0, t_1]$ such that almost everywhere on $[t_0, t_1]$ it holds

$$u_{\min} \leq u(t) \leq u_{\max},$$

where $u_{\min}, u_{\max}$ are given real numbers. Such a control is further denoted as an admissible one. Finally, $x \in R^n$ is the vector of state variables and $x_0 \in R^n$ is the given initial state of the system.

More general forms of bilinear systems are described in [31]. The following estimate for this continuous dependence was obtained in [8]:

$$\max \; \| x(t) - y(t) \|_{R^n} \; \leq K \; \max \; \left| \int_{t_0}^{t} (u_x(s) - u_y(s)) ds \right|, \tag{12}$$

where $x(t)$ and $y(t)$ are solutions of (11) for $u_x(t)$ and $u_y(t)$ respectively, $t_0 \leq t \leq t_1$ and K is a constant depending only on $A, B, c, t_0, t_1, u_{\min}$, and $u_{\max}$. Representation of trajectories of system (11) and estimate (12) are used in order to study some important properties of the so-called attainable set of bilinear systems.

4.2 Dependence of state trajectories of bilinear system on intermittent input signal

In this subsection I formulate Lemma 2, which is the most important theoretical result concerning the further application in microalgal biotechnology. Lemma 2, could be directly employed for the judgement about right role of the parameters of light/dark cycles (i.e. frequency and light-to-dark ratio) for photosynthetic production rate during flashing light experiments.

Lemma 2. Let us consider a constant function u_0 measurable on a closed interval

$[t_0, t_1]$ such that $u_0 \in [u_{\min}, u_{\max}]$, on $[t_0, t_1]$. Let us divide closed interval $[t_0, t_1]$ into k closed equal subintervals $[t_0 + (i-1)h, t_0 + (i-1)h + h_a]$, and into k closed equal subintervals $[t_0 + (i-1)h + h_a, t_0 + ih]$, $i = 1, 2, \ldots, k$, $h = (t_1 - t_0)/k$, and let $u_0 = \frac{h_a}{h} u_a + \frac{1-h_a}{h} u_b$, $0 < u_a < u_b$, $u_a \in [u_{\min}, u_{\max}]$, $u_b \in [u_{\min}, u_{\max}]$.

Then there exists a piecewise constant function $u^{**}(s)$ with the following properties:

1) $u^{**}(s) = u_a$ on each subinterval of the form $[t_0 + (i-1)h, t_0 + (i-1)h + h_a]$,
2) $u^{**}(s) = u_b$ on each subinterval of the form $[t_0 + (i-1)h + h_a, t_0 + ih]$,
3) for all $t \in [t_0, t_1]$

$$\left| \int_{t_0}^{t} u_0 \, \mathrm{d}s - \int_{t_0}^{t} u^{**}(s) \, \mathrm{d}s \right| \le (u_\mathrm{b} - u_\mathrm{a}) \frac{h_a(1-h_a)}{h}. \qquad (13)$$

Proof. The proof is performed in Section 4.4 of my thesis.

Remark. Lemma 2 – together with estimate (12) – not only establishes that the state trajectory of a bilinear system for a constant control signal u_0 can be approximated by the state trajectory corresponding to the intermittent control signal u^{**}, with an arbitrary previously defined precision, but even induce an important idea resulting in a simple strategy for optimal light regime finding (let see the next Subsection).

4.3 Optimal light regime proposal

Until now the direction of use of the Lemma 2 pointed from the intermittent input signal to the continuous one. For the specific reason of the optimisation of light regime parameters, the implications can be inverted. The reasoning is simple: If, with growing frequency of light/dark cycles, the right side of relation (13) tends to zero, and consequently the state trajectory x^{**} tends to the state trajectory x_0, we could work, while searching optimal light regime parameters, with more simple class of constant input signals, and then applicate the results to the intermittent light regime. Always when the *sufficiently high* frequency of light/dark cycles may be assured.

By this way, a dynamical problem is transformed to the static one. For the specific situation of the PSF model, the optimal, i.e. maximal, growth takes place when the irradiance has the value $I_{opt} = \sqrt{\frac{\gamma\delta}{\alpha\beta}}$ (see Subsection 4.2.1 of my thesis).

4.3.1 Optimal light-to-dark ratio determination

In order to extend the applications of the above mentioned reasoning, the so-called light/dark cycles will be generalised to a piecewise continuous function I^{**}, for

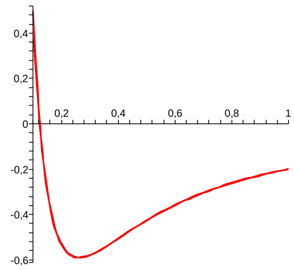

Figure 4: The dependency of the derivative of specific growth rate respective to ratio of average to maximal irradiance ϕ_b (while the $\phi_a = 0$). The frequency of Light/Dark cycles was 1 Hz. Both numerical simulations and plot were performed in Maple.

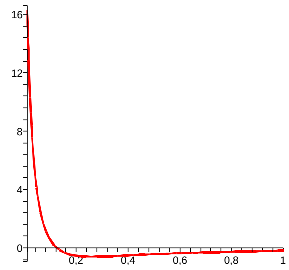

Figure 5: The dependency of the derivative of specific growth rate respective to ratio of average to maximal irradiance ϕ_b (while the $\phi_a = 0$). The frequency of Light/Dark cycles was 2 Hz. Note that both the optimal value of ratio ϕ_b and the parameter sensitivity are very different than where the frequency was 1 Hz.

the similar time course as described in Lemma 2: $0 < I_a < I_b$, $I_a \in [I_{\min}, I_{\max}]$, $I_b \in [I_{\min}, I_{\max}]$. Let us introduce the following notation:

$$\phi_a = \frac{h_a}{h} \tag{14}$$

and

$$\phi_b = \frac{1 - h_a}{h} \tag{15}$$

Note that $\phi_a + \phi_b = 1$. The cycle frequency ν is the inverse value of h.

Then, based on the Lemma 2, for the optimal performance of the photosynthetic factory, the next relation must be accomplished:

$$I_{opt} = \phi_a \ I_a + \phi_b \ I_b \tag{16}$$

The equation (16) represents a simple algebraic relation for finding the optimal ratio ϕ_a (and consequently $\phi_b = 1 - \phi_a$).

The interesting problem arise when the sensitivity to the changes of ratii ϕ_a and ϕ_b is investigated. The next two figures document the dependency of the derivative of specific growth rate respective to ratio ϕ_b (while the $\phi_a = 0$). The frequency of Light/Dark cycles is 1 Hz in Fig. 4, and 2 Hz in Fig. 5.

4.3.2 Minimal frequency determination

The role of frequency of intermittent (or flashing) light on the photosynthetic efficiency was, I mean, misunderstood and partly exaggerated in the past. Definitely,

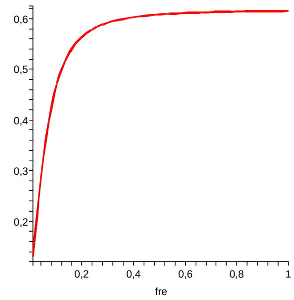

Figure 6: The dependency of the specific growth rate on the frequency of intermittent light respective to ratio ϕ_b (while the $\phi_a = 0$). The ratio $\phi_b = I_{av}/I_0$ is 1/10. The numerical simulations were performed in Maple.

there is not an optimal mysterious frequency for which the photosynthetic efficiency jumps to some extreme value. Nevertheless, some comments to the limits of frequencies interval should be done. The lower limit could not be zero, which corresponds to the infinitely large period and 'theorically' represents a continous light regime. This fact produces some mishmash in the numerical calculations.

Equally, the upper limit could not be infinitely large which *actually* represents a continous light regime, but taking into account the functional properties of the PSU, only periods comparable to the PSII turnover time can be used efficiently. Thus, the biggest frequency for the numerical calculations can be set up to 50 Hz.

The dependency of the specific growth rate on the frequency of intermittent light respective to ratio ϕ_b (while the $\phi_a = 0$) is drawn in the next figure.

An interesting and practically important question can be presented, viewing the figure 6: *What is the minimal frequency to ensure e.g. the 90% value of maximal growth rate corresponding to the actual ratio ϕ_b?* The solution of this problem can be readily solved numericaly, but there are also the analytical tools residing in the results of Section 4.5 of my thesis. This analysis is conceived to be effectuated in the near future.

Remark. Performing the numerical simulations for different combinations of light regime parameters was very exciting way to effectuate numerically the *flashing light experiments*. I very enjoed this activity facilitated by the MAPLE programming environment. The graphical results clearly showed: (i) the non-existence of an extreme value of specific growth rate for a particular value of frequency of L-D cycles, (ii) the value of ratio of specific growth rate for intermittent light regime (determined by h_a, h_b, u_a, u_b) to the specific growth rate for constant irradiance I_{av} ($I_{av} = 1/h \cdot [h_a u_a + h_b u_b]$) was growing for the growing frequency (or for disminishin h), but has *the limit superior equal to 1 !!*

This finding suprised me six mounth ago, but now I see that every experimental and theoretical result pointed to this *resumé*, just mentioned in two above statements. The anallytical proof and some theoretical explication of the general phenomena, which can be called the *dependence of state trajectory of bilinear system on input signal*, is presented in detail in Section 4.4 of my thesis.

4.4 Multicompartment/CFD approach

As stated before, a critical issue in the modeling of photobioreactors is the close interaction between fluid flow and the biochemical reactions (i.e. photosynthesis). In particular, light regime has a large effect on the corresponding local specific growth rate which, in turn, affects the productivity. In this chapter theoretically and in the next chapter practically, I demonstrate how a multicompartment/computational fluid dynamics (CFD) modeling approach can be applied to take account of these interactions (see also [1], [2], [45], [46]). The main advantage of the multicompartment/CFD approach is the division of one big problem (reaction and transport) in two simpler. Then the fluid flow is solved separately (and more specificaly) from the process dynamics. The approach to multicompartment modelling characterises the flow rates between adjacent zones. These can be calculated by means of steady-state CFD calculations. Also the other fluid mechanical quantity, such as the shear stress, that have important effects on the growth of microalgal cells (e.g. on the filamentous cyanobacteria *Spirulina*), can be determined within each compartment.

A structured model of PSF for biomass production in a continuous photobioreactor is used for our purpose. After the spatial discretization, the system of ODE on the basis of (Eq. 17) can be written. Therefore, the system can be solved both analytically and numericaly. The analytical solution, allowing the deeper comprehension of the interrelated phenomena, is presented in the case study in Section 7.1 of my thesis. The numerical solution, which is necessary in more complicated cases, is discussed in Section 7.2 of my thesis.

4.4.1 Spatial discretisation of PBR culture volume

The simple modular principle of spatial discretisation of PBR culture volume is the main advantage of the multicompartment approach.

The basic idea behind the spatial discretization, is the fact that the size of compartment volume can reflect the peculiarity of the process, i.e. the compartment volumes can be of several orders bigger that its for fluid flow calculation.

Also the other parts of the process unit, i.e. pumping device, degasser (or retention tank), heat exchanger, etc., can be naturally included into the system. The graphical scheme of a general photobioreactor spatial discretisation is shown in Fig. 7.

4.4.2 Governing equations of PBR model

The system of ordinary differential equations (Eq. 17), for $i = 1, .., n \cdot m$ (where $n \cdot m$ is the total number of compartments), then the initial condition (IC), the boundary condition (BC), and the model constants and parameters (which can change in time!), represent the mathematical formulation of the problem of microalgal growth in a general PBR. Let us consider again the (Eq. 17), with specific

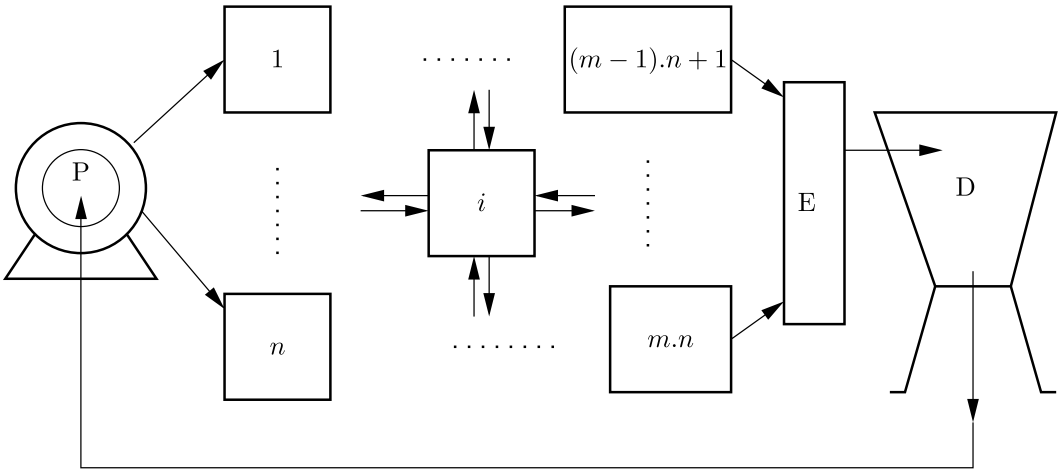

Figure 7: Scheme of spatial discretisation of a photobioreactor with recycle.

IC corresponding to the long time of incubation in dark , and BC corresponding to the respective mode of cultivation.

$$V_i \frac{\mathrm{d}c_i}{\mathrm{dt}} = \dot{V}\left[\sum_{j=1}^{N_i} c_j\ f_{ji} - \sum_{j=1}^{N_i} c_i\ f_{ij}\right] + V_i\ R_{c_i} \quad , \quad i \in 1, 2, ..., N_c \tag{17}$$

where R_{c_i} is defined e.g. by (Eq. 8, 9, 10).

IC (after long time in dark):

$$(c)_i(t_0) = (c_x)_i \begin{bmatrix} x_1(t_0) \\ x_2(t_0) \end{bmatrix}_i = \begin{bmatrix} 1 \\ 0 \end{bmatrix} \tag{18}$$

BC-BOM (batch operation mode): BC corresponding to the flowrates from/to outside of the system does not exists, the PBR is closed in sense of the material transport, the total flowrate $\dot{V}$ is the model parameter.

BC-COM (continuous operation mode): (i) The flowrate of the fresh medium at the inlet to some compartment (usually to the retention tank), (ii) the flowrate of the mixture (algal suspension) at the outlet of the PBR photic zone .

BC-FBM (fed-batch operation mode): The addition of the fresh medium in discrete instances.
The mathematical formulation has to be completed (closed) by the algebraic equation which determines the level of irradiance in each compartment (its average value, according to the main result of the section 4.4 of my thesis) as function of operating conditions (biomass concentration c_x and incident irradiance I_0). This problem was studied in detail in Chapter 3 of my thesis.

A peculiarity of the PSF model, intensively analysed in Chapter 4, consists in fact that the concentrations in the (Eq. 17), are state vectors with three components. Moreover, the bioreaction is modeled by the system of ODE in which all components are involved (see e.g. Eq. 8). Because the three components are linearly dependent, one state can be eliminated, and consequently the system of ordinary differential equations (Eq. 17), for $i = 1,..,n \cdot m$, actually represents $(2 \cdot n \cdot m)$ equations for $(2 \cdot n \cdot m)$ variables.

Very important remark 1.
By the system of ODE (17) time courses of the concentrations in each of N_i compartments are described. Let us now analyse what happens if concentration c were biomass concentration. According to the assumption accepted in the beginning of this chapter, the c_x is uniformly distributed along the PBR. Moreover, the order of specific growth rate μ is of 10^{-5} s, thus, assuming for an instant and for a small region in PBR the constant value of specific growth rate, we get

$$c_x(t + \Delta t) = c_x(t) \exp(\mu \Delta t) \tag{19}$$

which means that $c_x(t + \Delta t) \simeq c_x(t)$, for Δt comparable with characteristic time for cell transport between adjacent compartments. Afterwards, in the Eq. (17) the term c_x can be extracted from the sum. For the resting term, according to the continuity equation, holds

$$\sum_{j=1}^{N_i} (f_{ji} - \ f_{ij}) = 0 \tag{20}$$

which means the loss of sensivity of the whole process to the mixing.

In the literature, I have seen two solutions of this problem (in the modelling of algal growth in a PBR): (i) to declare that the radial mixing is negligible, and proceed with the simulation,[9] (ii) to adopt the Lagrangian approach.
In my thesis I selected the *third way*: the state description of the process (by the three-state model of PSF), where the states are very sensitive to the light gradient – characteristic times are in order of seconds and minutes (photoinhibition), and consequently the strong differences in concentrations of state components $(c_1,\ c_2,\ c_3)$, make meaningful the system of ODE 17. The time and spatial average of the state x_2 is then multiplied by a constant ($\kappa\ \gamma\ \simeq 5 \cdot\ 10^{-5}\ \mathrm{s}^{-1}$) which in fact represents the *scale jump* from PSF microscale to our human macroscale.

Very important remark 2.
The system of ODE 17, when the concentration c means the concentrations of a

[9] However there is the second stumbling block: viewing the equation 19, the time step Δt has to be suficiently big for breaking the relation $c_x(t + \Delta t) \simeq c_x(t)$ (in certain paper I have seen the time step Δ t=3600 s !!

state components c_1 or c_2 (c_3 is linearly dependent and be calculated whatever it wants), the reaction term involves the other state too. For that reason it seems the number of equations must be doubled. The other solution of this state interdependencies is the numerical method based on operator splitting. This topic is discussed in Chapter 6 of my thesis.

4.4.3 Analytical solution

The solution of the ODE system (Eq. 17) is readily available in simple cases, because the system (Eq. 17) represents the system of linear ODE. Moreover, for the continuous operation mode, the coefficients of the system matrix are constants. The analytical solution of this type of problem is presented in the case study in Section 7.1 of my thesis.

The advantage of the analytical solution is the straightforward possibility to optimise some operating conditions, e.g. the biomass concentration and the incident irradiance (in continuous operation mode). The optimisation of PBR design parameters is also more simple having a solution in a closed form.

For a big system, or for complicated or time dependent BC, it is preferable to obtain the solution numericaly, e.g. by the application of *operator splitting* (OS) method, see e.g. [25] and [17]. Although I did not conclude the numerical simulations of microalgal growth in such a complex system as the PBR with Fresnel lenses, I will briefly explain the idea of OS method in the next subsection.

4.5 Resumé

The original results of my thesis are enumerated as follows:

1. By the literature research, the elucidation of the terminology, methods and theoretical and experimantal approaches was achieved. Different disciplines involved in PBR modelling and design were firstly decoupled and then harmonicaly unified for serving to the common goal: elaboration of the optimal PBR design methodology. As a by-product, several serious problems in PBR modelling were detected: (i) the non-reliability of the PBR scale-up methodology, (ii) the difficult modelling of the microalgal growth in point of view of multiscale processes (i.e. fast transport process and slow algal growth), (iii) the non-existence of a structured model explaining the flashing light experiments, and further the right role of light regime parameters (i.e. frequency and ratio of light/dark cycles, and average value of irradiance), (iv) the non-existence of a structured model with distributed parameters, which could be used for growth simulations.

2. Development of *PSF model of microalgal growth*. The so-called model of photosynthetic factory was successfuly used to explain the right role of the synergic role of light/dark cycles frequency and the ratio between average

and incident irradiance. By means of functional analysis of a general bilinear system, the real importance of light regime parameters was declared by the next statement: "The resulting specific growth rate μ in certain volume of the algal culture is approaching to the limiting value when the extent of mixing in the corresponding compartment is growing. This value only depends on average irradiance in the compartment (is equal to the specific growth rate depicted from the static *P – I curve* for corresponding value of average irradiance)".

This is the main result of my thesis. After that, the experimental results of effects of intermittent light on microalgal growth published in [60] and [36], was simulated in PSF model with good qualitative agreement. Possessing the certainty about the limits of so-called flashing light enhancement, the original Terry's concept published in [60] was corrected and reformulated. Actually, the enhancement takes place, but due to the "light dilution" in dense algal culture thanks to good mixing.

3. Development of *Distributed parameter model of microalgal growth* in a general PBR. The second main result of my thesis is the discovery that the PBR spatial discretisation, according to the non-uniform irradiance distribution across the PBR, and the discretisation represented by the well mixed units (or compartments), plays a complementary role. The determination of a model of interconnected well mixed unit must preceed to the photometric studies. Then the application of first main result of my thesis indicates what is needed to calculate: only the average irradiance inside the compartment (and the irradiance in the surface corresponding to the next compartment).

   The third main result of my thesis is the solution of the multi-scale problem of the algal cell growth. As stated before, the growth process of microalgae is sensitive to the phenomena which occur in different time and spatial scales. The states of PSF model are sensitive to the light intermittencies and the multicompartment approach cope with the spatial effects.

4. *Model testing.*

   Only a simple case study, aiming to illustrate the methodology of the optimisation of PBR operating conditions and design parameters, was performed by means of numerical simulations. Nevertheless, the graphical outputs of the numerical simulations effectuated in MAPLE shows convincently the trends already seen in flashing light experiments. Ergo, the validity of PSF model implemented into well mixed compartments was proved. Also the equivalence of Eulerian and Lagrangian approaches was confirmed (or better formulated: the model shows the correct behaviour independently of the approach selected).

5 Conclusions and outlooks for further research

The results just presented contribute to modelling and simulation of microalgal growth in a photobioreactor operating under strong irradiances. In this context, the design and perfomance of the novel photobioreactor with Fresnel lenses, here described, represents a successful practical application of theoretical concept of "light dilution".

In order to model microalgal growth, firstly I have chosen an appropriate dynamic model, so-called *three-state model of photosynthetic factory*, able to cope with the different time scales of two relevant processes: growth and transport by convection and dispersion. Theoretical analysis of PSF model led me to the analytical proof of the role of frequency and ratio of the so-called light/dark cycles on photosynthetic growth rate. The results of well known flashing light experiments mark good agreement with the numerical simulations effectuated for the corresponding conditions using the Maple programming language, which confirms the validity of the main theoretical result of my thesis: *The resulting specific growth rate (μ) in the culture is approaching, while light/dark cycle frequency or the relative flow rate is growing, to the limiting value, which only depends on average irradiance in the culture.*

The next step in PBR modelling was the extension of lumped parameters model of PSF to the distributed parameters model of algal growth in a general bioreactor, including the transport phenomena, by application of *multicompartment/CFD approach*. The resulting model of PBR was able to cope with the multi-scale problem of the algal cell growth, due to its sensitivity to the phenomena which occur in different time and spatial scales.

The idea of spatial discretisation to the so-called *well mixed units* or *well mixed compartments* apparently goes against the discretisation imposed by the heterogenity of light distribution. The reconciliation of these two different phenomena was possible thanks the result of the theoretical analysis of PSF model as a bilinear system. This complementarity of not apparently concordant phenomena was very surprising. In fact, this conclusion justify the separate solution of the photosynthetical growth, which is determined on basis of an average irradiance in the well mixed compartment, and the transport by convection and dispersion. Also for this reasons, the operator-splitting method for numerical simulation was proposed.

Finaly, two case studies for flat panel PBR and tubular PBR was formulated. In the first simple study the numerical results showed good qualitative agreement with experimental results published in literature. Hence, the expected predictive capacity of general PBR model was confirmed. This validation of model structure makes possible the further developement of design methodology based on mathematical modelling and numerical simulations.

There are many directions for further work related to the presented thesis. The *multicompartment/CFD approach* gives the overall framework for simulation of microalgal growth in a photobioreactor, independently on the form of basic growth model with lumped parameters. The detailed solution of the influence of various transport mechanisms in direction of light gradient, i.e. in the radial direction in the case of a reactor with a cylindrical geometry, on the algal growth is far to be found. Neither is the influence of shear stress, however the multicompartment/CFD approach makes possible a natural inclusion of this pnenomena into the PBR model.

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Anotace

Předložená disertační práce se zabývá matematickým modelováním a simulací procesu růstu mikroskopických řas ve fotobioreaktorech. Mezi všemi vnějšími vlivy, které působí na průběh masové kultivace mikrořas, byly do modelu zahrnuty dva nejvýznamnější, určující světelný režim v řasové suspenzi:

(i) způsob dodávání a množství světelné energie,

(ii) hydrodynamické poměry ve fotobioreaktoru.

Dále byl přijat předpoklad, že struktura fotobioreaktoru je předem dána zvoleným typem reaktoru (tj. jeho geometrií) a konstrukční parametry patří mezi parametry modelu.

Pro modelování procesu fotosyntézy s uvažováním fotoinhibice byl vybrán dynamický model, tzv. model fotosyntetické továrny (PSF), publikovaný v práci Eilers and Peeters (1988) [11]. Přijatá matematická formulace procesu fotosyntézy, tj. bilineární systém s jedním skalárním vstupem (ozářeností), umožňuje řešit problematiku optimálního řízení, řiditelnosti systému a dosažitelnosti stavů. Použitím dříve publikovaných výsledků o spojité závislosti stavové trajektorie bilineárního systému na řízení (Čelikovský (1988) [8]), bylo dokázáno, že pro intermitentní režim osvětlení se stavová trajektorie blíží k stavové trajektorii při konstantní hodnotě vstupu, jež odpovídá časově zprůměrované hodnotě intermitentního vstupu. Přitom je specifická růstová rychlost při konstantní ozářenosti limitní hodnotou specifické růstové rychlosti při intermitentním režimu (má-li ovšem průměrná ozářenost shodnou hodnotu jako ozářenost konstantní) pro rostoucí frekvenci cyklů světlo/tma.

Pro modelování růstu mikrořas v celém fotobioreaktoru, jež je systémem s rozloženými parametry, je v této práci navržen tzv. *multicompartment/CFD* přístup, který umožňuje postihnout i vliv proudění řasových buněk v suspenzi (zvl. vliv radiálního promíchávání) a vliv nerovnoměrnosti rozložení ozářenosti v příčném řezu osvětlenou sekcí reaktoru na růst mikrořas.

Po implementaci modelu PSF do jednotlivých jednotek reaktoru a při uvažování ustáleného proudění (toky mezi jednotkami závisí jen na prostorové souřadnici a na velikosti celkového průtoku) byly dosaženy následující výsledky: (i) při zanedbatelném radiálním promíchávání došlo k ustanovení rovnovážného stavu a průměrná hodnota specifické růstové rychlosti odpovídala hodnotě dané integrací lokálních hodnot specifické růstové rychlosti přes celý objem osvětlené zóny reaktoru, dále: (ii) při nezanedbatelném vlivu radiálního promíchávání (tzn. při vzniku tzv. *Light/Dark cycles*) došlo také k ustanovení rovnovážného stavu, při kterém však průměrná hodnota specifické růstové rychlosti odpovídala hodnotě odečtené z grafu kinetické závislosti růstu (a sice kinetice s inhibicí substrátem - kinetice s *fotoinhibicí*) pro průměrnou hodnotu ozářenosti (úplné promíchávání způsobuje zprůměrování ozářenosti). Výsledky simulací jsou tak ve shodě s analytickým řešením a také s klasickými experimenty, tzv. *flashing light experiments* (viz např. Nedbal *et al.* (1996) [36], nebo Terry (1986) [60]).

Předložené původní výsledky disertace ukazují na výhody metodologie návrhu bioreaktorů spočívající ve spojení postupů matematického modelování-simulace-optimalizace ve srovnání s dosud převažující metodikou tzv. *scale-up* a otevírají tak prostor pro další teoretický i experimentální výzkum v oblasti návrhu řasových fotobioreaktorů, čímž se následně zvyšuje pravděpodobnost úspěšné komerční produkce mikrořas.