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Feedback linearizing control of light-to-microalgae ratio in artificially lighted photobioreactors

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Abstract: The present paper describes the design and the validation of a lumostatic controller for artificially lighted photobioreactors operated in discontinuous mode. The ratio between the incident light intensity and the biomass concentration, termed light-to-microalgae ratio, was selected as output variable, while its control was provided by manipulating the power supply of a light source and consequently the incident light intensity. The biomass yield on light energy was introduced in order to properly compare the batches operated under constant light intensities with the lumostatic batches. The results obtained in simulation show that a lumostatic batch can yield at least 10% more biomass per mole of supplied photons. The nonlinear controller, synthesized on the feedback linearizing technique, was implemented and validated on a laboratory torus photobioreactor inoculated with *Chlamydomonas reinhardtii* cells.

Keywords: light-to-microalgae ratio, photobioreactor, lumostat, microalgae, feedback linearization, process control, closed-loop control.

1. INTRODUCTION

During last decades the microalgae consolidated their role in food, biotechnological and environmental applications (Brennan and Owende, 2009) and whether they are cultivated in open or closed photobioreactors (PBR), naturally or artificially lighted, the processes have to be monitored and controlled in order to attain optimal performances and traceability. The most challenging feature of microalgae is their photosynthetic metabolism that is the use of light energy and inorganic carbon to constitute organic substances used further in cellular functions. Despite the fact that the photosynthetic microalgae have simple growth requirements and can bloom on renewable raw materials (i.e. solar light as source of energy, inorganic carbon from flue gasses or air and inorganic nutrients from wastes), there are still several obstacles to overcome for their cultivation to be economically feasible. In these environmental conditions the microalgal cultures are subjected to strong disturbances (i.e. day-night cycles, weather conditions, evaporation, contamination, etc) which make the photosynthetic productivity profiles extremely complex. On the other hand the artificially lighted PBRs result in higher repeatability of photosynthetic processes and therefore they are aimed for the cultivation of added value compounds (i.e. pigments, fatty acids, proteins, polycarbohydrates, stable isotopes, etc). These reactors are operated in continuous or discontinuous mode and even though the continuous cultures have unquestionable advantages, most scientific papers make reference to batch cultures. Nevertheless, the discontinuous cultures are characterized by a series of advantages being preferred since they do not require additional buffer capacities, are safer to contamination and are easier to be implemented.

The light is obviously the main factor that limits the photosynthetic growth of microalgae, creating a heterogeneous field which is responsible for the decrease of the growth rates along the depth of the culture. Thus, the radiative models, which describe the light attenuation inside photosynthetic cultures, are fundamental in PBRs studies (Cornet *et al.*, 1998).

The discontinuous cultures of microalgae are generally exposed to constant incident light intensities, the photonic energy received by individual cells decreasing as the biomass concentration increases. In the early stage of cultivation, when the number of cells is low, the photonic energy per individual cell may exceed its capacity of absorption, being dissipated as heat or fluorescence, but can also inhibit cell growth. This method of cultivation can be optimized by manipulating the incident light intensity so that the light energy absorption to be maximized. These types of controllers are termed *lumostats*. In this context, for artificially lighted PBRs operated in batch mode, the present paper brings forth the design and validation of a lumostatic controller, meant to increase the biomass yield per mole of supplied photons.

Although there are numerous papers regarding the modelling of light availability inside photosynthetic cultures, the papers related to the control of incident light intensity are very rare. However, some authors investigated the effect of the incident light intensity manipulation while controlling, by means of simple methods, output variables such as the photosynthetic activity (Eriksen et al., 1996; Marxen *et al.*, 2005), average irradiance (Suh and Lee, 2001), specific light uptake rate (Choi *et al.*, 2003; Lee *et al.*, 2006) or specific irradiation rate (Kang *et al.*, 2006; Yoon *et al.*, 2012).

The present paper is organised as follows: after a brief introduction, the experimental bench is presented, followed by the modelling of the synthetic growth of microalgae cultures and the design of the lumostatic controller along with its practical implementation.

2. MATERIALS AND METHOD

A strain of C. reinhardtii 137AH was used for the validation of the lumostatic controller and there was used a minimum growth medium (MGM) in order to maintain the strain in photoautotrophic growth conditions. The MGM consisted in: NaHCO₃ – 1.68 g·L⁻¹, NH₄Cl – 1.45 g·L⁻¹, MgSO₄·7H₂O – 0.28 g·L⁻¹, CaCl₂·2H₂O – 0.05 g·L⁻¹, KH₂PO₄ – 0.61 g·L⁻¹ and Hutner's trace elements – 1 mL·L⁻¹. The PBR employed in the validation experiment is a lab-scale torus-shape one that is described in detail elsewhere (Fouchard et al., 2008). The reactor is lighted from one side by an electroluminescent diode panel (calibrated with a LI-COR light meter LI-1400) whose incident light flux can be controlled through a voltage generator. A linear correlation between the incident light intensity and the voltage of the power supply was found $(R^2=0.999)$. The maximal incident light intensity of the LED panel is 800 µmol·m⁻²·s⁻¹. The reported incident light intensities were measured at the surface of the culture, behind the transparent panel. The turbidity of the culture was measured online by means of a Mettler Toledo® InPro8200 probe, connected to a Mettler Toledo® Trb8300 transmitter. The dry matter of the culture was analyzed daily by filtering known volumes of sample and weighting them before and after being dried in an oven, for 24h at 110 °C. The correlation between dry matter and turbidity was found to be linear ($R^2 = 0.999$). The pH was measured with a Mettler Toledo® Inpro 3253SG/120/Pt100 electrode (which also measures the temperature) connected to a Mettler Toledo® M400 transmitter. The pH was controlled with pure CO₂ at 7.5 as described in Ifrim et al., (2013). N_2 gas was also bubbled into the reactor as vector gas. The injected gas volumes were monitored through specific Bronkhörst® HIGH TECH EL-FLOW flow meters provided with proportional valves which allow the adjusting of the required rates in percents. The signals from the installation were collected through a DAQ board and registered on the process computer with a sampling time of 1s. The process computer hosts the control software conceived in LabVIEW® that allows Matlab® scripts to run in tandem.

3. MODELLING OF THE PHOTOSYNTHETIC GROWTH OF MICROALGAE

The photosynthetic organisms depend completely on the photosynthetic apparatus in order to comply with their metabolic requirements. They are using light, as source of energy, to convert CO_2 into organic substances. The photosynthetic growth of microalgae in PBRs results in a particular class of models in which the growth kinetics must be coupled with the availability of light. Early attempts to find a proper model to describe the light-limited growth of photosynthetic microalgae were based on the assumption that the rate at which the light energy is taken up is directly proportional to the rate of cell synthesis, expressing the specific growth rate, μ , as follows:

$$\mu = q_e X - \mu_s \tag{1}$$

where q_e is the specific light uptake rate, X is the biomass concentration and μ_s the specific maintenance rate. The specific light uptake rate was first defined by Van Liere and Mur (1979) as the total amount of energy absorbed by the culture divided by the total biomass present in the culture:

$$q_e = \frac{(q_0 - q_L)}{V} \cdot \frac{A}{V} \tag{2}$$

where q_0 is the average incident light intensity and q_L is the average output light intensity measured on the backside of the reactor. A and V are the lighted surface of the reactor and the volume of the culture, respectively. This formulation has indeed certain limitations because it gives a linear relation between the specific growth rate and the absorbed energy, neglecting phenomena such as photolimitation and photoinhibition. Furthermore, the light is attenuated inside the culture regardless of reactor's geometry or the source of light, creating a heterogeneous field. A typical case of light attenuation is presented in Fig. 1 for a rectangular photobioreactor lighted from one side.

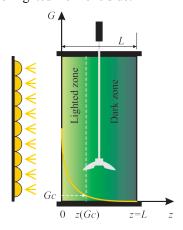


Fig. 1. Schematic representation of light attenuation inside a photobioreactor lighted from one side

The light gradient is responsible for the decreasing growth rates along the depth z of the culture. The mutual shading of the cells, when the light intensity is low or when the concentration of biomass is high, leads to the installation of a dark zone inside the culture where the cells are respiring. The available radiant light energy for which photosynthesis compensates the respiration (thus, the exchange rates for O₂ and CO₂ are equal to zero) is known as the compensation point, Gc. In order to characterize the attenuation of light inside photobioreactors, various concepts were approached starting from simple models such as the Lambert-Beer law all the way to rigorous radiative models such as the two-flux model which considers two phenomena, namely the absorption of light by pigments and the scattering of light by cells. A radiative model, simplified on a hypothesis of monodimensional attenuation, which was designed for rectangular reactors lighted on one side, was used also for the torus photobioreactor in question. The analytical expression of the spectral irradiance, G, has the following form (Pottier et al., 2005):

$$G(z) = 2q_0 \frac{(1+\alpha)e^{\delta(L-z)} - (1-\alpha)e^{-\delta(L-z)}}{(1+\alpha)^2 e^{\delta L} - (1-\alpha)^2 e^{-\delta L}}$$
(3)

with $\delta = X\sqrt{E_a(E_a+2bE_s)}$ the two-flux extinction coefficient and $\alpha = \sqrt{(E_a)/(E_a+2bE_s)}$ the linear scattering modulus. E_a and E_s are the mass absorption and the mass scattering coefficients while b is the backward scattering fraction and L is the depth of the photobioreactor.

The coupling of growth kinetics with radiative transfer can be approached in two manners which differ in the sense that the irradiance, G(z), calculated for each z along the reactor's depth, is introduced in the formulation:

- whether it is averaged $-\langle G(z) \rangle$ and used as a single value in the kinetic equation of the specific growth rate $\mu(\langle G(z) \rangle)$ (Molina Grima *et al.*, 1996);
- or used to compute local photosynthetic responses, $\mu(G(z))$ which are thereafter averaged $-\langle \mu(G(z))\rangle$ (Cornet *et al.*, 1998). $\langle \rangle$ denotes a special averaging.

One of the most employed models for expressing the specific growth rate of photoautotrophs is the Haldane type model:

$$\mu = \mu_0 \int_0^L \frac{G(z)}{K_I + G(z) + \frac{G^2(z)}{K_I}} dz - \mu_s \tag{4}$$

where μ_0 is related to the maximum specific growth rate $\mu_{max} - \mu_0 = (1 + 2\sqrt{K_I/K_{II}})$, K_I is the irradiance half-saturation constant and K_{II} is the irradiance inhibition constant. The evolution of the biomass concentration in discontinuous cultures can be thus easily computed, under the hypothesis of homogeneous culture's broth:

$$\frac{dX}{dt} = \langle \mu \rangle(t)X(t) \tag{5}$$

4. DESIGN OF LUMOSTATIC CONTROLLER

Even though q_e was not well received by the scientific community for modelling the growth of photosynthetic organisms, it remained a useful parameter for scale-up and control in PBR studies. In dense cultures the energy absorbed is close to the irradiated energy, therefore q_L will be always equal to 0. For the case in which a photobioreactor lighted on one side is used, the measurement of q_L can be carried out only at low biomass concentrations when the entire volume of the culture gets to be lighted. Certain authors neglected q_L naming the new parameter specific irradiation rate, q_i , which is practically the product between the light-to-microalgae ratio, $R_{q_0/X}$, and the surface-to-volume ratio, $R_{A/V}$ (Kang et al., 2006; Yoon et al., 2012). In most cases $R_{A/V}$ is constant (except when samples are taken) while $R_{q_0/X}$ varies in time. However, in order to obtain a simple I/O model, for control purposes, the $R_{A/V}$ can be neglected. The expression of $R_{q_0/X}$ considered for control will have the following form:

$$\frac{dR_{q_0/X}}{dt} = \frac{q_0(t)}{X(t)} - R_{q_0/X} \tag{6}$$

The utility of such model arises from the fact that both variables of the model can be measured online. Thus, $R_{q_0/X}$ will be the ratio between the incident light flux and the biomass concentration, and the control variable q_0 will have the same dynamics as the biomass, whereas its range will be given by its prescribed value. The more the biomass concentration increases, the more the incident light intensity

increases, up to a saturation value which depends of the strain proprieties and the light source characteristics.

The growth model (5) is not needed for the control of $R_{q_0/X}$ because its simple expression consists in two measurable variables (*i.e.* q_0 and X). However this model will be used to compare in simulation the differences between constant light batch cultures and lumostatic batches.

The control scheme of the lumostatic batch is presented in Fig. 2.

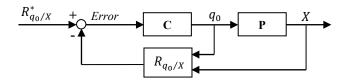


Fig. 2. Control scheme of light-to-microalgae ratio (C – controller, P – process)

As it can be seen in Fig. 2, the objective is to control the light-to-microalgae ratio in closed loop by actuating the incident light flux intensity. Considering that the experimental bench presented in section 2 is provided with a turbidity probe which can be associated with the biomass concentration and with a LED panel which can be set to a precise light intensity by imposing a known voltage on its power source, the $R_{q_0/X}$ can be calculated online making the automatic control issue easily approachable.

The control q_0 was synthesized on the feedback linearizing control technique (FLC) which presents the advantage, in comparison to the standard linear algorithms, of returning a linear closed loop unconditionally stable over a large set of operating conditions (Henson and Seborg, 1997). The FLC technique involves three steps, namely deriving a proper I/O model through appropriate manipulations (deriving output y with respect to time until the input u appears), defining a stable linear reference model of the tracking error and calculating the control action u so that the I/O model exactly matches the reference model (Bastin and Dochain, 1990). Model (6) is an I/O model that is appropriate for control purposes because the output variable, $R_{q_0/X}$, is an explicit function of the input variable, q_0 . The nonlinear model (6) rewritten in state-space form will be:

$$\begin{cases} \dot{x} = f(x) + g(x)u \\ y = h(x) \end{cases} \Leftrightarrow \begin{cases} \left[\dot{R}_{q_0/X}\right] = \left[-R_{q_0/X}\right] + \left[1/X\right]q_0 \\ y = R_{q_0/X} \end{cases}$$
(7)

where u, x and y are the input, state and output variables, while f(x), g(x) and h(x) are nonlinear functions. The relative degree, r, is a fundamental characteristic of a nonlinear system and practically represents the number of times the output, y, needs to be differentiated before the input, u, appears explicitly. q_0 appears explicitly after the first derivation of $R_{q_0/X}$ and therefore the relative degree of model (6) is equal to 1. The regulation error, $er = y^* - y$, is assumed to decrease according to the following stable linear time-varying first order dynamics:

$$\sum_{k=0}^{r} \lambda_{r-k} \frac{d^k}{dt^k} [y^*(t) - y(t)] = 0, \lambda_0 = 1$$
 (8)

where y^* represents the setpoint and λ is the pole of the system that impose the error dynamics. The coefficient λ is arbitrary with the exception that it must be chosen so that the differential equation (6) to be stable. Since $\delta = 1$ the reference model will be reduced to $der/dt = -\lambda er$.

The control u must be calculated as follows, so that the I/O model matches the reference model:

$$u(t) = \frac{1}{g(x)} \left[-f(x) + \sum_{k=0}^{r} \lambda_{r-k} \frac{d^k}{dt^k} [y^*(t) - y(t)] + \frac{d^r y^*}{dt^r} \right]$$
(9)

Considering that the setpoint, $R_{q_0/X}^*$, is always constant (and implicitly its derivative equal to zero) the control algorithm will have the following expression:

$$u(t) = q_0 = X(R_{q_0/X} + \lambda er) \tag{10}$$

with $er = R_{q_0/X}^* - R_{q_0/X}$. Knowing that $R_{q_0/X}$ is the ratio between q_0 and X, it can be said that the controller behaves as a nonlinear integrator that, after each iteration, adds to the previous q_0 a fraction proportional with X and er.

Selecting a proper setpoint for light-to-microalgae ratio is a delicate issue that depends on various factors such as PBR geometry, radiative transfer, characteristics of the lighting source, etc. To better understand the mechanism of lumostatic batches the input variable q_0 was plotted for a wide range $R_{q_0/X}^*$ as it can be seen in Fig. 3.

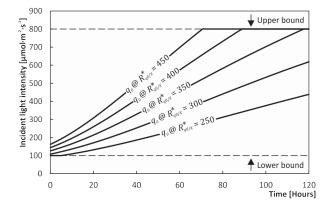


Fig. 3. Control variable, q_0 , evolution for a wide range of light-to-microalgae ratio setpoints, $R_{q_0/X}^*$.

The control is lower bounded at 100 μ mol m⁻² s⁻¹ to ensure a minimum level of light, regardless of the operating point. The true utility of the lower bound is that it prevents the photoinhibition during lag phase, allowing the controller to increase q_0 only when new biomass is forming, without requiring the intervention of a human operator. The upper bound (*i.e.* 800 μ mol m⁻² s⁻¹) is characteristic to the light source used for this study. However, if $R_{q_0/X}^*$ is too high, the control variable will be quickly saturated. Contrarily, if the setpoint is too small, lower biomass concentrations will be obtained at longer culturing periods. The measuring unit for $R_{q_0/X}$ is μ mol m·s⁻¹·kg⁻¹; yet, because it does not have consistent physical significance the light-to-microalgae ratio will be expressed hereinafter in arbitrary units. The simulations presented in Fig. 3 were made by integrating

model (5) and (6) whose parameters, which were identified elsewhere (Fouchard *et al.*, 2009; Ifrim *et al.*, 2013), are given in Appendix.

In order to properly compare a constant light batch and a lumostatic batch a performance index needs to be introduced, namely the biomass yield on light energy (Cuaresma *et al.*, 2011):

$$Y_{X/q_0} = \frac{(X - X_0)V}{q_0 A t} \tag{11}$$

where X_0 is the biomass concentration after inoculation and t is the cultivation time. To observe the behaviour of a lumostatic batch the best setpoint in the given configuration is considered to be $R_{q_0/X}^* = 350$ (Fig. 3). A lumostatic batch piloted at $R_{q_0/X}^* = 350$ is comparable, in terms of biomass produced per amount of time, with a constant light batch maintained at q_0 of app. $500 \, \mu \text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$, as it can be seen in Fig. 4a. The light-to-microalgae ratio is plotted in Fig. 4b for both batches. It can be observed that by using constant light the cells are exposed to high amounts of light energy in the first hours of cultivation and yet the light is rapidly attenuated at higher concentrations of biomass. The new formed biomass is practically obtained by multiplying the volumetric growth rate, μX , with the total volume of the culture and neglecting the inoculum concentration.

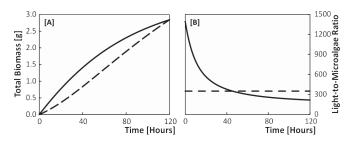


Fig. 4. Time evolution of A) new formed biomass, B) $R_{q_0/X}$.

- constant light batch and - - - - - lumostatic batch.

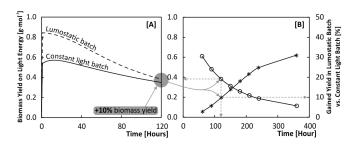


Fig. 5. A) Time evolution of Y_{X/q_0} in constant light and lumostatic batches, B) The Y_{X/q_0} and gained yield in lumostatic batches as a function of culturing time; Y_{X/q_0} in lumostatic cultures, Y_{X/q_0} - gained yield in lumostatic batch vs. constant light batch.

In Fig. 5a the biomass yield with light energy is plotted for both batches resulting that the lumostatic batch yields 10% more biomass per mole of supplied photonic energy. The more the culturing time increases, the more it increases the gained yield of a lumostatic batch over a constant light batch. The disadvantage is that at high culturing periods, when the

gained yield could reach up to 30%, the Y_{X/q_0} decreases and the plant might not be productive anymore (Fig. 5b). However, there is no general rule of choosing a proper $R_{q_0/X}^*$ because it is characteristic to each system and depends on many technological aspects, but it can be taken into consideration for further optimal control techniques.

5. PRACTICAL VALIDATION OF THE LUMOSTATIC CONTROLLER

In order to validate the light-to-microalgae ratio algorithm (10) the torus PBR was inoculated with cells of *Chlamydomonas reinhardtii* that were cultivated in discontinuous mode. The batch was conducted for app. 120 hours in lumostatic mode, $R_{q_0/X}$ being maintained at 350 (Fig. 6). $R_{q_0/X}$ was calculated online based on measurable biomass, X, and incident light intensity values, q_0 . The lumostatic controller tracked the prescribed value with a global accuracy of \pm 8.4 σ (\pm 2.4 % σ _r) that decreased to \pm 4.5 σ (\pm 1.3 % σ _r) during the last 80 hours. σ is the standard deviation and σ _r is the relative standard deviation.

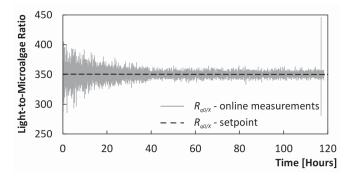


Fig. 6. Time variation of $R_{q_0/X}$ in a lumostatic batch

The biomass concentration was estimated online based on the signal of a turbidity probe. A moving average filter with a period of 60 samples (one minute) was also implemented for the noisy turbidity signal. Dry mass analyses were made once a day in order to verify the validity of the correlation under varying light intensities (Fig. 7).

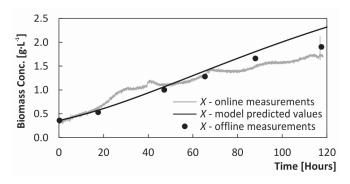


Fig. 7. Time variation of biomass concentration in a lumostatic batch

The photosynthetic growth model (5) fits well the experimental data only in the first 70 hours when the biomass reaches $\sim 1.4~{\rm g \cdot L^{-1}}$ and the incident light intensity is 500 ${\rm \mu mol \cdot m^{-2} \cdot s^{-1}}$. Above this point, the growth rate decreases in

comparison with the values predicted by the growth model. However, further studies must be made regarding the growth kinetics in order to increase the productivity of biomass. Nevertheless, the lumostatic controller is giving good results regardless of the growth kinetics as it turns out in practice.

Fig. 8 displays the control variable, q_0 , expressed both as voltage, which is the real parameter used by the power source of the LED panel and as light intensity given in μ mol·m⁻²·s⁻¹, which is used by the model. The two variables were found to be linearly dependent, the panel being operated in the range 17-24 V which corresponds to 100-800 μ mol·m⁻²·s⁻¹. Due to the slow biomass dynamics the incident light intensity was recalculated with the same periodicity of 60 samples (one minute). The tuning parameter λ was determined empirically as being equal to one, both in practice and simulation. Because $R_{q_0/X}$ is a simple ratio between q_0 and X, the incident light intensity profile overlaps the biomass, registering the same mismatch during the last 50 hours when compared with the photosynthetic growth model response.

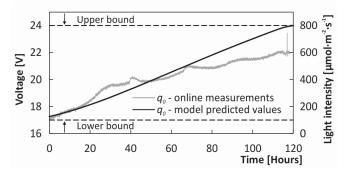


Fig. 8. Time variation of input variable q_0 in a lumostatic batch

In order to demonstrate that $R_{q_0/X}$ control leads the photosynthetic culture towards constant light conditions the average irradiance, $\langle G(z) \rangle$, was reconstructed from experimental data and then compared to model predicted values (Fig. 9).

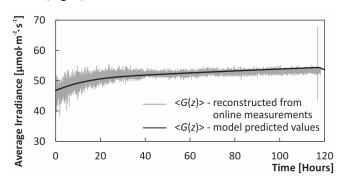


Fig. 9. Time variation of reconstructed $\langle G(z) \rangle$ vs. model predicted values in a lumostatic batch

By using the online measured values of q_0 (Fig. 8) and X (Fig. 7), the irradiance, G(z), was calculated with the radiative model (3) in 100 points inside the reactor and then averaged. Fig. 9 displays the offline reconstructed $\langle G(z) \rangle$ evolution which turns out to be quasi-constant during $R_{q_0/X}$

control and furthermore well predicted by the photoautotrophic growth model.

6. CONCLUSIONS

By using the FLC theory, a simple and reliable algorithm was synthesized to control the light-to-microalgae ratio through manipulation of the incident light intensity. It was demonstrated in simulation that the lumostatic batches can yield at least 10% more biomass per mole of supplied photons when compared with constant light batches. Although further kinetic studies must be made, the lumostatic controller was validated with good results on a torus PHB. The experimental data showed that the average irradiance inside the culture was maintained quasi-constant through this type of control.

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Appendix

Model parameters: absorption and scattering coefficients – $E_a = 172 \text{ m}^2 \cdot \text{kg}^{-1}$, $E_s = 870 \text{ m}^2 \cdot \text{kg}^{-1}$, b = 0.0008 (-); kinetic model parameters – $\mu_0 = 0.16 \text{ h}^{-1}$, $K_I = 120 \text{ μmol·m}^{-2} \cdot \text{s}^{-1}$, $K_{II} = 2500 \text{ μmol·m}^{-2} \cdot \text{s}^{-1}$, $\mu_s = 0.013 \text{ h}^{-1}$; photobioreactor geometry coefficients – $V = 1.47 \cdot 10^{-3} \text{ m}^3$, L = 0.04 m, $A = 3.75 \cdot 10^{-2} \text{ m}^2$. *Initial conditions* (t = 0) used for numerical simulation: $X(0) = 0.36 \text{ g·L}^{-1}$, $X_{new}(0) = 0 \text{ g}$, $R_{q_0/X}(0) = 277.78$, $Y_{X/q_0}(0) = 0 \text{ g·mol}^{-1}$.