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Modelling of Microalgae Culture Systems with Applications to Control and Optimization

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Abstract Mathematical modeling is becoming ever more important to assess the potential, guide the design, and enable the efficient operation and control of industrial-scale microalgae culture systems (MCS). The development of overall, inherently multiphysics, models involves coupling separate submodels of (i) the intrinsic biological properties, including growth, decay, and biosynthesis as well as the effect of light and temperature on these processes, and (ii) the physical properties, such as the hydrodynamics, light attenuation, and temperature in the culture medium. When considering high-density microalgae culture, in particular, the coupling between biology and physics becomes critical. This chapter reviews existing models, with a particular focus on the Droop model, which is a precursor model, and it highlights the structure common to many microalgae growth models. It summarizes the main developments and difficulties towards multiphysics models of MCS as well as applications of these models for monitoring, control, and optimization purposes.

Keywords Microalgae · Photobioreactors · Raceways · Modeling · Optimization · Biofuel · CO_2 mitigation

Contents

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1 Introduction

The renewal of phytoplankton-based processes over the last decade has mainly been driven by the great promises of these microscopic plants. Both microalgae and cyanobacteria show a great potential for industrial applications, including food, pharmaceuticals and cosmetics, chemicals, and even biofuel [74]. Contributing the most to this popularity is perhaps the fact that microalgae present high photosynthetic yields compared to terrestrial plants and that certain species can reach a very high lipid content, above 50 % dry weight [73]. With the prospect of achieving lipid productivities several-fold higher than those of terrestrial plants [111], many have started envisioning large-scale microalgae culture systems (MCS) for biodiesel production [22]. Nonetheless, these predictions are often based on crude extrapolations of the productivities obtained in the lab, where conditions differ drastically from those of outdoor production systems, and thus far they could not be confirmed experimentally on pilot- or larger-scale demonstration plants. In this context, mathematical modeling can be a great help to understand, and in turn remedy, the gap between lab-scale observations and the industrial-scale reality. Not only can these models be used for monitoring, control, and optimization of the actual production systems, but they could also drive the choice of a particular microalgae species that is best suited to the local environment or even to a particular season of the year.

Modeling of high-density MCS, together with their control and optimization, proves more challenging than that of bacterial or yeast culture systems. To a large extent, this added complexity stems from the wide range of mechanisms used by microalgae to respond to, or protect themselves from, light and other local environmental factors. This is particularly so in outdoor production systems, where microalgae are permanently subject to unsteady conditions, for example, due to diurnal light and temperature variations. The dynamics induced by this periodic

forcing are difficult to model accurately and also make it hard to devise effective control strategies. Another modeling challenge is tied to the way microalgae access light in order to sustain their growth. Because of light-absorption mechanisms and shadowing effects, a higher microalgae concentration will reduce the amount of light available to the culture. This sets an upper limit on the theoretical (steady) concentration of microalgae, whereby the average growth over the light column is exactly balanced by respiration; that is, the net growth rate is zero. However, predicting this limit is not at all straightforward as microalgae also undergo photoacclimation via the adaptation of their pigments to the available light [2, 65]. In other words, light attenuation, as driven by pigment concentration and cell size, is itself dependent on light. Moreover, under nitrogen-limited growth conditions—often used to stimulate the production of a valuable metabolite [90]—both the pigment composition and concentration vary [42, 96, 108] and the cells increase their size, further affecting light attenuation [102].

Advances in the dynamic modeling of microalgae populations are scattered across various fields, including oceanography, ecology, and biotechnology. An early dynamic model of phytoplankton was proposed by Riley [89] for describing phytoplankton populations on Georges Bank. Quite remarkably, Riley modeled both nutrient- and light-limited growth by considering an exponential decrease of light along depth. A growing number of kinetic models describing the rate of photosynthesis have been proposed ever since, ranging from simple hyperbolic expressions [3] to complex representations accounting for the photoinhibitory effects caused by an excess of light [79, 81, 101, 110]. The processes of nutrient uptake and nutrient-limited growth likewise have been described by a number of semiempirical models [19, 30–32], and these models have later integrated light-limitation effects in order to represent the nonlinear couplings between photosynthesis and nutrient limitation (especially nitrogen) [36, 42, 78].

Regarding MCS, perhaps the first dynamic model of a raceway pond was proposed by Sukenik et al. [105] within the scope of the Aquatic Species Program [99]. This model was later extended to encompass discrete-time photoacclimation dynamics [106]. Other, less detailed, models were also proposed in the meantime [8, 45, 49]. By and large, high-density MCS present many challenges and opportunities for their reliable modeling as well as in applications of these models for control and optimization purposes [10].

This chapter starts by outlining the principles and building blocks of microalgae culture models (Sect. 2). Models that describe the main processes involved in microalgae growth and bioaccumulation are reviewed in Sect. 3, namely carbon and nitrogen internalization (and loss), carbohydrate and lipid storage, and pigment adaptation. The focus in Sect. 4 is on the physical characteristics of MCS, including models of the light distribution, flow pattern, and temperature evolution. Following this review is a discussion about the combination of the biological and physical properties into the overall modeling of outdoor, possibly high-density, MCS together with open issues (Sect. 5). Finally, a number of monitoring, control, and optimization strategies that take advantage of the available mathematical models are considered (Sect. 6).

2 Building Blocks of Microalgae Culture Models

Within the context of bioprocesses, the most natural way of building a model that captures the main process dynamics is to consider conservation principles. Mass, energy, and momentum balances normally come in the form of ordinary differential equations (ODEs) for lumped systems, or partial differential equations (PDEs) in the case of distributed systems, for instance, due to imperfect mixing or advection. An important feature of balance-based models is the presence of conversion terms (e.g., to describe biochemical reactions) as well as transfer terms (e.g., to account for liquid–gas exchange).

In the context of MCS, the solutions to the balance equations characterize concentration fields for key species inside the culture medium (mass balance of nutrient and biomass), along with velocity (momentum balance) and temperature (energy balance) fields in the liquid phase. In addition to transport and accumulation terms, the other critical terms in these equations are those describing:

(i) **Intrinsic biological properties**, including the rates of nutrient internalization, microalgae growth and decay, and intracellular storage. These rates are dependent upon both current and past conditions in terms of culture medium concentration, light, temperature, and so on. Note, in particular, that this dependence is with respect to local conditions in nonhomogeneous culture media.

(ii) **Physical properties**, including the light transmitivity, the viscosity, and the mass and thermal diffusivities in the culture medium. All these properties depend on the (possibly local) compositions of the culture medium and the characteristics of the microalgae cells themselves. They are also dependent on the geometry and mode of operation of the reactor.

The fact that the aforementioned biological and physical properties are strongly coupled and span multiple timescales (ranging from milliseconds to days) makes high-fidelity simulation of an overall microalgae culture system particularly challenging. An important feedback mechanism here is the effect of light on the local growth rate, which modifies the concentration of microalgae, affecting the way light is absorbed and scattered in the culture medium in turn. Even the hydrodynamics in the reactor can have an effect on light distribution as the waves at the surface create a complex air–water interface that can punctually concentrate solar rays. Regarding timescales, light effects on microalgae growth range from milliseconds for photoproduction, to minutes or hours for photoinhibition and photoregulation, and to days or weeks for photoacclimation. On top of this, MCS are subject to various periodic forcings, such as diurnal cycles or mixing inside the reactor leading to fast light variations.

The next two sections give an overview of available models describing the biological and physical properties of MCS. Then, we discuss various ways of coupling these models.

3 Modeling of Intrinsic Biological Properties

The dynamics that underlie microalgae growth—including internalization of the main nutrients (N, P) and carbon (inorganic and/or organic) as well as the effect of light and temperature—can be described in numerous ways.

Detailed metabolic models have been developed by accounting for all available, yet still partial, knowledge about the metabolic pathways of particular microalgae species. These models usually rely upon the principle of *balanced growth*, which precludes internal storage of metabolites. This assumption dramatically reduces the number of kinetic parameters needed to describe the individual reactions in the model. Nonetheless, the fixation of inorganic carbon (CO_2 or bicarbonate) in autotrophic microalgae is dependent on the incoming photon flux, thus this assumption should therefore be limited to constant light culture conditions from a strict point of view. Despite these limitations, some authors have considered using metabolic modeling tools under transient light conditions [23, 57, 98, 113]. Others have expanded the approach to allow for internal storage and reuse of metabolites such as neutral lipids and carbohydrates [24, 58].

A second kind of growth model, often referred to as compartmental models, describes the physiological status of microalgae cells in terms of quotas for key components, such as nutrients, chlorophyll, lipid, carbohydrate, and so on. These macroscopic (semiempirical by nature) models are simpler than their metabolic counterparts, yet they do not call for any assumption about balanced growth. As such, they are well suited for coupling with detailed hydrodynamics models for flow and temperature computations (see Sect. 5), and they are equally well suited for development of model-based monitoring and control strategies (see Sect. 6).

The emphasis in the remainder of this section is more specifically on this latter class of models, focusing successively on nutrient-limited growth, light effects, and temperature effects, and assuming the other factors constant.

3.1 Nutrient-Limited Growth and Decay

In a classical batch experiment, microalgae continue to grow for several days after key nutrients are depleted from the culture medium. This apparent uncoupling between the processes of nutrient uptake (inorganic nitrogen, phosphorus, vitamins, etc.) and growth in microalgae is well documented [95], and it has led to the development of so-called quota models, which account for nutrient storage (pooling) inside the cells. For instance, the internal cell quota q_n of a limiting nutrient (substrate) s can be described by a simple mass-balance equation of the form:

$$\dot{q}_n = \rho(s, \cdot) - \mu(q_n, \cdot) q_n \,, \tag{1}$$

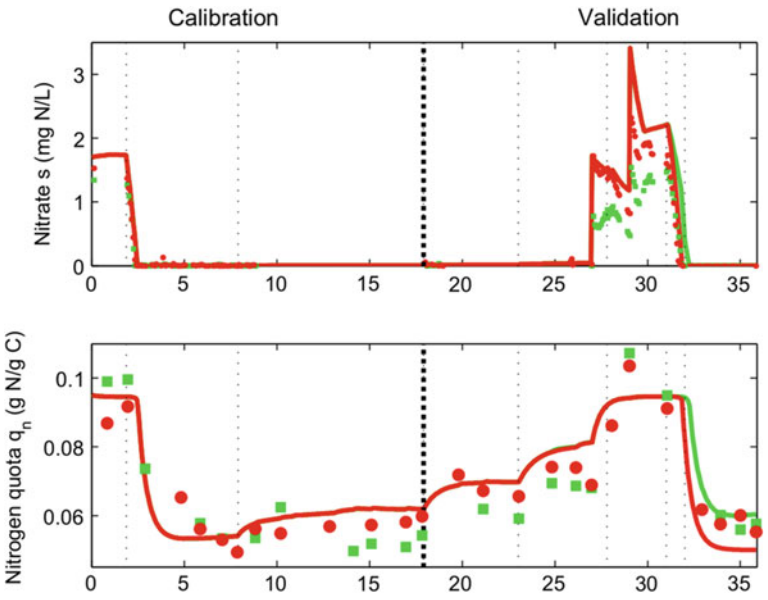

Fig. 1 Comparison between measurements and model predictions of the nitrate concentration (s) in the culture medium and the nitrogen quota (q_n) under nitrogen-limited conditions in a chemostat culture of *Isochrysis* aff. *galbana*. Reproduced from [67]

where ρ and μ denote the uptake rate of nutrient s and the corresponding nutrient-limited growth rate, respectively.[1] The quota q represents the intracellular amount of nutrient per unit cell mass. In the case where nitrogen is the limiting nutrient, for instance, the internal nitrogen quota can be defined in units of g(N) g(C)$^{-1}$.

Initially introduced to describe the limiting effect of Vitamin B_{12} on the growth rate of phytoplankton [30], the Droop model has also been found to predict the effect of macronutrient limitation accurately, including nitrogen or phosphorus limitation, and it has been widely validated [13, 31, 97, 109]; see, for instance, Fig. 1. Although applicable to a single limiting nutrient and constant light conditions only, the simplicity of the Droop model is a big help in practice as it enables detailed mathematical analysis [12, 14, 62]. Moreover, the meaning of its parameters makes it easy to relate to measurable quantities such as minimal and maximal internal quota and maximum growth rate.

The growth rate $\mu(q_n, \cdot)$ in the Droop model is expressed as an increasing function of the internal quota q_n:

$$\mu(q_n) = \mu_\infty \left(1 - \frac{Q_0}{q_n}\right), \tag{2}$$

[1] We use the notation $(\cdot)$ here to recall that these rates can also depend on other key parameters, such as light, temperature, and so on.

where Q_0 stands for the minimal cell quota, below which no growth is possible, and μ_∞ represents the growth rate at an hypothetical infinite quota. Alternative formulations have been proposed on the basis that a minimum (nonzero) internal quota is required in order for microalgae to grow. In a model due to Geider [42], for instance, the growth rate is simply a linear function of the internal quota, $\mu(q_n) = \mu_\infty (q_n - Q_0)$.

The uptake rate $\rho(s)$, on the other hand, is traditionally expressed in terms of Michaelis–Menten kinetics [19]:

$$\rho(s) = \rho_m \frac{s}{s + k_s}, \tag{3}$$

where k_s is the half-saturation constant for substrate uptake associated with the maximum uptake rate ρ_m. In Geider's model [42], an extra multiplicative term of the form $(Q_{max} - q_n)^p$ is appended to the uptake rate expression (3), usually with $p = 1$ [10, 17], so nutrient uptake comes to a stop as soon as the maximum internal quota Q_{max} is reached. Nonetheless, it is not hard to show that the original uptake rate expression (3) defines a maximum quota in a natural way, too. For instance, the internal cell quota is upper bounded by the value $Q_0 + \frac{\rho_m}{\mu_\infty}$ under nonlimiting nutrient conditions ($\rho(s) = \rho_m$) and using the growth expression (2).

In addition to uptake and growth rates, one must account for the loss of carbon via respiration. The corresponding rate of respiration can be expressed as the sum of a basal respiration rate and a term proportional to the cost of biosynthesis. The latter is typically assumed to be proportional to either the growth rate or the uptake rate [42, 92]. Although respiration is often accounted for indirectly as part of the "net" growth rate μ, it becomes necessary to distinguish the respiration rate from the growth rate more specifically when considering the effect of light. One possible approach involves defining the basal respiration as proportional to the cell concentration, while accounting for the rate of biosynthesis in the net growth rate. Note also that nitrogen is assumed to be released at the same rate as carbon in many models [42, 78], meaning that cell mortality and excretion are accounted for in the respiration rate.

3.2 TAG Synthesis

Among the various classes of lipids produced by microalgae, triacylglycerols (TAGs) are considered the preferred class in most applications, including algal-derived biofuel. Many microalgae strains have the ability to accumulate large quantities of lipids in the form of TAGs under environmental stress conditions such as nitrogen starvation. TAGs serve as energy and carbon storage compounds and as an electron sink in situations where the electron supply provided by photosynthesis exceeds the requirements for growth [54].

A number of models describing carbon storage in microalgae cells have become available in recent years [29, 47, 67, 77, 92]. Among them, the model in [67]

presents a simple structure based on the Droop model for representing TAG production in response to nitrogen deprivation and under constant light. It divides intracellular carbon into three pools, namely a functional pool (quota q_f), a sugar pool (quota q_s), and a TAG pool (quota q_l). The carbon fluxes between these pools lead to complex dynamics describing the storage and utilization of both sugars and TAGs. The way the model builds upon the Droop model is via a cascade structure, whereby the pool dynamics depends on the nitrogen quota q_n as well as the uptake rate ρ and growth rate μ:

$$\begin{aligned} \dot{q}_f &= -q_f\,\mu(q_n,\cdot) + (k_1 + k_3)\rho(s,\cdot) \\ \dot{q}_l &= [k_2\,q_n - q_l]\,\mu(q_n,\cdot) - k_3\,\rho(s,\cdot) \\ q_f + q_s + q_l &= 1, \end{aligned} \tag{4}$$

where k_1, k_2, and k_3 are (pseudo-)stoichiometric coefficients. This way, the model inherits the Droop model properties, taking advantage of its track-record validation, and only adding a limited number of extra parameters. Another interesting feature of the model is its ability to reproduce experimentally observed hysteresis in the dynamics of TAG accumulation, as shown in Fig. 2. This behavior contributes to making biolipid optimization complex and rather counterintuitive.

Other recent experimental studies have shown that TAG dynamics can become even more intricate when applying periodic light conditions in combination with

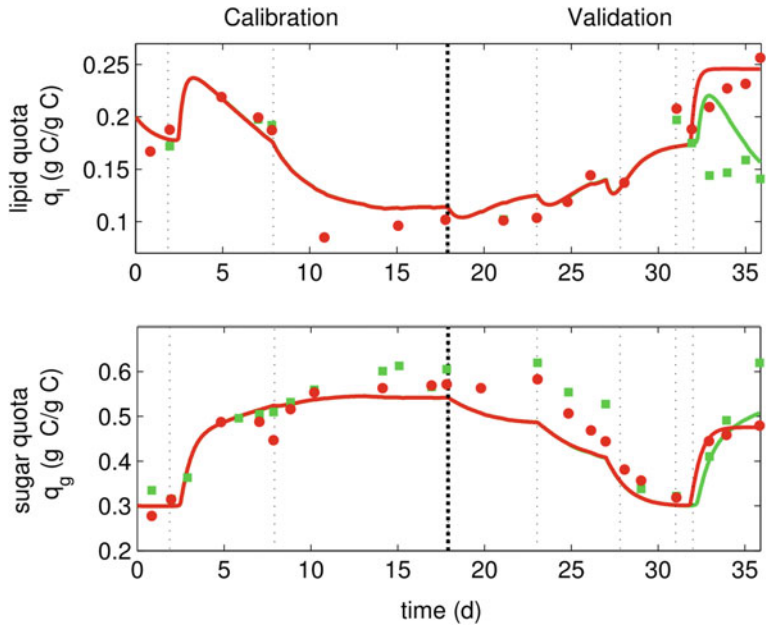

Fig. 2 Comparison between measurements and model predictions of the TAG (q_l) and sugar (q_g) quotas under nitrogen-limited conditions in a chemostat culture of *Isochrysis* aff. *galbana*. Reproduced from [67]

nitrogen deprivation. In particular, TAGs accumulate at a much slower pace than when exposed to continuous light; the lipids that are produced after a nitrogen starvation are consumed during dark phases (probably through a respiration process), thus maintaining the TAG pool at a low level [61]. Understanding how the mechanisms involved in cell synchronization interfere with the dynamics of TAG accumulation, and being able to model these interactions in turn reliably, clearly calls for further research.

3.3 Pigment Synthesis

A key mechanism used by microalgae to adapt their photosynthesis response to the available light involves modification of their pigment content. This photoacclimation strategy takes place at a timescale of days or weeks [25] and it can lead to a dramatic variation in the rate of photosynthesis. For instance, Fig. 3 shows PI-response curves for a microalgae culture preacclimated at different light irradiances, either normalized in terms of carbon or chlorophyll. On top of this, pigment synthesis can be strongly disrupted under nitrogen-limited growth as the pigment content is related to the protein content, which is itself related to the nitrogen status (nutrient quota q_n); this is especially so under complete nitrogen deprivation.

Geider et al. [42] were among the first to introduce chlorophyll as a state variable in their models, in addition to the carbon and nitrogen contents. They expressed the rate of pigment synthesis per carbon unit as proportional to the product between the rates of photosynthesis and nitrogen uptake.

More recently, Bernard [10] presented a model whereby chlorophyll is proportional to the cellular nitrogen content, used as a proxy of the actual protein content:

$$\theta = \gamma(I_0)\, q_n, \tag{5}$$

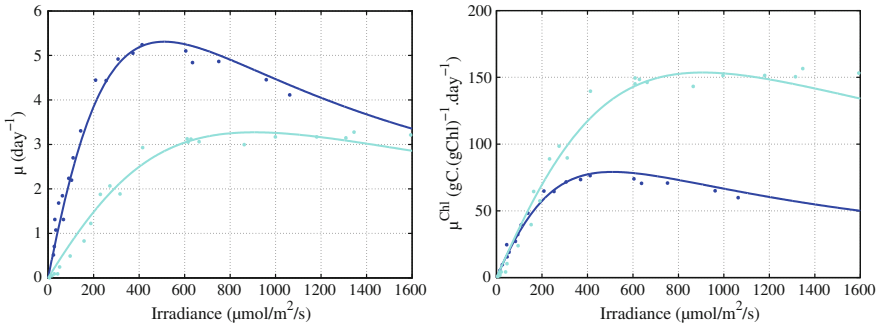

Fig. 3 Comparison between measurements and model predictions of the photosynthetic responses of the diatom *Skeletenonema costatum* under different acclimation states: I_0 = 50 μ mol m^{-2} s^{-1} (*dark points/lines*); I_0 = 1,200 μ mol m^{-2} s^{-1} (*light grey points/lines*). The photosynthesis rate is normalized by carbon (*left plot*) or chlorophyll (*right plot*). Data from [2]

where the chlorophyll quota θ represents the cellular chlorophyll-to-carbon ratio. In this expression, γ is a saturation function of the form

$$\gamma(I_0) := \gamma_{\mathrm{m}} \frac{k_{I_0}}{I_0 + k_{I_0}},$$

parameterized by γ_{m} and k_{I_0}. Moreover, I_0 is a conceptual variable representing the irradiance at which the cells are photoacclimated. In [10], the adaptation mechanism associated with I_0 is assumed to be driven simply by first-order dynamics:

$$\dot{I}_0 = \delta\, \mu(q_n, \cdot)\, [I - I_0], \tag{6}$$

with I the current light irradiance, and δ the photoacclimation rate constant. An alternative way to account for photoacclimation, without introducing the conceptual variable I_0, involves replacing (5) and (6) with:

$$\dot{\theta} = \delta'\, \mu(q_n, \cdot)\, [\gamma(I)\, q_n - \theta]. \tag{7}$$

In yet another model of photoacclimation, Geider [43] proposed the following alternative expression:

$$\theta = \frac{\mathrm{e}^{kT}}{(a - bT)\mathrm{e}^{kT} + cI}, \tag{8}$$

relating the chlorophyll quota θ to the current light irradiance and temperature.

3.4 Light-Limitation Effects

Notwithstanding its importance and significance, the Droop model, as well as other models derived from similar considerations, does not account for the effect of light on the growth rate. As such, it cannot be used directly to describe photolimited microalgae cultures.

Many photosynthesis models describe the effect of light in terms of PI–response curves. They use various proxies, such as the electron transfer rate (ETR), the O_2 production rate, the CO_2 consumption rate, or the growth rate itself. Although all these proxies do not involve the exact same mechanisms, the corresponding PI curves all have the same shapes.

Early photosynthesis rate models [3] considered simple hyperbolic expressions and did not account for photoinhibition by excess light. Such photoinhibition can be represented in a PI curve in either one of two main ways [79, 81, 101, 110]:

- The Platt model [81] is fully empirical and defines an exponential expression of the form $I\mathrm{e}^{-I}$, whereby the growth rate first increases with increasing light intensity up to a certain optimal irradiance, and then decreases from these maximal values as the light irradiance keeps increasing because of photoinhibition.

- The Han model [50], originating in the works of [33, 34, 59], has a stronger physical basis. It describes the chloroplasts in microalgae as arrays of photosynthetic units (PSUs), whereby each PSU is comprised of an antenna complex made up of pigments that is associated with the reaction center of a Photosystem II (RCII). The description of photoproduction and photoinhibition assumes that an RCII can be in either one of three states, namely open, closed, or damaged, and each RCII can transit from one state to another depending on the current light irradiance. An interesting property of the Han model is that the PI–response expression obtained by equilibrating the fast dynamics is of Haldane type.

The PI responses predicted by either of these representations are of course driven by other factors acting on growth, including nutrient limitation, pigment composition, and temperature. Nonetheless, a peculiarity of the chlorophyll-specific growth rate, $\mu^{\mathrm{chl}}(\cdot):=\mu(\cdot)/\theta$, is that the corresponding PI slope at vanishing light irradiance is typically independent of the photoacclimation light, and therefore independent of the chlorophyll quota θ. This property, which was perhaps first highlighted in [82], can be observed on the right plot of Fig. 3.

In particular, the foregoing constant initial slope property is key to understanding the way models that relate photoproduction/photoinhibition to the acclimation state are designed.

- The model by Geider and coworkers [65], which does not account for photoinhibition, expresses the (carbon-specific) growth rate as

$$\mu(I,\theta,q_{\mathrm{n}}) = \mu_{\max}(q_{\mathrm{n}})\left[1-\exp\left(\frac{-\alpha\theta I}{\mu_{\max}(q_{\mathrm{n}})}\right)\right], \tag{9}$$

  where $\mu_{\max}$ stands for the maximal growth rate (dependent on the nitrogen quota q_{n} and realized at high light irradiance); and α corresponds to the (constant) initial slope of

$$\frac{\mu(I,\theta,q_{\mathrm{n}})}{\theta}.$$

- The model by Bernard [10] is based on the Han model and can be expressed in the form

$$\mu(I,\theta,q_{\mathrm{n}}) = \mu_{\max}(q_{\mathrm{n}})\frac{I}{I+\frac{\mu_{\max}(q_{\mathrm{n}})}{\alpha\theta}\left(\frac{I}{I_{\mathrm{opt}}}-1\right)^2}, \tag{10}$$

  from which it is readily checked that the initial slope of $\frac{\mu(I,\theta,q_{\mathrm{n}})}{\theta}$ is again given by α.

At this point, both light- and nutrient-limitation effects can be combined by modulating the growth rate expressions (9) or (10) by a term dealing with substrate limitation, namely, the linear term $(q_n - Q_0)$ for the Geider model and Droop-like kinetics (2) for the Bernard model. Other models coupling chlorophyll production and

photosynthesis have been proposed [36, 40, 78], but they have not been used as much so far. More complex models have also been developed [38, 116], but being more accurate in the detail of the described mechanisms, they comprise more parameters and state variables that render their calibration/validation more difficult, too.

A situation under which the current light-effect models can become inaccurate and should be reconsidered is in the presence of fast-changing light regimes (flashing effect). The way photosynthesis reacts to high-frequency variations in light intensity remains the subject of active research. In order to capture these mechanisms, dynamic models describing the way photons are harvested (at a fast timescale) are required. These models typically work at a lower level by considering the concept of photosynthetic yield [20, 33, 34, 70, 115] and also try to account for nonphotochemical quenching (NPQ) regulation [75].

The effect of light flashes on microalgae growth has been studied experimentally, yet mainly for caricatural light patterns consisting in a succession of on/off periods at varying frequencies. Under such lighting protocols, the Han model—or related models—are found to represent the cell behavior well. We also note that for computational tractability reasons, the fastest timescale can be handled via singular perturbation techniques [21, 88] when considering some typical periodic light forcing [112].

3.5 Temperature-Limitation Effect

Like light, temperature too can play an important role in growth, and it is especially important to take its effect into account when considering outdoor MCS [87]. Two models have mainly been used to describe temperature effects on microalgae growth to date [15, 76].

The model in [15], based on the cardinal temperature model with inflection (CTMI) of [93], is detailed next. The main advantage of this model lies in its calibration simplicity, despite its empirical nature. The growth rate expression $\mu(\cdot)$ developed in the previous sections is simply modulated by a multiplicative switching function $\phi(T)$ taking values in the range $[0, 1]$:

$$\mu(\cdot)\,\phi(T)\,. \tag{11}$$

Moreover, the function $\phi(T)$ is given by

$$\phi(T) = \begin{cases} \dfrac{(T-T_{\max})(T-T_{\min})^2}{(T_{\text{opt}}-T_{\min})\left[(T_{\text{opt}}-T_{\min})(T-T_{\text{opt}})-(T_{\text{opt}}-T_{\max})(T_{\text{opt}}+T_{\min}-2T)\right]}, & \text{if } T \in [T_{\min},\ T_{\max}], \\ 0, & \text{otherwise,} \end{cases} \tag{12}$$

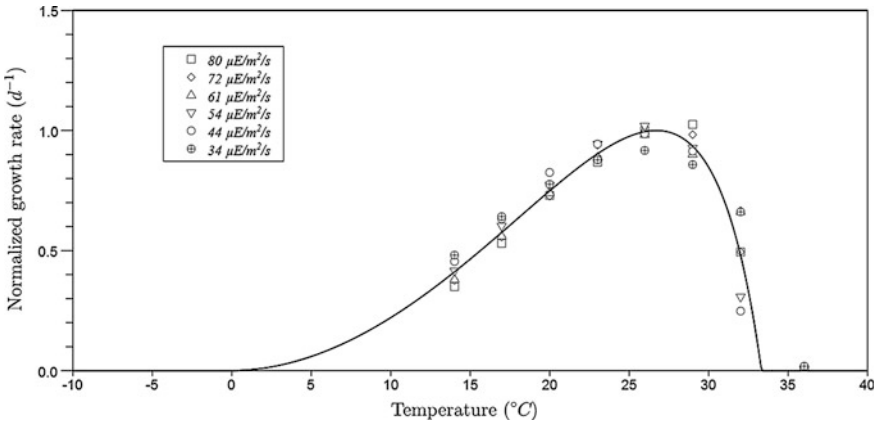

Fig. 4 Effect of temperature on the growth rate of *Nannochloropsis oceanica*: switching function $\phi(T)$ under various light irradiance and temperature conditions. Reproduced from [15]

with $T_{\min}$ and $T_{\max}$, the minimal and maximal temperatures, respectively, at which cells can grow; and T_{opt}, the optimal temperature for growth. Note that the following property must hold in order for the model to present the actual asymmetry that is experimentally observed:

$$T_{\text{opt}} > \frac{T_{\min} + T_{\max}}{2}. \tag{13}$$

Shown in Fig. 4 is a switching function whose parameters $T_{\min}$, $T_{\max}$, and T_{opt} are calibrated under different light and temperature conditions [15].

In addition to having a global effect on growth, temperature is usually assumed to have a similar effect on carbon and nutrient uptake, respiration, TAG synthesis, and the like. In accounting for temperature effects, it is therefore necessary to modulate the corresponding uptake, respiration, or biosynthesis rates, whose expressions have been given in previous subsections, in the same way as in (11).

4 Modeling of Physical Properties

4.1 Light Distribution

The light distribution in a microalgae culture decreases progressively in moving deeper into the culture medium due to photon absorption (mainly by pigments) and diffusion (mainly by the particles in the medium). Because of the complex multi-diffusive nature of the culture medium, classical theories such as the Mie theory do

not readily apply, thereby making the computations particularly challenging. Moreover, the optical properties are dependent on the light wavelength, explaining that green light will be mainly found in the darkest zones of the reactor. A large body of research has been devoted to high-fidelity simulation of the light distribution in such complex media; see, for example, Csogor et al. [28], Fernandez et al. [37], Suh and Lee [104]. In contrast, deriving analytical expressions, which can be used more easily for monitoring and control purposes, proves more difficult.

In the case of simple planar geometry with a 90° illumination angle with respect to the surface, one can use the Beer–Lambert law as a first approximation,

$$I(z,\cdot) = I_0 \exp(-\xi z), \tag{14}$$

where I_0 and $I(z)$ denote the irradiances at the surface level and at depth z, respectively, and ξ is the light attenuation parameter. The latter appears to be mainly correlated with the cell concentration x and the chlorophyll content θx, leading to the following simple approximation:

$$\xi = (a + b\theta)x + c, \tag{15}$$

with a, b, and c the specific light-attenuation coefficients due to biomass, chlorophyll, and background turbidity, respectively.

A key advantage of using the Beer–Lambert approximation is that it allows deriving analytical expressions of the average light and growth rate in simple photobioreactor configurations. Nonetheless, this model does not account for light backscattering, which can become significant in dense microalgae cultures. More accurate radiative transfer models based on the inherent optical properties of microalgae can be used in this context [27, 28, 39, 60, 84, 104].

In the remainder of this subsection, we consider a simple planar geometry of thickness L with a 90° illumination angle with respect to the surface. On application of the Beer–Lambert approximation, one can define the optical depth $\lambda = \xi L$, which reflects the actual amount of light energy absorbed by the culture medium. In particular, we have

$$\lambda = \ln\left(\frac{I_0}{I(L)}\right). \tag{16}$$

Under the additional assumption that all the concentrations are homogeneous (perfect mixing), the average irradiance received by the microalgae across the culture medium is given by

$$\overline{I} = \frac{I_0}{L}\int_0^L \exp(-\xi z)dz = \frac{I_0}{\lambda}[1 - \exp(-\lambda)]. \tag{17}$$

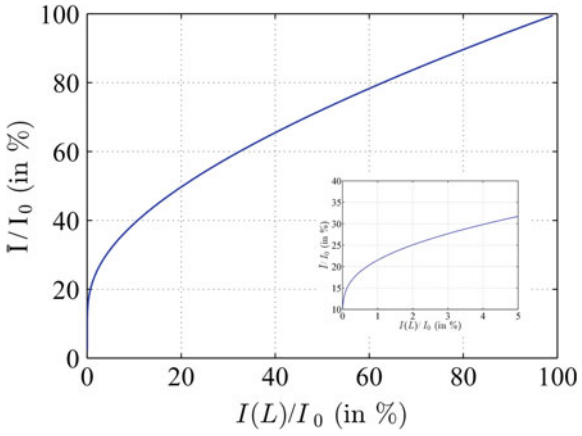

Fig. 5 Average light ratio $\frac{\bar{I}}{I_0}$ as a function of $\frac{I(L)}{I_0}$ in a simple planar geometry with 90° illumination angle

Finally, combining the two previous expressions gives

$$\frac{\bar{I}}{I_0} = \frac{1 - \exp(\lambda)}{\lambda} = \frac{\frac{I(L)}{I_0} - 1}{\ln\left(\frac{I(L)}{I_0}\right)}, \tag{18}$$

which does not depend explicitly on the light attenuation parameter ξ. The evolution of $\frac{\bar{I}}{I_0}$ versus $\frac{I(L)}{I_0}$ is represented in Fig. 5.

4.2 Microalgae Cell Trajectories

Because MCS are both mixed and optically thick at the same time, the microalgae cells are constantly crossing light gradients, which generates fast changes in the light irradiance received by a particular cell. In practice, the cells are exposed to light patterns with characteristic timescales that can be as small as a microsecond [83]. The ability to represent such fluctuations calls for detailed hydrodynamics simulations.

A number of studies have become available in recent years that use computational fluid dynamics (CFD) [71, 83, 85] for characterizing the hydrodynamics in MCS, for instance, based on commercial CFD codes such as ANSYS Fluent. In a second step, Lagrangian trajectories are obtained by integrating the computed velocity field. Although it has been applied mainly to simulate closed photobioreactors (PBRs), this approach has more recently been used to simulate open raceway ponds too [52]. It is because of their larger scale that raceway ponds are

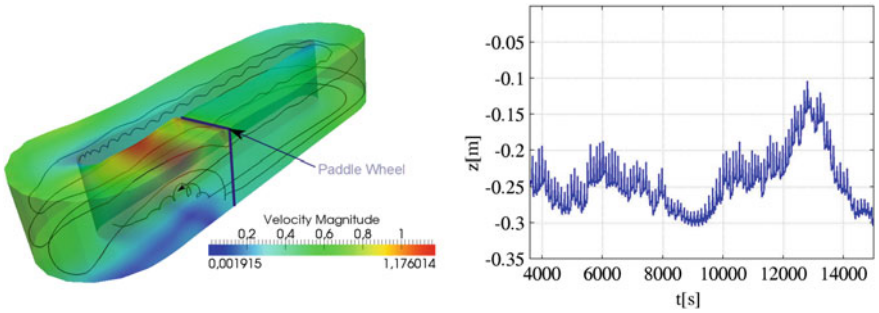

Fig. 6 3D Representation of the velocity field in a raceway pond along with a particular cell trajectory (*left plot*), and variation of the depth of a single cell over time (*right plot*). Reproduced from [52]

more computationally challenging to simulate than PBRs. Using a novel discretization scheme of the Navier–Stokes equations, the model in Bernard [10] has allowed reconstruction of the cell trajectories, and thus of the corresponding received light pattern, over longer time horizons. An illustration of the outcome of such simulations is shown in Fig. 6.

4.3 Temperature Variation

First-principles models describing temperature variations in MCS can be developed based on energy-balance considerations. These models account for the incoming solar energy on the one hand, and the energy dissipated through thermal re-emission, evaporation, and photosynthesis. In particular, this includes the direct and diffuse solar radiation, the radiation from the air and from the ground, the radiation of the culture medium, the evaporation flux, the heat flux in the CO_2-enriched bubbling gas, the conductive flux with the ground surface, and the convective flux at the surface.

Dynamic models that assume an homogeneous temperature of the culture medium have recently been proposed for both photobioreactors [5] and raceway ponds [6]. By taking into account the location, reactor geometry, light irradiance, air temperature, and wind velocity, these models can thus accurately predict the temperature evolution of the culture medium in outdoor facilities throughout a typical day, including the evaporative water losses. When not regulated, it has been shown that the temperature in open raceway ponds (which have a lower thermal inertia and where the benefit of evaporation is quite limited) can peak to values approaching 40 °C, thereby threatening microalgae cell survival. An illustration of these simulation results is presented in Fig. 7.

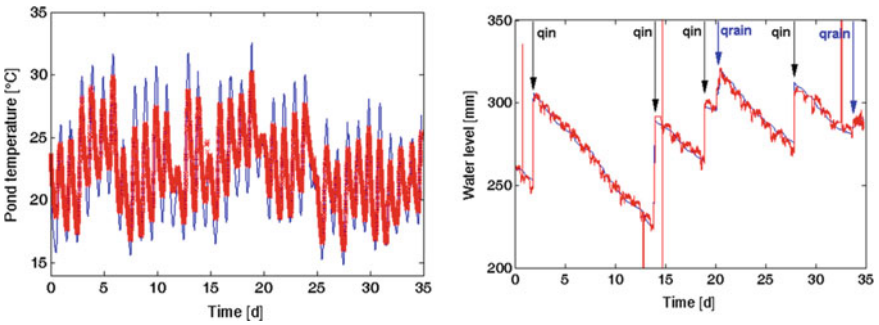

Fig. 7 Simulation of the temperature evolution in a raceway (*blue line*) using a model [6] and comparison with experimental measurements (*red lines*). On the *right-hand side*, the water level is also represented, with the indication of the water supply (*qin*) and rain (*qrain*)

5 Towards Multiphysics Models of Microalgae Culture Systems

5.1 Chemostat Culture

Under the assumption that the culture medium is perfectly mixed, that is, when the cells and limiting substrate concentrations are homogeneous, the main challenge involves characterizing the effect of the light gradient on the growth of the entire cell population. Note that, unlike light effects, the coupling on temperature is rather straightforward in the case where an homogeneous culture medium temperature is assumed.

The following equations describing the evolution of the cell concentration x and substrate concentration s, possibly coupled with the quota Eq. (1) in the classical Droop model, can be derived from mass-conservation principles:

$$\dot{s} = Ds_{\text{in}} - \rho(\cdot)x - Ds \tag{19}$$

$$\dot{x} = (\overline{\mu}(I_0, \cdot) - r)x - Dx, \tag{20}$$

where D stands for the dilution rate; s_{in}, the inlet substrate concentration; r, the basal respiration rate, and $\bar{\mu}(I_0, \cdot)$, the apparent specific growth rate at (surface) irradiance level I_0. It is precisely the determination of $\bar{\mu}(I_0, \cdot)$ that requires special attention here. When separating the timescales of mixing and photosynthesis, two limit situations can be distinguished:

(i) In the situation where mixing is much faster than the photosynthetic processes (infinite mixing assumption), all the cells respond to the mean light intensity $\overline{I}$, as discussed earlier in Sect. 4.1:

$$\overline{\mu}(I_0,\cdot) = \mu(\overline{I}(I_0,\cdot),\cdot)\,. \tag{21}$$

(ii) In the reverse situation where the photosynthetic response is now much faster than the mixing, the apparent specific growth rate corresponds to the average growth rate over the culture depth. For instance,

$$\overline{\mu}(I_0,\cdot) = \frac{1}{L}\int_0^L \mu(I(z,I_0,\cdot),\cdot)\,\mathrm{d}z = \frac{1}{L}\int_{I_0}^{I(L)} \frac{\mu(I,\cdot)}{I}\,\mathrm{d}I, \tag{22}$$

in the case of a simple planar geometry of thickness L with a 90° illumination angle. The rightmost term in (22) provides yet another interpretation of $\overline{\mu}(I_0,\cdot)$ as the average of the yield $\eta(I,\cdot) := \frac{\mu(I,\cdot)}{I}$ over the range of irradiance.

Assumption (ii) has been the basis for the theory of the "light-limited chemostat" [55, 56]. Experimental evidence that the microalgae cells respond to all irradiance levels within the light gradient range can be found in Huisman et al. [55], whereby photosynthetic efficiency is shown to increase with decreasing light intensity, and therefore with culture depth, using fluorescence measurement techniques.

Under this assumption, it can be shown [11] that, if the specific growth rate $\mu(I,\cdot)$ attains its maximal value $\mu_{\max}$ at I_{opt}, then the average growth rate $\overline{\mu}(I_0,\cdot)$ itself will attain its maximal value (which is lower than $\mu_{\max}$) at $I_{\mathrm{opt}}\sqrt{\frac{I_0}{I(L)}}$. Because the latter irradiance level is always greater than I_{opt}, a microalgae culture system with either a higher cell content or a larger water depth shall always be less prone to photoinhibition.

The foregoing considerations are illustrated in Fig. 8 displaying the apparent specific growth rate for various impinging lights I_0 and optical depths ranging from $\lambda = 0$ (limit case that no shading effect occurs) to $\lambda = 10$ (almost complete light attenuation). For example, $\lambda = 3$ corresponds to 95 % of the light being absorbed, either due to a high culture concentration or a thick culture. Note that for such optical depth levels (or higher), the apparent growth rate can be approximated well with Monod-type kinetic rates, keeping in mind that the Monod half-saturation constant should be biomass related. Of course, this does not mean that the microalgae are not subject to photoinhibition effects, but rather that these effects are not directly visible due to the averaging process. Such averaging induces a clear loss of productivity at higher optical depth nonetheless.

5.2 Open Questions

The analysis of a simple chemostat culture in the previous subsection has distinguished two limit cases in order to estimate the apparent growth rate, either as the growth rate at the mean irradiance level, or as the average growth rate across the

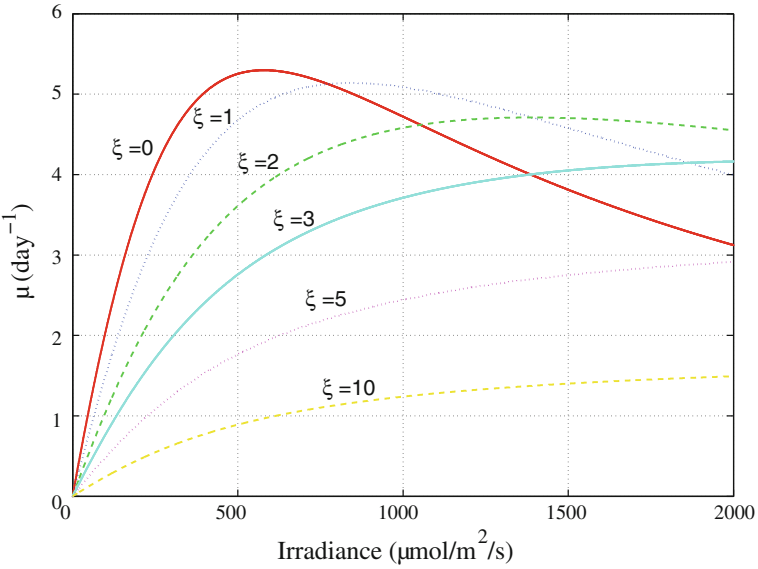

Fig. 8 Apparent specific growth rate (22) in a simple planar chemostat geometry as a function of the impinging light I_0 and for various optical depths λ

culture depth. The reality is probably halfway, yet predicting exactly where it occurs calls for a new paradigm.

The light pattern received by a microalgae cell is strongly influenced by the hydrodynamics in the culture medium [64, 80, 85, 91]. Both the average light and the variation frequency between light and dark phases can indeed differ greatly under different flow regimes, culture concentrations, and so on. This is illustrated in Fig. 9 comparing the simulated light patterns received by a microalgae cell in a tubular photobioreactor at two different cell concentration levels. Although fast-scale photosynthesis models, such as the Han model (see Sect. 3.4), can in principle

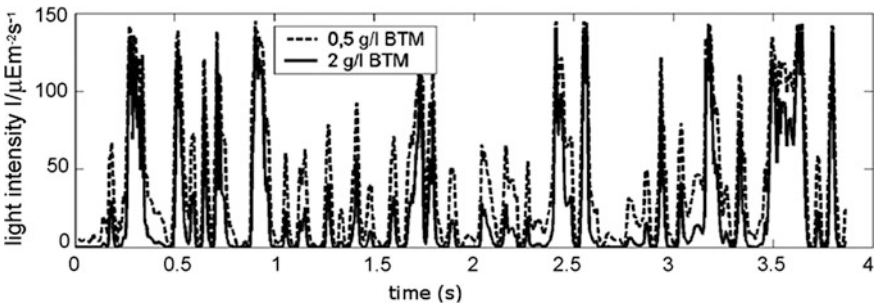

Fig. 9 Simulated light patterns received by a microalgae cell in a tubular photobioreactor at two different cell concentration levels: 0.5 g L^{-1} (*dashed*) and 2 g L^{-1} (*solid*). Reproduced from [80]

be used with this kind of light signal in order to predict the indirect effect of the hydrodynamics on microalgae growth, these models have not been validated with realistic light signals as of yet.

In addition to their effects on fast photosynthetic processes, one should also consider the combined effects of the light gradient and flow mixing on the dynamics of photoacclimation (see Sect. 3.3). The question of the light irradiance at which the cells are photoadapted—that is, which irradiance value I to use in Eq. (6)—is indeed crucial and it still remains open [80, 85, 91, 114].

Finally, in order to predict the daily productivity of a microalgae culture system, one has to combine multiple biological and physical models acting across a wide range of timescales. These models have characteristic timescales ranging from milliseconds for the light reactions, to minutes for the photodamage effect, and to days for the growth and photoacclimation processes. Such an integration has been investigated in Esposito et al. [35], where a coupled Han–Geider model is embedded within a large eddy simulation (LES) framework. Likewise, a coupled Han–Droop model [51] was used for the purpose of raceway light pattern reconstruction and experimental simulation in Hartmann et al. [52].

By and large, the approach that involves coupling detailed hydrodynamics simulators to fast timescale models describing the photosynthetic response holds much promise for a better understanding and prediction of the actual productivity of MCS. However, the added complexity and high computational burden has not yet been compensated by more accurate predictions in the authors' opinion.

6 Towards Model-Based Optimization and Control of Microalgae Culture Systems

6.1 Model-Based Operations Optimization

In parallel with the development of accurate mathematical models describing MCS, an increasing number of studies involving the application of systematic model-based optimization techniques in order to gain process operational insights have started to emerge. A natural optimization criterion in this context appears to be maximizing biomass productivity per unit area, referred to as surface productivity hereafter. Of course, alternative optimization criteria can be considered in the case where TAG synthesis or other biosynthesis is to be optimized.

The following considerations are for a simple planar geometry of thickness L, and our initial focus is for a steady-state microalgae culture under constant impinging light I_0 (and constant temperature). Using the Beer–Lambert law (14) as a first approximation and assuming that the light attenuation parameter ξ varies linearly with the microalgae concentration x, it is not hard to see that the optical depth λ is a function of the cell concentration per unit area $\varsigma := xL$ only (see Sect. 4.1). It follows that the apparent specific growth rate $\bar{\mu}(I_0, \cdot)$ given by (22) is itself a function of ς, and the net surface productivity P can be expressed as

$$P(I_0, \varsigma, \cdot) = (\bar{\mu}(I_0, \varsigma, \cdot) - r)\varsigma. \tag{23}$$

In other words, only the cell concentration per unit area matters according to the metric P: a thin culture (small L) with high cell concentration x is equivalent to a thick culture (large L) with low cell concentration x. Also note that the net surface productivity P in (23) attains its maximum for a finite, nonzero value ς^*, due to the productivity approaching zero for either very small or very large values of ς: in the case of a small cell concentration per unit area, this is because only a tiny fraction of the impinging light is actually absorbed, whereas there is nearly no residual light inside the culture medium when the cell concentration becomes too big.

Using the property that the apparent specific growth rate is approximated closely by Monod-type kinetics in dense cultures having an optical depth $\lambda \geq 3$ (see Sect. 5.1 and Fig. 8), it can be shown [56] that the optimal cell concentration per unit area ς^* maximizing surface productivity is such that the remaining light at depth L corresponds to the compensation light, namely the light at which the growth rate matches the basal respiration rate. Similar conclusions are obtained in [26, 107] by means of different approaches.

Accounting for the light periodicity in determining the optimal productivity introduces yet another layer of complexity. The optimization of cell productivity on a diurnal basis is considered in Akhmetzhanov et al. [1], whereas optimization on account of high-frequency light variations due to mixing and light gradient effects is investigated in Celikovsky et al. [21].

More recently, optimal operation strategies for continuous microalgae cultures have been investigated in Grognard et al. [46]. There the optimal operation is mainly determined by the need to reach a periodic regime, whereby the cell concentration at the end of a 24-h period matches the initial cell concentration, and it is no longer possible for the cell concentration $\varsigma(t)$ to track the optimal cell concentration ς^* for a given impinging light level perfectly. Instead, it is shown that the optimal trajectory $\tilde{\varsigma}(t)$ should be such that it approaches ς^* on average.

6.2 Monitoring and Control

In practice, the goal is often to maintain the culture conditions in such a way that the productivity of microalgae is close to a maximum. A number of simple control actions can be performed that do not interfere with the characteristic timescales of microalgae growth:

- pH Regulation can be achieved via controlling the injection rate of inorganic carbon (typically in the form of gaseous CO_2), for instance, using online pH measurements and a simple PI or PID controller [9, 18]. Alternatively, an MPC controller reducing the CO_2 losses in outdoor photobioreactors was proposed in Garcia Sanchez et al. [41].

- Regulation of the microalgae concentration can be achieved via a feedback control loop measuring the cell density, for example, using an NIR light transmittance sensor, and controlling the injection of fresh growth medium [94]. Turbidostats can also be used for this purpose [72].
- Temperature regulation can be achieved by means of low-level controllers, for instance, by maintaining the culture medium temperature near the optimal growth temperature T_{opt} (see Sect. 3.5). In doing so, one must make sure that the energetic penalty incurred by the temperature control does not overrun the corresponding productivity increase nonetheless. For instance, it has been estimated [5] that the energy needed to downregulate the temperature of a photobioreactor in California to 25 °C (the temperature without control can raise above 40 °C) is about 1.8 GJ $year^{-1}$ m^{-2}, representing the equivalent of 0.3 oil-barrel per meter-square.

A major bottleneck for the implementation of more advanced control and online optimization strategies in MCS appears to be the lack of online sensors that can monitor the biological activity. The idea of using software sensors, also known as observers, that infer key unmeasured bio/chemical variables based on readily available online measurements and a mathematical model of the system, is quite appealing in this context. A high-gain observer was developed in Bernard et al. [16] for estimating both the internal quota and the remaining nutrient concentration. More recently, an interval observer providing confidence intervals on the same inferred quantities and taking into account the discrete nature of the measurements was presented in Goffaux et al. [44]. Regarding TAG synthesis too, a nonlinear interval observer was designed in Mairet et al. [68] for monitoring the internal TAG content of microalgae.

Early work involving the online estimation of microalgae growth, and their subsequent use in closed-loop control strategies in an objective to maximize microalgae productivity, can be traced back to the early 1980s [7, 48]. Since then, other authors have investigated the use of inorganic carbon [4] or oxygen production [103] as a proxy to estimate microalgae growth.

Concerning closed-loop control finally, a simple and near-optimal strategy that enables microalgae productivity levels very close to their theoretical limit was recently presented in [69]. This strategy was derived based on the actual model-based optimal control strategy and involves controlling the optical depth in a raceway pond by applying a dilution rate proportional to the productivity rate. The latter can be inferred based on a software sensor that exploits direct measurements of either the CO_2 injection rate or the O_2 production rate, with the proportionality factor evaluated using online parameter estimation. The proposed strategy could be validated numerically against a model coupling a high-fidelity physical model of an open raceway pond with a biological model accounting for light and temperature limitation effects, photoacclimation, and internal storage of carbon in the form of

TAGs and carbohydrates. Moreover, a mathematical analysis supports the design of this controller by demonstrating its (global) stability with respect to the uncertain initial conditions.

Despite a number of recent advances [53, 66, 69], the availability of robust controllers remains scarce. This can be attributed to the complexity and high-nonlinearity of Droop-like models, that make the derivation and analysis of the controllers particularly arduous. However, the continued development of reliable dynamic models of MCS presents many opportunities for optimization-based control techniques, including nonlinear model predictive control (NMPC) and dynamic real-time optimization.

7 Conclusions

Industrial exploitation of microalgae is just starting, motivated by their huge potential, and by the diversity of innovative applications [86, 100]. However, such photosynthetic organisms whose energy-harvesting strategy is strongly related to light, are more difficult to model than more classical microorganisms (bacteria, yeasts, or fungi). They have a strong aptitude to store nutrients, which motivates the use of quota models (typically the Droop model) that are more complex than the classical Monod model. The microalgae biomass contributes to attenuate light, inducing then a strong coupling between biology (microalgae growth) and physics (radiative transfer properties and hydrodynamics).

Some models exist that can describe separately some of these processes, but there is a clear incentive to develop integrated predictive models, which could realistically predict the behavior of a microalgae culture system, especially in a context of bioenergy production from solar energy. It takes a lot of effort to validate these models over long periods of time, due to the need for extensive measurement datasets. Eventually, these models will support monitoring and optimization of MCS and will guide the development of this promising technology. They will also realistically support quantification and optimization of the reachable productivities, depending on species, type of culture process, period of the year, and location and, therefore, calibrate the corresponding investments. They will also contribute to improve the environmental impact assessment [63] by better quantifying the balance between the requested energy to maintain the algae in suspension and inject CO_2, and the recovered energy through biofuel production.

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