

# A Macromodel for Outdoor Algal Mass Production

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A model describing growth of an outdoor algal (*Spirulina platensis*) culture was developed. The model can simulate biomass production, pH, growth rate, oxygen evolution, and CO<sub>2</sub> fixation rate. It was calibrated and validated against experimental data obtained by a novel automatic data logger/controller instrumentation which can number most vital parameters of the culture including on line estimation of oxygen production rate (OPR). The importance of understanding light distribution through the pond and its effects on the photosynthesis and respiration processes are emphasized. A maximum yield of about 38 g day<sup>-1</sup> m<sup>-2</sup> under optimal conditions is predicted. The present model can also be a useful tool for optimization of algal mass production sites.

## INTRODUCTION

The advantages of describing complex systems such as biological processes by mathematical modelling have been previously discussed.<sup>1-3</sup> Mathematical models can give a better understanding of the biological system and its interaction with the environment by providing detailed information on the biological, physical, and chemical processes involved.<sup>3,4</sup> Models can also be used in the prediction of the behavior of the system under different environmental or operational conditions, as well as for optimization and control studies.<sup>5,6</sup>

Although algal cultures are relatively simple ecosystems to model mathematically, and several models have been proposed,<sup>7,8</sup> only a few of them have been developed specifically for outdoor algal ponds.<sup>9,10</sup> The outdoor models proposed so far have two basic handicaps:

1. The models were developed under the assumption that the results obtained from laboratory experiments could be directly extrapolated to outdoor conditions. Experience has demonstrated that this assumption is not always true.
2. The lack of sufficient data for statistical validation of the model. The reasons for this deficiency could be found in the low quantity and quality of data that

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could be obtained with traditional methods that were generally employed to test models, i.e., either radioactive carbon determination methods<sup>11</sup> or those which rely on the estimation of dry weight, chlorophyll or protein.<sup>12</sup> The low sampling rate at which this data is collected (generally not more than one or two samples per day) is the result of the difficulties inherent in these classical analytical methods.<sup>11,12</sup>

The low sampling rate is a real limitation in the validation of the models, since according to Nyquist's theorem,<sup>13</sup> in order to reconstruct a sampled system which is frequency limited, it is necessary to sample it at a rate not less than twice the maximum systems frequency. This implies, for example, that if the growth rate in an algal pond is determined by the daily increase in dry weight, it will be impossible to recover the daily trajectory of the biological process.

In recent articles,<sup>14-16</sup> we have reported the successful application of a new estimation technique for on-line determination of oxygen production rate (OPR) for estimating the growth rate of an outdoor algal culture. As the method also gives the diffusion rate between the atmosphere and the culture, it has been demonstrated<sup>16</sup> that it can be used to study the dissolved oxygen and carbon dioxide exchange rates between the pond and the atmosphere.

An algal culture model which is based on on-line OPR estimation data will not suffer from the shortcomings described above, since: (1) OPR measurements are automatically performed by a new microprocessor data logger/controller system which also measures other relevant parameters, such as light, pH, and temperature, etc. necessary for calibration and validation of the model; (2) the sampling rate for OPR estimation is very high (eight times per day) relative to the data obtained by classical methods.

## MODEL THEORY

As a result of economic and other practical considerations, most algal production plants are outdoor process making use of the free solar energy. The utilization of natural light as an energy source implies a system that is subject to environmental changes and to two well defined operating zones created by the light-dark cycle. The algal growth rate

( $\mu$ ) is thus a dependent variable function of a number of environmental and physiological variables (light, temperature, pH, nutrients, etc).

Algal growth kinetic<sup>9</sup> is similar to the kinetic of other microorganisms and is usually described by a first-order kinetic model:

$$\frac{dC(t)}{dt} = \mu(l, T, S, \text{pH}, \dots) \cdot C(t) \quad (1)$$

where  $C(t)$  is the biomass concentration at time  $t$  ( $\text{mg L}^{-1}$ ) and  $\mu(l, T, S, \text{pH}, \dots)$  is the algal growth rate ( $\text{time}^{-1}$  units) as a function of light intensity ( $l$ ), temperature ( $T$ ), substrate concentration ( $S$ ), pH, etc. It is assumed that the pond is a well mixed photobioreactor, so that  $C(t)$  is homogeneous throughout the pond, and that no temperature or gas gradient exist throughout the culture volume.

As already pointed out by Goldman,<sup>9</sup> the common goal of virtually all outdoor mass culture production systems is to optimize biomass yield. To accomplish this, nutrients are supplied in excess so that algal growth is regulated by two factors: sunlight and temperature. As will be demonstrated later, in order to obtain maximum yield, it is necessary to optimize the use of the light energy available by the correct selection of the pond operational conditions (pond depth, biomass concentration, temperature, etc). When light energy is available, photosynthesis can take place, and an increase in biomass is expected. In the dark, respiration prevails and the algae consume part of the organic carbon accumulated during the light period, decreasing thereby the biomass. Equation (1) can therefore be rewritten as:

$$\frac{dC(t)}{dt} = \mu_{\text{eff}} C(t) \quad (2)$$

where

$$\begin{aligned} \mu_{\text{eff}} &= \mu_p - \mu_r \\ \mu_p &= \begin{cases} f(T, l); & l > 0 \\ 0 & ; l = 0 \end{cases} \\ \mu_r &= f(T) \end{aligned}$$

where  $\mu_{\text{eff}}$  is the effective growth rate;  $\mu_p$  is the photosynthetic growth rate;  $\mu_r$  is the respiration growth rate; and  $l$  is light intensity.

### Determination of Growth Rate

The exchange of oxygen and carbon dioxide between the atmosphere and an outdoor algal culture can be described by the Film Model.<sup>17</sup> Assuming that the growth conditions of the culture do not change appreciably during a given experiment, the net OPR can be used as an indicator of the net photosynthetic/respiration rate. The dissolved oxygen (DO) concentration in the culture can be described by the following equation<sup>14,15</sup>:

$$\frac{d\text{DO}(t)}{dt} = -\frac{\text{DO}_2}{zH} [\text{DO}(t) - \text{DO}(\text{atm})] + \sum_{i=1}^n F_{\text{phi}} - \sum_{j=1}^m F_{rj} \quad (3)$$

where  $\text{DO}_2$  is the molecular diffusion coefficient of  $\text{O}_2$ ;  $z$  is the thickness of the solution boundary layer;  $\text{DO}(\text{atm})$  is the dissolved oxygen concentration at the surface solution film;  $H$  is the depth of culture in the pond;  $\sum_{i=1}^n F_{\text{phi}}$  is the sum of all oxygen production sources; and  $\sum_{j=1}^m F_{rj}$  is the sum of all the oxygen consumption sources. As has already been stated,<sup>15</sup> this equation can be used to design an estimator to estimate  $(\text{DO}_2/zH)$  and net consumption/production rate.

The solution of differential eq. (3) is an exponential function:

$$\begin{aligned} \text{DO}(t) &= [\text{DO}(0) - A]e^{K_L t} + A \\ A &= \text{DO}(\text{atm}) + \frac{\text{OPR}}{K_L} \\ \text{OPR} &= \sum_{i=1}^n F_{\text{phi}} - \sum_{j=1}^m F_{rj} \\ K_L &= -\frac{\text{DO}_2}{zH} \end{aligned} \quad (4)$$

This equation predicts an exponential decrease/increase in DO with a time constant  $K_L^{-1}$  and a final value  $A$ . It can be shown that a necessary condition for the system observability is that  $d\text{DO}(t)/dt \neq 0$ , this condition can be fulfilled if the system is perturbed from steady state by a forced excitation of DO from its dynamic equilibrium. DO will follow an exponential path when returning to its quasi steady state. Hence, an estimator of the form

$$Y = aX + b \quad (5)$$

where

$$\begin{aligned} Y &= \frac{d\text{DO}(t)}{dt} \\ a &= -\frac{\text{DO}_2}{zH} \\ b &= \text{OPR} \\ X &= [\text{DO}(t) - \text{DO}(\text{atm})] \end{aligned}$$

can be utilized. OPR can then be correlated with other parameters like optical density (OD) or dry weight by integration.<sup>14-16</sup> Assuming constant growth conditions and only one species of alga, the biomass concentration in the pond at a given time ( $t_i$ ) it is related to OPR by:

$$C(t_i) = K \sum_0^{t_i} \text{OPR}(t) dt + C(0) \quad (6)$$

where  $K$  is a constant whose value depends on the average C:N:P ratio of the algae and  $C(0)$  is the initial biomass concentration. Equation (6) can be expressed in a discrete form:

$$C(t_n) = K \sum_{i=0}^n \text{OPR}_i \Delta t - C(0) \quad (7)$$

where  $\text{OPR}_i$  is the OPR value obtained from the  $i^{\text{th}}$  experiment and  $\Delta t$  is the time between experiments. OPR can

also be correlated to algal growth by differentiating eq. (6) and dividing by the biomass concentration:

$$\mu = \frac{1}{C(t)} \frac{dC(t)}{dt} = \frac{K \cdot \text{OPR}}{C(t)} \quad (8)$$

### Gas Exchange

The OPR method yields an independent estimate for the gas exchange factor ( $\text{DO}_2/z$ ) which can then be used to estimate the fluxes of both oxygen and carbon dioxide through the surface boundary layer of the pond. If the relationship between oxygen production and carbon dioxide production/consumption ( $Q$ ) is known, OPR can be used to estimate the net carbon production/consumption rate ( $F_c$ ):

$$F_c = Q \cdot \text{OPR} \quad (9)$$

Considering that  $(\text{DO}_2/z) \approx (\text{DCO}_2/z)$ ,<sup>16</sup> the rate of change of inorganic carbon in the algal culture can be described in a form similar to that used for the dissolved oxygen concentration (eq. 3):

$$\frac{d\text{TCO}_2}{dt} = -\frac{\text{DCO}_2 \cdot \alpha\text{CO}_2}{zH} [\text{pCO}_2(t) - \text{pCO}_2(\text{atm})] - F_c \quad (10)$$

where

$\text{TCO}_2$  is the concentration of total  $\text{CO}_2$  ( $\text{mol cm}^{-3}$ );

$\text{DCO}_2$  is the molecular diffusion of  $\text{CO}_2$  ( $\text{cm}^2 \text{s}^{-1}$ );

$\alpha\text{CO}_2$  is the solubility of  $\text{CO}_2$  ( $\text{g mol cm}^{-3} \text{atm}^{-1}$ );

$\text{pCO}_2$  is the partial pressure of  $\text{CO}_2$  ( $\text{g atm}$ ); and

$\text{pCO}_2(\text{atm})$  is the partial pressure of  $\text{CO}_2$  ( $\text{g}$ ) at the surface film ( $\text{atm}$ ).

The partial pressure of carbon dioxide and the concentration of total carbon can be calculated from carbonate alkalinity and pH using the apparent constants of the carbonate systems, ( $K'_1$  and  $K'_2$ )<sup>18</sup>:

$$\text{pCO}_2 = \frac{CA \cdot a_H^2}{K'_1 \cdot \alpha\text{CO}_2 [a_H + 2K'_2]} \quad (11)$$

$$\text{TCO}_2 = CA \left[ \frac{a_H K'_1 + K'_1 K'_2 + a_H^2}{a_H K'_1 + 2K'_1 K'_2} \right] \quad (12)$$

The model assumes that the driving force of the  $\text{CO}_2$  exchange process is dependent, as in the case of  $\text{O}_2$  on the partial pressure gradient across the  $z$  layer, and neglects all kinetics and migration effects. That is, the carbonate system in the bulk of solution is considered in equilibrium at any instant.<sup>18</sup> Although eq. (10) can theoretically be used for estimating  $F_c$ , practical considerations suggest that simpler and better estimations can be obtained by estimating OPR<sup>16</sup>.

$$\frac{d\text{TCO}_2}{dt} = -\frac{\text{DCO}_2 \cdot \alpha\text{CO}_2}{zH} [\text{pCO}_2(t) - \text{pCO}_2(\text{atm})] - Q \cdot \text{OPR} \quad (13)$$

This equation can then be used to determine the state of the carbonate system as well as  $\text{CO}_2$  exchange rates, at any

time. The relationship between  $\text{TCO}_2$  and pH can be derived from eq. (12) in the form of a second-order equation:

$$a_H^2 + ba_H + c = 0 \quad (14)$$

where

$$b = K'_1 - \left[ 1 - \frac{\text{TCO}_2}{CA} \right]$$

$$c = K'_1 K'_2 \left[ 1 - 2 \frac{\text{TCO}_2}{CA} \right]$$

It is thus clear that the ratio  $\text{TCO}_2/CA$  is the master variable that controls pH, and that the apparent dissociation constants of carbonic acid ( $K'_1, K'_2$ ) determine the state of the carbonate system. The values of  $\alpha\text{CO}_2$  and of DO ( $\text{atm}$ ) can be calculated by the experimental equation derived by Weiss,<sup>19</sup> in which the solubility is expressed as a nonlinear function of salinity and temperature.

### Dark Cycle

At dark  $\mu_{\text{eff}} = \mu_r$ , and a net respiration process causes a decrease in biomass. The influence of temperature on respiration has been examined in detail elsewhere,<sup>20</sup> and an Arrhenius type relationship was suggested:

$$\mu_r = \mu_{rm} e^{-K/T} \quad (15)$$

in which  $\mu_{rm}$  is the maximum respiration rate and  $K$  and  $T$  are in Kelvin degrees.

From field and laboratory measurements it was found that respiration is significantly inhibited when the temperature falls below  $15^\circ\text{C}$ , most likely due to an overall slow down of enzymatic activity. We have found that over the temperature range of interest, eq. (15) can be approximated by a linear function:

$$\mu_r = \begin{cases} aT + b; & 15 < T \leq 40^\circ\text{C} \\ 0; & T < 15^\circ\text{C} \end{cases} \quad (16)$$

where  $T$  is in degrees centigrade and  $a$  is a constant.

### Light Cycle

Assuming that there is a linear relationship between OD and biomass concentration and that Beer-Lambert law is applicable, light intensity at a given depth  $x$  in the pond is related to surface light intensity  $l(0)$  by:

$$l(x) = l(0)e^{-\epsilon C(t)x} \quad (17)$$

where  $\epsilon$  is the extinction coefficient.

Optical density is usually related to light attenuation along a path length of 1 cm, as defined by:

$$\text{OD} = \log \left[ \frac{l(x)}{l(x+1)} \right] \quad (18)$$

where  $x$  is measured in cm. Hence,

$$\text{OD}(t) = C(t)\epsilon 0.434 \quad (19)$$

and

$$l(x) = l(0)e^{-2.3OD(t)x} \quad (20)$$

The photosynthetic active layer  $X_D(t)$  can be calculated from eq. (20) as:

$$x_D(t) = -\frac{1}{2.3OD(t)} \ln \left[ \frac{l_D}{l(0)} \right] \quad (21)$$

where  $l_D$  is the dark intensity (or the minimum light intensity) necessary for active biomass growth. The thickness of the dark zone ( $Z_D$ ) is thus:

$$Z_D = H - x_D \quad (22)$$

where  $H$  is the pond's depth. Assuming that respiration (similar to night respiration) is taking place in the dark zone [eq. (15)] we obtain:

$$\mu_r = \begin{cases} [aT + b] \left[ \frac{Z_D}{H} \right]; & 15 \leq T \leq 40^\circ\text{C} \\ 0; & T < 15^\circ\text{C} \end{cases} \quad (23)$$

Neglecting light inhibition and assuming that the system is far below saturation (from a kinetic point of view) then:

$$\mu_{p,x} = ql(x) \quad (24)$$

where  $\mu_{p,x}$  is the growth rate function at depth  $x$ .

Equation (2) can therefore be rewritten for the case of  $\mu_{\text{eff}} = \mu_p$  as:

$$\left( \frac{dC(t)}{dt} \right)_x = ql(x)C(t) \quad (25)$$

Replacing eqs. (20) and (24) in eq. (25), we obtain:

$$\mu_{p,x} = \frac{1}{C(t)} \frac{dC(t)}{dt} = ql(0)e^{-2.3OD(t)x} \quad (26)$$

Integrating through the pond depth ( $H$ ):

$$\begin{aligned} \mu_p &= \int_0^H ql(0)e^{-2.3OD(t)x} dx \\ &= \frac{ql(0)}{2.3OD(t)H} (1 - e^{-2.3OD(t)H}) \end{aligned} \quad (27)$$

For sufficiently large  $H$  eq. (27) reduces to:

$$\mu_p \cong \frac{ql(0)}{2.3OD(t)H} \quad (28)$$

Finally, the temperature effect on the growth rate is approximated by an equation of the form:

$$\mu_p(l, T) \cong \frac{ql(0)}{2.3OD(t)H} \left[ \frac{1 - (T - T_{\text{op}})^2}{\Delta T^2} \right] \quad (29)$$

where  $T_{\text{op}}$  is the optimum growth temperature (about  $33^\circ\text{C}$ ) and  $\Delta T$  is the difference between the maximum and minimum growth temperatures ( $10$ – $35^\circ\text{C}$ ). The curve is parabolic with a maximum at  $T_{\text{op}}$ .

## EXPERIMENTAL

### Instrumentation

The present study was carried out with an automatic data-logging/control system. The parameters measured during the study were pH, DO, OPR, OD, light intensity, and water and air temperatures and over a short period of time wind velocity. The electrode signals were sent to a microcomputer via a special purpose interface controller. The interface comprised four channels each with signal conditioners for DO, light intensity, OD, temperature, three very high impedance analog inputs, and 16 control relays. The interface controller was connected to the microcomputer via two 8-bit I/O ports plus an additional edge-sensitive input line. The microcomputer employed was a Commodore model CMD-64, to which a complex interface adaptor (CIA) had been added. Analog-to-digital conversion was obtained by first converting the analog signal to a proportional frequency signal and then counting the frequency pulses over a predetermined period of time. The advantages of this conversion method are not only its low cost but also its ability to attenuate interfering noise by the inherent integration method. The major disadvantage of the method is the slow conversion rate (about one sample per second). However, this did not prove to be a problem since the rate of change of the phenomena under study was rather slow (time constant of hours). The system's power supply had a battery backup to ensure uninterrupted operation in case of power failure. The system had graphics capabilities and was programmed to perform automatic self testing and calibration before every set of measurements. The data were recorded by a tape recorder and were later transferred to a floppy disk for further data processing and plotting.

### Algae and Growth Conditions

The algae *Spirulina platensis* was cultivated in Zarouk's medium.<sup>21</sup> The outdoor cultivation pond at the Sde Boker Campus of the Ben Gurion University had a surface area of  $2.5 \text{ m}^2$  and a water depth of 12 cm. The pond was stirred with a motor driven paddle.

### Analytical Methods

Dry weight, chlorophyll, phosphate, and nitrate were measured in aqueous samples from the culture pond. Dry weight and chlorophyll were measured as described by Vonshak.<sup>21</sup> Phosphate and nitrate were analyzed spectrophotometrically. Light attenuation of the samples were measured with a Klett-Summerson colorimeter, model 3074-A110 fitted with a green filter. All experimental work was carried out at the Microalgal Biotechnology Laboratory at the Desert Research Institute (Sde Boker, Israel).

## DO Perturbation

The proposed OPR estimation algorithm was applied after monitoring the DO concentration transient following perturbation of the DO level. The perturbation was induced by bubbling air through the pond and hence changing the DO concentration in the medium. Bubbling was carried out automatically every 3 h (a total of 8 experiments per day) for about 15 min after which normal operation was resumed. The data sampling rate during the transient period, following the air bubbling period, was once every 3 min. The sampling rate at other times was once every 15 min.

## Data Processing

Data processing was carried out on-line on the CMD-64 microcomputer. All data processing programs were written in BASIC. Further analysis was carried out on a microcomputer system similar to the one used for this monitoring operation.

## RESULTS AND DISCUSSION

The data employed for the model calibration were obtained from experiments performed between June and November (summer/autumn in Israel) of the years 1984–1986.<sup>15</sup> (Table I) During the experimental periods, the performance of the outdoor culture was monitored continuously. Air bubbling through the culture changed the DO level to about oxygen saturation level. When the bubbling was stopped the DO returned to the quasi-steady state level (Fig. 1) with a time constant of  $(DO_2/zH)^{-1}$ , which was evaluated and used in the model. The OPR and the gas exchange coefficient were estimated on-line [eq. (5)]. The estimated  $DO_2/z$  were in the range of 0.06 to 0.1  $cm\ min^{-1}$  from which the film thickness ( $z$ ) was calculated to be in the range of 1 to 16  $\mu m$  for the oxygen time constant range [eq. (4)] of about

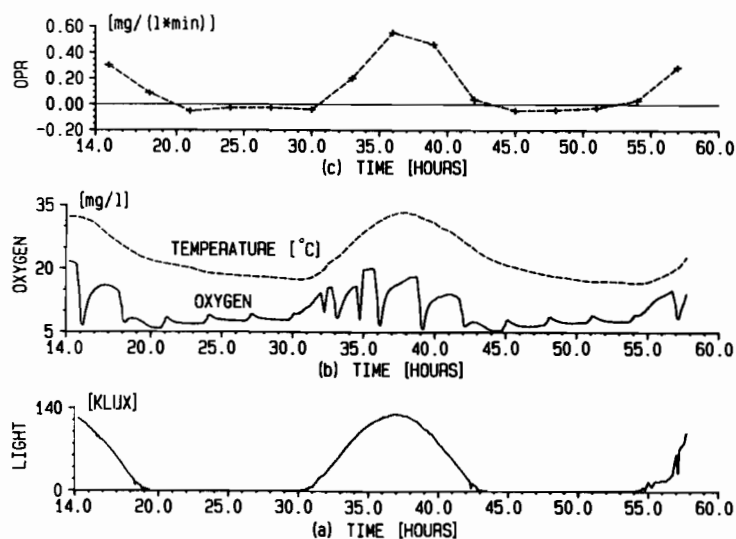
**Table I.** Constants and conversion constants used in simulation runs.

(1)	Equation (3) $\frac{DO_2}{z} = 0.09\ cm\ min^{-1}$
(2)	Equation (1b) $a = -0.0231\ min^{-1}\ ^\circ C^{-1}$ $b = 0.343\ min^{-1}$
(3)	Equation (24) $q = 5.679\ min^{-1}\ klux^{-1}$
(4)	Equation (29) $Top = 33^\circ C$ $\Delta T = 25^\circ C$
(5)	Equation (12) $CA = 200\ meq/L$
(6)	100 Klett units = 0.2 OD
(7)	100 klux = 2200 $\mu E\ m^{-2}\ s^{-1}$ (photosynthetic active range)

8 to 15 min. The question of kinetic enhancement can thus be excluded in the present case.<sup>16,22</sup> This supports our conjecture that the only  $CO_2$  flux that should be considered is the one associated with the molecular diffusion which is linearly proportional to the  $pCO_2$  gradient across the  $z$  layer.

A block diagram of the model is shown in Figure 2. The input parameters are pond depth, initial biomass concentration, carbonate alkalinity, initial pH, salinity,  $DO_2/z$ , maximum and minimum temperatures, maximum light intensity, and light hours. The temperature and light intensity can also be provided as external data. The output of the model simulation include the parameters: biomass changes expressed in terms of OD and dry weight, OPR, pH,  $pCO_2$ ,  $TCO_2$ ,  $HCO_3^-$ ,  $CO_3^{2-}$ , DO, and biomass production per  $m^2$ . Several of the parameters simulated by the model for a pond depth of 12 cm are given in Figure 3. All the model variables (biomass increase,  $TCO_2$ , etc) were simulated for a standard volume of 1 L. Total amounts (i.e., total yield) are obtained by multiplying the simulated values by the total pond volume. The effective collecting area ( $A_{eff}$ ) for light energy is:

$$A_{eff} = \frac{V_{eff}}{H} \quad (30)$$



**Figure 1.** Typical data recorded by the automatic monitoring and control system in the outdoor algal pond: (a) light intensity, (b) temperature and DO, and (c) OPR. Perturbations in DO concentration were produced by bubbling air for 15 min every 3 h.



The model was tested against the outdoor data for biomass concentration between 400 to 2000 mg dry weight L<sup>-1</sup>. The dry weight increase measured analytically, estimated from OPR [eq. (5)], and simulated by the model are shown in Table II and Figure 4. Model confidence limits, based on  $\chi^2$  tests were calculated to be greater than 85%. The good agreement suggest that the model reasonably follows the process of increase in biomass.

As the organic carbon produced by photosynthesis represents approximately half of the total biomass, it's supply rate must be higher than the supply rate of other nutrients (i.e. phosphorus, nitrogen, etc.), pointing out the importance of understanding the carbon supply system in algal cultures. Simulation of pH change during a typical growth period is shown in Figure 5. During the night, pH decrease due to the evolution of carbon dioxide by respiration [Fig. 5(c)]. During light hours photosynthesis takes place and inorganic carbon is consumed, resulting in a pH increase. A general

trend of TCO<sub>2</sub> consumption and pH increase can be observed (Fig. 5). Since the influence of pH on algal growth rate is a second order effect (as opposed to other parameters such as light or temperature), the model does not describe this relationship. There is, however, a build-in capability to facilitate the addition of this module in the future (Fig. 2).

Algal biomass production is affected by several parameters, including biomass concentration, light intensity, pond depth, and temperature. Some parameters, such as light intensity, cannot be controlled, but it is possible to design a pond in such a way as to obtain maximum utilization of solar energy available. Two parameters: OD and pond depth [eq. (28)], can be adjusted to maximize the yield. To study this problem several simulations were performed. First, the relationship between light intensity, OD, and growth rate was examined for different values of light intensity and initial biomass concentration for a 24-h period. The net daily growth and production were calculated [Fig. 6(a)]. It can

Table II. Experimental results.

Date	Dry weight (mg/L) measured	Biomass increment (mg/L)				
		From dry weight (mg/L)	From OPR (mg/L)	deviation <sup>b</sup> (%)	From model (mg/L)	deviation <sup>b</sup> (%)
7/20/84	1545					
7/22/84	1789	243.5	239	-1.64	237	-2.48
7/22/84 <sup>a</sup>	1585					
7/22/84	2029	444	429	-3.19	418	-5.84
7/25/84	1330					
7/26/84	1626	296	217	-26.38	241	-18.26
7/28/84	1833	207	192	-7.21	187	-9.29
7/28/84 <sup>a</sup>	1530					
7/30/84	1678	148	158	6.87	165	11.51
7/30/84 <sup>a</sup>	1163					
7/31/84	1360	197	199	1.41	197	0.13
8/1/84	1689	329	284	-13.68	322	-1.96
8/1/84 <sup>a</sup>	1321					
8/3/84	1638	317	375	20.42	334	7.23
8/14/84	1201					
8/15/84	1303	102	115	13.28	140	36.93
8/16/84	1517	204	212	3.94	189	-7.14
8/21/84	1542					
8/22/84	1658	116	137	18.56	130	11.97
8/28/84	530					
8/29/84	773	243	267	10.12	220	-9.56
8/30/84	991	218	220	1.22	183	-15.73
9/2/84	1417	426	503	18.11	424	-0.50
9/3/84	1586					
9/4/84	1680	94	108	15.33	89	-4.92
9/5/84	1768	88	86	-1.65	83	-5.57
9/10/84	440					
9/11/84	709	269	281	4.51	295	9.66
9/13/84	610					
9/14/84	956	346	354	2.37	345	0.32
7/14/85	640					
7/15/85	944	304	288	-4.96	275	-9.63
8/27/85	620					
8/29/85	1130	510	568	11.52	489	-4.10
9/8/85	428					
9/10/85	726	296	290	-1.96	319	7.76

<sup>a</sup> After dilution with Zarouk medium.

<sup>b</sup> Percent relative to dry weight measured.

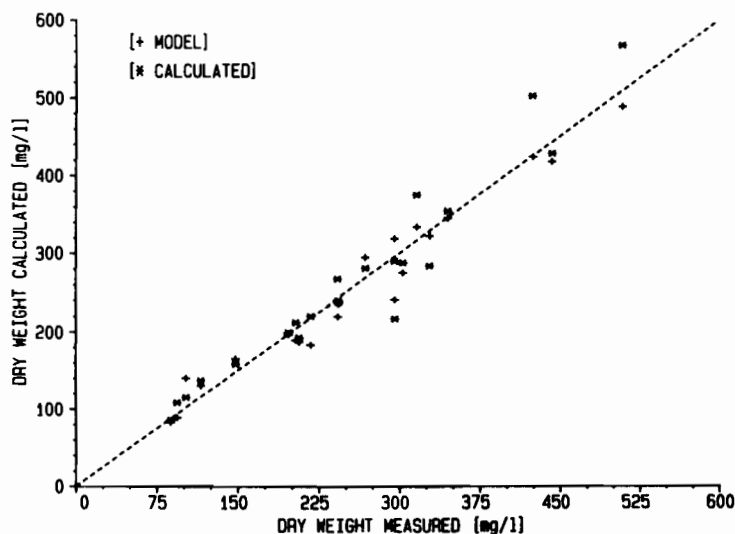


Figure 4. Biomass (dry weight) calculated from OPR estimation method (\*) and form model simulation (+) vs. dry weight measured by conventional analytical method.

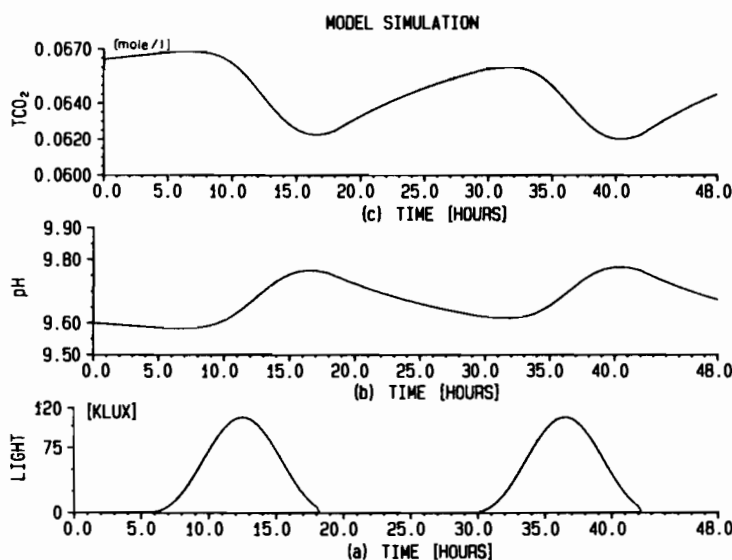


Figure 5. Model simulation of carbon evolution: (a) light intensity, (b) pH, and (c)  $\text{TCO}_2$ .

be seen that increased light intensity increased the growth rate and that an increase in OD lowered it. Since the model does not consider light saturation [eq. (24)], growth rate is directly proportional to the light intensity. Similar simulations were performed for different pond depths. [Fig. 6(b)] pointing out that at smaller depth values an increase in growth rate is predicted.

Biomass production (given in  $\text{g m}^{-2} \text{ day}^{-1}$ ) as a function of light intensity, OD and depth is shown in Figure 7. The maximum production predicted by the model is about  $38 \text{ g m}^{-2} \text{ day}^{-1}$ , a value that agrees well with several previous results obtained independently.<sup>9,23</sup> As light intensity increases the optimum OD is higher [Fig. 7(a)]. The maximum yields for different pond depths were about the same,

but the OD required to reach the maximum increases as the depth decreases [Fig. 7(b)]. It should be noted that although the yield was found to be about the same for each depth, total pond volumes were different. For example, the volume of a 9-cm-deep pond would be about 40% less than that of 15-cm-deep pond. Hence, for the 9-cm case, the production costs could be drastically reduced. Furthermore, higher biomass concentration levels facilitates a more efficient filtration and separation of the algae.<sup>23</sup>

The effect of temperature on biomass growth was also studied by model simulation. Production was evaluated as a function of the maximum and minimum daily temperatures at optimum OD and pond depth for different light levels. It was found that higher night temperature always

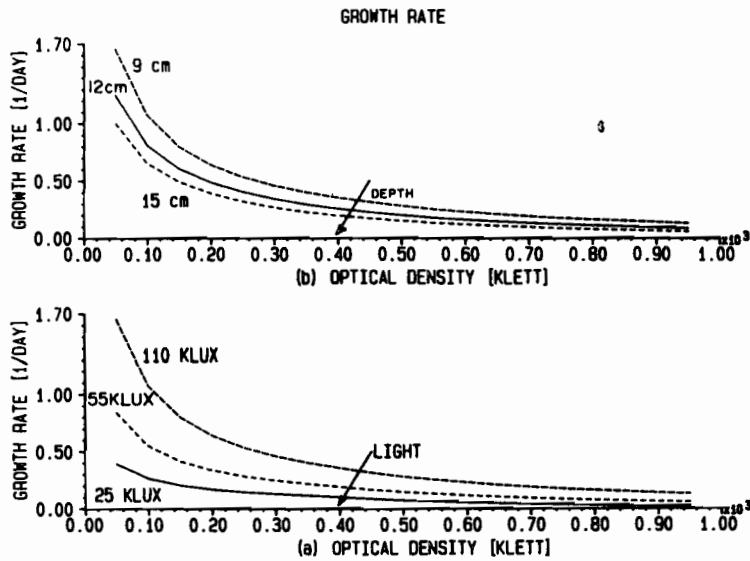


Figure 6. Algal growth rate over a 24-h period as a function of OD: (a) for different light intensity values (pond depth 9 cm) and (b) for different pond depth values (light intensity is 110 klux). Temperature range is 15–33°C.

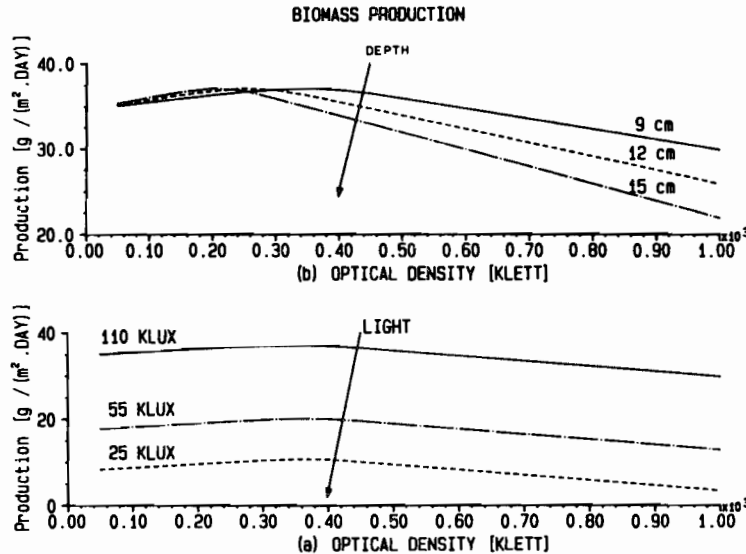


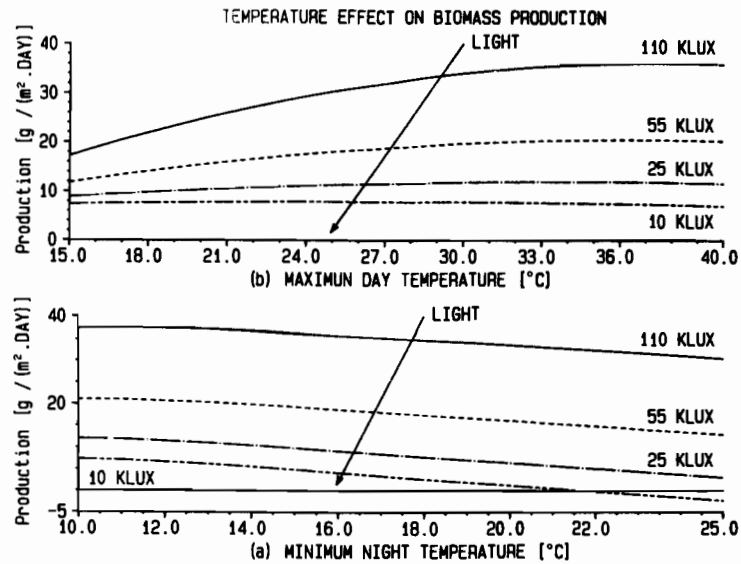
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increased the production loss. At low light intensity and high temperatures a negative production growth rate was found [Fig. 8(a)]. These results support previous experiments which show that high night temperature will reduce overall productivity. From the model simulation we can conclude that as long as there is no damage to the algal cells it is not desirable to artificially increase the night temperature.<sup>23,24</sup>

The diurnal temperature cycle plays an important role when light intensity levels are high [Fig. 8(b)]. Optimum daily production is found for a maximum day temperature of 32 to 36°C. At low light levels, when algal growth is light limited, temperature has little effect on biomass produc-

tion. In this case the photosynthetic activity is low [eq. (28)] and respiration rate is relatively high. The optimal maximum daily temperature is found to be about 24°C [Fig. 8(b)].

Although light saturation phenomenon was found in the laboratory at low biomass concentrations, it was not observed in outdoor ponds at higher biomass concentration levels. The difference in behavior could be explained by the different experimental conditions. In the laboratory, when experiments for determining the  $\mu = f(I)$  curves are performed, low biomass concentrations and low depth are employed to reduce light attenuation and obtain an almost constant light intensity throughout the medium. In this case each individual cell in the medium receives almost the same



**Figure 8.** Temperature effect on biomass production for different maximum diurnal light intensity values as a function of (a) minimum night temperature (maximum day temperature 33°C), (b) maximum day temperature (minimum night temperature 15°C); pond depth is 9 cm.

light energy level. However, since high biomass levels are usually used outdoors and since the culture is well mixed, individual cells are randomly exposed to different light levels. Hence, although the surface light intensity could be higher outdoors than that of the laboratory, for which saturation is observed, the mean light energy available to the individual cells is lower [eq. (27)]. The effect of mixing on light distribution and the possibility of employing very intensive mixing in order to enhance the flashing light effect,<sup>25</sup> has been studied by several researchers.<sup>26-28</sup> Although, several models describing the algal motion in the pond have been proposed,<sup>29-31</sup> they have not been experimentally validated. The present model assumes constant stirring and does not consider the stirring effect. The mixing rate question thus remains open until more experiments are performed to elucidate this problem.

## CONCLUSIONS

The results presented in this article show that the present model can describe on a macroscale an outdoor pond of *S. platensis*, and can therefore help in prediction and optimization of the process. The model can be used to select optimum biomass density and can estimate the pond's productivity. Although more research is necessary in order to define the optimum operational conditions for maximal productivity, several conclusions already emerge and can be used to optimize the biomass production. Since night respiration consumes between 10 and 50% of the biomass produced during the day,<sup>15</sup> it is obviously necessary to reduce the respiration losses in order to conserve biomass. This can be achieved either by reducing the temperature during the night period (ideally below 15°C), and/or by reducing the biomass concentration.

Notwithstanding, the question of the feasibility of implementing the proposed solutions, it is important to realize that this model can provide valuable information about several parameters interacting in the ecosystem. It can also be helpful in the design of mass production systems, eventually leading to the development of "smart" controllers with a built in ability to seek the most economical growth conditions, helping in eventually establishing a better production system of algal biomass. More experimental work on the effect of turbulence and photoinhibition is required in order to try and incorporate those two parameters into the model.

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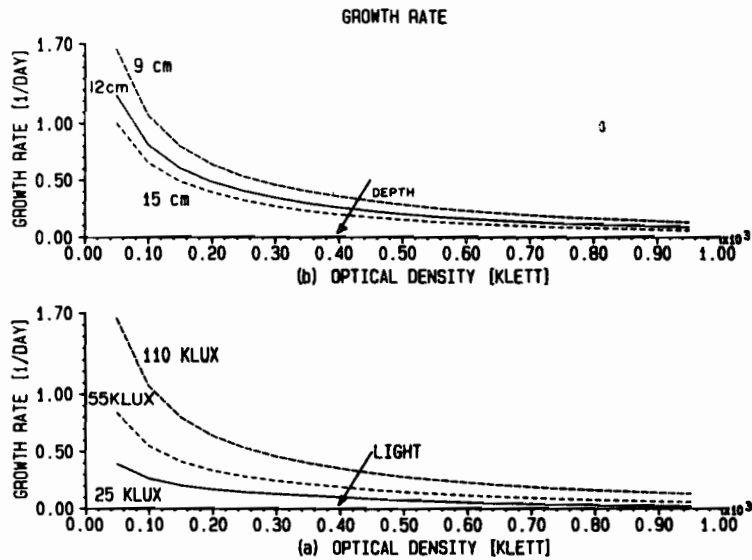


Figure 6. Algal growth rate over a 24-h period as a function of OD: (a) for different light intensity values (pond depth 9 cm) and (b) for different pond depth values (light intensity is 110 klux). Temperature range is 15–33°C.

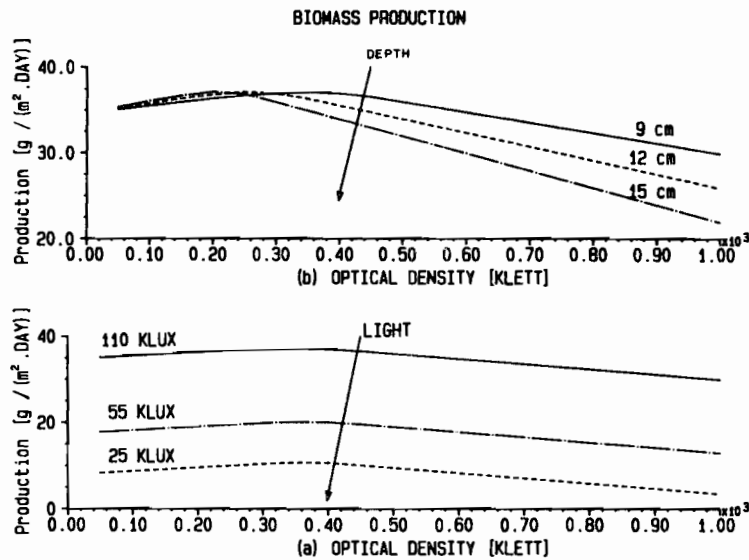


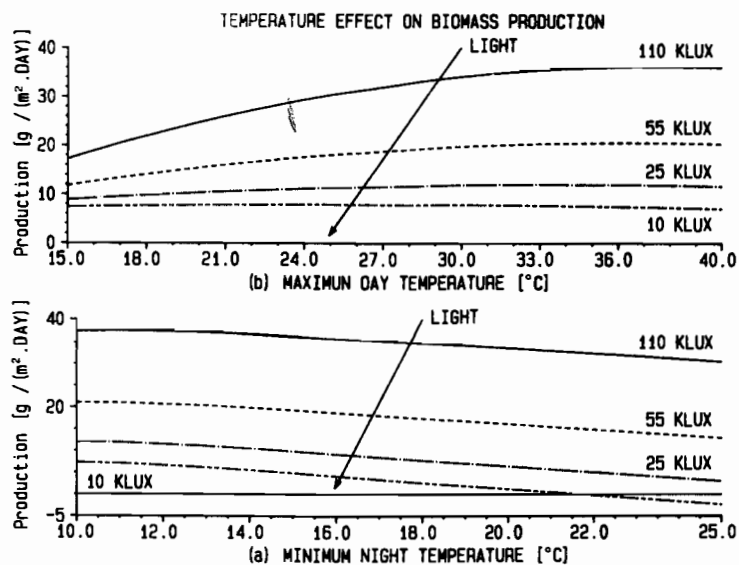
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